5th Edition

Nutrition in Pediatrics

Basic Science • Clinical Applications



Duggan • Watkins • Koletzko • Walker

NUTRITION
IN PEDIATRICS

BASIC SCIENCE • CLINICAL APPLICATIONS

FIFTH EDITION

Christopher Duggan, MD, MPH
Editor in Chief
Professor of Pediatrics
Harvard Medical School
Director, Center for Nutrition
Division of Gastroenterology, Hepatology, and Nutrition
Boston Children's Hospital
Professor in the Department of Nutrition
Harvard T.H. Chan School of Public Health
Boston, Massachusetts

John B. Watkins, MD
Emeritus Associate Professor of Pediatrics
Harvard Medical School
Division of Gastroenterology, Hepatology and Nutrition
Boston Children's Hospital
Boston, Massachusetts

Berthold Koletzko, MD, PhD
Head, Division of Metabolic and Nutritional Medicine
Dr. Von Hauner Children's Hospital
University of Munich Medical Centre
München, Germany

W. Allan Walker, MD
Conrad Taff Professor of Nutrition
Director, Division of Nutrition
Harvard Medical School
Boston, Massachusetts
Member, Mucosal Immunology and Biology Research Center
Massachusetts General Hospital
Charlestown, Massachusetts

2016
PEOPLE'S MEDICAL PUBLISHING HOUSE—USA
SHELTON, CONNECTICUT

People's Medical Publishing House-USA
2 Enterprise Drive, Suite 509
Shelton, CT 06484
Tel: 203-402-0646
Fax: 203-402-0854
E-mail: info@pmph-usa.com



© 2016 Christopher Duggan, John B. Watkins, Berthold Koletzko, W. Allan Walker

All rights reserved. Without limiting the rights under copyright reserved above, no part of this publication may be reproduced, stored in or introduced into a retrieval system, or transmitted, in any form or by any means (electronic, mechanical, photocopying, recording, or otherwise), without the prior written permission of the publisher.

16 17 18 19/PMPH/9 8 7 6 5 4 3 2 1

ISBN-13 978-1-60795-180-3
ISBN-10 1-60795-180-0
eISBN-13 978-1-60795-273-2

Printed in the China by People's Medical Publishing House (PMPH).
Editor: Linda Mehta; Copyeditor/Typesetter: diacriTech; Cover designer: Allison Dibble

Library of Congress Cataloging-in-Publication Data

Names: Duggan, Christopher (Christopher P.), editor. | Watkins, John B., 1938-, editor. | Koletzko, B. (Berthold), editor. | Walker, W. Allan, editor.

Title: Nutrition in pediatrics : basic science, clinical applications / [edited by] Christopher Duggan, John B. Watkins, Berthold Koletzko, W. Allan Walker.

Other titles: Nutrition in pediatrics (Walker)

Description: Fifth edition. | Shelton, Connecticut : People's Medical Publishing House-USA, 2016. | Includes bibliographical references and index.

Identifiers: LCCN 2016006518 (print) | LCCN 2016007253 (ebook) | ISBN 9781607951803 | ISBN 1607951800 | ISBN 9781607952732 (eISBN) | ISBN 9781607952732 ()

Subjects: | MESH: Child Nutrition Disorders | Child Nutritional Physiological

Phenomena | Adolescent Nutritional Physiological Phenomena | Infant Nutrition Disorders | Infant Nutritional Physiological Phenomena

Classification: LCC RJ53.D53 (print) | LCC RJ53.D53 (ebook) | NLM WS 115 | DDC 615.8/54083—dc23

LC record available at http://lccn.loc.gov/2016006518

Notice: The authors and publisher have made every effort to ensure that the patient care recommended herein, including choice of drugs and drug dosages, is in accord with the accepted standard and practice at the time of publication. However, since research and regulation constantly change clinical standards, the reader is urged to check the product information sheet included in the package of each drug, which includes recommended doses, warnings, and contraindications. This is particularly important with new or infrequently used drugs. Any treatment regimen, particularly one involving medication, involves inherent risk that must be weighed on a case-by-case basis against the benefits anticipated. The reader is cautioned that the purpose of this book is to inform and enlighten; the information contained herein is not intended as, and should not be employed as, a substitute for individual diagnosis and treatment.

Sales and Distribution

Canada
Login Canada
300 Saulteaux Cr.
Winnipeg, MB R3J 3T2
Phone: 1.800.665.1148
Fax: 1.800.665.0103
www.lb.ca

Foreign Rights
John Scott & Company
International Publisher's Agency
P.O. Box 878
Kimberton, PA 19442, USA
Tel: 610-827-1640
Fax: 610-827-1671
rights@johnscottco.us

United Kingdom, Europe, Middle East, Africa
Eurospan Limited
3, Henrietta Street, Covent Garden
London WC2E 8LU, UK
Tel: Within the UK: 0800 526830
Outside the UK: +44 (0)20 7845 0868
http://www.eurospanbookstore.com

Singapore, Thailand, Philippines, Indonesia, Vietnam, Pacific Rim, Korea
McGraw-Hill Education (Asia) 1 International Business Park
#01-15A, The Synergy
Singapore 609917
Tel: (65) 6863-1580
Fax: (65) 6862-3354

(65) 6863-1580
mghasia.sg@mheducation.com
www.mheducation.asia

Australia, New Zealand, Papua New Guinea, Fiji, Tonga, Solomon Islands, Cook Islands
Woodslane Pty Limited
10 Apollo Street
Warriewood NSW 2102
Australia
Tel: 612-8445-2300

Fax: 612-9997-5850
www.woodslane.com.au

India, Bangladesh, Pakistan, Sri Lanka, Malaysia
Jaypee Brothers Medical Publishers Pvt. Ltd.
4838, 24 Ansari Road, Darya Ganj
New Delhi-110002, India
Phone: +91 11 23272143
Fax: +91 11 23276490
www.jaypeebrothers.com

People's Republic of China
People's Medical Publishing House
International Trade Department
No. 19, Pan Jia Yuan Nan Li
Chaoyang District
Beijing 100021, P.R. China
Tel: 8610-67653342
Fax: 8610-67691034
www.pmph.com/en

DEDICATIONS

To the continued successful growth and nourishment of Michael, Brendan, and Emily Duggan and in gratitude for many teachers and students in the field of pediatric nutrition.

—Christopher Duggan, MD, MPH

To the children, parents, and staff of the growth and nutrition program whose shared knowledge, caring, and expertise enriches us all and to our grandchildren: Mariposa, Charlotte, Lillie, Gwen, and their “Pinkhouse” partners in “Friendship.”

—John B. Watkins, MD

To a wonderful group of teachers, colleagues, and friends that I have been privileged to work with and to learn from, and to my family, especially my wife Sibylle, for all their patience and support.

—Berthold Koletzko, MD, PhD

To my youngest grandchild William McDonald Walker (Mac), the paradigm of healthy nutrition in children.

—W. Allan Walker, MD

CONTENTS

CONTRIBUTORS
PREFACE

PART I: GENERAL CONCEPTS

CHAPTER 1 Nutrition: The Driving Force for Health, Disease Prevention, and Development
Robert D. Baker and Susan S. Baker

CHAPTER 2 Clinical Assessment of Nutritional Status
Babette S. Zemel, Asim Maqbool, and Irene E. Olsen

CHAPTER 3 Laboratory Assessment of Nutritional Status
Clodagh M. Loughrey and Ian S. Young

CHAPTER 4 Body Composition and Growth
Kenneth J. Ellis

CHAPTER 5 Carbohydrates
Clemens Kunz, Lena Geiger, and Christoph Fusch

CHAPTER 6 Proteins and Amino Acids
Johannes B. van Goudoever

CHAPTER 7 Macronutrient Requirements for Growth: Fats and Fatty Acids
Kathrin Krohn, Hans Demmelmair, and Berthold Koletzko

CHAPTER 8 Trace Elements
Nancy F. Krebs and K. Michael Hambidge

CHAPTER 9 Iron
Amina Khambalia, Ashley Aimone, and Stanley H. Zlotkin

CHAPTER 10 Vitamins
Roland Kupka and Eduardo Villamor

CHAPTER 11 Dietary Reference Intakes in the United States
Alice H. Lichtenstein and Stephen R. Daniels

CHAPTER 12 Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals
Mandana Amir Shaghaghi, Peter J.H. Jones, and Philip M. Sherman

PART II: PHYSIOLOGY AND PATHOPHYSIOLOGY

CHAPTER 13 Energy Requirements in Health and Disease
Jean Louis Bresson

CHAPTER 14 Severe Acute Malnutrition: Pathophysiology, Clinical Consequences, and Treatment
Mary E. Penny

CHAPTER 15 Childhood Malnutrition: Prevention and Control at the National Level
Gerardo Weisstaub, Ana María Aguilar, Magdalena Araya, Ann Hill, and Ricardo Uauy

CHAPTER 16 Community Nutrition and Its Impact on Children: Highly Industrialized Countries
Johanna Dwyer, Kristy Hendricks, Janice H.Y. Chow, and Melanie Reyes

CHAPTER 17 Nutritional Epidemiology
Carine M. Lenders, Emily Keefe, and Walter C. Willett

CHAPTER 18 Drug Therapy and the Role of Nutrition
Kathleen M. Gura and Lingtak-Neander Chan

PART III: PERINATAL NUTRITION

CHAPTER 19 Nutrition and Gene Expression
Mona Bajaj-Elliott and Ian R. Sanderson

CHAPTER 20 Nutrition and Epigenetics
Sang-Woon Choi and Simonetta Friso

CHAPTER 21 Nutrition and the Humoral Regulation of Growth
William E. Russell and J. Marc Rhoads

CHAPTER 22 Gastrointestinal Development: Implications for Infant Feeding
Josef Neu, Martha Douglas-Escobar, and Sandra Fucile

CHAPTER 23 Diet, Environment, and Gut Microbiota in the Metagenomic Era
Carlotta De Filippo and Paolo Lionetti

CHAPTER 24 Nutrition and Neuropsychological Development
Maureen M. Black and Kristen M. Hurley

CHAPTER 25 Maternal Nutrition and Pregnancy Outcome

Theresa O. Scholl and Xinhua Chen

CHAPTER 26 Human Growth and Disease in Later Life
David J.P. Barker

CHAPTER 27 Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism
William W. Hay, Jr., Laura D. Brown, Stephanie R. Wesolowski, and Paul J. Rozance

CHAPTER 28 Amino Acid Nutrition in Utero: Placental Function and Metabolism
Timothy R.H. Regnault and Frederick C. Battaglia

CHAPTER 29 Human Milk: Nutritional Properties
Marcia H. Monaco and Sharon M. Donovan

CHAPTER 30 Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut
Anne Donnet-Hughes, Eduardo J. Schiffrin, and W. Allan Walker

CHAPTER 31 Approach to Breastfeeding
Robert M. Lawrence and Ruth A. Lawrence

CHAPTER 32 The Low-Birthweight Infant: Inpatient Care
Thibault Senterre and Alexandre Lapillonne

CHAPTER 33 Nutritional Management of Preterm Infants Post-Discharge
Mandy Brown Belfort

CHAPTER 34 The Term Infant
Ekhard E. Ziegler and Susan J. Carlson

CHAPTER 35 Complementary Feeding and Health Outcomes
Mary S. Fewtrell

PART IV: OBESITY

CHAPTER 36 Energy and Substrate Regulation in Obesity
Daniel J. Hoffman and Susan B. Roberts

CHAPTER 37 Evaluation and Management of Childhood Obesity
Alison G. Hoppin and Janey S. Pratt

CHAPTER 38 The Metabolic Syndrome in Children and Adolescents
Martin Wabitsch

CHAPTER 39 Childhood Obesity Prevention
John J. Reilly and Adrienne H. Hughes

PART V: NUTRITION IN LOW- AND MIDDLE-INCOME COUNTRIES

CHAPTER 40 The Nutrition Transition and Global Child Health
Benjamin Caballero

CHAPTER 41 Infant and Young Child Feeding in Low- and Middle-Income Countries
Souheila Abbeddou and Kenneth H. Brown

CHAPTER 42 Child Nutrition and Economic Development
William A. Masters

CHAPTER 43 Global Interventions in Child Health
Zulfiqar Ahmed Bhutta and Jai K. Das

CHAPTER 44 Pediatric HIV Infection
Colleen Hadigan and Anna Coutsoudis

CHAPTER 45 Obesity in Developing Economies
Sumathi Swaminathan and Anura Kurpad

PART VI: NUTRITIONAL ASPECTS OF SPECIFIC DISEASE STATES

CHAPTER 46 Feeding Difficulties
Peter B. Sullivan

CHAPTER 47 Failure-to-Thrive: Growth Failure in the Outpatient Setting
Jennifer A. Woo Baidal and Susanna Y. Huh

CHAPTER 48 Developmental Disabilities
Kelly A. Dougherty and Babette S. Zemel

CHAPTER 49 Inborn Errors of Metabolism and the Liver
Charles P. Venditti and Gerard T. Berry

CHAPTER 50 Inborn Errors of Fasting Adaptation—Glycogen Storage Disease
David A. Weinstein and Katalin M. Ross

CHAPTER 51 Acute and Chronic Kidney Disease: Nutrition in Renal Disease
Nancy M. Rodig

CHAPTER 52 Inflammatory Bowel Disease
Francesca Penagini and Rob Heuschkel

CHAPTER 53 Celiac Disease
Dascha C. Weir and Ciaran P. Kelly

CHAPTER 54 Food Allergies
Julie Wang, Jennifer M. Maloney, and Hugh A. Sampson

CHAPTER 55 Exocrine Pancreas
Kevin Gaskin and Karen Blumenthal

CHAPTER 56 Acute and Chronic Liver Disease
Deirdre A. Kelly, Susan Protheroe, and Sara Clarke

CHAPTER 57 Cancer Prevention
A. Lindsay Frazier

CHAPTER 58 Cancer Treatment
Lori J. Bechard and Christopher Duggan

CHAPTER 59 Diabetes Mellitus
Joseph I. Wolfsdorf, Sanjeev Mehta, Maryanne Quinn, and Karen Y. Warman

CHAPTER 60 Diarrheal Diseases
Kerri Gosselin and Christopher Duggan

CHAPTER 61 Intestinal Failure, Short Bowel Syndrome, and Intestinal Transplantation
Daniel Kamin

CHAPTER 62 The Critically Ill Child
Eric A. Sparks, Jeremy G. Fisher, Faraz A. Khan, Nilesh M. Mehta, and Tom Jaksic

CHAPTER 63 Hyperlipidemia and Cardiovascular Disease
Sarah D. de Ferranti

CHAPTER 64 Diet and Dental Disease
Rosalyn Sulyanto and James H. Shaw

CHAPTER 65 Adolescent Eating Disorders
Kendrin R. Sonneville and Alison E. Field

CHAPTER 66 Adolescence: Advancing Bone Health
Keith J. Loud and Catherine M. Gordon

PART VII: APPROACH TO NUTRITIONAL SUPPORT

CHAPTER 67 Enteral Nutrition and Formulas
Zoë McCallum and Julie E. Bines

CHAPTER 68 Parenteral Nutrition
Corina Hartman, Arie Riskin, and Raanan Shamir

CHAPTER 69 Nutrition in the Athlete
Kathryn E. Ackerman and Madhusmita Misra

CHAPTER 70 Special Diets
Sharon Evans, Tracey Johnson, Katharina Dokoupil, and Anita MacDonald

CHAPTER 71 Nutrition Support Teams
Virginie Colomb and Cécile Lambe

APPENDICES

APPENDIX I Nutritional Assessment
Melanie V. Connolly

APPENDIX II Nutritional Requirements
Melanie V. Connolly

APPENDIX III Enteral Products
Melanie V. Connolly

INDEX

CONTRIBUTORS

Souheila Abbeddou, MSc, PhD
Postdoctoral Scholar, Department of Nutrition
University of California, Davis
Davis, California
Infant and Young Child Feeding in Low- and Middle-Income Countries (41)

Kathryn E. Ackerman, MD, MPH
Division of Sports Medicine
Boston Children's Hospital
Harvard Medical School Neuroendocrine Unit
Massachusetts General Hospital
Boston, Massachusetts
Nutrition in the Athlete (69)

Anna María Aguilar, MD, MSc
Universidad Mayor de San Andrés
La Paz, Bolivia
Childhood Malnutrition: Prevention and Control at the National Level (15)

Ashley Aimone, MSc, RD
Registered Dietitian
University of Toronto
Toronto, Ontario, Canada
Iron (9)

Magdalena Araya, MD, PhD
Instituto de Nutrición y Tecnología de los Alimentos (INTA)
University of Chile
Santiago, Chile
Childhood Malnutrition: Prevention and Control at the National Level (15)

Jennifer A. Woo Baidal, MD, MPH
Assistant Professor of Pediatrics at CUMC
Director of Pediatric Weight Management,
Division of Pediatric Gastroenterology, Hepatology, and Nutrition
New York–Presbyterian Morgan Stanley Children's Hospital
New York, New York
Failure-to-Thrive: Growth Failure in the Outpatient Setting (47)

Mona Bajaj-Elliott, PhD
University College London
Institute of Child Health
London, United Kingdom
Nutrition and Gene Expression (19)

Robert Baker, MD, PhD
Professor of Pediatrics
University at Buffalo
Buffalo, New York
Nutrition: The Driving Force for Health, Disease Prevention, and Development (1)

Susan S. Baker, MD, PhD
Professor of Pediatrics
University at Buffalo
Buffalo, New York
Nutrition: The Driving Force for Health, Disease Prevention, and Development (1)

David J. P. Barker, MD, PhD, FAC*
MRC Lifecourse Epidemiology Unit
University of Southampton
Southampton, United Kingdom
Knight Cardiovascular Institute and Moore Institute
Oregon Health and Science University
Portland, Oregon
Human Growth and Disease in Later Life (26)
**Deceased*

Frederick C. Battaglia, MD
Professor Emeritus
University of Colorado School of Medicine
Aurora, Colorado

Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism (27)

Lori J. Bechard, PhD, Med, RD, LDN
Instructor in Pediatrics
Center for Nutrition
Division of Gastroenterology, Hepatology and Nutrition
Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
Cancer Treatment (58)

Mandy Brown Belfort, MD, MPH
Assistant in Medicine
Assistant Professor of Pediatrics
Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
Nutritional Management of Preterm Infants Post-Discharge (33)

Gerard T. Berry, MD
Director, Metabolism Program
Professor of Pediatrics
Harvard Medical School
Boston Children's Hospital
Division of Genetics
Boston, Massachusetts
Inborn Errors of Metabolism and the Liver (49)

Zulfiqar Ahmed Bhutta, MBBS, PhD, FRCP, FRCPCH, FCPS
Husein Lalji Dewraj Professor and Head Division of Women & Child Health
The Aga Khan University
Karachi, Pakistan
Global Interventions in Child Health (43)

Julie Bines, MBBS, MD, FRACP
Professor of Paediatrics
Department of Paediatrics
University of Melbourne
Victoria, Australia
Enteral Nutrition and Formulas (67)

Maureen M. Black, PhD
Professor of Pediatrics
Department of Pediatrics
University of Maryland School of Medicine
Baltimore, Maryland
Nutrition and Neuropsychological Development (24)

Jean Luis Bresson, MD, PhD
Physiologie
Hôpital Necker-Enfants Malades
Paris, France
Energy Requirements in Health and Disease (13)

Kenneth H. Brown, MD
Distinguished Professor Emeritus
Department of Nutrition
University of California
Davis, California
Infant and Young Child Feeding in Low- and Middle-Income Countries (41)

Laura D. Brown, MD
Department of Pediatrics
Perinatal Research Center
University of Colorado School of Medicine
Aurora, Colorado
Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism (27)

Benjamin Caballero, MD, PhD
Professor of Pediatrics and International Health
Johns Hopkins Bloomberg School of Public Health
Johns Hopkins University School of Medicine
Baltimore, Maryland
The Nutrition Transition and Global Child Health (40)

Susan J. Carlson, MMSc, RD
Department of Food and Nutrition Services
University of Iowa Hospital
Iowa City, Iowa

The Term Infant (34)

Lingtak-Neander Chan, PharmD, BCNSP
Associate Professor of Pharmacy
School of Pharmacy
University of Washington
Seattle, Washington
Drug Therapy and the Role of Nutrition (18)

Xinhua Chen, MD
Assistant Professor of Medicine
Beth Israel Deaconess Medical Center
Harvard Medical School
Boston, Massachusetts
Maternal Nutrition and Pregnancy Outcome (25)

Sang Woon Choi, MD, PhD
Adjunct Scientist
Vitamins and Carcinogenesis Laboratory
Jean Mayer USDA Human Nutrition Research Center on Aging
Tufts University
Boston, Massachusetts
Nutrition and Epigenetics (20)

Janice H. Y. Chow, MS, RD
Clinical Dietitian
San Mateo Medical Center
San Mateo, California
Community Nutrition and Its Impact on Children: Highly Industrialized Countries (16)

Sara Clarke, BSc (Hons)
The Liver Unit
The Department of Gastroenterology and Nutrition
Birmingham Children's Hospital
Birmingham, United Kingdom
Acute and Chronic Liver Disease: Nutrition in Renal Disease (56)

Virginie Colomb, MD, PhD
Department of Pediatric Gastroenterology Nutrition Hôpital Necker-Enfants Malades
Paris, France
Nutrition Support Teams (71)

Melanie Connolly, MSc, RD, CNSC
Adjunct Professor, Nutrition
Northeastern University
Boston, Massachusetts
Regis College
Weston, Massachusetts
Framingham State University
Framingham, Massachusetts
Appendix I: Nutritional Assessment
Appendix II: Nutritional Requirements
Appendis III: Enteral Products

Anna Coutsoudis, PhD
Department of Pediatric and Child Health
School of Clinical Medicine
University of KwaZulu-Natal
Glenwood, Durban, South Africa
Pediatric HIV Infection (44)

Stephen R. Daniels, MD, PhD
Professor and Chairman, Department of Pediatrics,
University of Colorado School of Medicine
Pediatrician-in-Chief
L. Joseph Butterfield Chair in Pediatrics
Children's Hospital Colorado
Aurora, Colorado
Dietary Reference Intakes in the United States (11)

Jai Das, MD, MBA
Division of Woman and Child Health
Aga Khan University
Karachi, Parkistan
Global Interventions in Child Health (43)

Sarah D. de Ferranti, MD, MPH
Director, Preventive Cardiology
Department of Cardiology

Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
Hyperlipidemia and Cardiovascular Disease (63)

Carlotta De Filippo, DSc, PhD
Department of Food Quality and Nutrition
Fondazione Edmund Mach-Research and Innovation Centre
San Michele All'adige TN, Italy
Diet, Environment, and Gut Microbiota in the Metagenomic Era (23)

Hans Demmelmair, PhD
Division of Metabolic Diseases and Nutritional Medicine
Dr. von Hauner Children's Hospital
University of Munich Medical Centre
München, Germany
Macronutrient Requirements for Growth: Fats and Fatty Acids (7)

Katharina Dokoupil, MSc
Department of Metabolism and Nutrition
Dr. von Hauner Children's Hospital
University of Munich
Munich, Germany
Special Diets (70)

Anne Donnet-Hughes, PhD
Nestlé Research Center
Lausanne, Switzerland
Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut (30)

Sharon M. Donovan, PhD, RD
Professor and Melissa M. Noel Endowed Chair in Nutrition and Health
Departments of Food Science and Human Nutrition
University of Illinois
Urbana, Illinois
Human Milk: Nutritional Properties (29)

Kelly A. Dougherty, PhD
Division of Gastroenterology, Hepatology and Nutrition
Department of Pediatrics
The Children's Hospital of Philadelphia
University of Pennsylvania
Philadelphia, Pennsylvania
Developmental Disabilities (48)

Martha Douglas-Escobar, MD, FAAP
Assistant Professor
Department of Pediatrics
University of California San Francisco
San Francisco, California
Gastrointestinal Development: Implications for Infant Feeding (22)

Christopher Duggan, MD, MPH
Professor of Pediatrics
Harvard Medical School
Professor in the Department of Nutrition
Harvard TH Chan School of Public Health
Director, Center for Nutrition
Division of Gastroenterology and Nutrition
Boston Children's Hospital
Boston, Massachusetts
Cancer Treatment (58)
Diarrheal Diseases (60)

Johanna Dwyer, DSc, RD
Frances Stern Nutrition Center
Tufts Medical Center
Department of Medicine and Community Health
Tufts University School of Medicine
USDA HNRC on Aging at Tufts University
Medford, Massachusetts
Community Nutrition and Its Impact on Children: Highly Industrialized Countries (16)

Kenneth J. Ellis, PhD
Professor of Pediatrics and Nutrition
Baylor College of Medicine
Houston, Texas
Body Composition and Growth (4)

Sharon Evans, MSc, RD
Research Dietitian
Dietetic Department
Birmingham Children's Hospital
Birmingham, United Kingdom
Special Diets (70)

Mary S. Fewtrell, MA, MD, FRCPCH
Childhood Nutrition Research Centre
Institute of Child Health
University College London
London, United Kingdom
Complementary Feeding and Health Outcomes (35)

Alison E. Field, ScD
Professor of Pediatrics
Division of Adolescent Medicine
Boston Children's Hospital
Department of Epidemiology
Harvard TH Chan School of Public Health
Channing Division of Network Medicine
Brigham and Women's Hospital
Harvard Medical School
Boston, Massachusetts
Adolescent Eating Disorders (65)

Jeremy G. Fisher, MD
General Surgery Resident
The Center for Advanced Intestinal Rehabilitation
Department of Surgery
Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
The Critically Ill Child (62)

A. Lindsay Frazier, MD, ScM
Associate Professor in Pediatrics
Harvard Medical School
Associate Professor in Pediatric Oncology
Dana-Farber Cancer Institute
Research Associate in Nutrition
Harvard TH Chan School of Public Health
Boston, Massachusetts
Cancer Prevention (57)

Simonetta Friso, MD, PhD
Department of Medicine
University of Verona School of Medicine
Verona, Italy
Nutrition and Epigenetics (20)

Sandra Fucile, OTR, PhD
Department of Pediatrics
Division of Neonatalogy
University of Florida
Gainesville, Florida
Gastrointestinal Development: Implications for Infant Feeding (22)

Christoph Fusch, MD
Department of Paediatrics
McMaster University
Hamilton, Ontario, Canada
Carbohydrates (5)

Kevin Gaskin, MD, FRACP
James Fairfax Professor of Paediatric Nutrition
Head, Department of Gastroenterology
University of Sydney Medical School
The Children's Hospital at Westmead
Westmead, Australia
Exocrine Pancreas (55)

Lena Geiger, MSc
Institute of Nutritional Science
Justus Liebig University Giessen
Giessen, Germany
Carbohydrates (5)

Catherine M. Gordon, MD, MSc

Director and Rauh Chair, Division of Adolescent and Transition Medicine
Director, Proctor Scholar Program
Professor, Department of Pediatrics
University of Cincinnati College of Medicine
Cincinnati Children's Hospital
Cincinnati, Ohio
Adolescence: Advancing Bone Health (66)

Kerri Gosselin, MD
Assistant Professor of Pediatrics
Division of Gastroenterology and Nutrition
University of Massachusetts Medical School
Worcester, Massachusetts
Diarrheal Diseases (60)

Johannes B. van Goudoever, MD, PhD
Professor and Head, Department of Paediatrics
University of Amsterdam Faculty of Medicine
Head of Paediatrics, VU Medical Center and AMC Emma Children's Hospital
Amsterdam, the Netherlands
Proteins and Amino Acids (6)

Kathleen M. Gura, PharmD, BCNSP, FASHP, FPPAG, FASPEN
Manager, Clinical Research
Department of Pharmacy
Clinical Pharmacist GI/Nutrition
Boston Children's Hospital
Associate Professor of Clinical Pharmacy
Massachusetts College of Pharmacy and Health Sciences University
Boston, Massachusetts
Drug Therapy and the Role of Nutrition (18)

Colleen Hadigan, MD, MPH
Laboratory of Immunoregulation
National Institute of Allergy and Infectious Disease
National Institutes of Health
Bethesda, Maryland
Pediatric HIV Infection (44)

K. Michael Hambidge, MD, ScD
Professor Emeritus of Pediatrics
Department of Pediatrics
University of Colorado
Aurora, Colorado
Trace Elements (8)

Corina Hartman, MD
Acting Director and Head of Nutritional Support
Pediatric Gastroenterology, Hepatology, and Nutrition Institute
Schneider Children's Medical Center of Israel
Petah Tikva, Israel
Parenteral Nutrition (68)

William W. Hay, Jr., MD
Professor of Pediatrics
Anschutz Medical Campus
University of Colorado School of Medicine
Perinatal Research Center
Aurora, Colorado
Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism (27)

Kristy Hendricks, DSc, RD
Hood Center for Children and Families
Community Research Program
The Geisel School of Medicine Dartmouth
Lebanon, New Hampshire
Department of Public Health and Community Medicine
Tufts Medical School
Medford, Massachusetts
Community Nutrition and Its Impact on Children: Highly Industrialized Countries (16)

Rob Heuschkel, MBBS, MRCPCH
Consultant Paediatric Gastroenterologist
Department of Paediatric Gastroenterology, Hepatology & Nutrition
Addenbrookes Hospital
Cambridge University Hospitals
NHD Foundation Trust
Cambridge, United Kingdom
Inflammatory Bowel Disease (52)

Ann Hill, PhD
Emeritus Professor
London School of Hygiene and Tropical Medicine
University of London
London, United Kingdom
Childhood Malnutrition: Prevention and Control at the National Level (15)

Daniel J. Hoffman, PhD
Associate Professor
Department of Nutritional Sciences
Rutgers University
Piscataway, New Jersey
Energy and Substrate Regulation in Obesity (36)

Alison G. Hoppin, MD
Associate Director for Pediatric Programs
MassGeneral Hospital for Children's Weight Center
Assistant Professor of Pediatrics
Harvard Medical School
Boston, Massachusetts
Evaluation and Management of Childhood Obesity (37)

Adrienne H. Hughes, BSc, PhD
Physical Activity for Health Group
School of Psychological Sciences & Health
University of Strathclyde
Glasgow, Scotland
Childhood Obesity Prevention (39)

Susanna Y. Huh, MD, MPH
Assistant Professor of Pediatrics
Center for Nutrition
Division of Gastroenterology, Hepatology and Nutrition
Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
Failure-to-Thrive: Growth Failure in the Outpatient Setting (47)

Kristen M. Hurley, PhD
Assistant Professor
Department of International Health
Division of Human Nutrition
Johns Hopkins Bloomberg School of Public Health
Baltimore, Maryland
Nutrition and Neuropsychological Development (24)

Tom Jaksic, MD, PhD
W. Hardy Hendren Professor of Surgery
Harvard Medical School
Vice-Chairman of Pediatric General Surgery
Surgical Director
Center for Advanced Intestinal Rehabilitation
Boston Children's Hospital
Boston, Massachusetts
The Critically Ill Child (62)

Tracey Johnson, BSc, RD
Dietetic Department
Birmingham Children's Hospital
Birmingham, United Kingdom
Special Diets (70)

Peter J. H. Jones, PhD
Director, Richardson Centre for Functional Foods and Nutraceuticals
University of Manitoba
Winnipeg, Canada
Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals (12)

Daniel Kamin, MD
Director, GI Inpatient Consultation Services
Division of Gastroenterology, Hepatology and Nutrition
Boston Children's Hospital
Assistant Professor of Pediatrics
Harvard Medical School
Boston, Massachusetts
Intestinal Failure, Short Bowel Syndrome, and Intestinal Transplantation (61)

Emily Keefe, RN, MSN, FNP
Graduate Student in MSN and FNP Programs

School of Nursing and Health Sciences
Simmons College
Boston, Massachusetts
Nutritional Epidemiology (17)

Ciarán P. Kelly, MD
Professor of Medicine
Harvard Medical School
Chief, Herrman L. Blumgart Internal Medicine Firm
Director of Gastroenterology Training
Medical Director, Celiac Center
Beth Israel Deaconess Medical Center
Boston, Massachusetts
Celiac Disease (53)

Deirdre A. Kelly, MD, FRCP, FRCPI, FRCPHC
Professor of Paediatric Hepatology
The Liver Unit
Birmingham Children's Hospital
Birmingham, United Kingdom
Acute and Chronic Liver Disease: Nutrition in Renal Disease (56)

Amina Khambalia, PhD
Research Fellow in Epidemiology
Kolling Institute of Medical Research
Northern Clinical School
The University of Sydney
New South Wales, Australia
Iron (9)

Faraz A. Khan, MD
Administrative Chief Resident
Department of Surgery
Wayne State University
Detroit, Michigan
The Critically Ill Child (62)

Berthold Koletzko, MD, PhD
Professor of Paediatrics
Division of Metabolic Diseases and Nutritional Medicine
Dr. von Hauner Children's Hospital
Ludwig Maximilian University
University of Munich Medical Centre
Munich, Germany
Macronutrient Requirements for Growth: Fats and Fatty Acids (7)

Nancy F. Krebs, MD, MS
Professor of Pediatrics
Section Head, Nutrition
University of Colorado College of Medicine
Denver, Colorado
Trace Elements (8)

Kathrin Krohn, MD
Division of Metabolic Diseases and Nutritional Medicine
Dr. von Hauner Children's Hospital
Ludwig Maximilian University
Munich, Germany
Macronutrient Requirements for Growth: Fats and Fatty Acids (7)

Clemens Kunz, PhD
Professor of Nutritional Science
Institute of Nutritional Science
Justus Liebig University Giessen
Giessen, Germany
Carbohydrates (5)

Roland Kupka, DSc
Senior Advisor, Micronutrients
UNICEF, United Nations
New York, New York
Adjunct Assistant Professor of Nutrition
Harvard University
Boston, Massachusetts
Vitamins (10)

Anura Kurpad, MD, PhD, FAMS
Professor and Head, Division of Nutrition

St. John's Research Institute
Professor of Physiology
St. John's Medical College
Bangalore, India
Obesity in Developing Economies (45)

Cécile Lambe, MD
Pediatric Gastroenterology and Nutrition Department
Hôpital Necker-Enfants Malades
Paris, France
Nutrition Support Teams (71)

Alexandre Lapillonne, MD, PhD
Department of Neonatology
Hôspital Necker-Enfants Malades
Paris, France
The Low-Birthweight Infant: Inpatient Care (32)

Robert M. Lawrence, MD, FAAP, FABM
Clinical Professor of Pediatrics
Pediatric Immunology, Rheumatology, and Infectious Diseases
Department of Pediatrics
HIV Care for Children and Adolescents
General Pediatric Infectious Disease
Assoc. Director of Pediatric Residency Program
Florida/Caribbean AIDS Education Training Center
Faculty
University of Florida College of Medicine
Gainesville, Florida
Approach to Breastfeeding (31)

Ruth A. Lawrence, MD, FAAP, FABM, FAAC
Professor of Pediatrics and Obstetrics and Gynecology
University of Rochester
School of Medicine
Rochester, New York
Approach to Breastfeeding (31)

Carine M. Lenders, MD, MS, ScD
Associate Professor of Pediatrics
Boston University School of Medicine
Boston, Massachusetts
Nutritional Epidemiology (17)

Alice H. Lichtenstein, DSc
Gershoff Professor of Nutrition Science and Policy
Friedman School, Tufts University
Director and Senior Scientist
Cardiovascular Nutrition Laboratory
JM USDA Human Nutrition Research Center on Aging
Boston, Massachusetts
Dietary Reference Intakes in the United States (11)

Paolo Lionetti, MD, PhD
Associate Professor of Paediatrics,
Department of Neuroscience, Psychology, Pharmacology and Child's Health
University of Florence
Meyer Children's Hospital
Florence, Italy
Diet, Environment, and Gut Microbiota in the Metagenomic Era (23)

Keith J. Loud, MDCM, MSc
Chief, Section of General Academic Pediatrics
Children's Hospital at Dartmouth
Associate Professor of Pediatrics
Geisel School of Medicine at Dartmouth
Hanover, New Hampshire
Adolescence: Advancing Bone Health (66)

Clodagh M. Loughrey, MD, MRCP, FRCPath
Consultant Chemical Pathologist
Belfast Trust
Belfast, Northern Ireland
Laboratory Assessment of Nutritional Status (3)

Anita MacDonald, BSc, PhD, RD
Consultant Metabolic Dietitian
Birmingham Children's Hospital

Birmingham, United Kingdom
Special Diets (70)

Jennifer M. Maloney, MD
Division of Allergy and Immunology
Clinical Instructor of Pediatrics
Mount Sinai School of Medicine
New York, New York
Food Allergies (54)

Asim Maqbool, MD
Attending Physician
Division of Gastroenterology, Hepatology, and Nutrition
Children's Hospital of Philadelphia
Associate Professor of Pediatrics
Perelman School of Medicine
University of Pennsylvania
Philadelphia, Pennsylvania
Clinical Assessment of Nutritional Status (2)

William A. Masters, PhD
Professor of Nutrition
Department of Food and Nutrition Policy
Friedman School of Nutrition Science and Policy
Tufts University
Boston, Massachusetts
Child Nutrition and Economic Development (42)

Zoë McCallum, MBBS, FRACP
Department of Gastroenterology and Clinical Nutrition
Royal Children's Hospital
Melbourne, Australia
Enteral Nutrition and Formulas (67)

Nilesh M. Mehta, MD, DCH, MRCPCH
Director, Critical Care Nutrition
Associate Medical Director, MSICU
Children's Hospital Boston
Associate Professor in Anesthesia
Harvard Medical School
Boston, Massachusetts
The Critically Ill Child (62)

Sanjeev Mehta, MD, MPH
Director of Quality and Assistant
Investigator Section on Genetics and Epidemiology
Joslin Diabetes Center
Staff Physician, Joslin Clinic
Instructor in Pediatrics
Harvard Medical School
Boston, Massachusetts
Diabetes Mellitus (59)

Madhusmita Misra, MD, MPH
Professor of Pediatrics
Neuroendocrine Unit
Pediatric Endocrine Unit
Massachusetts General Hospital for Children
Harvard Medical School
Boston, Massachusetts
Nutrition in the Athlete (69)

Marcia H. Monaco, PhD
Departments of Food Science and
Human Nutrition and Pediatrics
University of Illinois
Urbana, Illinois
Human Milk: Nutritional Properties (29)

Josef Neu, MD
Professor and Director, Neonatology Fellowship Training Program
University of Florida College of Medicine
Gainsville, Florida
Gastrointestinal Development: Implications for Infant Feeding (22)

Irene E. Olsen, PhD, RD, LDN
Adjunct Assistant Professor
University of Pennsylvania School of Nursing
Children's Hospital of Pennsylvania

Philadelphia, Pennsylvania
Clinical Assessment of Nutritional Status (2)

Francesca Penagini, MD
Department of Pediatrics
Luigi Sacco Hospital
Universita' degli Studi di Milano
Milan, Italy
Inflammatory Bowel Disease (52)

Mary E. Penny, MBChB
Senior Researcher
Instituto de Investigación Nutricional
Lima, Peru
Severe Acute Malnutrition: Pathophysiology, Clinical Consequences, and Treatment (14)

Janey S. Pratt, MD
Co-Director, Massachusetts General Hospital Weight Center
Director, MGH Nutrition Support Unit
Assistant Professor of Surgery
Massachusetts General Hospital
Boston, Massachusetts
Evaluation and Management of Childhood Obesity (37)

Susan Protheroe, MD, MRCP, FRCPHC
Consultant Paediatric Gastroenterologist
Honorary Senior Clinical Lecturer in Paediatrics and Child Health
Department of Gastroenterology and Nutrition
Birmingham, United Kingdom
Acute and Chronic Liver Disease: Nutrition in Renal Disease (56)

Maryanne Quinn, MD
Assistant in Medicine (Endocrinology)
Boston Children's Hospital
Assistant Professor of Pediatrics
Harvard Medical School
Boston, Massachusetts
Diabetes Mellitus (59)

Timothy R. H. Regnault, MD, PhD
Associate Professor
Department of Obstetrics and Gynaecology
University of Western Ontario
London, Ontario, Canada
Amino Acid Nutrition in Utero: Placental Function and Metabolism (28)

John J. Reilly, BSc, PhD
Professor of Physical Activity and Public Health Science
School of Psychological Sciences & Health
University of Strathclyde
Glasgow, Scotland
Childhood Obesity Prevention (39)

Melanie Reyes, MS, RD, LD
Registered Dietitian and Personal Trainer
Life Time Fitness
Austin, Texas
Community Nutrition and Its Impact on Children: Highly Industrialized Countries (16)

J. Marc Rhoads, MD
Professor of Pediatrics
Division of Pediatric Gastroenterology
The University of Texas
Health Science Center at Houston
Houston, Texas
Nutrition and the Humoral Regulation of Growth (21)

Arieh Riskin MD, MHA
Director of the Neonatal Intensive Care Unit
Department of Neonatology
Bnai Zion Medical Center
Clinical Associate Professor
Rappaport Faculty of Medicine
Technion, Israel Institute of Technology
Haifa, Israel
Parenteral Nutrition (69)

Susan B. Roberts, PhD
Senior Scientist and Director
Energy Metabolism Laboratory

Tufts University
Boston, Massachusetts
Energy and Substrate Regulation in Obesity (36)

Nancy M. Rodig, MD
Assistant Professor of Pediatrics
Harvard Medical School
Division of Nephrology
Boston Children's Hospital
Boston, Massachusetts
Acute and Chronic Kidney Disease (51)

Katalin M. Ross, RD, LDN
Registered Dietitian
Glycogen Storage Disease Program
University of Florida Health
Gainesville, Florida
Inborn Errors of Fasting Adaptation—Glycogen Storage Disease (50)

Paul J. Rozance, MD
Associate Professor of Pediatrics
Perinatal Research Center
University of Colorado School of Medicine
Aurora, Colorado
Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism (27)

William E. Russell, MD
Cornelius Vanderbilt Chair
Professor of Pediatrics
Professor of Cell and Developmental Biology
Director, Division of Endocrinology and Diabetes
Vanderbilt University School of Medicine
Nashville, Tennessee
Nutrition and the Humoral Regulation of Growth (21)

Hugh A. Sampson, MD
Professor of Pediatrics
Division of Allergy and Immunology
Department of Pediatrics
Mount Sinai School of Medicine
New York, New York
Food Allergies (54)

Ian R. Sanderson, MD, MSc, MRCP
Professor of Paediatric Gastroenterology
Blizard Institute
Centre for Immunobiology
Barts and The London School of Medicine & Dentistry
London, United Kingdom
Nutrition and Gene Expression (19)

Eduardo J. Schiffrin, MD, PhD
Clinical Development Unit
Nestec Ltd.
Vers-chez-les-Blanc
Lausanne, Switzerland
Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut (30)

Theresa O. Scholl, PhD, MPH
Professor of Obstetrics and Gynecology
Department of Obstetrics and Gynecology
University of Medicine and Dentistry of New Jersey SOM
Stratford, New Jersey
Maternal Nutrition and Pregnancy Outcome (25)

Thibault Senterre, MD, PhD, FRCPC
Centre Hospitalier Universitaire de Liège
Centre Hospitalier Régional de la Citadelle
University of Liège
Herstal, Belgium
The Low-Birthweight Infant: Inpatient Care (32)

Mandana Amir Shaghaghi, MSc, RD
Richardson Centre for Functional Foods
University of Manitoba
Winnipeg, Canada
Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals (12)

Raanan Shamir, MD
Sackler Faculty of Medicine

Tel Aviv University
Institute of Gastroenterology, Nutrition and Liver Diseases
Co Chair, Schneider Children's Medical Center of Israel
Petah Tikva, Israel
Parenteral Nutrition (68)

James H. Shaw, MD, PhD
Professor of Nutrition Emeritus
Harvard University School of Dental Medicine
Boston, Massachusetts
Diet and Dental Disease (64)

Philip M. Sherman, MD, FRCPC
Research Institute
Hospital for Sick Children
Professor of Pediatrics
University of Toronto
Toronto, Ontario, Canada
Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals (12)

Kendrin R. Sonneville, ScD, RD, LDN
Assistant Professor, Human Nutrition Program
University of Michigan School of Public Health
Ann Arbor, Michigan
Adolescent Eating Disorders (65)

Eric A. Sparks, MD
Surgery Resident
Center for Advanced Intestinal Rehabilitation
Department of Surgery
Boston Children's Hospital
Harvard Medical School
Boston, Massachusetts
The Critically Ill Child (62)

Peter B. Sullivan, MA, MD, FRCP, FRCPCH
Associate Professor in Paediatrics
Children's Hospital
Oxford University Hospitals NHS Trust
Oxford, United Kingdom
Feeding Difficulties (46)

Rosalyn Sulyanto, DMD, MS
Attending in Pediatric Dentistry
Boston Children's Hospital
Boston, Massachusetts
Diet and Dental Disease (64)

Sumathi Swaminathan, PhD
Assistant Professor
Division of Nutrition
St. John's Research Institute
Bangalore, India
Obesity in Developing Economies (45)

Stephanie R. Thorn, PhD
Department of Pediatrics
Perinatal Research Center
University of Colorado School of Medicine
Aurora, Colorado
Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism (27)

Ricardo Uauy, MD, PhD
Professor of Pediatrics
Pontifical Catholic University of Chile
Santiago, Chile
Childhood Malnutrition: Prevention and Control at the National Level (15)

Charles P. Venditti, MD, PhD
Investigator, National Human Genome Research Institute
Director, Organic Acid Research Unit
National Institutes of Health
Bethesda, Maryland
Inborn Errors of Metabolism and the Liver (49)

Eduardo Villamor, MD, DrPh
Professor of Epidemiology
University of Michigan School of Public Health
Department of Epidemiology
Ann Arbor, Michigan

Vitamins (10)

Martin Wabitsch, MD, PhD
Head, Division of Pediatric Endocrinology and
Diabetes Endocrine Research Laboratory
Department of Pediatrics and Adolescent Medicine
Ulm, Germany
The Metabolic Syndrome in Children and Adolescents (38)

W. Allan Walker, MD
Conrad Taff Professor of Nutrition
Professor of Pediatrics
Harvard Medical School
Chief, Mucosal Immunology Laboratory
Pediatric Gastroenterology and Nutrition Unit
Massachusetts General Hospital for Children
Charlestown, Massachusetts
Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut (30)

Julie Wang, MD
Division of Allergy and Immunology
Associate Professor of Pediatrics
Mount Sinai School of Medicine
New York, New York
Food Allergies (54)

Karen Y. Warman, MS, RD
Clinical Dietitian Specialist, Diabetes Program
Boston Children's Hospital
Boston, Massachusetts
Diabetes Mellitus (59)

John B. Watkins, MD
Emeritus Associate Professor of Pediatrics
Harvard Medical School
Division of Gastroenterology, Hepatology and Nutrition
Boston Children's Hospital
Boston, Massachusetts
Volume Editor

David A. Weinstein, MD, MMSc
Professor of Endocrinology
Director, Glycogen Storage Disease Program
Associate Program Director for Research
University of Florida College of Medicine
Gainesville, Florida
Inborn Errors of Fasting Adaptation—Glycogen Storage Disease (50)

Dascha C. Weir, MD
Instructor in Pediatrics
Harvard Medical School
Associate Director, The Celiac Disease Program
Department of Gastroenterology, Hepatology, and Nutrition
Boston Children's Hospital
Boston, Massachusetts
Celiac Disease (53)

Gerardo Weisstaub, MD, MSc
Instituto de Nutrición y Tecnología de los Alimentos (INTA)
University of Chile
Santiago, Chile
Childhood Malnutrition: Prevention and Control at the National Level (15)

Walter C. Willet, MD, DrPH
Frederick John Stare Professor of Epidemiology and
Nutrition
Chair, Department of Nutrition
Department of Epidemiology
Harvard T. H. Chan School of Public Health
Professor of Medicine
Harvard Medical School
Boston, Massachusetts
Nutritional Epidemiology (17)

Joseph I. Wolfsdorf, MB, BCh
Director, Diabetes Program
Clinical Director and Associate Chief, Division of Endocrinology
Boston Children's Hospital
Professor of Pediatrics

Harvard Medical School
Boston, Massachusetts
Diabetes Mellitus (59)

Ian S. Young, BSc, MD, FRCP, FRCPath
Professor of Medicine
School of Medicine, Dentistry, and Biological Sciences
Director, Centre for Public Health
Associate Medicine Director, Belfast Health and Social Care Trust
Centre of Excellence for Public Health
Queen's University Belfast
Belfast, Ireland
Laboratory Assessment of Nutritional Status (3)

Babette S. Zemel, PhD
Research Professor of Pediatrics
Division of Gastroenterology, Hepatology and Nutrition
The Children's Hospital of Philadelphia
Philadelphia, Pennsylvania
Clinical Assessment of Nutritional Status (2)
Developmental Disabilities (48)

Ekhard E. Ziegler, MD
Professor, Department of Pediatrics
University of Iowa Hospitals and Clinics
Iowa City, Iowa
The Term Infant (34)

Stanley H. Zlotkin, OC, MD, PhD, FRCPC
Professor, Nutritional Sciences and Paediatrics
University of Toronto
Chief, Centre for Global Child Health
The Hospital for Sick Children
Toronto, Ontario, Canada
Iron (9)

PREFACE

This fifth edition of *Nutrition in Pediatrics: Basic Science and Clinical Applications* is our continuing effort to establish a comprehensive and accessible approach to pediatric nutrition for a wide range of clinicians, scientists, and—most importantly—trainees. An important addition to the textbook has been the inclusion of Professor Berthold Koletzko to our Editorial Board. As Professor of Pediatrics at the University of Munich and Head of the Division of Metabolic and Nutritional Medicine at Hauner Children's Hospital, Dr. Koletzko brings a wealth of clinical and research expertise to this text. In addition, his global collaborations have allowed us to substantially broaden our team of authors, making this version of the text a truly international one.

The fifth edition builds on the foundations of the several previous versions, with updated chapters on the broad themes of General Concepts, Physiology and Pathophysiology, Perinatal Nutrition, Obesity, Nutritional Aspects of Specific Disease States, and Approach to Nutritional Support. Owing to the recognition that nutritional issues overwhelmingly affect children living in resource-poor countries, we have also added a new section entitled "Nutrition in Low- and Middle-Income Populations." Expert authors in this field have contributed reviews on the topics of the nutrition transition, complementary feeding, economic development, HIV disease, and the occurrence of obesity in developing economies.

Although all of our contributions are excellent, notable additions to this 5th edition include an outstanding leadoff chapter on the role of nutrition for health, disease prevention, and development; new contributions on macronutrients, epigenetics, and the microbiome; and several significantly revised and updated classic chapters. As in past editions, comprehensive appendices are included that serve the reader by collating important tools for nutritional assessment, nutritional requirements, and enteral products.

The editors are grateful to Ms. Linda Mehta for her many efforts in the publication of this textbook.

Finally, we sincerely thank the dozens of authors who contributed countless hours of writing and research for the completion of this text. Their expertise and labors constitute the heart of this book.

—Christopher Duggan,
for the editors
Berthold Koletzko
John B. Watkins
W. Allan Walker

Nutrition: The Driving Force for Health, Disease Prevention, and Development

1

Robert D. Baker, MD, PhD and Susan S. Baker, MD, PhD

A SWEEPING LOOK AT NUTRITION

Nutrition is the underpinning of life itself. Good nutrition has consequences for health, societal well-being, economic stability, and advancements. Poor nutrition has a direct and measurable impact on all aspects of human functioning. Nutrition is the driving force for much of human and societal evolution. This fundamental role of nutrition has been recognized since ancient times. More recently, evolutionary biologists have recorded the importance of nutrition on evolution, and the basic requirements for the human organism have been identified.

This book deals with the scientific and clinical practice of pediatric nutrition. That it is now into its fifth edition implies that readers and users of this book are convinced of the importance of nutrition to the growing and developing child. In this first chapter, we invite “devotees” of pediatric nutrition to take a very broad look at the importance of food and to consider food as a driving force for the evolution of the human race, as a major part of our culture, as an economic force, and as a means toward future change.

From prehistory forward, food and the search for food have been

central to human evolution. In examining the expansion of humans from their African origins throughout the Middle East, Europe, Asia, the Far East including the Pacific Islands, across the bridge to North America, and south through Central America to include all of South America, one of the most striking aspects of this amazing migration is the rapidity with which it took place. After existing in Africa for 5–6 million years, the great migration started about 1 million years ago and was essentially complete about 20,000 years ago.[1] The migration was not a single sweep, but occurred in waves and fits and starts. The reasons for man's moving were undoubtedly multifactorial, but prominent among these factors had to be food availability. The move from "gathers" to "hunter-gathers" required the acquisition of tools, weapons, and necessitated cooperation, but also ushered in conflict. The beginning of agriculture, in the fertile crescent, not only provided a more stable source of nutrition but also allowed humans to establish settlements, which required rules and patterns of behavior far different from the behavior of hunter-gathers. Among many other results of "settlements" was the possibility of specialization; most would be farmers but there were also warriors, carpenters, builders, teachers, and eventually philosophers and scientists.[1] The move toward agriculture was gradual. Man first learned that by tending plants that bore fruit, the yield could be increased. Some tribes in New Guinea still practice this early form of agriculture. They clear completing plants from food-bearing plants and then leave returning months later to harvest the enhanced fruit production.[1] Wheat and other grains were the first food plants to be domesticated. The cultivation of pulses in addition to grains allowed both protein and carbohydrate needs to be largely supplied through plants. Domesticated animals became a readily available and dependable source of protein. Initially in the Middle East but then spreading, communities began domesticating animals; first sheep and then goats and finally cattle.

LACTOSE INTOLERANCE

About 7000–8000 years ago, these Middle Eastern farmers began consuming milk.[2] This created a genetic dilemma for humans. Lactose is the disaccharide in most mammalian milk, including human milk.[3] Human infants, to survive, must be able to digest and use lactose. Before the introduction of dairy products into the human food chain, there was no advantage of being able to digest lactose beyond the age of weaning. So, for most of the human race, the gene that codes for lactase becomes progressively less active during childhood. However, with the introduction

of milk into the human diet, an evolutionary pressure for persistent lactase activity was created. Humans dealt with this dilemma in two ways. The lifelong use of dairy products spread from the Middle East north to Europe and Scandinavia where the persistence of the lactase gene was common. An added advantage for human at northern latitudes was lactose's positive effect on calcium absorption, partially making up for decreased vitamin D due to lack of exposure to sunlight. Dairy as a food source also spread from the Middle East into Eastern Europe and Asia. Here, the gene of persistence of lactase was less common, but man developed a system to decrease the lactose by fermenting milk to make yogurts and cheeses.[2]

THE POTATO FAMINE

The results of agriculture have not always been beneficial. The potato was first domesticated 7000 years ago in the Andes, where thousands of varieties are still grown.[4] Potatoes were introduced to Europe after the Spanish conquest of South America in the 1500s. After an initial slow start in Europe, it became a staple food and is estimated to be responsible for 25% of the European population growth between 1700 and 1900. The limited varieties of potatoes grown in Europe created a setup for disease. The Great Irish Potato Famine began in 1845 when a fungus, *Phytophthora infestans,* caused potato harvest failure.[5] In Ireland, where a third of the population was entirely dependent on potatoes for food, over a million people died and a million emigrated.[6] This diaspora forever changed the history of Ireland, Great Britain, and the United States. In Ireland, so great was the suffering and death that the famine has entered in the folk psyche of the people. It triggered the growth of Irish nationalism and soured the Irish–English relationship. It furthered the split between the Catholic and Protestant islanders. Great Britain is still dealing with the aftermath of the famine and the sense among the Irish that they were abandoned. In the United States, one need only look at surnames on the ballot in any election to verify to what extent the Irish have entered into our national heritage.

THE DUST BOWL

The Great Plains were originally known as the "Great American Desert," because this region received an average of less than 10 inches of precipitation a year and was considered unacceptable for agriculture. However, a combination of huge pressure to settle in the West and a string

of unusually wet years induced farmers, mostly immigrants, into this area.[7] Poor farming techniques such as deep plowing of virgin top soil, no cover crops, no crop rotation, and lack of wind breaks created a few very profitable years followed by a collapse of the agriculture when the wet years gave way to more normal rainfall. The result was soil erosion on a vast scale leading to huge dust storms called “black rollers.”Dust from the Great Plains reached East Coast cities. Hundreds of thousands of people were displaced. Hunger became so severe that tumble weed (an invasive species inadvertently introduced from the Ukrainian steppes) was used for feeding cattle and even for human consumption.[7]

THE GREEN REVOLUTION

The green revolution refers to the widespread application of modern farming techniques, improved seeds, fertilizers, and pesticides, to areas where traditional, small-scale farming was practiced. This movement began in the 1940s in Mexico, but became vitally important in the 1960s in India. In 1961, India faced a huge food shortage. The prospect of massive food shortages and famine frightened the Indian government and the world. The Indian government adopted the green revolution methods, in particular, use of a variety of rice developed at the International Rice Research Institute along with fertilizers and pesticides. Yields were up to 10 times that of traditionally grown rice crops. Some say that over a billion people, worldwide, have been saved from starvation because of the green revolution.[8] Green revolution techniques have gained widespread use. Famine, suffering, and death have been avoided. But the green revolution is not without its critics. The green revolution is heavily dependent on herbicides, pesticides, fertilizers, and monoculture. Hence, reliance on petrochemical products necessarily increased. While the crop yield dramatically increased so did the energy input needed to produce crops. In addition, the world population continues to increase, leading some to propose the Malthusian argument that we have merely postponed starvation and famine that will surely come. The green revolution has led to widespread deforestation and dramatic loss of biodiversity. So the verdict on the green revolution is not yet in.[9]

GENETICALLY MODIFIED FOODS

Genetically modified (GM) foods have been genetically engineered via DNA modification. Currently, there are many examples of GM foods on

the market. There is a general consensus that GM foods do not add risk to the consumer. The DNA of GM foods is largely broken down and inactivated in the gastrointestinal (GI) tract. To date, there have been no reports of allergy induced by conformational changes within the structure of GM foods. Likewise, there have been no reports of toxicity. However, there are concerns. Could GM crops become locally invasive? The evidence is against this as almost always the GM crop is less "fit" than its non-GM counterpart, so although it can escape the field, it rarely survives in the wild. To date, the major aim of developing a GM crop is to increase the yield. Examples of this are Bt. In this case, a gene from the bacteria, *Bacillus thuringiensis*, is introduced into the plant genome. This gene codes for a toxin that attacks the Lepidoptera larvae, allowing for naturally occurring bacteria to infect and kill the larva. In the case of corn, the initial target was the European Corn Borer.[10] Since first introduced, Bt crops have been further modified to include other insect pests. The modified varieties have become significantly more sophisticated than the original Bt varieties. Another aim of GM technology has been to improve the quality of the crop. An example of this is "golden rice," a variety of rice genetically modified to contain beta-carotene and thus serves as a tool to fight vitamin A deficiency.[11] As with the Green revolution, there has been substantial opposition to genetically altering the food chain. The opposition mainly centers around fears of unintended environmental consequences and fear that a few large multinational corporation will control all access to seeds.

FOOD USED FOR GOOD AND EVIL

In writing about Ethiopian refugees in 1986, Nevin Scrimshaw wrote: "In Ethiopia today as in Biafra in 1969 and frequently throughout history, enforced hunger has been a weapon to crush political resistance."[12] A US policy of using food as political weapon could not have been more clearly stated than it was by Henry Kissinger in a memo written in 1974, where he stated that food aid could be withheld from third world countries that did not implement birth control as demanded by the US government.[13] Recently, we have witnessed an example of food as a political weapon when the Obama administration stopped food shipments to North Korea in response to the launch of a missile into space by North Korea.[14] One can argue the propriety of such a use of food; one can question the effectiveness in achieving the desired results, but it is clear that food is a weapon in world politics.

Food is also a means to achieve laudable ends. Treating vitamin A deficiency with “golden rice” is a desirable goal. Fortifying fish sauce with iron in Vietnam has greatly reduced the prevalence of iron-deficiency anemia.[15] Salt in the United States has been fortified with iodine since 1924. Adding iodine to salt virtually eliminated goiter resulting from iodine deficiency in the United States.[16] The addition of folate to wheat has reduced the occurrence of neural tube defects.[17]

The wide distribution and relative lack of safety measures (most of the regulations are adhered to and monitored by the producer and thus largely voluntary) in our food supply have opened the way to accidental contamination, but also to allow an opening to those who would like to harm large numbers of people. A challenge for the future is to devise safety measure that will ensure that health-promoting, uncontaminated food is delivered from the point of origin to the table.

FOOD BECOMES A SCIENCE

As discussed earlier, knowledge of food and how it relates to mammalian health was accumulating throughout history and was a driving force for health and development. However, nutritional sciences as currently understood probably began with the advances made during the mid-18th century in France.[18] Studies conducted during this time focused on understanding macronutrients and how they were incorporated into human tissue, that is, how ingested plant or animal products, or air, as it was known to contain nitrogen, were transformed into body parts. The first observations that dietary carbohydrate could be converted to fat were made, and it was established that animals generated heat and that this heat could be quantified. During the early 1900s, proteins were observed to hydrolyze into amino acids and short-order proteolytic enzymes were discovered, setting the stage for understanding the essentiality of specific amino acids.

Atwater, known as the father of nutritional sciences, initially focused on protein nutrition and also built the first calorimeter and was able to show that the heat produced by an individual is the same as that produced by the combustion of the same quantities of nutrients. He also established the estimates of metabolizable energy from protein, carbohydrate, and fat in mixed diets as 4, 4, and 9 kcal/g, respectively. These values retain their validity today.

For several decades, the focus of nutritional sciences was on the requirements for protein and energy, how they were related, and how

disease or trauma would alter them.[19,20] The essentiality of specific nutrients and deficiency states caused due to their absence from the diet were established. The list of essential nutrients is long, but vitamins, minerals, lipids, and specific amino acids were identified and synthesized. Products were developed that could be consumed to prevent or treat deficiency states.

During the late 20th century, the life-saving option of parenteral nutrition, delivery of nutrients that bypassed an absent or nonfunctional GI tract to sustain life and promote growth and development, was discovered and used clinically.[21–23] However, as the dangers of this option became apparent and the importance of directly nourishing the GI epithelial cells was discovered, parenteral nutrition as a common therapy for moderately ill patients or as a preparation for surgery in patients with an intact GI tract lost favor. Currently, parenteral nutrition is used only when the GI tract cannot deliver the required nutrition. Technical strides permitted the development of appliances, formulas, and experience, so enteral nutrition became the preferred route to deliver nutrients, even when the GI tract was compromised, to support ill children and promote health. The enteral route of nutrition support acknowledged the importance of the GI tract as a modulator of nutrition and immune function. Most recently, the complicated and dynamic interaction of nutrients, metabolism, and the GI microbiome has received attention.[24,25]

CHANGING VIEW OF NUTRIENTS

As these concepts were incorporated into the practice of nutrition, additional observations suggested that diets could provide therapeutic options and nutrients might play a role in treating disease. For example, the relative content of dietary fat and carbohydrate can have a desirable effect on the respiratory quotient and assist those with pulmonary disease.[26,27] Some nutrients, specifically, omega-3 polyunsaturated fatty acids, nucleotides, vitamin D, and specific amino acids singly or in combination have been shown to modulate the immune system within defined disease states or specific situations.[18,28–34] This has led to the concept of nutrients as pharmacological agents. Indeed, enteral nutrition support can have a positive effect on the induction and maintenance of remission in inflammatory bowel disease; however, no specific nutrient or combination of nutrients that can treat inflammatory bowel disease has been identified. To date, except for deficiency states, no single nutrient has withstood the scrutiny demanded of pharmaceutical agents to demonstrate

the efficacy for treatment of a specific condition or disease.

OVERNUTRITION

Although nutrition prescribed "correctly" can have a positive effect on health, it can also cause harm. Recent observations have brought to light the concept of overnutrition as a disease state.

According to the Centers for Disease Control, overweight and obesity are labels for ranges of weight that are greater than what is generally considered healthy for a given height. The terms also identify ranges of weight that have been shown to increase the likelihood of certain diseases and other health problems.

Growth charts for children and adolescents aged 2–19 years are used to determine the body mass index (BMI), weight/height^2, for age and sex percentile. Overweight is defined as a BMI at or above the 85th percentile and lower than the 95th percentile for children of the same age and sex.[35] Obesity is defined as a BMI at or above the 95th percentile for children of the same age and sex.[35]

The concern for obese and overweight children and the focus of resources on this disease state by health and government agencies developed because of observations that childhood obesity is associated with risk factors for chronic diseases that include cardiovascular disease, insulin resistance and type 2 diabetes, sleep apnea, fatty liver disease, joint and musculoskeletal problems, and a greater risk of social and psychological problems than nonobese children. These observations are magnified by the fact that obese children are likely to become obese adults and adult obesity is associated with serious, chronic health conditions.[36]

OVERVIEW OF THIS BOOK

The next chapters in this book address the current body of knowledge in nutrition for children. The basic biochemistry and metabolism of the science of nutrition is presented in a manner that makes this information clinically useful. Specifically, the book addresses nutritional strategies to support the sickest of children, to provide for normal growth and development for normal infants and children and those born with metabolic errors, to prevent diseases, both in childhood and later in adulthood, and to support normal growth and development for children who have specific disease states. The knowledge contained in this volume will be directly applicable to the practice of medicine to the primary care

physician, those who work in intensive care units and serve nutrition support services.

REFERENCES

1. Diamond J. *Guns, Germs, and Steel: The Fates of Human Societies*. Paper back ed. New York, NY: W. W. Norton and Company;1999:480.
2. McMichael T. *Human Frontiers, Environments and Disease: Past Patterns, Uncertain Futures*. Cambridge: Cambridge University Press;2001:413.
3. Wojcik KY, Rechtman DJ, Lee ML, et al. Macronutrient analysis of a nationwide sample of donor breast milk. *J Am Diet Assoc*. 2009;109(1):137–140.
4. Weatherford JM. *Indian Givers: How the Indians of the Americas Transformed the World*. New York, NY: Fawcett Columbine;1988:69.
5. Volk T. This month's fungus is Phytophthora infestans, cause of late blight of potato and the Irish potato famine. *Tom Volk's Fungus of the Month for March 2001*; 2001. http://botit.botany.wisc.edu/toms_fungi/m2001alt.html (accessed March 29, 2013).
6. Laxton E. *The Famine Ships: The Irish Exodus to America (1846–1851)*. New York, NY: Bloomsbury;1997:192.
7. Egan T. *The Worst Hard Time: The Untold Story of Those Who Survived the Great American Dust Bowl*. Boston, MA: Houghton Mifflin Harcourt;2006:352.
8. Matthews C. *Save and Grow Farming Model Launched by FAO*. Food and Agriculture Organization of the United Nations, Media Center; 2011. http://www.fao.org/news/story/en/item/80096/icode/ (accessed March 30, 2013).
9. Jain HK. *Green Revolution: History, Impact and Future*. Houston, TX: Studium Press; 2010.
10. Broderick N, Raffa K, Handelsman J. Midgut bacteria required for *Bacillus thuringiens is* insecticidal activity. *PNAS*. 2006;103(41):15196–15199.
11. Ye X, Al-BabiliS, Kloeti A, et al. Engineering the provitamin A (beta-carotene) biosynthesis pathway into (carotenoid-free) rice endosperm. *Science*. 2000;287(5451):303–305.
12. Scrimshaw N. Consequences of hunger for individuals and societies. *Fed Proc*. 1986;45:2421–2426.
13. Implications of Worldwide Population Growth For U. S. Security and Overseas Interests (The Kissinger Report), USAID. Washington, DC: USAID; 1974.
14. Corbett US Using Kissinger "Food as a Weapon" Plan to Starve North Korea. 2012. http://www.corbettreport.com/us-using-kissinger-food-as-a-weapon-plan-to-starve-north-korea (accessed March 30, 2013).
15. Mannar V, Gallego E. Iron fortification: country level experiences and lessons learned. *J Nutr*. 2002;132:856S–858S.
16. Howard M. When it rains it pours: endemic goiter, iodized salt, and David Murray Cowie MD. *Am J Publ Health*. 1987;77(2):219–229.
17. Castilllo-Landellotti C, Tur J, Uauy R. Impact of folic acid fortification of flour on neural tube defects: a systematic review. *Public Health Nutr*. 2012;31:1–11.
18. Carpenter KJ. A short history of nutritional science: part 4 (1945–1985). *J Nutr*. 2003;133(11):3331–3342.
19. Munro HN, Allison JB. *Mammalian Protein Metabolism*. I to IV. New York, NY: Academic Press, Inc.; 1964.
20. Waterlow JC, Garlick PJ, Millward DJ. *Protein Turnover in Mammalian Tissues and in the Whole Body*. New York, NY: Elsevier/North-Holland Biomedical

Press; 1978.
21. Wilmore DW. Rhoads lecture. The practice of clinical nutrition: how to prepare for the future. *JPEN J Parenter Enteral Nutr*. 1989;13(4):337–343.
22. Wilmore DW, Dudrick SJ. Growth and development of an infant receiving all nutrients exclusively by vein. *JAMA*. 1968;203(10):860–864.
23. Dudrick SJ, Wilmore DW, Vars HM, Rhoads JE. Long-term total parenteral nutrition with growth, development, and positive nitrogen balance. *Surgery*. 1968;64(1):134–142.
24. Zhu L, Baker SS, Gill C, et al. Characterization of gut microbiomes in nonalcoholic steatohepatitis (NASH) patients: a connection between endogenous alcohol and NASH. *Hepatology*. 2013;57(2):601–609.
25. Smith MI, Yatsunenko T, Manary MJ, et al. Gut microbiomes of Malawian twin pairs discordant for kwashiorkor. *Science*. 2013;339(6119):548–554.
26. Askanazi J, Weissman C, Rosenbaum SH, et al. Nutrition and the respiratory system. *Crit Care Med*. 1982;10(3):163–172.
27. Shikora SA, Benotti PN. Nutritional support of the mechanically ventilated patient. *Respir Care Clin N Am*. 1997;3(1):69–90.
28. Jolliet P, Pichard C. Immunonutrition in the critically ill. *Intensive Care Med*. 1999;25(6):631–633.
29. McCowen KC, Bistrian BR. Immunonutrition: problematic or problem solving? *Am J Clin Nutr*. 2003;77(4):764–770.
30. Galban C, Montejo JC, Mesejo A, et al. An immune-enhancing enteral diet reduces mortality rate and episodes of bacteremia in septic intensive care unit patients. *Crit Care Med*. 2000;28(3):643–648.
31. Kelly D, Wischmeyer PE. Role of L-glutamine in critical illness: new insights. *Curr Opin Clin Nutr Metab Care*. 2003;6(2):217–222.
32. Garrel D, Patenaude J, Nedelec B, et al. Decreased mortality and infectious morbidity in adult burn patients given enteral glutamine supplements: a prospective, controlled, randomized clinical trial. *Crit Care Med*. 2003;31(10):2444–2449.
33. Mayer K, Fegbeutel C, Hattar K, et al. Omega-3 vs. omega-6 lipid emulsions exert differential influence on neutrophils in septic shock patients: impact on plasma fatty acids and lipid mediator generation. *Intensive Care Med*. 2003;29(9):1472–1481.
34. Mayer K, Gokorsch S, Fegbeutel C, et al. Parenteral nutrition with fish oil modulates cytokine response in patients with sepsis. *Am J Respir Crit Care Med*. 2003;167(10):1321–1328.
35. Spear BA, Barlow SE, Ervin C, et al. Recommendations for treatment of child and adolescent overweight and obesity. *Pediatrics*. 2007;120(suppl 4):S254–S288.
36. Prevention, C.f.D.C.a. Basics about childhood obesity. April 27, 2012 [cited January 31, 2013].

Clinical Assessment of Nutritional Status

2

Babette S. Zemel, PhD, Asim Maqbool, MD, and Irene E. Olsen, PhD, RD

The clinical assessment of nutritional status is an integral part of patient care because nutritional status affects the body's response to illness. Attention to this is especially important in pediatric patients because the nutritional requirements to support normal growth and maturation are combined with those of coexisting medical illness. This combination creates for clinicians the unique challenge of estimating and meeting the nutritional needs of pediatric patients in the clinical setting as discussed throughout this chapter and book.

The assessment should allow for the early detection of both nutrient deficiencies and excesses. There is no single nutrition measurement that is optimal; therefore, a combination of different measures is required. Growth is an important indicator of health and nutritional status of a child and requires a growth chart for interpretation; a variety of growth charts are currently available and will be reviewed in this chapter. Each growth measurement performed needs to be accurate and obtained at age-appropriate intervals in order to identify at-risk patients or monitor a patient's clinical response to nutritional therapy.

During infancy, childhood, and adolescence, profound changes in growth and body composition occur. Therefore, clinicians must understand normal growth to recognize abnormal patterns and the nutritional changes that occur with acute and chronic disease. With the epidemic of pediatric

obesity, the proper identification of the overweight or obese patient is also important. A brief nutritional screening assessment may be used to identify patients in need of an in-depth assessment. A typical nutritional screening includes a brief medical and dietary history (including feeding ability), anthropometric measurements (e.g., weight, stature), and possibly laboratory data. A full nutritional assessment includes more detailed medical and dietary histories (including a measure of dietary intake), a complete physical examination, further anthropometric and body composition measurements, sexual and skeletal maturation, laboratory data, and the estimation of nutritional requirements. In addition to the clinician's global assessment of the child based on these objective data, his/her clinical judgment is important in determining the nutritional status.[1]

MEDICAL HISTORY

Medical history is a central component of the nutritional assessment. Past and present medical information, including the duration of the current illness, relevant symptoms, diagnostic tests and therapies (e.g., chemotherapy, radiation), and medications, is documented. Because nutritional abnormalities are often associated with certain disease states, it is essential to identify underlying medical conditions and the concomitant medication history. Medications can cause nutritional deficiencies (e.g., methotrexate as a competitive antagonist of folic acid metabolism) and drug–nutrient interactions (e.g., phenytoin and tube feedings; Table 2-1). Drug–nutrient interactions may occur between drugs (prescription and nonprescription) and foods, beverages, and dietary and vitamin/mineral supplements. Alterations in drug metabolism and absorption by food or pharmacologic interactions may be clinically significant.[2] Past medical history includes previous acute and chronic illness, hospitalizations, and surgical procedures. The history of past growth patterns (with previous growth charts, as possible), onset of puberty (for the child and other family members), and a developmental history (including feeding abilities) may also be included. Family history should include a medical history and the family's social and cultural background, especially as related to diet therapy and the use of alternative and complementary medicine. The review of systems includes oral motor function, dental development, and gastrointestinal symptoms such as vomiting, gastroesophageal reflux, diarrhea, and constipation.

TABLE 2-1 Examples of Some Common Drug–Nutrient Interactions

Drug	Nutrient
Amphotericin B	Hypokalemia, hypomagnesemia
Antacids	Vitamin D and iron deficiency, hypophosphatemia
Phenobarbital	Vitamin D deficiency
Cholestyramine	Vitamin A, D, E, and K malabsorption
Cyclosporin	Elevated triglycerides, hypokalemia, hypomagnesemia
H2 blockers	Iron deficiency
Methotrexate	Folate deficiency
Phenytoin	Folate deficiency
Corticosteroids	Hyperglycemia, hypophosphatemia
Sucralfate	Hypophosphatemia
Sulfasalazine	Folate deficiency
Trimethoprim	Folate deficiency
6-Mercaptopurine	Purine metabolism (DNA synthesis/repair)

PHYSICAL EXAMINATION

Physical examination includes anthropometrics (see later), including weight, length or stature, head circumference, and upper arm measures. The frequency of measurements of well children follows the recommendations of the American Academy of Pediatrics (AAP).[3] For the preterm infant, weight should be measured daily, and length and head circumference weekly. For newborns, weight, length, and head circumference should be measured at birth and again at 3–5 days of age. For healthy children up to 2 months of age, all three measures should be obtained monthly, then every two months from 2–6 months of age, and every 3 months from 6–24 months of age. For children aged 24–36 months, weight and length/height should be assessed every 6 months and annually thereafter. The pattern of measurement for hospitalized patients depends on the age of the patient, illness, and the degree of nutritional intervention. Nutritional assessments for patients with complex chronic disease states should be conducted every one to two months and less often in those with milder disease (every 6–12 months). The general physical examination includes an assessment of the patient's general condition and close examination of skin, hair, and teeth (see Appendix Table I-8). This includes an assessment for pallor, clinical assessment of body fat stores,

wasting of muscle mass, edema, rash, thinning of hair, and evidence of specific nutritional deficiencies. Examples of specific signs include the flag sign or the loss of hair color associated with a period of malnutrition followed by recovery with a return of normal hair color and texture. Examination of specific organ systems and obtaining medical record information is helpful in assessing the severity of the underlying disease process. It is also important to consider the clinician's clinical judgment in the assessment of nutritional status.[1] Documenting sexual development by Tanner staging (Appendix I) is a routine part of the nutritional assessment of adolescents (see later). For a summary of signs and symptoms of specific nutritional abnormalities, see Appendix Table I-8.

DIETARY HISTORY

The dietary history is an essential component of the nutritional assessment. The dietary history provides information not only on the amount and quality of the food consumed but also on the eating patterns and behaviors of the family. This part of the nutritional assessment should detail the number of meals, snacks, and beverages consumed; special foods eaten by the child and family; vitamin and mineral supplements ingested regularly; food allergies; intolerances; and unusual feeding behaviors. The child and family are asked about psychosocial factors that impact on food selection and intake, including family history, socioeconomic status, and use of the Special Supplemental Nutrition Program for Women, Infants, and Children and supplemental food programs, parent/caretaker's perception of the child's nutritional status, and religious and cultural considerations. Food-related factors may affect dietary intake and include food allergies, intolerances, self-imposed and prescribed diets, and feeding skills. These factors are also noted in the assessment.

The assessment of dietary intake of breastfed infants is more difficult because the volume of milk consumed cannot be directly measured. An estimate is obtained by weighing the infant before and after feeds and using a conversion factor of 1 mL volume of breast milk consumed for each gram of weight gained. In formula-fed infants, the clinician should inquire about both the amount and type of formula consumed and the details of the method of preparation (concentrates, powders, modular additives).

The quantity and quality of dietary intake are assessed by prospective food records (with weighed or estimated food portions), retrospective 24-hour recalls (previous 24 hours or of a "typical" 24-hour period), or food

frequency questionnaires.[3] The prospective food records are usually carried out for three to seven days (including a combination of weekend and weekdays) and provide the most accurate assessment of actual intake. However, food records are used most often in the research setting because they are labor intensive and time consuming. As available, these records are analyzed and compared to the dietary reference intakes (DRIs) (see later) using a computerized nutrient analysis program. A limitation of food records is that parents tend to forget to record all foods eaten or modify feeding practices to be more healthy, which may lead to incorrect estimates of intake.[4] The retrospective 24-hour diet recall provides a quicker assessment of dietary intake. For a 24-hour recall, the child/parent is asked to recall what and how much the child ate and drank over the past 24 hours. Recall accuracy depends on the child/parent's memory and their ability to estimate portion sizes. Also, because this is only one day of intake, it may not be representative of the usual intake. When the child's intake is affected by acute illness, a 24-hour recall of a "typical day" is more useful to estimate usual intake. The 24-hour recall tends to underestimate usual energy intake yet may overreport the intake of infants and toddlers.[5]

Another way of assessing dietary intake is the food frequency questionnaire method. These questionnaires collect information on both the frequency and the amount consumed of specific foods and are useful in the clinical setting to identify usual eating patterns. A limitation of the food frequency questionnaires is that the amounts of food and thus intake of energy and some nutrients are often overreported.[5] All of these methods of dietary assessment are somewhat limited owing to gaps in the nutrient databases, which lack information about bioavailability, presence of inhibitors and enhancers of absorption, and nutrient availability of specific nutrients of interest.[6]

The most commonly used method of dietary assessment in hospitalized patients is the calorie count. This is a variation of the prospective food records as the amount of food consumed from a known quantity of food (as specified by a menu or list) is recorded. The accuracy of the calorie count assessments is limited by the number of individuals required for the completion of these forms throughout a 24-hour period (e.g., the dietitian, the nurse for each shift, the child's family, the child). However, calorie counts are a useful part of nutritional assessment follow-up because these provide a rough assessment of the patient's appetite, intake, and compliance with nutrition recommendations.

ANTHROPOMETRICS AND BODY COMPOSITION

At a minimum, nutritional assessment of a child includes a measured weight, length or height, and head circumference (birth to 3 years of age), and these measurements are followed up over time to assess short- and long-term growth and nutritional status. For children with chronic disease, a mid-upper arm circumference (MUAC) and triceps skinfold (TSF) thickness are also part of the assessment to determine body fat and protein stores. In addition, a dual energy x-ray absorptiometry (DXA) scan may be added to more thoroughly assess body composition (percent fat, lean body, and fat mass) and bone mineral density (BMD) (see Chapter 4).

Accurate and reliable anthropometric and body composition measurements require proper equipment and techniques. Training and practice in anthropometric technique cannot be overemphasized. For all growth measures, three independent measures should be obtained. The current convention is to use the average of three measurements. The clinician's assessment for a child depends on the quality of these data. Equipment requirements for each measure are discussed below.

Weight

Weight is a measure of overall nutritional status. Optimal interpretation requires additional information regarding the age, sex, and height/length of the child. Weight is determined using a digital or beam balance scale. Until the child is approximately 24 months or can cooperate and stand independently, a pan version of the scale is used. Weight should be measured in light or no clothing and without a diaper for infants. It is important that the scale is zeroed prior to each measurement and is calibrated using known weights at least monthly or on movement of the scale.[3] Weights are recorded to the nearest 0.01 kg in infants and 0.1 kg in older children.

Stature: Length or Height

A measure of stature is important for monitoring long-term nutritional status. Recumbent length is measured using a length board for children from birth to 2 years. The measurement of length requires two individuals. The first person positions the infant straight on the board so that the infant's head is against the headboard and positioned with the Frankfort plane parallel to the headboard.[3] The Frankfort plane is the alignment from the lower margin of the orbit to the upper margin of the auditory meatus as

shown in Figure 2-1. The second person extends the infant's legs with the knees flattened and adjusts the moveable footboard such that the heels lie flat against the movable footboard.[3] For children aged 2 years and older who are able to stand independently and cooperate, height is measured using a stadiometer, with a moveable headboard at a fixed 90-degree angle to the back of the stadiometer. The child is measured barefoot or in thin socks and in minimal clothing to allow the observer to check for correct positioning. For the measurement, the child stands erect, with feet close together, heels, buttocks, and back of head touching the stadiometer, and looking ahead with the Frankfort plane parallel to the floor.[3] Asking the child to inhale deeply just prior to obtaining the measurement assists in attaining fully erect posture. Because length overestimates the height by 0.7 cm,[7] it is essential to record the method of measurement during the transition from recumbent length to standing height. The change to standing height is also accompanied by the transition to pediatric (2–18 years) growth charts (see later). Both length and height measurements are recorded to the nearest 0.1 cm.

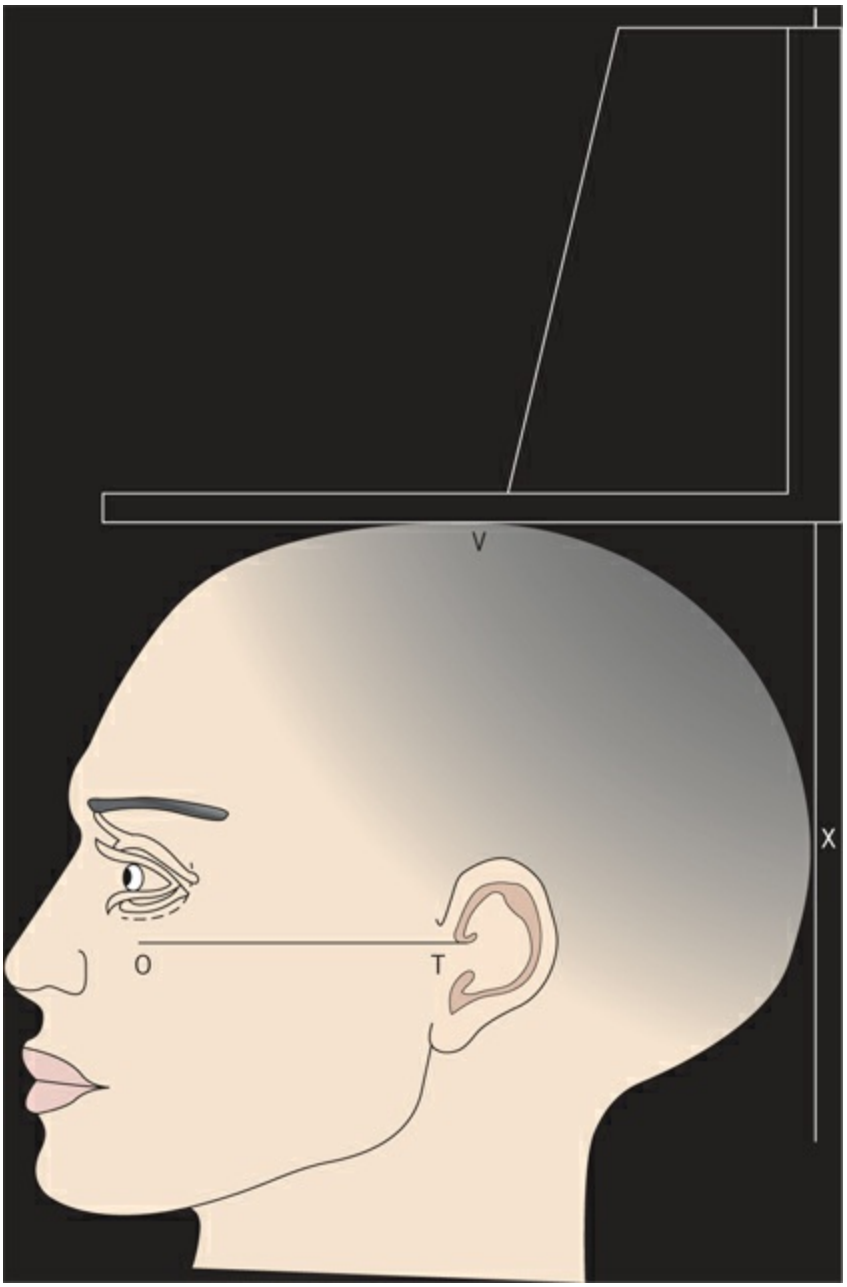


Figure 2-1 The Frankfort plane is the horizontal line extending from the auditory meatus (T) to the lower margin of the orbit (O). Reprinted with permission from Anthropometry Illustrated, Roww WD, Carr RV, and Carter JEL, Turnpike Electronic Publications, Inc.

Among children for whom the stature measurements are not possible owing to physical constraints (e.g., contractures, nonambulatory), alternative measures are available. Upper arm and lower leg lengths provide reliable and valid indexes of stature in children.[8,9] These

measurements are conducted using sliding calipers. All measurements are recorded to the nearest 0.1 cm.

The shoulder to elbow length is used for the upper arm measurement in infants and young children (birth to 24 months).[8,9] This measurement is obtained with the arm bent to a 90-degree angle, and the measurement is taken from the superior lateral surface of the acromion to the inferior surface of the elbow. For older children (2–18 years), the arm should hang in a relaxed position at the side, and the distance between the superior lateral surface of the acromion and the proximal tip of the radius is measured. The lower leg length is measured as the knee-to-heel length for infants and young children (0–24 months) and as a tibia length in older children (2–18 years). For infants and young children, the superior surface of the knee to the heel is measured while the leg is bent at a right angle at the hip, knee, and ankle. In older children, the tibia length is measured from the proximal medial border of the tibia plateau to the medial malleolus; ideally, the leg being measured is crossed over the opposite knee.[3] Alternatively, knee-to-heel length can be measured in older children and used to predict stature.[10,11] The right-side extremity should be measured for these alternative stature estimates.[3] If there are asymmetric extremity abnormalities, the measurement should be taken on the least affected side and the side of measurement noted in the chart.

Head Circumference

Head growth, primarily owing to brain development, is most rapid during the first 3 years of life. Routine measurement of head circumference (the frontal occipital circumference) is a component of the nutritional assessment in children up to age 3 and longer in children who are at high nutritional risk. Head circumference is a less-sensitive indicator of short-term nutritional status than weight and height because brain growth is generally preserved in cases of nutritional stress. Head circumference is not a helpful nutritional status measure in children with hydrocephalus, microcephaly, or macrocephaly.

Head circumference is measured using a flexible, nonstretch tape measure. The circumference should be taken at the maximum distance around the head, which is found by placing the measuring tape above the supraorbital ridge and extending around the occiput.[3] Care should be taken to keep the tape measure flat against the head and parallel on both sides. The measurements should be recorded to the nearest 0.1 cm.

GROWTH CHARTS AND TABLES

Serial measurements are essential for the optimal assessment of short- and long-term growth and nutritional status. A number of growth charts and tables are available for the comparison of weight, stature, and head circumference with reference populations by age and sex. Weight is also assessed relative to a child's stature using growth charts that evaluate weight for length/height, weight/height[2] (body mass index [BMI]) for age, or weight/length[3] (ponderal index) for age; these measures assess "relative weight" or weight independent of stature. The types of charts and tables available for clinical assessment in infants and children are reviewed. Several of these charts are included in Appendix I.

Premature Infant Growth Charts

Several types of charts are available for growth assessment of premature infants in the neonatal intensive care unit (NICU). Intrauterine growth charts[12–15] are preferred over postnatal growth charts[16–19] as the pattern and rate of normal intrauterine growth are the standard for growth of premature infants.[20] Postnatal growth charts are based on actual, not ideal, growth of premature infants over time so that these can serve as a useful adjunct curve in a nutritional assessment. Growth measurements are plotted using corrected gestational age of 12–36 months, depending on the child's size and growth.

The three most widely used intrauterine growth charts in US NICUs include the Lubchenco,[12,13] Fenton,[15] and Olsen[14] charts. All include weight-, length-, and head circumference-for-age curves. The newest and first gender-specific set of charts from Olsen et al.[14] are becoming a clinical standard of care as these were based on more recent data (1998–2006) and a large sample of birth data that represents the racial distribution of US births. The Fenton unisex growth charts[15] have a popular "hybrid" curve presentation that joined intrauterine data with postnatal data and span from 22–50 weeks gestational age. However, the intrauterine portions of these curves were created from mixed sources of non-US data: Canadian data[21] were used for weight-for-age, and a combination of Swedish data[22] and Australian data[23] were used for the length- and head circumference-for-age curves. The postnatal portion of the Fenton curves used the Centers for Disease Control and Prevention (CDC) 2000 data[24] and later the World Health Organization (WHO) data.[25] Finally, the Lubchenco growth charts,[12,13] while historically a NICU standard of care,

are now less representative of the growth of infants in the US NICUs. However, this set of charts has the advantage of including a measure of relative weight (ponderal index-for-age) that may be helpful in the growth assessment of premature infants (see "Weight-for-Length/Height" section).

The Olsen curves define new cutoffs for the classification of infants as small-for-gestational age (or <10th percentile) or large-for-gestational age (>90th percentile), and the cutoffs vary from existing curves used in NICUs. For example, the Olsen curves tend to have a lower average size than the Lubchenco curves at younger gestational ages and a higher average size at the older gestational ages; differences from the Fenton curves vary based on age, gender, and growth measurement.[14] The relationship between these newly defined high-risk groups and health outcomes is under investigation.

Once a preterm infant reaches 40 weeks corrected gestational age, their growth should be monitored on the WHO growth charts[25] until 2 years of age and then switched to the CDC 2000 growth charts[24] for 2–20 years of age as recommended by the CDC for infants born at full term. Former premature infants are plotted on these charts based on their corrected gestational age (as mentioned earlier). Although all premature infants may not achieve "good" placement on these growth charts, these charts provide an appropriate goal for growth. Also available as adjunct assessment tools are two series of postnatal growth charts: the Infant Health and Development Program charts for low-birth-weight (LBW, 1501–2500 g) and very low-birth-weight (VLBW, <1500 g) premature infants for boys[16,17] and girls[18,19] and the National Institute of Child Health and Human Development Neonatal Research Network Growth Observational Study[26] projected growth charts and tables for VLBW infants (available online at http://neonatal.rti.org). These charts provide a comparison of how a LBW or VLBW premature infant grows relative to two reference populations of similar infants. Although these postnatal growth charts are based on older data, more recent charts do not exist. As noted earlier, postnatal charts represent actual, not ideal, patterns of growth for former premature infants and may be used in conjunction with but not in place of the WHO growth charts.

CDC, NCHS, and WHO Growth Charts

2000 CDC Growth Charts

In 2000, the CDC and National Center for Health Statistics (NCHS) released an updated set of growth charts called the CDC growth charts (see

Appendix I, Figures I-9 to I-18).[24] These charts are available for boys and girls aged 0–36 months for weight, length, and head circumference by age and weight for length, and aged 2–20 years for weight, height, and BMI for age and weight for height. Further details on the CDC growth charts, including data exclusions, are reviewed in the 2000 CDC report.[24] The CDC growth charts are available on the Internet (www.cdc.gov/growthcharts).

2006 WHO Growth Charts

The WHO undertook the multicenter growth reference study to generate international growth reference standards based on a large healthy sample of infants and children. Data were gathered from Brazil, Ghana, India, Norway, Oman, and the United States. The sample was selected to reflect healthy children living under conditions (breastfed, nonsmoking environments) favorable for fully achieving growth according to their growth potential. The data combined a longitudinal follow-up from birth to 24 months as well as a cross-sectional survey of the children between birth and 71 months. The growth standards derived are prescriptive rather than descriptive; that is, they represent how children should grow as opposed to how they grew at a particular time and place, thereby underscoring that the new growth charts are consistent with growth outcomes occurring under the best health practices. The 2006 WHO reference standards include weight for age, length/height for age, weight for length/height, and BMI by age for children from birth to 60 months.[3] The CDC recommends the use of these charts to monitor the growth of infants and children (breast and bottle, or infant formula fed), birth to 2 years of age, and the use of the CDC 2000 growth charts for children aged 2–20 years.[27]

When comparing the 2000 CDC growth charts to the 2006 WHO growth charts, several differences are notable. The CDC growth charts used a population-based sample that included all children regardless of health status or feeding mode. On average, the CDC sample was heavier and shorter children than that of the WHO growth study. For this reason, the 5th–95th percentile is considered as the normal range. In contrast, the WHO growth charts are based upon a healthy population of breastfed infants, and two standard deviations above and below the median is considered as the normal range. Generally, the recommended cutoffs for these two respective growth charts, the WHO (two standard deviations from the mean for age, or the 2.3rd percentile) and CDC cutoff values (5th and 95th percentiles), are fairly similar when screening for shortness and overweight in the first two years of life,[28] although some age-specific

differences occur. For example, the use of the WHO growth charts results in a higher prevalence of underweight during the first 6 months of life and a lower prevalence thereafter.[29] Another difference between the charts is that the WHO weight for length curves span from 45 to 110 cm, whereas the CDC weight for length curves span from 45 to 103 cm. The extended range for length in the WHO weight for length curves facilitates the assessment of tall children who are unable to stand (for whatever reason). As a consequence of these differences in study design, selection of the sample, and resulting growth curves, slightly fewer US children will be identified as underweight using the WHO charts, and the slower growth among breastfed infants during ages 3–18 months will appear within the normal range. Finally, gaining weight more rapidly than indicated on the WHO charts might signal early development of overweight.[27]

In summary, there are age-specific differences in the WHO and CDC growth charts owing to differences in the study design and the samples used to generate the reference ranges.[29] Accordingly, the cutoffs used to identify children at risk of nutritional complications are different for these two sets of growth charts. The WHO charts are recommended for growth assessment up to the age of 2 years, and the CDC 2000 charts are recommended for children aged 2–20 years. These growth curves are included in Appendix I and Figures I-1–I-18.

Incremental Growth Velocity

Reference data are also available for incremental growth velocity for boys and girls for weight, stature, and head circumference from 0 to 18 years (see Appendix I and Tables I-4 and I-5).[30–35] These data are presented according to the time interval between measurements (1, 3, or 6 months) and are used to evaluate growth velocity percentile (3rd–97th or 5th–95th percentiles). New incremental growth standards for breastfed infants from the WHO are available for weight, length, and head circumference.[35] These detailed tables offer weight increments (grams) and velocity (grams per day) by birth weight group for the first 60 days of life, weight increments (grams) in 1-, 2-, 3-, 4-, and 6-month time intervals, and length (centimeters) and head circumference (centimeters) increments over 2-, 3-, 4-, and 6-month intervals up to 24 months of age. Because of the rapidly changing rate of growth during the first 2 years of life, the presentation by time intervals between measurements allows for more accurate and convenient assessment. Calculating growth velocity and comparing it to standards based on a larger or smaller measurement interval can yield

inaccurate assessment. Downloadable tables can be found at http://www.who.int/childgrowth/standards/en/.

In clinical practice, the incremental tables for weight, length, and head circumference[30,33–34] are helpful in the assessment of former premature infants and other children with growth failure from any cause. The growth increments are easily divided into daily, weekly, or monthly weight gain goals. Assessment of incremental growth is more sensitive in detecting growth faltering or catch-up growth than the use of growth charts that assess static growth status at one point in time based on cross-sectional data. However, incremental growth measurements are extremely sensitive to measurement error, so attention to measurement technique is required.

Growth charts for the assessment of height and height velocity in relation to the stage of sexual maturity based on US reference data are also available (see Appendix I and Figures I-23 and I-24).[36] These charts provide height growth for early, middle, and late maturers by sex and age at which peak height velocity was reached and explain some of the variation in growth related to different stages of puberty. The height velocity charts are often used in the care of children with poor growth and chronic illnesses.

Special Growth Charts

Although the WHO and CDC growth charts are recommended for the growth and nutritional assessment of all children, a number of disease-specific charts have been published (e.g., achondroplasia, Brachmann-de Lange syndrome, cerebral palsy, Down syndrome, Marfan syndrome, myelomeningocele, Noonan's syndrome, Prader–Willi syndrome, sickle cell disease, Silver–Russell syndrome, Turner's syndrome, Williams syndrome; see Appendix I and Table I-7). The weight-for-age and height-for-age growth charts for boys and girls with Down syndrome of ages 0–36 months and 2–18 years were published in the 1980s.[37,38] However, due to improvements in care of children with Down syndrome, these charts no longer represent the growth of contemporary children. Until new growth charts become available, the AAP recommends using the growth charts for typical children to monitor growth in children with Down syndrome.[39] Many other disease- or syndrome-specific growth charts are based on small samples of children and include children with suboptimal nutritional status. Disease-specific charts may be helpful to use in conjunction with the WHO or CDC growth charts for comparison to "peers."

A set of growth charts is also available for the assessment of alternate

stature measures,[8,9] including upper arm length (infants, 0–24 months; girls, 3–16 years; boys 3–18 years) and lower leg length (infants, 0–24 months; girls, 3–16 years; boys, 3–18 years). Similar to other measures of stature, these linear growth measures are used along with weight to help determine a child's nutritional status.

ASSESSMENT OF ANTHROPOMETRICS

Nutritional status indices are essential for the clinical interpretation of growth measurements. Every nutritional assessment requires one or more of the following indices for interpretation.

Percentiles for Age and Sex

When each growth measure is plotted on a growth chart, a percentile or rank of the individual compared to the reference population is determined. For example, the 25th percentile weight for age means that the individual weighs the same or more than 25% of the reference population of the same age and sex, and the 75th percentile weight for age means that the individual weighs the same or more than 75% of the reference population of the same age and sex.[40] Percentiles are easily interpreted and used clinically. Available growth charts provide reference growth of children ranging from the 5th to 95th percentiles or the 3rd to 97th percentiles, depending on the chart. In clinical practice, the WHO and CDC recommended cutoffs should be used for screening and follow-up of healthy children. Weight-for-age and height-for-age percentiles are also used to screen for malnutrition using published classifications (see Appendix I and Table I-6). BMI percentiles are used to identify patients at risk for under- or overnutrition; however, the Waterlow criteria are still used by many practitioners. Height-for-age is an indicator of long-term nutritional status, with low height-for-age reflecting stunting. Height-for-age is generally interpreted as short (<5th percentile), normal (5th–95th percentile), and tall (>95th percentile) for children aged 2–20.

Genetic Growth Potential: Midparental Height

In the assessment of a child's stature, it is helpful to estimate the genetic potential for stature as determined by the biologic parents' adult height.[41] This is important to distinguish whether the child is achieving their genetic potential for growth based on family genetic background or whether disease and/or poor nutrition might be affecting their growth. An

adjustment for parental height is used for a child's length (0–36 months) or height (3–18 years) and is based on the mean of the height of both biological parents. This allows adjustments to the child's stature for tall or short parents. The corrections are based on the Fels Institute and older NCHS data.[42] Parental height adjustment is appropriate for use with most of the parents and children in the United States; however, it should not be used when the parents do not meet their genetic potential for height (e.g., in situations of poor health and/or nutritional status during the parental childhood or adolescence).[41]

Weight for Length/Height

Weight relative to length/height provides information on growth and nutritional status that is different from weight-for-age or length/height-for-age alone; it helps to determine and classify nutritional status in the individual patient.[43] Although there is no routinely used measure of relative weight in preterm infants, there is evidence to suggest that there should be.[44] For children aged 0–2 years, the CDC recommends that weight-for-height is assessed by determining a percentile on the WHO growth charts.[25] Weight-for-height is generally interpreted as underweight (<5th percentile using the CDC charts; <3rd percentile using the WHO charts), within normal variation (5th–95th percentile using the CDC charts; 3rd–97th percentile using the WHO charts), and overweight (>95th percentile using the CDC charts; >97th percentile using the WHO charts) and is used in screening healthy children. Weight-for-height measures are also used for screening classification of protein–calorie malnutrition (see Appendix I and Table I-6).

BMI is another measure of weight relative to height. The CDC growth charts provide BMI for age and sex from age 2 to 20 years[24,45] and is the recommended assessment tool for children aged 2 years and older. The WHO growth charts also include infant BMI graphs; however, these are not widely used; the WHO BMI-for-age curves range from birth to 2 years of age and 2–5 years of age. Because both weight and height in children change over time, there is no fixed BMI value for the diagnosis of obesity in children as there is for adults (e.g., BMI ≥ 30). The BMI percentile for age must be used for interpretation. In the United States, the 85th and 95th BMI percentiles for age and sex are used to define "overweight" and "obesity" in children.[3] A comparison of BMI-for-age and weight-for-height to predict underweight and overweight in children and adolescents (2–19 years) showed good agreement, especially in young children (2–5

years of age), but BMI had the greatest sensitivity to detect overweight and obesity across the entire age range.[46] From the International Obesity Task Force, cutoff points for BMI to define overweight and obesity in children based on cross-sectional growth studies from six countries (Brazil, Great Britain, Hong Kong, the Netherlands, Singapore, the United States) are also available.[47]

Differences in BMI at the lower percentiles of the reference range largely reflect lean mass, whereas at upper percentiles of BMI, differences are largely due to fat mass.[48] BMI is not a perfect measure of adiposity, since height is not fully independent of weight in children. Stature has been postulated to affect BMI, and in populations with taller stature, BMI may overestimate the prevalence of obesity.[49] One study comparing techniques to evaluate adiposity (BMI, isotopic dilution, skinfold equations) in stunted and nonstunted children concluded that BMI predicted percent body fat in both groups of children.[50] An additional study showed that the relationship between BMI and adiposity varied across different populations.[51] BMI should be interpreted with caution in certain clinical conditions including edema, pregnancy, in individuals with a high tumor burden, and in individuals with increased lean body mass such as in well-developed athletes. BMI is a screening tool to identify children and adolescents at risk for overweight or obesity, and further assessments to determine adiposity should be considered as clinically indicated.

Percent Ideal Body Weight

Historically, percent of ideal body weight (IBW) was used to classify the degree of over- or undernutrition into clinical classification: >120% IBW as obese; 110%–120% IBW as overweight; 90%–110% IBW as normal range; 80%–90% IBW as mild wasting; 70%–80% IBW as moderate wasting; and <70% IBW as severe wasting. IBW is also used as a clinical weight goal in the nutritional rehabilitation of a child.

Percent IBW should be used with caution. A recent study[52] compared the use of BMI percentiles and percent IBW to screen for malnutrition in children with cystic fibrosis using data from the cystic fibrosis patient registry. Among children with stature between 25th and 75th percentiles, both percent IBW and BMI yielded similar estimates of ideal weights, and in the classification of malnutrition. However, compared with the BMI, percent IBW underestimated the severity of malnutrition in children with short stature (height <25th percentile), whereas the opposite trend was

found for the subjects with tall stature (height >75th percentile). These findings support the use of BMI in lieu of percent IBW for the classification of pediatric malnutrition, especially for children aged 2 years and older. Currently, BMI charts are preferred in the classification of overweight and obesity. The use of the BMI charts for classification of degrees undernutrition is not as well established. The WHO uses weight-for-height cutoffs of <–2SD for moderate undernutrition and <–3SD for severe undernutrition[53] and has proved to be an effective screen for identifying children at risk of nutrition-related morbidity and mortality.[54]

Percent Weight Loss

The percent of usual body weight loss is an important clinical indicator of nutritional status and nutritional risk. Percent weight loss is calculated as [(previous weight – current weight)/previous weight × 100]. A 5% or greater weight loss in one month may be considered an indicator of nutritional risk in children. However, it is important to also look at the absolute weight loss as opposed to the percent weight loss alone, as this may be additionally informative and clinically meaningful. For obese children, change in BMI is recommended for monitoring weight management.[55]

Growth Velocity

Growth velocity (change in the growth parameter over time) is useful to detect a change in nutritional status and to monitor the effectiveness of nutritional and medical therapy. As discussed earlier, age- and sex-specific charts and tables are available for the evaluation of weight, stature, and head circumference growth over time.[7,30–34,36]

Body Composition

In children with many acute and chronic diseases, the nutritional assessment requires measurement of body composition (body fat and protein stores) in addition to weight, stature, and head circumference (see Chapter 4) for a detailed discussion of this topic). The measures described below also are useful to perform in critically ill children for whom weight and length/height measurements cannot be performed easily, and in those conditions with significant fluid shifts and fluid retention.

Mid-Upper Arm Circumference

MUAC can be used as a measurement of growth, an index of energy and

protein stores and can provide information on fat patterning.[3] The measurement is taken at the midpoint of the upper arm, located halfway between the lateral tip of the acromion and the olecranon when the arm is flexed at a 90-degree angle (measured and marked). For the MUAC measurement, the child should be upright with the arm relaxed by the side. A flexible, nonstretchable measuring tape is placed perpendicular to the long axis of the arm. The measuring tape should encircle the arm without compressing the soft tissue, and MUAC should be recorded to the nearest 0.1 cm.[3] This measurement should be taken in triplicate and used as an average.

Triceps Skinfold Thickness

The TSF thickness is an indicator of subcutaneous fat (energy) stores and total body fat and provides information on fat patterning.[3] For the TSF measurement, the child should be upright with the arm relaxed at the side. The TSF thickness is measured at the midpoint of the upper arm (defined earlier) over the center of the triceps muscle on the back of the arm (measured and marked beforehand). The anthropometrist lifts the skinfold with the thumb and index finger, approximately 1 cm above the marked midpoint, and places the calipers at the marked point. Two to three seconds after the handles of the calipers are released, the measurement is taken and the calipers are removed. This measurement should be taken in triplicate, used as an average, and recorded to the nearest 0.1 cm.[3]

Reference data (age and sex specific) are available for the assessment of mid-upper arm circumference (MAC) and TSF thickness[56] (see Appendix I, Figures I-34, I-35, I-38, and I-39, and Tables I-1 and I-2). The MAC and TSF measurements are used to calculate upper arm muscle area and fat area.[56,57] These are clinical indicators of total body stores of muscle and fat.

$$\text{Upper arm muscle area (cm}^2\text{)} = [\text{MUAC (cm)} - (\text{TSF (cm)} \times \pi)]^2/(4 \times \pi),$$
where $\pi = 3.14$.

$$\text{Upper arm fat area (cm}^2\text{)} = \text{upper arm area (cm}^2\text{)} - \text{upper arm muscle area (cm}^2\text{)},$$
where upper arm area $(\text{cm}^2) = \text{MUAC}^2/(4 \times \pi)$.

Dual Energy X-Ray Absorptiometry

DXA is a noninvasive, rapid, indirect, low-radiation measurement that is used to assess BMD. Bone fragility is of a concern in a variety of pediatric disorders associated with altered bone mineral acquisition or increased fracture risk. A small number of pediatric disorders involve primary bone diseases, such as osteogenesis imperfecta or idiopathic juvenile osteoporosis, where the disorder directly targets bone formation. Secondary bone disorders involve diseases that affect the skeleton through other disease mechanisms such as inflammation, malabsorption, low physical activity, endocrine disturbances, hematological disorders, and poor dietary intake, or disease treatments such as glucocorticoid therapy.[58]

DXA scans can be performed on the lumbar spine, proximal and distal femur, forearm, and whole body. The recommended sites for children and adolescents are the lumbar spine and whole body; when these measurement sites are not feasible, the distal forearm and proximal femur should be considered. For older adolescents, the proximal femur may be a useful measure to obtain for long-term follow-up, since the lumbar spine and proximal femur are the preferred sites in adults.[59] DXA scans provide information on bone mineral content (BMC) in grams per centimeter or areal BMD (aBMD) in grams per square centimeter in different skeletal regions or the whole body. Although primarily used for the assessment of bone health, whole-body scans also provide body composition measures of fat-free mass, fat mass, and percent body fat.[60] Smoothed percentiles for percent body fat assessed by DXA are now available for children aged 8–19 years.[61]

As with other growth-related outcomes, BMC and aBMD values used to assess bone health must be compared with the reference data in healthy age- and sex-matched children and adolescents to interpret bone health status. The results for BMC and aBMD are expressed as a Z-score (standard deviation score), determined by the comparison of an individual child's BMC or aBMD with the reference database. A Z-score of 0 is the median (similar to the 50th percentile on a growth chart) for the reference data, with +1, +2, –1, and –2 representing plus and minus 1 and 2 standard deviations from the reference median. The International Society of Clinical Densitometry[62] defines low BMC or aBMD as a Z-score ≤ −2, adjusted for age, sex, and body size, as appropriate. Osteoporosis in children cannot be made solely on the basis of low BMC or aBMD. Osteoporosis is defined as low BMC or aBMD in the presence of a clinically significant fracture history. A "clinically significant fracture history" consists of a long bone fracture of the lower extremities, a vertebral compression fracture, or two

or more long bone fractures of the upper extremities.[62]

Recently, the ability to assess bone health in children aged 5–20 years has been improved by the availability of reference ranges based on a large multicenter, multiethnic sample in the United States from the National Institutes of Health Bone Mineral Density in Childhood Study.[63] Sex- and age-specific reference ranges are provided for African American and non-African American for BMC and aBMD of the total body, total body less head, lumbar spine, proximal femur, femoral neck, and radius. African American children and adults have higher bone density than other ethnic groups.[64,65] Age, sex, and ancestry group reference ranges for the lateral distal femur are also available[66]; this measurement is particularly helpful for children with metal implants or contractures for whom traditional measurement sites are not feasible. The use of ancestry group-specific reference ranges (African American vs. non-African American) is used as an indicator of whether a child is reaching their genetic potential for BMC or aBMD. As recommended by the International Society for Clinical Densitometry, adjustment of BMC/aBMD values for growth in children is preferred since DXA measurements are influenced by growth status. Several techniques have been proposed.[67,68] The least biased technique requires the calculation of height-for-age Z-score, and the BMC/aBMD adjustment is based on whether a child is tall for age or short for age.[69]

The advantages of the DXA are the low radiation exposure, fast scan time, and noninvasive nature. The precision of the instrument is excellent. The radiation dose is small (<1 mrem, Hologic Delphi Clinical Bone Densitometer product specifications) or less than that received during a standard airline flight across the United States.[70] A limitation of DXA in the assessment of body composition is that it provides a two-dimensional assessment of three-dimensional structures and is unable to distinguish between cortical and trabecular bone. However, it is the most widely used clinical technique for bone health assessment. The timing of initial DXA scans and frequency of follow-up measurements depends on the clinical needs, particularly the risk factors for poor bone acquisition, disease-specific patterns of bone loss, and medical therapies.[58] Patients with poor BMD measurements are rarely measured less than six months after the baseline assessment. Bone health in children and adults is altered by weight-bearing physical activity and intake/absorption of calcium and vitamin D. Other nutrients such as protein, vitamin K, magnesium, and zinc[71–73] have also been implicated in bone health.

Cortical and trabecular bone can be measured using quantitative

computed tomography (QCT). This technique describes volumetric BMD (grams per cubic centimeter) and also differentiates between cortical and trabecular bone. Specialized peripheral QCT and high-resolution peripheral QCT scanners have been developed to decrease radiation exposure for the peripheral skeleton, and the measure derived indices of bone structure and strength for the research setting.[74] Although peripheral quantitative computed tomography (pQCT) has provided insights into the nature of bone deficits in the children with chronic disease, lack of standardization of measurement sites and inadequate pediatric reference data limit the use of these techniques in clinical care.[75]

Air Displacement Plethysmography

Air displacement plethysmography method of body composition assessment is safe, noninvasive, and fast. There are two commercially available plethysmographs from Life Measurement, Inc. (Concord, CA): the BOD POD® for use in children and adults[76] and the PEA POD® for use in infants up to 6 months of age.[77,78] A pediatric option for the BOD POD now permits assessment of children aged 2–5 years.[79] The method requires the subject to sit (BOD POD) or lie (PEA POD) within a closed chamber in minimal, close fitting, or no clothing. The subject's total body volume is estimated indirectly by measuring the volume of air that he/she displaces inside an enclosed chamber.[76] This volume is used to compute body density (body mass or weight/body volume) and in turn fat mass (kilogram), fat-free mass (kilogram), and percent body fat. Overall, the BOD POD[76,79] and PEA POD[77] provide reliable and valid estimates of body composition; however, neither device is widely available in the clinical setting at this time.

Sexual and Skeletal Maturation

The tempo of growth and body composition changes are influenced by sexual/pubertal and skeletal maturation. Many factors, such as genetics and disease processes, can affect sexual and skeletal maturation. Overnutrition and undernutrition can also affect the tempo of maturation and are therefore important in the nutritional assessment of an individual patient and their growth potential. The physically immature child with likely growth delay has the potential to catch up to the size of his/her peers once maturity advances.

Sexual maturity is assessed using the Tanner staging system by the clinician physical examination[80] or as a pubertal self-assessment form

completed by the child/parent.[81–84] Pubic hair in both males and females should be assessed using the Tanner Pubic Hair stages 1–5. Breast development in girls and genital development or testes size in boys are also categorized into stage 1–5 based on the classifications of Tanner (see Appendix I and Figures I-19 to I-22).

Skeletal maturation (or bone age) is the second method for the assessment of physical maturity. Bone age is assessed by a left hand-wrist radiograph and scored using the standards developed by Greulich and Pyle[3] or the newly revised TW3 method developed by Tanner and colleagues[3] Bone age provides a measure of "how far a given individual has progressed along his or her road to full maturity"[3] regardless of chronologic age. Sexual and skeletal maturity provide a measure of physical maturity and are valuable in formulating the nutritional assessment of children and adolescents.

Laboratory Tests

Laboratory testing is a helpful but less essential part of a nutritional assessment in most children and is presented in detail in Chapter 3. Nutritional information can be obtained from plasma, serum, urine, stool, hair, and nail samples. The latter two are rarely used clinically. Depending on the underlying medical condition and related nutritional problems from the history and physical examination, a focused laboratory assessment may be obtained. Serum albumin and prealbumin reflect the adequacy of protein and calorie intake. Because the half-life of albumin is approximately 20 days, it also reflects long-term protein stores. The shorter half-life of prealbumin (two to three days) is a better short-term indicator of calorie and protein intake. However, the usefulness of prealbumin in the hospitalized patient may be limited by the fact that it is decreased in the setting of stress, sepsis, and acute illness. Checking a C-reactive protein level may help identify when the low prealbumin level is related to stress. Anemia can be attributable to multiple nutritional deficiencies (e.g., iron, vitamin B12, folate, vitamin C, protein, and vitamin E), and a careful analysis of the red blood cell indices and peripheral blood smear will help to determine what further nutritional laboratory tests should be obtained (e.g., iron studies, vitamin levels). In premature infants, nutritional anemias can be attributable to iron, vitamin E, and copper deficiencies. Nutritional tests to check for bone health may include serum, calcium, phosphorus, alkaline phosphatase, magnesium, and 25-hydroxyvitamin D. Additional information on bone health may be

obtained from a parathormone level, radiography, and DXA scan. Specific vitamin and mineral levels can be checked when deficiency or excess states are suspected. A urine analysis, along with serum electrolytes, is useful in assessing the hydration status of the patient. See Appendix Table I-8 for a list of selected clinical findings related to nutritional inadequacies and Chapter 3 for a more in-depth look at laboratory assessment of nutritional status.

NUTRITIONAL REQUIREMENTS

The estimation of nutritional requirements is the last step in a nutritional assessment. Recommendations for energy and protein intake, as well as specific vitamins and minerals, are needed for patient care (see Chapters 8–10). The history (medical and dietary), physical examination, and anthropometric and laboratory data obtained are used to help estimate these nutritional requirements. These provide a starting point for nutritional therapy and are modified over time based on the patient's ongoing health status and response to nutritional therapy. The adequacy of the nutritional therapy provided should be vigilantly monitored in children with failure to thrive and obesity and in those patients with conditions requiring enteral or parenteral nutrition.

The DRIs[85] provide updated protein recommendations (see Appendix II; also see Chapter 6). Adequate intakes (AIs) for protein in infants 0–6 months are based on the mean protein intake of healthy breastfed infants.[3] Nitrogen intake, nitrogen balance (the minimum protein intake necessary to maintain nitrogen balance), rates of protein deposition, and efficiency of protein use all influence protein requirements. For individuals 7 months through 18 years of age, the protein recommendations are based on a combination of these factors, plus a safety factor to account for individual variation.[85] The DRIs provide protein recommendations (grams per kilogram per day) by age from birth to 8 years and by age and sex starting from 9 years. Individualized needs can be estimated by multiplying the age/sex appropriate grams of protein per kilogram per day by the body weight (in kilograms). As previously noted, DRIs may be further individualized based on the child's nutritional, medical, and growth needs and should be adjusted over time based on clinical status and response to nutritional intervention.

There are a number of methods to estimate energy needs of children in the clinical setting, including the DRIs for estimates of total energy needs,[85] the WHO[86] and Schofield[87] prediction equations for estimates of

resting energy expenditure (REE), and a direct measurement of REE (see below). The DRIs include estimated average requirements, AIs, and tolerable upper intake levels for most nutrients; see Chapter 11 for details.[3] The DRIs are used in Canada and the United States, and nutrient intakes at the suggested levels promote nutrient function, biological and physical well-being, and disease prevention. DRIs are available for vitamins, minerals (see Chapters 8–10), energy, and macronutrient recommendations (see Chapters 5–7). Generally, the DRIs provide for the nutritional needs of the healthy individual and population. Therefore, DRIs may require adjustments in the clinical setting because they do not address energy or nutrient requirements for individuals who are malnourished or have acute/chronic disease.

Both the Food and Nutrition Board DRI and the Food and Agricultural Organization of the WHO recommendations for energy requirements in children 0–2 years are estimated from total energy expenditure (TEE) measurements (by the double-labeled water methods) and estimated energy needs for tissue deposition for growth "at rates consistent with good health."[85,86] For children at this young age, estimated energy requirements (EERs) vary based on weight. The EERs for children aged 3–8 years and 9–18 years are also from TEE and energy deposition costs (20 and 25 kcal/day, respectively) and are based on age, weight, height, and level of physical activity (see Appendix II). The important role of moderate physical activity in achieving and maintaining the appropriate energy balance for optimal health is emphasized in these new recommendations. Levels of physical activity are categorized into four levels: sedentary, low active, active, and very active.[3] EER equations are also available for use in children aged 3 years and above who are at "risk of overweight" defined as a BMI >85th percentile and "overweight" as a BMI >95th percentile.[3] DRIs provide an estimate of total energy needs in kilocalories per day and may be adjusted based on nutritional, medical, and growth needs of the individual patient.

The WHO and Schofield REE prediction equations offer another method to estimate energy requirements. These equations are based on the evaluation of several thousand children and are clinically useful. The WHO equations (see Appendix II and Table II-15) estimate REE by sex, age, and weight groups and approximate the basal metabolic rate.[88] Total daily energy needs are then estimated by multiplying the estimated REE by a factor to adjust for physical activity, medical status, and/or the need for catch-up growth. The Schofield equations (see Appendix II and Table II-13) use sex, age, weight, and height of the child and may more

accurately predict REE in children with altered growth and body composition (i.e., failure to thrive and obesity).[89] Schofield REE estimates are also adjusted for the patient's activity, stress, and growth needs (see Table 2-2) to approximate total daily energy needs.

The current DRI for EER provides additional guidance beyond the 1985 FAO/WHO/UNU report for energy requirements with respect to catch-up growth in undernourished or stunted children[90] based on limited data. Similarly, the 2001 FAO/WHO/UNU report on Human Energy Requirements does not specifically address energy requirements for children recovering from moderate or severe malnutrition or chronic illnesses.[91] Direction with respect to energy replacement to allow for twice the normal growth rate for children aged 6–24 months is provided. Similar to the new WHO growth charts, the emphasis in the WHO report is prescriptive as opposed to descriptive. Thus, it indicates how individuals should grow in response to appropriate energy intake in optimal conditions rather than reflect how they were observed to grow with inadequate energy intake or excess.

TABLE 2-2 Disease and Physical Activity Factors for Adjustment of Resting Energy Expenditure (REE)

REE Factor	Disease and Activity Factors
×1.0–1.1	Well-nourished children, sedated on ventilator, extracorporeal membrane oxygenation; minimal stress
×1.3	Well-nourished children with decreased activity, minor surgery, mild-to-moderate sedation; minimal stress
×1.5	Ambulatory child with mild-to-moderate stress Inactive child with sepsis, cancer, trauma, extensive surgery Minimally active child with malnutrition and catch-up growth requirements
×1.7	Active child with catch-up growth requirements Active child with severe stress

Source: Adapted from Mascarenhas MR, Tershakovec AM, Stallings VA. Parenteral and enteral nutrition. In: Wyllie R, Hyams JS, eds. *Pediatric Gastrointestinal Disease*. Philadelphia: WB Saunders; 1999:741–757.

Resting Energy Expenditure and Indirect Calorimetry

Prediction equations to estimated energy needs in children are not designed to address the special needs of children with complex health

conditions. The indications for performing indirect calorimetry for REE measurements are to determine energy requirements for medically and nutritionally complex patients. These studies can be performed in critically ill and intubated patients, provided cuffed endotracheal tubes are used to measure gas exchange precisely.

The REE approximates basal metabolic rate and can be determined by indirect calorimetry. This technique measures gas exchange, specifically, the oxygen consumed and carbon dioxide produced as a product of the oxidation of a dietary substrate. The amount of heat or energy produced from the oxidation of a specific dietary substrate is determined by the ratio of carbon dioxide exhaled to the oxygen inhaled and is referred to as the respiratory quotient (RQ). Complete oxidation of pure glucose or carbohydrate will yield equal amounts of carbon dioxide produced for oxygen consumed, resulting in an RQ of 1. For fats and mixed meals, the RQ is less than 1, since they are oxidized less than a carbohydrate only meal.

Following consumption of a mixed meal for dinner in which the macronutrient proportions are known, an age appropriate overnight fast precedes the measurements of REE. Studies are performed early in the morning in a rested, awake state, with minimal preceding physical activity/exertion. For children who are unable to cooperate or limit their movement, the test is sometimes performed in a sedated state. For infants, REE is usually measured while sleeping after a feed. The study subject lies still on a bed and gas exchange is measured usually via a ventilated hood device. The studies typically take 30–60 minutes to complete. The REE values obtained are then compared to the WHO/Schofield equations[87,88] to calculate a REE as a percent of predicted values based on age, sex, and body size. The EER can also be calculated using a physical activity level (PAL) to estimate the needs for physical activity.

Table 2-2 provides an example of PAL adapted to estimate energy requirements in children with different types of illnesses and corresponding expected physical activity. Further research is required regarding REE adjustment factors for disease states and PAL needed, and their ability to determine the energy needs of children with difference severities of malnutrition and in the context of acute and chronic illnesses.

In summary, for the estimation of energy needs in the clinical setting, an REE based on indirect calorimetry is preferred for children with complex medical and nutritional needs. If an REE measurement cannot be obtained, then the WHO or Schofield equations are recommended. These estimates of REE are then adjusted based on activity, stress, and growth

for an estimate of total daily energy needs for the individualized patient. As mentioned earlier, the new DRIs for total energy recommendations may replace these estimates in the clinical setting. However, these estimates of TEE are not as reliable as those values obtained by more accurate research methods (e.g., doubly labeled water).[92] Finally, it should be remembered that all estimates are guidelines for the initiation of nutritional therapy. Adjustments in the nutritional regimen are made over time based on objective measures, such as weight gain, laboratory data, and medical condition.

CONCLUSION

In pediatric care, adequate nutrition must support both usual nutrient requirements and nutrients needed for optimal growth and development. However, nutritional status depends in part on current and past illnesses, and a child's response to illness is affected by his/her nutritional status. Thus, understanding and addressing the nutritional status of a hospitalized child are important components of clinical care. No single measure provides an adequate assessment of nutrition status. A nutritional assessment often includes the input of many members of the health care team. This chapter provides the factors to be considered in the nutritional assessment and monitoring of a hospitalized child. Initial nutritional goals are adjusted over time as the child's medical and nutrition status change to provide optimal care.

REFERENCES

1. Baker JP, Detsky AS, Wesson DE, et al. Nutritional assessment: a comparison of clinical judgement and objective measurements. *N Engl J Med*. 1982;306:969–972.
2. Maka DA, Murphy LK. Drug-nutrient interactions: a review. *AACN Clin Issues*. 2000;11:580–589.
3. Tanner J, Healy M, Goldstein H, Cameron N. *Assessment of Skeletal Maturity and Prediction of Adult Height (TW3) Method*. London: W.B. Saunders; 2001.
4. Ziegler P, Briefel R, Clusen N, Devaney B. Feeding Infants and Toddlers Study (FITS): development of the FITS survey in comparison to other dietary survey methods. *J Am Diet Assoc*. 2006;106:S12–S27.
5. Ziegler P, Hanson C, Ponza M, et al. Feeding infants and toddlers study: meal and snack intakes of Hispanic and non-Hispanic infants and toddlers. *J Am Diet Assoc*. 2006;106:S107–S123.
6. Dwyer J. Nutritional status: dietary assessment. In: *Encyclopedia of Human Nutrition*. San Diego, CA: Academic Press; 1999.
7. WHO Multicentre Growth Reference Study Group. *WHO Child Growth Standards: Growth Velocity Based on Weight, Length and Head Circumference:*

Methods and Development. Geneva: World Health Organization; 2009.
8. Spender QW, Cronk CE, Charney EB, Stallings VA. Assessment of linear growth of children with cerebral palsy: use of alternative measures to height or length. *Dev Med Child Neurol*. 1989;31:206–214.
9. Stallings VA, Zemel BS. Nutritional assessment of the disabled child. In: Sullivan PR, Rosenbloom L, (eds.) *Clinics in Developmental Medicine*. London: MacKeith Press; 1996.
10. Stevenson RD. Use of segmental measures to estimate stature in children with cerebral palsy. *Arch Pediatr Adolesc Med*. 1995;149:658–662.
11. Chumlea WC, Guo SS, Steinbaugh ML. Prediction of stature from knee height for black and white adults and children with application to mobility-impaired or handicapped persons. *J Am Diet Assoc*. 1994;94:1385–1388, 1391; quiz 9–90.
12. Lubchenco LO, Hansman C, Boyd E. Intrauterine growth in length and head circumference as estimated from live births at gestational ages from 26 to 42 weeks. *Pediatrics*. 1966;37:403–408.
13. Lubchenco LO, Hansman C, Dressler M, Boyd E. Intrauterine growth as estimated from liveborn birth-weight data at 24 to 42 weeks of gestation. *Pediatrics*. 1963;32:793–800.
14. Olsen IE, Groveman SA, Lawson ML, et al. New intrauterine growth curves based on United States data. *Pediatrics*. 2010;125:e214–e224.
15. Fenton TR. A new growth chart for preterm babies: Babson and Benda's chart updated with recent data and a new format. *BMC Pediatr*. 2003;3:13.
16. Casey PH, Kraemer HC, Bernbaum J, et al. Growth status and growth rates of a varied sample of low birth weight, preterm infants: a longitudinal cohort from birth to three years of age. *J Pediatr*. 1991;119:599–605.
17. The Infant Health and Development Program. Enhancing the outcomes of low-birth-weight, premature infants. A multisite, randomized trial. The Infant Health and Development Program. *JAMA*. 1990;263:3035–3042.
18. Guo SS, Roche AF, Chumlea WC, et al. Growth in weight, recumbent length, and head circumference for preterm low-birthweight infants during the first three years of life using gestation-adjusted ages. *Early Hum Dev*. 1997;47:305–325.
19. Guo SS, Wholihan K, Roche AF, et al. Weight-for-length reference data for preterm, low-birth-weight infants. *Arch Pediatr Adolesc Med*. 1996;150:964–970.
20. American Academy of Pediatrics Committee on Nutrition, ed. *Pediatric Nutrition Handbook*. 5th ed. Chicago, IL; 2004.
21. Kramer MS, Platt RW, Wen SW, et al. A new and improved population-based Canadian reference for birth weight for gestational age. *Pediatrics*. 2001;108:E35.
22. Niklasson A, Ericson A, Fryer JG, et al. An update of the Swedish reference standards for weight, length and head circumference at birth for given gestational age (1977–1981). *Acta Paediatr Scand*. 1991;80:756–762.
23. Beeby PJ, Bhutap T, Taylor LK. New South Wales population-based birthweight percentile charts. *J Paediatr Child Health*. 1996;32:512–518.
24. Kuczmarski RJ, Ogden CL, Grummer-Strawn LM, et al. CDC growth charts: United States. *Adv Data*. 2000;314:1–27.
25. de Onis M. *WHO Child Growth Standards: Length/Height-for-Age, Weight-for-Age, Weight-for-Length, Weight-for-Height and Body Mass Index-for-Age: Methods and Development*. Geneva: World Health Organization.
26. Ehrenkranz RA, Younes N, Lemons JA, et al. Longitudinal growth of hospitalized very low birth weight infants. *Pediatrics*. 1999;104:280–289.
27. Grummer-Strawn LM, Reinold C, Krebs NF. Use of World Health Organization and CDC growth charts for children aged 0–59 months in the United States.

MMWR Recomm Rep. 2010;59:1–15.
28. Mei Z, Ogden CL, Flegal KM, Grummer-Strawn LM. Comparison of the prevalence of shortness, underweight, and overweight among US children aged 0 to 59 months by using the CDC 2000 and the WHO 2006 growth charts. *J Pediatr*. 2008;153:622–628.
29. de Onis M, Garza C, Onyango AW, Borghi E. Comparison of the WHO child growth standards and the CDC 2000 growth charts. *J Nutr*. 2007;137:144–148.
30. Baumgartner RN, Roche AF, Himes JH. Incremental growth tables: supplementary to previously published charts. *Am J Clin Nutr*. 1986;43:711–722.
31. Guo SM, Roche AF, Fomon SJ, et al. Reference data on gains in weight and length during the first two years of life. *J Pediatr*. 1991;119:355–362.
32. Roche AF, Himes JH. Incremental growth charts. *Am J Clin Nutr*. 1980;33:2041–2052.
33. Roche AF, Mukherjee D, Guo SM, Moore WM. Head circumference reference data: birth to 18 years. *Pediatrics*. 1987;79:706–712.
34. Roche AF, Guo S, Moore WM. Weight and recumbent length from 1 to 12 mo of age: reference data for 1-mo increments. *Am J Clin Nutr*. 1989;49:599–607.
35. WHO Expert Committee. Physical status: the use and interpretation of anthropometry. Report of a WHO Expert Committee. *World Health Organ Tech Rep Ser*. 1995;854:1–452.
36. Tanner JM, Davies PS. Clinical longitudinal standards for height and height velocity for North American children. *J Pediatr*. 1985;107:317–329.
37. Cronk C, Crocker AC, Pueschel SM, et al. Growth charts for children with Down syndrome: 1 month to 18 years of age. *Pediatrics*. 1988;81:102–110.
38. Cronk CE. Growth of children with Down's syndrome: birth to age 3 years. *Pediatrics*. 1978;61:564–568.
39. Bull MJ. Health supervision for children with Down syndrome. *Pediatrics*. 2011;128:393–406.
40. WHO Expert Committee on Physical Status. Physical status: the use and interpretation of anthropometry. Report of a WHO Expert Committee. *World Health Organ Tech Rep Ser*. 1995;854:1–452.
41. Himes JH, Roche AF, Thissen D, Moore WM. Parent-specific adjustments for evaluation of recumbent length and stature of children. *Pediatrics*. 1985;75:304–313.
42. Hamill PV, Drizd TA, Johnson CL, et al. Physical growth: National Center for Health Statistics percentiles. *Am J Clin Nutr*. 1979;32:607–629.
43. Mascarenhas MR, Zemel B, Stallings VA. Nutritional assessment in pediatrics. *Nutrition*. 1998;14:105–115.
44. Olsen IE, Lawson ML, Meinzen-Derr J, et al. Use of a body proportionality index for growth assessment of preterm infants. *J Pediatr*. 2009;154:486–491.
45. Kuczmarski RJ, Flegal KM. Criteria for definition of overweight in transition: background and recommendations for the United States. *Am J Clin Nutr*. 2000;72:1074–1081.
46. Mei Z, Grummer-Strawn LM, Pietrobelli A, et al. Validity of body mass index compared with other body-composition screening indexes for the assessment of body fatness in children and adolescents. *Am J Clin Nutr*. 2002;75:978–985.
47. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition for child overweight and obesity worldwide: international survey. *BMJ*. 2000;320:1240–1243.
48. Freedman DS, Ogden CL, Berenson GS, Horlick M. Body mass index and body fatness in childhood. *Curr Opin Clin Nutr Metab Care*. 2005;8:618–623.

49. Bellizzi MC, Dietz WH. Workshop on childhood obesity: summary of the discussion. *Am J Clin Nutr*. 1999;70:173S–175S.
50. Hoffman DJ, Sawaya AL, Martins PA, et al. Comparison of techniques to evaluate adiposity in stunted and nonstunted children. *Pediatrics*. 2006;117:e725– e732.
51. Eckhardt CL, Adair LS, Caballero B, et al. Estimating body fat from anthropometry and isotopic dilution: a four-country comparison. *Obes Res*. 2003;11:1553–1562.
52. Lai HJ, Shoff SM. Classification of malnutrition in cystic fibrosis: implications for evaluating and benchmarking clinical practice performance. *Am J Clin Nutr*. 2008;88:161–166.
53. de Onis M, Blössner M. *WHO Global Database on Child Growth and Malnutrition*. Geneva: World Health Organization; 1997.
54. Dale NM, Grais RF, Minetti A, et al. Comparison of the new World Health Organization growth standards and the National Center for Health Statistics growth reference regarding mortality of malnourished children treated in a 2006 nutrition program in Niger. *Arch Pediatr Adolesc Med*. 2009;163:126–130.
55. Cole TJ, Faith MS, Pietrobelli A, Heo M. What is the best measure of adiposity change in growing children: BMI, BMI %, BMI z-score or BMI centile? *Eur J Clin Nutr*. 2005;59:419–425.
56. Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr*. 1981;34:2540–2545.
57. Zemel BS. Anthropometric assessment of nutritional status. In: Altschuler S and Liacouras C (eds.) *Pediatric Gastroenterology*. London: Churchill Livingstone; 1998.
58. Bishop N, Braillon P, Burnham J, et al. Dual-energy X-ray aborptiometry assessment in children and adolescents with diseases that may affect the skeleton: the 2007 ISCD Pediatric Official Positions. *J Clin Densitom*. 2008;11:29–42.
59. Baim S, Binkley N, Bilezikian JP, et al. Official Positions of the International Society for Clinical Densitometry and executive summary of the 2007 ISCD Position Development Conference. *J Clin Densitom*. 2008;11:75–91.
60. Ellis KJ. Human body composition: in vivo methods. *Physiol Rev*. 2000;80:649–680.
61. Ogden CL, Li Y, Freedman DS, et al. Smoothed percentage body fat percentiles for U.S. children and adolescents, 1999–2004. *Natl Health Stat Report*. 2011;43:1–7.
62. Rauch F, Plotkin H, DiMeglio L, et al. Fracture prediction and the definition of osteoporosis in children and adolescents: the ISCD 2007 Pediatric Official Positions. *J Clin Densitom*. 2008;11:22–28.
63. Zemel BS, Kalkwarf HJ, Gilsanz V, et al. Revised reference curves for bone mineral content and areal bone mineral density according to age and sex for black and non-black children: results of the bone mineral density in childhood study. *J Clin Endocrinol Metab*. 2011;96:3160–3169.
64. Kalkwarf HJ, Zemel BS, Gilsanz V, et al. The bone mineral density in childhood study: bone mineral content and density according to age, sex, and race. *J Clin Endocrinol Metab*. 2007;92:2087–2099.
65. Looker AC, Wahner HW, Dunn WL, et al. Proximal femur bone mineral levels of US adults. *Osteoporos Int.* 1995;5:389–409.
66. Zemel BS, Stallings VA, Leonard MB, et al. Revised pediatric reference data for the lateral distal femur measured by Hologic Discovery/Delphi dual-energy X-ray absorptiometry. *J Clin Densitom*. 2009;12:207–218.
67. Crabtree NJ, Kibirige MS, Fordham JN, et al. The relationship between lean body

mass and bone mineral content in paediatric health and disease. *Bone*. 2004;35:965–972.
68. Hogler W, Briody J, Woodhead HJ, et al. Importance of lean mass in the interpretation of total body densitometry in children and adolescents. *J Pediatr*. 2003;143:81–88.
69. Zemel BS, Leonard MB, Kelly A, et al. Height adjustment in assessing dual energy x–ray absorptiometry measurements of bone mass and density in children. *J Clin Endocrinol Metab*. 2010;95:1265–1273.
70. Stallings VA. Bone densitometry. In: Altschuler S and Liacouras C (eds.) *Clinical Pediatric Gastroenterology*. Philadelphia, PA: Churchill Livingstone; 1998.
71. Bonjour JP, Gueguen L, Palacios C, et al. Minerals and vitamins in bone health: the potential value of dietary enhancement. *Br J Nutr*. 2009;101:1581–1596.
72. Aaseth J, Boivin G, Andersen O. Osteoporosis and trace elements—an overview. *J Trace Elem Med Biol*. 2012;26:149–152.
73. Bonjour JP. Dietary protein: an essential nutrient for bone health. *J Am Coll Nutr*. 2005;24:526S–536S.
74. Zemel BS. Quantitative computed tomography and computed tomography in children. *Curr Osteoporos Rep*. 2011;9:284–290.
75. Zemel B, Bass S, Binkley T, et al. Peripheral quantitative computed tomography in children and adolescents: the 2007 ISCD Pediatric Official Positions. *J Clin Densitom*. 2008;11:59–74.
76. Fields DA, Goran MI, McCrory MA. Body-composition assessment via air-displacement plethysmography in adults and children: a review. *Am J Clin Nutr*. 2002;75:453–467.
77. Ellis KJ, Yao M, Shypailo RJ, et al. Body-composition assessment in infancy: air-displacement plethysmography compared with a reference 4-compartment model. *Am J Clin Nutr*. 2007;85:90–95.
78. Urlando A, Dempster P, Aitkens S. A new air displacement plethysmograph for the measurement of body composition in infants. *Pediatr Res*. 2003;53:486–492.
79. Fields DA, Allison DB. Air-displacement plethysmography pediatric option in 2–6 years old using the four-compartment model as a criterion method. *Obesity* (*Silver Spring*). 2012;20:1732–1737.
80. Tanner JM. *Growth at Adolescence: With a General Consideration of the Effects of Hereditary and Environmental Factors Upon Growth and Maturation from Birth to Maturity*. Oxford, UK: Blackwell Scientific Publications; 1962.
81. Duke PM, Litt IF, Gross RT. Adolescents' self-assessment of sexual maturation. *Pediatrics*. 1980;66:918–920.
82. Morris N, Udry J. Validation of self administered instrument to assess stage of adolescent development. *J Youth Adolesc*. 1980;9:271–280.
83. Neinstein LS. Adolescent self-assessment of sexual maturation: reassessment and evaluation in a mixed ethnic urban population. *Clin Pediatr* (*Phila*). 1982;21:482–484.
84. Schall JI, Semeao EJ, Stallings VA, Zemel BS. Self-assessment of sexual maturity status in children with Crohn's disease. *J Pediatr*. 2002;141:223–229.
85. Food and Nutrition Board TIoM. *Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids (Macronutrients)*. Washington, DC: National Academies Press; 2002.
86. FAO/WHO/UNU EC. Energy and protein requirements. In: *Technical Report Series #724*. Geneva, Switzerland: World Health Organization; 1985:71–112.
87. Schofield WN. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr*. 1985;39(suppl 1):5–41.

88. FAO/WHO/UNU Expert Consultation. *Energy and Protein Requirements: Technical Report Series*. Geneva: World Health Organization; 1985.
89. Sentongo TA, Tershakovec AM, Mascarenhas MR, et al. Resting energy expenditure and prediction equations in young children with failure to thrive. *J Pediatr*. 2000;136:345–350.
90. Chattopadhyay S, Lahiri K, Bharati G. Changes in free polyamines and related enzymes during stipule and pod wall development in Pisum sativum. *Indian J Exp Biol*. 2002;40:945–949.
91. FAO Food and Nutrition Technical Report Series 1. *Human Energy Requirements: Report of a Joint FAO/WHO/UNU Expert Consultation*. Rome: United Nations Univeresity, World Health Organization, Food and Agriculture Organization of the United Nations; 2004.
92. Das S, Roberts S. Energy metabolism. In: Bowman BA, and Russell, RM, (eds.). *Present Knowledge in Nutrition*. Washington, DC: International Life Sciences Institute (ILSI); 2001.

3

Laboratory Assessment of Nutritional Status

Clodagh M. Loughrey, MD, MRCP, FRCPath, MA and Ian S. Young, BSc, MD, FRCP, FRCPath

Nutritional assessment in pediatrics is primarily a clinical process, based on analysis of recent dietary intake and anthropometric measurement methods (as outlined in Chapter 2: Clinical Assessment of Nutritional Status). Laboratory assessment of nutritional status plays a complementary role that may be critical in specific situations, including:

- identification of suboptimal micronutrient status;
- objective and quantitative confirmation of nutritional deficiencies suggested by clinical assessment;
- provision of baseline and serial data with which to monitor response to nutritional interventions, particularly important in prevention of the refeeding syndrome (see Chapter 13: Protein–energy malnutrition);
- provision of clues to the underlying cause of growth failure, which may be related to undernutrition (e.g., malabsorption) or have alternative explanations (e.g., endocrine disorders, chronic kidney disease);
- epidemiological studies, which seek to identify associations

between disease and nutritional biomarkers.

Table 3-1 indicates the range of tests involved in assessing nutritional status. Table 3-2 lists age- and gender-based reference ranges for Exactly which supporting laboratory tests are used, when repeated and how often, will depend on the clinical status of the patient and whether monitoring or evaluation of nutritional support is also indicated.[2]

Effective use of the laboratory service demands an awareness of nonnutritional factors that can influence results. After a discussion of potentially confounding factors in the interpretation of laboratory data, we review laboratory assessment of protein and energy status, micronutrient status, and hematological and immunological testing relevant to nutrition, before finishing with a look at technologies that are likely to dominate nutritional testing in the laboratory in the foreseeable future.

POTENTIAL CONFOUNDERS OF INTERPRETATION

Interpretation of laboratory tests must take account of the range of biological and analytical factors that can influence results.[3] Some examples of relevance to nutritional assessment are given to illustrate these factors.

Biological Variation

Interindividual variation refers to differences in true concentrations of any nutrient between individuals. These may be dependent on several parameters unrelated to diet, including physiological, pathological, genetic, and environmental influences. Analytes that are significantly influenced by factors that cannot be controlled will often be unreliable indicators of intake. Examples include:

TABLE 3-1 Components of Assessment of Nutritional Status in Clinical Pediatrics

Clinical assessment	• Clinical history • Subjective global assessment[1] • Physical examination
Anthropometric assessment	• Height for age percentiles/Z-scores • Weight for age percentiles/Z-scores • Body mass index percentiles/Z-scores

	• Triceps skinfold and mid-arm circumference percentiles
Dietary assessment	• 24-hr recall • 72-hr diary • Semiquantitative food frequency questionnaire
Laboratory assessment	• CBC (Hb, hematocrit, total and differential WCC) • Albumin (or prealbumin if available)[a] + CRP; total protein • Renal function/hydrational status: BUN, creatinine, sodium • Potassium, phosphate, magnesium[a] • Liver function tests[a] • Fasting lipids[a] • Glucose[a] • Calcium, alk phos, vitamin D • Iron studies, vitamin B_{12} and folate • Selenium, zinc, copper • Prothrombin time • Other vitamins, other minerals • Nitrogen balance studies

Abbreviations: CBC, complete blood count; CRP, C-reactive protein; WCC, white cell count.

[a]Tests of particular relevance to monitoring in nutritional support

TABLE 3-2 Reference Ranges of Select Nutritional Laboratory Values

Nutrient	Age	Males	Either	Females
Albumin (g/dL)	< 5 d (<2.5kg)		2.0–3.6	
	< 5 d (>2.5kg)		2.6–3.6	
	1–3 y		3.4–4.2	
	4–6 y		3.5–5.2	
	>6 y		3.7–5.6	
Alkaline phosphatase (U/L)	1–30 d	75–316		48–406
	31–365 d	82–383		124–341
	1–3 y	104–345		108–317
	4–6 y	93–309		96–297
	7–9 y	86–315		69–325

	10–12 y	42–362		51–332
	13–15 y	74–390		50–162
	16–18 y	52–171		47–119
Calcium (mg/dL)	1–30 d	8.5–10.6		8.4–10.6
	31–365 d	8.7–10.5		8.9–10.5
	1–3 y	8.8–10.6		8.5–10.4
	4–6 y	8.8–10.6		8.5–10.6
	7–9 y	8.7–10.3		8.5–10.3
	10–12 y	8.7–10.2		8.6–10.2
	13–15 y	8.5–10.2		8.4–10.0
	16–18 y	8.4–10.3		8.6–9.8
Calcium, ionized (mg/dL)	0–1 mo		3.9–6.0	
Whole blood	1–6 mo		3.7–5.9	
	1–17 y		4.9–5.5	
Carnitine, total, fasting	1–12 mo		15–39	
(µmol/L)	1–7 y		18–37	
	7–15 y		31–43	
Cerulosplasmin (mg/L)	1–30 d	77–253		33–275
	31–365 d	154–484		154–429
	1–3 y	253–561		286–539
	4–6 y	286–561		264–539
	7–9 y	253–517		231–484
	10–12 y	209–506		209–484
	13–15 y	198–495		209–462
	16–18 y	198–451		220–495
Cholesterol (mg/dL)	1–30 d	54–151		62–155
	31–182 d	81–147		62–141

	183–365 d	76–179		76–216
	1–3 y	85–182		108–193
	4–6 y	110–217		106–193
	7–9 y	110–211		104–210
	10–12 y	105–223		105–218
	13–15 y	91–204		108–205
	16–18 y	82–192		92–234
Copper (µg/dL)	0–6 mo		38–104	
	6–12 mo		24–152	
	1–2 y		76–193	
	2–4 y		87–187	
	4–6 y		56–191	
	6–10 y		117–181	
	10–14 y		87–182	
	14–18 y		75–187	
Ferritin (ng/mL)	1–30 d	6–400		6–515
	1–6 mo	6–410		6–340
	7–12 mo	6–80		6–45
	1–5 y	6–60		6–60
	6–19 y	6–320		6–70
Folic acid (nmol/L)	0–1 y	16.3–50.8		14.3–51.5
	2–3 y	5.7–34.0		3.9–35.6
	4–6 y	1.1–29.4		6.1–31.9
	7–9 y	5.2–27.0		5.4–30.4
	10–12 y	3.4–24.5		2.3–23.1
	13–18 y	2.7–19.9		1.7–16.3
Glucose (mg/dL)	Outside neonatal period		70–126	
Glutathione peroxidase activity, U/L (serum)	Newborn (term)		180–890	
	1–5 y		554–985	

	6–15 y		567–1153	
	16 y - adult		780–1269	
Hemoglobin (g/dL)	1–3 d	12.5–16.6		12.7–16.4
	4–7 d	12.5–16.3		12.6–15.3
	8–14 d	11.9–15.7		12.7–14.9
	15–30 d	11.6–14.2		11.6–14.3
	31–60 d	10.2–12.7		11.1–13.7
	2–6 mo	10.5–13.0		10.7–13.4
	6 mo –2 y	10.4–12.5		10.8–12.6
	2–6 y	11.0–12.8		11.1–12.9
	6–12 y	11.0 13.3		11.3–13.4
	12–18 y	11.0–14.3		11.3–13.4
Homocysteine, total (µmol/L)	2 mo–10 y		3.3–8.3	
	11–15 y		4.7–10.3	
	16–18 y		4.7–11.3	
Insulin-like growth factor-1 (IGF-1) (ng/mL)	1–30 d	2–56		7–92
	1–6 mo	2–82		5–72
	6–12 mo	2–54		8–75
	1–3 y	17–116		12–135
	4–6 y	21–160		13–176
	7–9 y	65–207		53–253
	10–12 y	69–196		75–357
	13–15 y	80–404		66–344
	16–18 y	96–383		99–364
Iron (µg/dL)	0–90 d	72–203		75–235
	4–12 mo	23–142		60–192
	1–3 y	25–126		55–162

	4–10 y	15–128		28–122
	11–14 y	32–107		25–102
	15–18 y	30–130		25–107
Iron-binding capacity (µg/dL)	0–90 d	155–330		165–275
	4–12 mo	150–380		250–455
	1–3 y	215–420		160–415
	4–10 y	185–415		260–385
	11–14 y	265–410		250–420
	15–18 y	270–415		285–410
Magnesium (mg/dL)	1–30 d	1.7–2.4		1.7–2.5
	1–12 mo	1.6–2.5		1.9–2.4
	1–3 y	1.7–2.4		1.7–2.4
	4–6 y	1.7–2.4		1.7–2.2
	7–9 y	1.7–2.3		1.6–2.3
	10–12 y		1.6–2.2	
	13–15 y		1.6–2.3	
	16–18 y		1.5–2.2	
Mean corpuscular volume	1–3 d	94.0–106.3		89.7–105.4
MCV (μm^3)	4–7 d	87.1–96.5		86.5–93.8
	8–14 d	87.1–94.8		87.4–92.2
	15–30 d	88.0–95.2		88.4–93.3
	31–60 d	86.5–92.1		85.7–91.6
	2–6 mo	79.6–86.3		82.0–87.0
	6 mo –2 y	75.6–83.1		76.6–83.2
	2–6 y	76.8–83.3		77.7–84.1
	6–12 y	78.2–83.9		79.5–85.2
	12–18 y	80.8–86.6		82.1–87.7

	> 18 y	83.5–90.2		82.7–89.4
Osmolality (plasma) (mOsm/kg)	1 mo –18 y		280–300	
Phosphorous (mg/dL)	1–30 d	3.9–6.9		4.3–7.7
	1–12 mo	3.5–6.6		3.7–6.5
	1–3 y	3.1–6.0		3.4–6.0
	4–6 y	3.3–5.6		3.2–5.5
	7–9 y	3.0–5.4		3.1–5.5
	10–12 y	3.2–5.7		3.3–5.3
	13–15 y	2.9–5.1		2.8–4.8
	16–18 y	2.7–4.9		2.5–4.8
Prothrombin time			11–15 s	
Prealbumin (transthyretin) (mg/dL)	1–40 d	3.2–15.9		4.2–14.4
	41–90 d	2.7–17.6		2.5–21.9
	3–9 mo	7.3–27.9		5.3–25.0
	10–24 mo	6.7–28.5		7.3–33.7
	2–10 y	6.9–31.2		8.0–35.2
	11–15 y	6.3–33.5		8.6–40.7
	16–18 y	8.0–41.6		13.7–44.1
Protein, total (g/dL)	0–2 mo	4.0–7.6		3.6–7.0
	2–6 mo	4.0–7.0		4.0–7.6
	6–12 mo	4.2–7.9		4.6–7.8
	1–6 y	6.0–8.0		6.0–7.8
	7–9 y	6.3–8.1		6.3–8.1
	10–19 y	6.4–8.6		6.4–8.6
Retinol-binding protein	0–30 d	0.91–5.6		1.01–5.75
(RBP) (mg/dL)	1 mo –18 y	1.09–8.7		0.98–6.76
Selenium (µg/dL)	0–<6 mo		0.8–4.4	
	6–<12 mo		0–6.9	
	1–<2 y		1.5–7.3	

	2–<4 y		0.9–10.4	
	4–<6 y		0.6–12.5	
	6–<10 y		2.4–11.8	
	10–<14 y		2.4–11.4	
	14–<18 y		2.1–11.8	
Soluble transferrin receptor (mg/L)	6 mo–2 y		1.37–2.85	
	2–6 y		1.05–3.05	
	6–12 y		1.16–2.72	
	12–18 y		0.97–2.60	
	>18 y		0.84–2.32	
Transferrin (mg/dL)	1–30 d	86–174		83–176
	1–6 mo	94–271		111–258
	>6–12 mo	152–296		126–303
	1–3 y	167–304		129–317
	4–6 y	172–291		149–331
	7–9 y	129–293		159–305
	10–12 y	115–316		158–313
	13–15 y	147–310		164–324
	16–18 y	165–289		154–344
Triglycerides (mg/dL)	0–12 mo		42–295	
	1–5 y		40–242	
	6–10 y		41–282	
	11–14 y		41–312	
	15–20 y		42–214	
Urea nitrogen (mg/dL)	1–30 d	4–12		3–17
	1–12 mo	2–13		4–14

	1–3 y	3–12		3–14
	4–6 y	3–16		4–14
	7–9 y	4–16		4–16
	10–12 y	5–18		5–16
	13–15 y	7–18		4–15
	16–18 y	5–20		4–15
Vitamin A (retinol) (µg/dL)	Preterm neonates		13–46	
	Term neonates		18–50	
	1–6 y		20–43	
	7–12 y		20–49	
	13–19 y		26–72	
Vitamin B_{12} (pmol/L)	0–1 y	216–891		168–1117
	2–3 y	195–897		307–892
	4–6 y	181–795		231–1038
	7–9 y	200–863		182–866
	10–12 y	135–803		145–752
	13–18 y	158–638		134–605
Vitamin D (as 25-hydroxy D) (ng/mL)[a]	1–30 d	3.3–33.4		1.9–32.0
	1 mo–1 y	7.4–53.3		11.6–48.2
	1–3 y	6.9–46.8		11.3–48.9
	4–12 y	4.6–37.4		2.8–36.7
	13–18 y	2.0–31.4		1.8–28.3
Vitamin E (µg/mL)	Pre-term neonates		0.5–3.5	
	Full-term neonates		1.0–3.5	
	2–5 mo		2.0–6.0	
	6–12 mo		3.5–8.0	
	1–6 y		3.0–9.0	
	7–12 y		4.0–9.0	

	13–19 y		6.0–10.1	
Zinc (µg/dL)	0–5 d		65–140	
	1–5 y		67–118	
	6–9 y		77–107	
	10–14 y	76–101		79–118
	15–19 y	64–104		60–101

[a] Although there is no consensus on optimal 25-hydroxy vitamin D levels, many experts define deficiency as a serum level less than 20 ng/mL and insufficiency as a serum level less than 30 ng/mL Ref "Evaluation, Treatment, and Prevention of Vitamin D Deficiency: An Endocrine Society Clinical Practice Guideline *Journal of Clinical Endocrinology & Metabolism*, July 2011, 96(7): 1911–1930.
Source: Adapted from Soldin SJ, Wong EC, Brugnara C and Soldin OP, *Pediatric Reference Intervals*, 7th ed. Washington, DC: AACC Press, 2011.

- Age: The use of an appropriate age-matched reference range is particularly important for some analytes, for example, alkaline phosphatase, low-density lipoprotein (LDL) cholesterol (see Table 3-2 for age-related reference intervals of a variety of analytes of potential value in nutritional assessment).
- Sex: After puberty, males exhibit greater activity in muscle enzymes due to increased muscle mass.
- Hydration: Serum concentrations of sodium, the predominant extracellular ion, are influenced to a large extent by extracellular water content (total body stores of electrolytes that are predominantly intracellular, e.g., potassium, phosphate, and magnesium, are also poorly reflected by serum levels).
- Genetic factors: Predisposition to elevated blood lipids, bilirubin, and glucose.
- Physiological factors: Diminished tissue stores of specific nutrients may be masked when blood levels are maintained by homeostatic mechanisms (e.g., calcium); the reliance of vitamin E for transport of LDL particles (vitamin E levels in plasma are strongly correlated with cholesterol concentrations, and it is thus appropriate to correct for these).
- Geographical variation (due to seasonal variation) in plasma levels of vitamin D, the major source of which is sun-exposed skin; also the altitude effect on hemoglobin due to reduced PO_2.

Intraindividual variation refers to temporal differences in true concentrations of any nutrient in a single individual and may relate to time of sampling in relation to factors such as meals, time of day, and season.

Analytes influenced solely or mainly by such controllable factors may make better nutritional indicators. An awareness of the potential influence of these factors on test outcome remains necessary for correct interpretation of test results, for example, when making day-to-day comparisons, blood should ideally be taken at the same time of day. Factors potentially influencing evaluation of laboratory parameters of nutritional status include:

- Diurnal variation of serum iron, zinc, and growth hormone
- Seasonal variation of some vitamins, notably vitamins D and C (latter due to the availability of fruit and vegetables)
- Menstrual cycle: cyclical variations in plasma lipids, proteins, calcium, creatinine, and iron have been noted
- Postprandial elevation of plasma triglycerides, iron, and some amino acids (notably the branched-chain amino acids: leucine, isoleucine, and valine)
- Recent versus longer term dietary exposure (e.g., in assessment of iron status and some vitamins, different tests can reflect different lengths of exposure)
- Exercise, which can affect blood lipids, free fatty acids, serum lactate, blood urea nitrogen, creatinine, and urine constituents, as well as muscle enzymes, depending on duration and intensity of the physical activity
- Obesity: Serum concentrations of blood cholesterol and triglycerides are correlated with fat mass
- Prolonged application of a tourniquet causes potential elevation of serum ionized calcium
- Postural effect on plasma volume: Increased hydrostatic pressure in the upright position causes increased concentrations (up to 8%) of all plasma proteins, including enzymes, hormones, calcium, hemoglobin, and other protein-bound compounds; bed rest has the opposite effect
- Effect of disease (notably when associated with an acute inflammatory process): Metabolic stress has a very significant effect on plasma proteins and some micronutrients (see sections on plasma proteins and micronutrients and Chapter 64: The Critically Ill Child for more detail).

Preanalytical variation refers to differences in the measured level of an analyte due to factors that may influence this at any stage from when the blood sample is drawn to its entrance into an analyzer system, for

example, sample collection, transport, separation, and storage.

- Sample collection proximal to the site of an intravenous infusion ("drip-arm sample") or via an indwelling central venous catheter will result in dilution of the blood with constituents of the infusion and can result in, for example, low sodium, high glucose, high potassium. Ideally an alternative limb should be selected and, if practical, the infusion should be shut off for three minutes prior to obtaining the specimen.
- Stress associated with blood drawing, particularly in children, may result in elevated blood glucose, growth hormone, cortisol, and prolactin. Higher blood lactate and lower pH may also be noted. Physical restraint may cause increased muscle enzyme activity.
- Capillary blood collection or the use of a very fine-bore needle can result in hemolysis that can affect both actual and measured levels of many blood constituents. Other potential interferents include sample lipemia, in patients receiving fat emulsion preparations, and icterus, common in neonates. Prolonged time before sample separation will have an effect similar to sample hemolysis, due to erythrocyte membrane "leakage."
- Appropriate collection tubes are of particular importance in the estimate of blood trace metal levels, which, being of low micromolar concentrations or less, are particularly susceptible to contamination by inappropriate vial constituents, including glass and rubber.
- Some analytes, such as ammonia, blood gases, gastrin, parathyroid hormone, homocysteine, lactate, and pyruvate, are unstable at room temperature and require the blood sample to be transported on ice and separated as soon as possible. This is facilitated by advance notice to the laboratory that the sample is being collected. Vitamin A is light sensitive, so samples need to be transported rapidly to the laboratory while being shielded from light to inhibit degradation.

Analytical variation refers to differences in measured levels of an analyte due to factors involved in the actual biochemical analysis.[4] Issues to consider include analytical sensitivity, specificity, precision and accuracy, and the source of the reference interval. Imprecision may arise due to methodological problems such as fluctuations in ambient temperature or automated fluid dispensing, or bias due to different

methods being used (important to consider when comparing results obtained from different laboratories) or to a problem with a batch of reagents. However, advances in instrumentation and methodology have led to improved precision and sensitivity and have considerably reduced much of the random error associated with biochemical analysis. This is generally the least significant error relating to a given analysis, with a coefficient of variation of <5% for most automated assays. Rigorous checks are made by laboratory staff to ensure that results are not issued unless control specimens fall strictly within an acceptable range of values.

Standardization of Laboratory Tests

Many nutritional biomarkers are poorly standardized so that significant between-laboratory differences may exist. Standard reference materials (SRMs) should be used where available to calibrate manufacturer or laboratory standards. Reference intervals and decision limits derived from published literature may not be valid in the absence of standardization. It is important for all laboratories to maintain good internal quality control, to participate in appropriate external quality assurance programs for each analyte, to ensure that quoted reference intervals are appropriate for each assay that they use, and that guidance is provided to clinicians about correct clinical decision limits.

Close liaison between clinical and laboratory staff will provide advice specific to each laboratory and facilitate accurate interpretation of results. However, some general points may be made, and the remainder of this chapter will review the laboratory assessment of macronutrient status followed by status of individual micronutrients.

SPECIFIC TESTS OF NUTRITIONAL STATUS

Although measurement of a nutrient in a single serum or plasma sample at a single point in time may not always be representative of its body stores, distribution, or functional metabolic significance, it is still the most practical means of evaluating availability of many nutrients. For some (e.g., folate and selenium), serum concentrations are indicative only of recent intake, whereas serum levels of vitamins D and B_{12} reflect total body stores; for others, notably plasma proteins, there is a very poor correlation between adequacy of protein intake and plasma concentrations. Various other approaches have been employed. Functional assays based on measurement of activity of an enzyme, which is dependent on a specific

micronutrient, are useful in some situations. Urine measurement is generally not helpful as excretion of micronutrients is more likely to reflect very recent intake than whole body stores. Measurement of nutrients in red cells, white cells, or other tissue stores (including hair and nails) may be a more reliable index of total body status for some nutrients, but the difficulty in isolating cell populations means that these are not often used in the clinical setting. Analysis of hair and nails is greatly subject to external contamination, for example, by shampoo and is generally not used outside the research setting, where some value may be derived from easier accessibility of samples. Similarly, assessment of stores via metabolic balance techniques and isotopic measures of nutrient status are largely research tools.

Macronutrient Assessment

Protein

Apart from water, protein is the most abundant chemical compound in the body. All human cells and tissues contain fundamental structural and functional proteins. These proteins are in a continuous state of flux, constantly being turned over through synthesis and degradation. In childhood, protein synthesis exceeds degradation so that there is net deposition of protein in new tissue. Newborn infants synthesize about 12g protein/kg/d, which falls to about 6g/kg/d by 1 year of age, and 4g/kg/d by adulthood. Insufficient dietary protein intake in childhood manifests with growth retardation.

There is no way of directly measuring the body's protein content. However, most protein is present in lean tissue (~43% in muscle and 15% in skin, in adult males), and the estimation of fat-free body mass has been used as a surrogate measure.

Somatic Protein

Assessment of lean body mass is most readily carried out in the clinical setting by anthropometric measures such as mid-arm muscle circumference (see Chapter 2: Clinical Assessment of Nutritional Status). It can also be estimated relatively inexpensively by bioelectric impedance, although this is subject to influence by factors including hydrational status. More complex methods include magnetic resonance imaging, underwater weighing, assessment of total body water, total body potassium, and total body nitrogen (see Chapter 4: Body Composition and Growth). Stable isotope studies can also be used to look at relative rates of protein

synthesis and degradation.

Because muscle creatine is directly metabolized to creatinine that is freely excreted in urine, urinary creatinine has been used as a biochemical indicator of total muscle mass. Analysis of 24-hour excretion of creatinine is compared to height-standardized reference values to give a creatinine-height index (CHI), which is expressed as a percentage of the reference value. This quick and inexpensive test has been used as a marker of muscle protein status,[5] although it is also influenced by factors that cause increased protein catabolism and by impaired renal function. A deficit of <15% is classified as mild, 15%–30% as moderate, and >30% as severe depletion in muscle mass. Urinary 3-methyl-histidine excretion may also be used as a marker of tissue breakdown, particularly in the absence of protein intake.

Nitrogen Balance

Nitrogen (N) balance is one of the oldest methods of nutritional assessment. Although subject to well-described limitations,[6] it remains an accessible tool for estimating adequacy of protein intake.

N balance studies have classically been used to define amino acid requirements,[7] since inadequate ingestion of an essential amino acid will result in negative N balance. Children (as well as others who should be actively gaining lean body mass) should normally be in positive N balance, whereas healthy adults may be said to be in nitrogen equilibrium if N loss is within 5% of N intake. Negative N balance can result from inadequate energy intake, inadequate protein intake, or catabolic stress and lean body mass breakdown. Positive N balance implies adequate energy and/or protein intake. It should be noted, however, that the mere demonstration of positive N balance does not disclose any information about protein distribution throughout the body, nor about the accumulation of lean body mass, and this test should not be used in isolation.

Urea is the main excretory product of N metabolism; approximately 85% of the body's N is lost in the urine. Other sources of N loss include fecal losses, integumental losses (e.g., desquamating skin, sweat, hair and nail growth), and miscellaneous losses (e.g., tissue fluid, saliva, vomitus, blood drawing, and menstrual losses). These losses can be high in patients with wounds or burn injuries. Since most proteins contain 16% N, dietary protein intake is customarily divided by a factor of 6.25 to estimate N intake.

The equation for calculating N balance is

$$\begin{aligned}\text{N balance} &= \text{N intake} - \text{N output}\\ &= (\text{24h dietary protien intake in grams}/6.25)\\ &\quad - \text{24 hrs UNN} - \text{Factor}\end{aligned}$$

where UUN is urine urea nitrogen (in grams), and Factor is the allowance made for uncollected N loss in stool, skin, and miscellaneous sources. In adults, this factor is 2–4 g per day, and in children, an estimate of 10 mg/kg/d may be used. If there are other significant unmeasured N losses (such as high output ileostomy), this approximation becomes invalid.

Recent work has focused on the development of stable isotope-labeled amino acid kinetic studies that do not depend on the same assumptions as nitrogen balance studies and may give more accurate estimations of amino acid requirements.[8] However, these techniques are at present confined to the research arena, and the assessment of dietary protein adequacy in the clinical setting remains inherently difficult.

Plasma Proteins

Proteins synthesized by the liver have long been used to assess protein status, decreased blood concentrations being presumed to reflect a reduced supply of amino acid precursors or decreased hepatic (and other visceral) mass. However, plasma proteins are subject to influences more dominant than nutritional status, particularly in the presence of an acute inflammatory process, and also fluid shifts. (See chapter 64: The Critically ill Child for a description of altered protein turnover in metabolic stress.) Plasma proteins may be classified according to whether their concentration increases or decreases in the setting of acute infection or catabolism (see Table 3-3). The concentrations of positive acute-phase proteins are increased in infectious or other catabolic illnesses due to preferential secretion by the liver. Conversely, negative acute-phase proteins, made up of labile protein stores that are catabolized early in starvation complicated by metabolic stress, are decreased in these circumstances. The magnitude of a positive acute-phase response is attenuated in protein–energy malnutrition.[9]

Albumin

Albumin is the most abundant protein in blood and is the most commonly used biochemical marker to assess protein status. However, its use for this purpose has significant limitations. Serum albumin is a poor marker of uncomplicated protein malnutrition (“simple” starvation); it is characteristically normal or even raised in severe advanced anorexia

nervosa. More than half of body albumin is extravascular (primarily in skin and muscle), and maintenance of normal plasma levels can occur from mobilization of these stores despite prolonged dietary protein inadequacy. Combined with its long half-life of 20 days, these factors make serum albumin a relatively insensitive marker of nutritional status and an insensitive measure of nutritional recovery. Nonetheless, hypoalbuminemia is a reasonable predictor of morbidity and mortality in hospitalized patients.

Hypoalbuminemia can be seen in situations of decreased synthesis (e.g., liver disease), increased losses (e.g., nephrosis, protein-losing enteropathy, burn injuries), or increased losses to extravascular spaces (e.g., acute catabolic stress with capillary leak). Fluid overload can also dilute albumin concentrations, and bed rest can decrease by 0.5 g/dL over several days due to redistribution into extracellular fluid.

TABLE 3-3 Plasma Proteins and the Acute Phase Response

Positive Acute Phase Proteins	Negative Acute Phase Proteins
C-reactive protein	Albumin
Fibrinogen	Prealbumin
Haptoglobin	Retinol-binding protein
Ferritin	Transferrin
Ceruloplasmin	
Alpha-1-antitrypsin	
Alpha-1-glycoprotein	

Prealbumin

Prealbumin, so named because of its proximity to albumin on an electrophoretic strip, is a transport molecule for thyroxine; hence its alternative name, transthyretin. It circulates in plasma in a 1:1 ratio with retinol-binding protein. It is subject to the same influences as serum albumin concentration, which limits its clinical utility; most notably, it is a negative acute-phase reactant. Like albumin, a single measurement may be considered to be a better indicator of those at a risk of malnutrition than those who are malnourished.[10] However, its short half-life (two days), small body pool, and high ratio of essential to nonessential amino acids make it a better measure of visceral protein status, and it may be more sensitive than albumin as a measure of nutritional recovery. In some studies, changes in prealbumin concentration have correlated with changes in nitrogen balance,[11] and it has been suggested that a weekly increase of

>40 mg/L in prealbumin concentrations reflects a switch away from hepatic synthesis of acute-phase proteins to visceral proteins.[12]

Retinol-Binding Protein

Retinol-binding protein (RBP) has similar properties to prealbumin in that it has a small body pool and a rapid response to protein–energy depletion and repletion. Its half-life is 12 hours. Since RBP is excreted by the kidneys, levels are high in renal failure. RBP levels drop in vitamin A deficiency and, as with albumin and prealbumin, with infectious or other catabolic stresses.[13]

Transferrin

Transferrin is another plasma protein sometimes used to assess visceral protein status. It is synthesized primarily in the liver and has a half-life of eight days. Transferrin concentrations are decreased in all situations, which lower serum albumin, as well as with steroid therapy, iron overload, and anemia of chronic disease. Increased concentrations are seen in pregnancy, oral contraceptive use, and iron deficiency anemia.

Reference ranges for these plasma proteins are given in Table 3-2.

Other Biochemical Indicators of Protein Status: Amino Acid Profiles

Quantitative amino acid analysis can be carried out by laboratories that possess an amino acid analyzer. In practice, actual amino acid concentrations in plasma are not as helpful as they might seem, since individual amino acids are subject to considerable physiological variation and their levels poorly reflect release or uptake by tissues. However, concentrations of the essential branched-chain amino acids such as leucine, isoleucine, and valine tend to be lower in protein–energy malnutrition and relatively higher in infection and injury. This may reflect the fact that branched-chain amino acids, unlike most others, are only minimally metabolized by the liver and are therefore available for muscle protein synthesis, which is not prominent in infection or injury.

Energy

Quantitative assessment of energy status of an individual is largely an exercise in metabolic balancing in that an estimation of energy requirements, based on measured expenditure, is compared to dietary calorific intake. It is also informative to have some perception of which

specific nutrients the body is using for energy sources, and the interpretation of this requires an understanding of how energy is normally provided. For detailed information on energy metabolism and expenditure, see Chapter 13: Energy Metabolism.

Energy Production From Nutrients

In humans, all energy is supplied by the hydrolysis of phosphate bonds in adenosine triphosphate (ATP). This is provided by the following related biochemical pathways:

1. Glycolysis (Embden–Meyerhof pathway)
2. Tricarboxylic acid (Krebs) cycle
3. Electron transport (respiratory chain)
4. Beta-oxidation of fatty acids

The substrate for glycolysis is glucose, either exogenous, from dietary carbohydrate, or derived from the breakdown of glycogen stores. The usual substrate for the Krebs cycle is acetyl CoA derived from glycolysis via pyruvate, and the Krebs cycle provides substrate for ATP production via respiratory chain reactions. However, fatty acid and some amino acids can also enter the Krebs cycle via acetyl CoA when intake is surplus or if there is a deficit in glucose availability through dietary insufficiency or depletion of glycogen stores. The main source of energy in a healthy diet is glucose derived from carbohydrate, with a smaller contribution from fat and a minor contribution from protein. Diets consisting of excess fat or protein in relation to carbohydrate will provide a greater proportion of energy in the form of fatty acids and some amino acids. Inadequate provision of energy from dietary sources triggers the mobilization of fat stores and nitrogen wasting for energy production.

Estimates of Energy Requirements

Plasma glucose, which is normally regulated by insulin and other hormones, is not a good indicator of energy status. Hypoglycemia in childhood is not a nutritional disorder but an indication for metabolic investigation. Hyperglycemia, which may be accompanied by glycosuria, may be due to frank diabetes mellitus or to a more temporary state of insulin resistance, not infrequently noted in acute metabolic stress and/or in association with parenteral feeding.

In estimating whether an individual's energy intake meets requirements, it is generally more helpful to use nonlaboratory-based procedures to assess energy expenditure. Total energy expenditure (TEE)

equals the sum of energies of physical activity, growth, obligatory losses of energy in stool and urine, diet-induced thermogenesis, and the basal metabolic rate (BMR). There are several equations for estimating energy expenditure (see Appendix II), although all are associated with a significant margin of error.

Estimates of TEE, however, are of limited value in critically ill patients, in whom formal assessment of resting energy expenditure (REE), which approximates BMR, is likely to be of most relevance. Indirect calorimetry (IC) estimates resting energy expenditure based on oxygen consumption and carbon dioxide production, providing a real-time, reproducible measurement in an individual patient with an acceptable degree of accuracy. Meticulous attention must be paid to potential confounding factors, particularly in ventilated patients. This procedure can be carried out at the bedside in a few minutes using portable gas analyzers. Recent data have suggested that targeting ICU patients at a high risk of metabolic alterations is an efficient manner of utilizing IC.[14]

Clinical Utility of the Respiratory Quotient. Calculation of the respiratory quotient (RQ), which is the ratio of carbon dioxide production to oxygen consumption, also helps to predict energy balance in addition to giving some insight into the type of fuel being utilized for energy production. Specific fuels have specific and constant respiratory quotients. Oxidation of glucose yields an RQ of 1.0, while oxidation of fat and proteins yield RQs of 0.7–0.8. With lipogenesis, the RQ can exceed 2.0; therefore, any measured RQ > 1.0 suggests that calories are being provided in excess of requirements.

Although the human body stores energy mainly in the form of fat, normal diets supply most of the energy required as carbohydrate (50%–55%), with a smaller contribution from fat (25%–40%), and only approximately 10% or less from protein. In starvation, the body tries to compensate for a lack of nutrient intake by reducing energy expenditure to divert substrate into maintaining essential functions such as the central nervous system. Initially, energy is provided by glycogen stores and amino acids derived from proteins, but the fuel source then switches primarily to fatty acids, ketones, and glycerol, with reduced protein catabolism to conserve lean body mass. This results in a marked production of ketone bodies, with a less significant urinary nitrogen loss. IC would give a low respiratory quotient of 0.6–0.7, indicating that the primary fuel source is fat. In contrast, however, in a hypermetabolic state, energy expenditure may be dramatically increased and catabolism may be rampant, with

indiscriminate concurrent breakdown of glycogen stores, body fat, and lean body mass. This state is indicated biochemically by significant urinary nitrogen losses with relatively less dramatic ketone body production. A higher RQ of 0.8–0.9 indicates a mixed fuel source.

Overfeeding Syndrome and the RQ. Postoperative stress, sepsis, trauma, burns, and chronic disease may sometimes be associated with a hypermetabolic state in which resting energy expenditure may be considerably increased. However, the nature of the metabolic stressor causes a variable response, even among subjects with similar type and severity of injury, and any increased energy demands may be offset by decreased physical activity. Accurate assessment of requirements is necessary, not only to meet increased needs with the aim of limiting protein catabolism for energy production but also to avoid causing hyperglycemia, which is often apparent in hypermetabolic patients, particularly in those receiving carbohydrate supplementation. This results in lipogenesis, as indicated by a high RQ (>1.0) and causes substantially increased CO_2 production. Overfeeding has been implicated in respiratory compromise, causing ventilatory dependence, hepatic steatosis, and increased risk of mortality. Patients with severe injury and those at extremes of life, that is, infants, especially preterm infants, and the elderly, are at particular risk.

Lipids and Fatty Acids

Fatty acids are classified according to carbon chain length and the presence or absence of double bonds. Long chain fatty acids (LCFAs) are the most common fatty acid configuration found in nature and have a backbone of 12–20 carbon atoms. Saturated LCFAs, which have no double bonds (e.g., stearic acid, C18:0), and monounsaturated LCFAs, which have one double bond (e.g., oleic acid, C18:1), can be synthesized in vivo from medium-chain fatty acids and are found in large quantities in adipose tissue triglycerides, where they constitute an important energy reserve. Both excess and insufficient intake of dietary fat can cause clinical problems, although in developed countries, by far the greater problem results from overconsumption of energy-dense foods with high saturated fat content. Part IV of this textbook comprehensively details the clinical consequences of pediatric obesity.

Plasma lipids and lipoproteins are poor indicators of nutritional status, being strongly influenced by genetic and environmental factors. Although plasma lipids tend to rise in obesity, elevations are also associated with

disease states, including hepatic, renal, and thyroid disorders, as well as diabetes mellitus. In addition, plasma low-density lipoprotein, which transports cholesterol, decreases in response to acute metabolic stress, with an accompanying drop in plasma total cholesterol. Normal and abnormal lipid metabolism is discussed in Chapter 65: Dyslipidemia.

Clinical deficiency states of essential fatty acids (EFAs) have been well documented. Although polyunsaturated long-chain fatty acids can also be mobilized for energy, their main physiological functions are structural and metabolic, for example, synthesis of cholesterol and some hormones, cell wall phospholipids, fat-soluble vitamins, and prostaglandins. LCFAs are further classified as ω-9 (omega-9, n-9), ω-6, or ω-3, depending on the number of carbon atoms between the terminal methyl group and its nearest double bond (see Chapter 7: Fats and Fatty Acids). The human organism can introduce double bonds in the ω-9 position only; ω-6 and ω-3 fatty acids must be supplied preformed in the diet and thus are essential nutrients. The EFAs in humans are linoleic acid ($C_{18:2}$ ω-6) and linolenic acid ($C_{18:3}$ ω-3), which by chain elongation can be metabolized, respectively, to arachidonic acid (AA; $C_{20:4}$ ω-6) and to eicoisapentanoic acid EPA ($C_{20:5}$ ω-3) and docosahexaenoic acid (DHA; $C_{22:6}$ ω-3), although these are more efficiently provided in the diet and are termed conditionally essential fatty acids. Arachidonic acid and DHA comprise the main lipid moiety of neural membrane structure. ω-6 fatty acids are the precursors of eicosanoids and thromboxane and may be classed as proinflammatory, whereas the ω-3 group is the basis for leukotrienes and some cytokines and may be classed as anti-inflammatory.

Infants are much more susceptible to EFA deficiency due to their limited body stores and rapid growth, resulting in high requirements. Three relatively common clinical scenarios of EFA deficiency are (1) prolonged fasting, or reliance on lipid-free parenteral nutrition, (2) extended use of a formula with a predominance of medium-chain fatty acids, and (3) fat malabsorption. The clinical signs of linoleic acid deficiency include poor growth and a desquamating skin rash, or redness and oozing in skin creases (deficiencies of zinc and biotin should also be considered with such skin features); however, it is much more common to diagnose EFA deficiency by biochemical profile.

Laboratory assessment of fatty acid status is complex. Most individual fatty acids can now be measured using sophisticated chromatographic techniques; results are generally expressed as a percentage of the total fatty acids. When EFA deficiency occurs, the nonessential fatty acid,

eicosatrienoic acid ($C_{20:3}$ ω-9) increases in serum; its three double bonds make it a "triene."Conversely, arachidonic acid ($C_{20:4}$ ω-9, i.e., a "tetraene") is reduced. A triene to tetraene ratio of >0.4 is consistent with EFA deficiency.[15] However, clinical features may be apparent at this level, and some laboratories use a lower cutoff.

Carnitine

Carnitine is a small, water-soluble molecule recognized to play two major physiological roles: (1) facilitating the transport of long-chain fatty acids, as acylcarnitines, into the mitochondrion for generation of energy and (2) facilitating the removal of short- and medium-chain fatty acids that accumulate inside the mitochondrion. The main dietary sources of carnitine are red meat and dairy products, but it can be synthesized endogenously from peptide-bound lysine and methionine. In strict vegetarians, this latter source provides more than 90% total available carnitine. It is stored mainly in the muscle (98%); the liver stores comprise 1.6% and a very minor proportion exists in extracellular fluid.[16]

Carnitine is generally considered a conditionally essential nutrient, although a clinical deficiency state has not yet been well defined in subjects who do not possess one of the rare primary inherited defects. Acquired carnitine deficiency is most likely to develop where there is reduced intake, reduced synthesis, increased requirements, or increased excretion. Both strict and lacto-ovo vegetarian children have lower plasma carnitine than their omnivorous peers (about 30% and 20%, respectively). Newborn infants, who depend heavily on lipids as a concentrated fuel source for rapid growth in early life, also have relatively low plasma and tissue carnitine, probably due to immature synthetic pathways and a lower renal threshold for excretion.[17] This may be of particular relevance in preterm infants, whose carnitine stores are very low compared to term infants. Critical illness has been reported to cause increased carnitine excretion, and parenteral nutrition formulas do not normally contain carnitine. Thus, the preterm sick infant who is maintained exclusively on intravenous feeding is likely to be at a greatest risk for functional carnitine deficiency, which may conceivably manifest with defective energy-dependent processes (e.g., hypoglycemia, respiratory distress, delayed growth, hyperlipidemia, and developmental delay). However, administration of carnitine to preterm infants has not been shown to result in improved weight gain compared to controls.

Plasma carnitine, constituting a very small proportion of total body

stores, does not necessarily reflect tissue levels, though plasma and urine are the most amenable to sampling. Carnitine is present both as free carnitine and bound to fatty acids as acylcarnitine esters. Both total and free carnitine must be measured, since reductions in total and/or free acylcarnitines, as well as altered ratios, have been described in both hereditary and acquired carnitine deficiencies. The recent advent of tandem mass spectrometry has enabled the identification of abnormal acylcarnitines, which has been of great value in expanded newborn screening for inherited metabolic disorders and may also help broaden our understanding of disordered carnitine metabolism.

Micronutrients and Major Minerals

Classical deficiency states of most micronutrients are well described but rare. See Chapter 8: Trace Elements and Chapter 10: Vitamins. Much more common are suboptimal levels that are not overtly associated with disease, and it may be in this arena that the value of laboratory assessment of nutrient status is greatest.

Childhood micronutrient requirements are weight and age dependent. Although tables of reference intake in health are widely available, little is known about the effects of disease on whether, how, and to what extent requirements might change.

Vitamin deficiency is more common among lower socioeconomic groups and those on a very restricted diet for therapeutic (e.g., phenylketonuria management) or other reasons; in adulthood, alcoholics and the elderly are at relatively greater risk. A recent observation has been the development of fat-soluble vitamin deficiency in aggressive obesity management either by pharmacological pancreatic lipase inhibition (orlistat, now sold over the counter) or by pancreatico-biliary diversion. Bariatric surgery for severe obesity associated with comorbidities in adolescence is increasingly recognized to be more effective than medical management. Data concerning late nutritional complication rates in children are not yet available, but it seems likely that the same spectrum of micronutrient deficiencies will be apparent in children as are recognized in adults after bariatric surgery (particularly when intestinal bypass is employed). In addition to fat-soluble vitamin deficiency, decreased vitamin B_{12}, iron, calcium, thiamine, and folate have all been recognized to occur.[18]

Clinical aspects of micronutrient and mineral deficiencies are given in detail in Chapter 8: Trace Elements, Chapter 9: Iron, and Chapter 10:

Vitamins. This chapter focuses on laboratory assessment of total body status of vitamins and specific issues that may influence interpretation of results. Similar descriptions relating to assessment of trace metals are given in Chapter 8, while Chapter 9 comprehensively details the laboratory assessment of iron status.

Reference intervals for vitamins and minerals, as well as various other laboratory nutritional parameters, are given in Table 3-2. As with plasma proteins, when interpreting plasma levels of micronutrients, it is important that care is exercised with results obtained during metabolic stress. In addition to the potential for whole body deficiencies of specific micronutrients via reduced intake, increased requirements, and increased losses, it is widely accepted that acute illness results in difficulty with interpretation of serum levels of micronutrients due to redistribution of vitamins and trace elements for various reasons, including acute-phase response of some carrier molecules (see Table 3-4). Monitoring of trends and magnitude of the acute-phase response (e.g., by measuring C-reactive protein concentrations) will facilitate interpretation of blood micronutrient levels. It has been suggested that plasma micronutrient concentrations can only be reliably interpreted if the C-reactive protein is <20 mg/L (plasma zinc), <10 mg/L (plasma selenium and vitamins A and D), or <5 mg/L (vitamins B_6 and C).[19]

TABLE 3-4 Effects on Plasma Concentrations of Micronutrients by Metabolic Stress

Micronutrient	Direction of Change	Reason for Change
Iron	Decreased	↓ Transferrin
Copper	Increased	↑ Ceruloplasmin
Zinc	Decreased	↓ Albumin; redistribution
Selenium	Decreased	↓ Selenoprotein P
Vitamin A	Decreased	↓ Retinol-binding protein
Vitamin B_6	Decreased	? Redistribution
Vitamin C	Decreased	↑ Transfer into tissues
Vitamin D	Decreased	? Redistribution
Vitamin E	Decreased	↓ LDL

In circumstances that make interpretation of serum concentrations difficult, indicators of function may be better predictors of status. Changes in serum concentration of micronutrients may occur rapidly in response to

recent ingestion or in the presence of the acute-phase response, whereas tissue stores or functional indicators may remain adequate. For examples, see iron, B vitamins, vitamins D and K, selenium, and copper.

Fat-Soluble Vitamins

Vitamin A

Vitamin A includes retinol/retinal/retinoic acid/retinyl ester, which are fat-soluble 20-carbon compounds that differ in their chemical group at the C15 terminal. They are yellowish oils, which are sensitive to oxygen and ultraviolet light. Retinol, the principal active form of vitamin A, can undergo reversible oxidation to the biologically similar retinal; further oxidation leads to retinoic acid that has less biological activity. Vitamin A is stored principally as retinyl palmitate. Some dietary carotenoids (~50 of them have been identified) are classified as provitamin A, since they are cleaved to produce retinol; these include β-carotene, α-carotene, and β-cryptoxanthine. The most common carotenoid in human plasma β-carotene is also the most effective provitamin A; it is a water-insoluble orange or purple solid that oxidizes in air to inactive metabolites.

The most widely used laboratory assessment of vitamin A status is high-performance liquid chromatography (HPLC) using plasma or serum.[20] Plasma concentration does not fall off until hepatic stores are critically low, so it is a late indicator. Compared to earlier spectrophotometric methods, HPLC is highly accurate, reproducible (between-batch coefficient of variation [CV], a maximum of 10%), specific, and sensitive (lower limit of detection <0.07 μmol/L). It also has the advantage that β-carotene and vitamin E can be measured at the same time. RBP has a close relationship with serum retinol levels and its use has been suggested as an alternative biomarker of vitamin A status that can be easily measured by immunoassay.[21]

Vitamin D (25-OH-Cholecalciferol)

Vitamin D status is best assessed by measuring serum levels of 25-OH cholecalciferol.[22] 25-OH D_2 and D_3 are generally considered biologically equivalent, so it is important to use an assay that measures both forms. The predominant form may vary in different countries, depending on supplement use and the preponderance of different dietary sources. Recently, epi- forms of both 25-OH D_2 and D_3 have been recognized (stereoisomers differing at only one stereo center). Their biological significance and activity is currently uncertain, but they generally account

for less than 10% of total serum cholecalciferol, although the proportion may be greater in children.[23] Clinical cutoffs for deficiency and insufficiency have been suggested in a number of guidelines, although this remains a controversial area. Vitamin D metabolites have recently been recognized as negative acute-phase reactants,[24] and levels show significant seasonal variation in nonequatorial countries depending on sunlight exposure, and these factors need to be taken into account when interpreting levels.

25-OH cholecalciferol can be measured by immunoassay or by liquid chromatography-mass spectroscopy (LC-MS). LC-MS is considered to be the gold-standard approach and allows the separation of the various forms of circulating vitamin D described earlier, if this is desired. LC-MS can also be utilized to measure the biologically active form of vitamin D (1,25-OH cholecalciferol), although the clinical usefulness of this measurement is not proven.[22]

Immunoassays show variable accuracy and bias in relation to MS-based assays. Depending on the antibodies used, immunoassays may differ in how they recognize the various forms of circulating vitamin D, and this in part explains the differences between them.[25] It is particularly important for laboratories to be aware of the accuracy of the immunoassay they use at critical decision limits.

Vitamin E (alpha-Tocopherol)

Vitamin E status is best assessed by measurement of circulating levels of alpha-tocopherol, the predominant form of vitamin E in serum. Gamma-tocopherol is present in serum in smaller amount, and other circulating tocopherols and tocotrienols are generally insignificant. Vitamin E circulates mainly within lipoproteins, of which low-density lipoprotein is the most prominent, and levels are closely related to serum lipids. Vitamin E is a negative acute-phase reactant because lipoprotein levels fall significantly as part of the acute-phase response. Measurement of serum cholesterol may therefore aid in the interpretation of vitamin E measurements, and the expression of results as an alpha-tocopherol:cholesterol ratio has been proposed.[26] Vitamin E is measured using a chromatographic approach, most commonly high- performance liquid chromatography, where alpha- and gamma-tocopherol are measured along with retinol and carotenoids.[27]

Vitamin K

Phylloquinone (K_1) is the main vitamin K produced by plants and is the main human dietary form. Vitamin K_2 (menaquinone) is produced by colonic gram-positive bacteria, although its contribution to vitamin K status is not clear. Synthetic vitamin K used in human nutrition is mostly derived from vitamin K_3 (menadione). Its relatively low plasma concentration (~1000-fold less than vitamins A and E) has caused difficulties with direct laboratory measurement, and the pragmatic solution has been to utilize functional measurement.[28]

The role of vitamin K has been best defined as a cofactor in the gamma-carboxylation of glutamyl residues in specific proteins to gamma-carboxyglutamyl (gla) residues. Important gla proteins include prothrombin as well as other clotting factors, which have led to measurement of clotting time, primarily prothrombin time (PT), as a proxy for vitamin K status. This is assessed by adding tissue thromboplastin to recalcified plasma, and the clotting time is compared with a normal control sample. Normal PT is 10–14 seconds: in vitamin K deficiency, this will rise by at least two seconds and can be greater than 30 seconds. Immunological measurement of undercarboxylated prothrombin (protein induced by vitamin K absence: PIVKA II) is more sensitive than tests of clotting and may be useful in the diagnosis of vitamin K deficiency that leads to bleeding in neonates.

Another gla protein, osteocalcin, which is present in bone matrix, has been shown to be affected by vitamin K deficiency; although any clinical implications for bone health are not yet clear, measurement of undercarboxylated osteocalcin (ucOC) may be an even better marker of vitamin K status. Direct measurement of plasma phylloquinone by HPLC[29] shows the best correlation with dietary intake, but requires several steps and is laborious, and thus not widely used. Between-batch CV is typically 11%–18%, with lower limit of detection around 50 pmol/L. Some relation to plasma triglycerides has been reported, and it may be useful to report vitamin K as a ratio to triglycerides.

Water-Soluble Vitamins

Vitamin B_1 (Thiamine)

The vitamin B_1 family consists of thiamine and its phosphate esters: thiamine monophosphate (TMP), thiamine diphosphate (TDP), and thiamine triphosphate. The principal biologically active form of thiamine is the diphosphate, TDP, a required coenzyme for the essential

decarboxylation reactions by which carbohydrates, fats, and alcohol are metabolized to produce energy. The majority of the total thiamine content of whole blood is found in erythrocytes as TDP. Because TDP concentrations are depleted from erythrocytes at a rate similar to depletion from other organs, measurement of TDP in erythrocytes or whole blood provides a good indication of the adequacy of body stores.

Historically, the most widely used test of thiamine status was measurement of the activity of the thiamine-dependent enzyme transketolase in red cells or whole blood and its increase when additional thiamine is added.[30] From these two measures, an activation coefficient can be calculated, with the normal range for erythrocyte transketolase activation coefficient (ETKAC) being <1.25. Currently, if a biochemical measurement of thiamine status is required, the measurement of thiamine diphosphate in whole blood or erythrocytes is generally preferred, most frequently by HPLC.[31] However, if thiamine deficiency is suspected, administration of thiamine should not be delayed while waiting for the results of biochemical tests.

Vitamin B_2 (Riboflavin)

Riboflavin is essential for the function of flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD), which act as coenzymes for a wide variety of oxidative enzymes that remain bound to the enzymes during the oxidation-reduction reactions. Riboflavin status is most commonly assessed using the erythrocyte glutathione reductase activation coefficient assay (EGRAC).[32] EGRAC reflects the availability of the FAD cofactor for glutathione reductase in red blood cells. Although EGRAC is generally considered to be a functional measure of riboflavin status, the exact relationship between EGRAC values and any specific function has not been well established. An activity coefficient (EGRAC)>1.30 is indicative of biochemical riboflavin deficiency. Riboflavin and its coenzyme derivatives, flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD), can be detected with high sensitivity using high-pressure liquid chromatography,[33] although these measures are not widely used for clinical purposes. As for thiamine, if deficiency is suspected, administration of riboflavin should not be delayed while waiting for the results of biochemical tests.

Vitamin B_3 (Niacin)

Niacin is required for the function of nicotinamide adenine dinucleotide

(NAD), and related metabolites and severe deficiency lead to the clinical condition of pellagra.

When biochemical assessment of niacin status is required, this has been most frequently achieved by the quantification of the major urinary niacin metabolites 1-methylnicotinamide (1-MN) and 1-methyl-2-pyridone-5-carboxamide (2-PYR), and sometimes the more minor metabolite 1-methyl-4-pyridone-5-carboxamide, typically by HPLC in timed urine collections.[34] Niacin status is usually expressed as the concentration ratio 2-PYR:1-MN or as the concentrations of the individual metabolites relative to creatinine. Spot urine samples may also be more conveniently used to assess deficiency. Measurement of whole blood or erythrocyte NAD or NADPH has also been recommended, using enzymatic assays, but some studies have suggested that these are a less reliable indicator of niacin deficiency.[35]

Vitamin B_6

Pyridoxal phosphate (PLP) is the main active form of vitamin B_6 and is involved as a cofactor in a wide range of enzymatic reactions. Measurement of PLP in serum of plasma is currently the most widely used biochemical measure of B_6 status,[36] with values <10 nmol/L generally considered to be indicative of vitamin B_6 deficiency. Urinary B_6 metabolites, specifically urinary 4-pyridoxic acid, have been utilized more rarely. Erythrocyte alanine aminotransferase (EALAT) activity coefficient was used historically but has largely been replaced by plasma PLP measurement.

Vitamin B_{12}

The serum concentration of vitamin B_{12} which constitutes true insufficiency is poorly defined. Macrocytosis is a well-known but late sign of B_{12} deficiency. Vitamin B_{12} is a cofactor in the remethylation of homocysteine, and a raised plasma homocysteine can demonstrate a metabolic “block” due to B_{12} deficiency. However, B_{12} is not the only cofactor involved in this pathway: folate deficiency in the presence of a common genetic polymorphism will also result in a raised homocysteine. A more specific functional test of B_{12} deficiency is urinary methylmalonic acid: this metabolite is not normally seen, but appears in urine if there is a block in the conversion of methylmalonyl CoA to succinyl CoA, for which

B_{12} is the only cofactor.[37] However, the most sensitive indicator of B_{12} status is currently considered to be serum holotranscobalamin measured by immunoassay, using a locally determined cutoff which is assay-dependent.[38]

Folate

Folate status is most commonly assessed by measurement of serum folate, although red cell folate may also be used.[39] Red cell folate reflects longer term folate status, but also shows greater analytical variability due to the sample preparation steps that are required. In addition, the red cell assay is more expensive and less convenient, and genetic variation of methylenetetrahydrofolate reductase (MTHFR) influences red cell folate values significantly.[40] For all of these reasons, serum folate measurement is the most widely used approach.

Vitamin C

Plasma ascorbate is the most commonly used biomarker of vitamin C status and can be measured by either enzyme assay with fluorimetric detection[41] or chromatographic techniques, most commonly HPLC.[42] Ascorbate is unstable in serum or plasma, so samples need to be stabilized by the addition of metaphosphoric acid shortly after sample collection before analysis. Clinical symptoms of vitamin C deficiency are rare if plasma ascorbate is >10 μmol/L. Leucocyte ascorbate has been suggested as a better indicator of body stores of vitamin C but is much more cumbersome to measure, and reference intervals are not well described.

Minerals and Trace Elements

Assessment of the status of key transition elements, including iron, selenium, zinc, copper, molybdenum, and manganese is described in the subsequent chapters.

Other Laboratory Indicators of Nutritional Status

Hematological Tests

The complete blood count is readily available and may yield useful early, though nonspecific, nutritional indicators.

Anemia may be attributable to deficiencies of iron, vitamin B_{12}, or folate, or a combination, and red cell indices may be helpful in discriminating between these. The total lymphocyte count (TLC), the

product of the total leucocyte count and the percentage of lymphocytes on a peripheral blood film, has been correlated with general nutritional status. Decrease in TLC to <15,000/uL is frequently noted in malnutrition and may be the only abnormal laboratory parameter noted in uncomplicated malnutrition, such as due to anorexia nervosa. It has been suggested that TLC 800–1200/uL represents a moderate nutritional deficit, while TLC < 800/uL indicates severe malnutrition.[43] However, lymphopenia is not specific for malnutrition and is not invariably present.

Immunological Tests

Cutaneous anergy— delayed or absence of response to subcutaneous administration of antigens such as *Candida albicans*, tetanus, diphtheria, and so forth—is consistently to be demonstrated in moderate to severe malnutrition, although lack of specificity is again an issue, as well as the requisite delay in result. There is growing recognition of the specific effects of nutrient status on immunocompetence, and sophisticated immunological testingis beginning to find a role in nutritional research, particular in intervention studies.[44]

Immune defense mechanisms can be organized into three main groups: physical defenses (mucosal epithelium and secretions), cellular responses, and humoral or circulating defenses (i.e., antibodies). Innate immunity comprises the generic cellular responses, including phagocytes, neutrophils, natural killer cells, and the complement system as well as the mucosal barriers to pathogens. Adaptive immunity is derived from the host response to recognition of nonself-antigens and involves B lymphocytes, which generate circulating antibodies, and T lymphocytes, which mediate cellular immunity resulting in immunological memory. Specific micronutrients have been shown to work in synergy to support these different components[45] (Table 3-5).

Despite the important roles that individual micronutrients play in maintaining immunocompetence, specific tests of immune competence, as stand-alone observations, have not found an important role in nutritional assessment. Although diet is one of the major exogenous factors involved, immune function tests are also influenced by a spectrum of endogenous and other exogenous factors unrelated to nutritional status, such as gender, age, lifestyle (e.g., smoking/exercise), psychological stress, and hormonal factors (e.g., menstrual cycle).[46] Also, the clinical relevance of small changes in single markers of immune function is far from clear: there is considerable redundancy in the immune system, such that change in functional capacity of one component due to nutritional deficiency may be

compensated for by a change in the functional capacity of another which is not dependent on the deficient nutrient.

TABLE 3-5 Specific Micronutrients Involved in Different Groups of Immune Defense Mechanisms

Mucosal Barriers	Cellular Defenses	Antibody Production
Vitamin A	Vitamin A	Vitamin A
Vitamin C	Vitamin B_6	Vitamin B_6
Vitamin E	Vitamin B_{12}	Vitamin B_{12}
Zinc	Vitamin C	Vitamin D
	Vitamin D	Vitamin E
	Vitamin E	Folic acid
	Folic acid	Zinc
	Iron	Copper
	Zinc	Selenium
	Copper	
	Selenium	

The observation that specific dietary constituents may exert a variety of positive effects on the immune system has led to investigation over the past 1–2 decades of the potential role of "nutritional immunomodulation" in ill patients. As a result, immune function tests may find a more useful role in the assessment of the effects of dietary manipulation on immune response. Such tests include in vivo measurement of response to controlled exposure to attenuated or killed microorganisms; this functional assessment has clear clinical relevance. Ex vivo assessments of immune function involve in vitro assays following in vivo dietary modification; although of less direct clinical relevance, these assays may shed light on mechanisms of effects noted in in vivo experiments and may in addition be more sensitive. Measurement of circulating cells and factors give less functional information in that they do not assess immune response to a challenge. A review by Albers et al.[46] scores a variety of immune function markers for their usefulness, based on biological relevance, biological variability, and practical feasibility and gives most weight to four in vivo markers and least to circulating markers. It is recommended that in vivo assessments are used and can be supported by the medium-scoring markers that will give additional information. If only the medium-scoring markers

are available, full combinations of markers should be used to provide the most reliable conclusions.

Emerging Technologies

The emergence of genome-based approaches may in years to come revolutionize the approach to the assessment of nutritional status.[47] However, currently, these techniques are mainly utilized for research purposes. Given the broad impact of many dietary constituents on physiological function, an integrated approach that includes assessment of the genome, epigenome, transcriptome, proteome, and metabolome may ultimately be required. This will present very considerable challenges in terms of data handling and interpretation, which will require substantial processing power and intelligent systems to aid in interpretation.

Genomics

Genetic variation may influence absorption, metabolism, elimination, or biological effects of nutrients.[48] For instance, genetic variation of the methylenetetrahydrofolate reductase gene significantly affects red cell folate status at a given level of folate intake. In turn, the intake of key nutrients may impact on gene expression, altering the transcriptome or epigenome through, for instance, altered methylation (in the case of folate). In the future, it is possible that genotype-specific dietary recommendations may be made.

Proteomics

As a consequence of impact on gene expression, intake of key dietary constituents impacts on the proteome and patterns of protein expression may act as dietary signatures. Dietary components may influence protein expression or posttranslational modification of proteins, and limited availability of cofactors may lead to modification of protein function, including enzyme activity, regulatory properties, or molecular recognition.[49]

Measurement of expression or activity of key individual proteins, through activity assay (for instance, glutathione reductase for selenium) or measurement of mass by immunoassay (for instance, holotranscobalamin in the case of vitamin B_{12}) is well established. Broader assessment of the proteome through mass spectroscopy-based techniques or two-dimensional gel electrophoresis is currently mainly a discovery technique and is limited by relatively poor reproducibility and high coefficients of variation.

Quantitative mass spectroscopy using stable isotope-labelled standards is beginning to gain traction for individual proteins but is currently beyond the scope of most routine laboratories.

Metabolomics

The metabolome is particularly attractive as a marker of dietary intake and nutritional status, since there is a close relationship between diet and metabolism. Large numbers of metabolites can be measured simultaneously using modern analytical approaches, and alterations in specific metabolites or patterns of change characteristic of certain nutritional deficiencies or excess might be easily identified.[50]

Important challenges in relation to assessment of the metabolome include defining preanalytical conditions for sample collection, and the influence of metabolites of pharmaceutical agents, particularly in the context of polypharmacy. In general, currently, urine is the preferred matrix rather than serum or plasma. Two main analytical approaches are used—mass spectrometry and nuclear magnetic resonance.

CONCLUSION

Laboratory testing is a significant component of nutritional assessment but is best interpreted in conjunction with clinical, dietary, and anthropometric evaluations. Laboratory assessment of nutritional status in children may be most meaningful in the investigation of suboptimal micronutrient status, but also plays an important role in gauging response to nutritional support and detection of complications. Clinicians must be aware of any limitations of specific laboratory tests, including limitations posed by the specific methodologies and instrumentation utilized by a particular laboratory. Appropriate liaison between clinical and laboratory staff allows more effective use of laboratory services, for example, by allowing meaningful discussion regarding test availability, response time, or clinical utility of newer tests and technologies to meet clinical and budgetary requirements.

REFERENCES

1. Detsky AS, McLaughlin AR, Baker JP, et al. What is subjective global assessment of nutritional status? *JPEN J Parenter Enteral Nutr.* 1987;11:8–13.
2. Gidden F, Shenkin A. Laboratory support of the clinical nutrition service. *Clin Chem Lab Med.* 2000;38(8):693–714.
3. Young DS. Preanalytical variables and biological variation. In: Burtis CA,

Ashwood ER, Bruns DE, eds. *Tietz Textbook of Clinical Chemistry and Molecular Diagnostics.* Missouri: Elsevier; 2012.
4. Blanck HM, Bowman BA, Cooper GR, et al. Laboratory issues: use of nutritional biomarkers. *J Nutr.* 2003;133:888S–894S.
5. Scrimshaw NS, Viteri FE. INCAP studies of kwashiorkor and marasmus. *Food Nutr Bull.* 2010;31(1):3441.
6. Kopple JD. Uses and limitations of the balance technique. *JPEN J Parenter Enteral Nutr.* 1987;11:79S–85S.
7. Joint WHO/FAO/UNU Expert Consultation. Protein and amino acid requirements in human nutrition. *World Health Organ Tech Rep Ser.* 2007;935:1–265.
8. Conley TB, McCabe GP, Lim E, et al. Age and sex affect protein metabolism at protein intakes that span the range of adequacy: comparison of leucine kinetics and nitrogen balance data. *J Nutr Biochem.* 2013;24:693–699.
9. Cruickshank AM, Hansell DT, Burns HJ, Shenkin A. Effect of nutritional status on acute phase protein response to elective surgery. *Brit J Surg.* 1989;76(2);165–168.
10. Shenkin A. Serum prealbumin: is it a marker of nutritional status or of risk of malnutrition? *Clin Chem.*2006;52(12):2177–2179.
11. Casati A, Muttini S, Leggieri C, et al. Rapid turnover proteins in critically ill ICU patients. Negative acute phase proteins or nutritional indicators? *Minerva Anestesiol.* 1998;64:345–350.
12. Bernstein L, Bachman T, Meguid M, et al. Measurement of visceral protein status in assessing protein and energy malnutrition: standard of care. Prealbumin in Nutritional Care Consensus Group. *Nutrition.* 1995;11:169–171.
13. Duggan C, Colin AA, Agil A, et al. Vitamin A status in acute exacerbations of cystic fibrosis. *Am J Clin Nutr*. 1996;64:635–639.
14. Mehta NM, Bechard LJ, Leavitt K, Duggan C. Cumulative energy balance in the pediatric intensive care unit: role of targeted indirect calorimetry. *JPEN J Parenter Enteral Nutr* 2009;33:336–344.
15. Holman RT, Johnson SB, Hatch TF. A case of human linoleic acid deficiency involving neurological abnormalities. *Am J Clin Nutr.* 1981;34:1534–1539.
16. Walter JH. L-carnitine. *Arch Dis Child.* 1996;74:475–478.
17. Shenai, JP, Borum PR. Tissue carnitine reserves of newborn infants. *Pediatr Res.* 1984;18:679–682.
18. Malinowski SS. Nutritional and metabolic complications of bariatric surgery. *Am J Med Sci.* 2006;331(4):219–225.
19. Duncan A, Talwar D, McMillan DC, et al. Quantitative data on the magnitude of the systemic inflammatory response and its effect on micronutrient status based on plasma measurements. *Am J Clin Nutr.* 2012;95(1):64–71.
20. Furr HC. Analysis of retinoids and carotenoids: problems resolved and unsolved. *J Nutr.* 2004;134(1):281S–285S.
21. Fujita M, Brindle E, Rocha A, et al. Assessment of the relative dose-response test based on serum retinol-binding protein instead of serum retinol in determining low hepatic vitamin A stores. *Am J Clin Nutr.* 2009;90(1):217–224.
22. Lai JF, Franke AA. Analysis of circulating lipid-phase micronutrients in humans by HPLC: review and overview of new developments. *J Chromatogr B Analyt Technol Biomed Life Sci.* 2013;931:23–41.
23. Yazdanpanah M, Bailey D, Walsh W, et al. Analytical measurement of serum 25-OH-vitamin D(3), 25-OH-vitamin D(2) and their C3-epimers by LC-MS/MS in infant and pediatric specimens. *Clin Biochem.* 2012. http://dx.doi.org/10.1016/j.clinbiochem.2012.11.030

24. Waldron JL, Ashby HL, Cornes MP, et al. Vitamin D: a negative acute phase reactant. *J Clin Pathol.* 2013;66(7):620–622. doi: 10.1136/jclinpath-2012–201301.
25. Ong L, Saw S, Sahabdeen NB, et al. Current 25-hydroxyvitamin D assays: do they pass the test? *Clin Chim Acta.* 2012 11;413(13–14):1127–1134.
26. Thurnham DI, Davies JA, Crump BJ, et al. The use of different lipids to express serum tocopherol: lipid ratios for the measurement of vitamin E status. *Ann Clin Biochem.* 1986;23(pt 5):514–520.
27. Aust O, Sies H, Stahl W, Polidori MC. Analysis of lipophilic antioxidants in human serum and tissues: tocopherols and carotenoids. *J Chromatogr.* 2001;936(1-2):83–93.
28. Sokoll LJ, Sadowski JA. Comparison of biochemical indexes for assessing vitamin K nutritional status in a healthy adult population. *Am J Clin Nutr.* 1996;63(4):566–573.
29. Azharuddin MK, O'Reilly DS, Gray A, Talwar D. HPLC method for plasma vitamin K_1: effect of plasma triglyceride and acute-phase response on circulating concentrations. *Clin Chem.* 2007;53(9):1706–1713.
30. Mataix J, Aranda P, Sánchez C, et al. Assessment of thiamin (vitamin B_1) and riboflavin (vitamin B_2) status in an adult Mediterranean population. *Br J Nutr.* 2003;90(3):661–666.
31. Lynch PL, Young IS. Determination of thiamine by high-performance liquid chromatography. *J Chromatogr A.* 2000;881(1-2):267–284.
32. Hill MH, Bradley A, Mushtaq S, et al. Effects of methodological variation on assessment of riboflavin status using the erythrocyte glutathione reductase activation coefficient assay. *Br J Nutr.* 2009;102(2):273–278.
33. Petteys BJ, Frank EL. Rapid determination of vitamin B_2 (riboflavin) in plasma by HPLC. *Clin Chim Acta.* 2011;412(1-2):38–43.
34. Terry RC, Simon M. Determination of niacin metabolites 1-methyl-5-carboxylamide-2-pyridone and N-1-methylnicotinamide in urine by high-performance liquid chromatography. *J Chromatogr.* 1982;232(2):261–274.
35. Creeke PI, Dibari F, Cheung E, et al. Whole blood NAD and NADP concentrations are not depressed in subjects with clinical pellagra. *J Nutr.* 2007;137(9):2013–2017.
36. Bisp MR, Bor MV, Heinsvig EM, et al. Determination of vitamin B_6 vitamers and pyridoxic acid in plasma: development and evaluation of a high-performance liquid chromatographic assay. *Anal Biochem.* 2002;305(1):82–89.
37. Sally PS. Vitamin B_{12} Deficiency. *N Engl J Med.* 2013;368:149–160.
38. Herrmann W, Obeid R. Utility and limitations of biochemical markers of vitamin B_{12} deficiency. *Eur J Clin Invest.* 2013;43(3):231–237.
39. Pfeiffer CM, Hughes JP, Lacher DA, et al. Estimation of trends in serum and RBC folate in the U.S. population from pre- to postfortification using assay-adjusted data from the NHANES 1988–2010. *J Nutr.* 2012;142(5):886–893.
40. Huang Y, Khartulyari S, Morales ME, et al. Quantification of key red blood cell folates from subjects with defined MTHFR 677CT genotypes using stable isotope dilution liquid chromatography/mass spectrometry. *Rapid Commun Mass Spectrom.* 2008;22(16):2403–2412.
41. Vuilleumier J, Keck E. Fluorometric assay of vitamin C in biological materials using a centrifugal analyser with fluorescence attachment. *J Micronutrient Analysis.* 1989;5:25–34.
42. Omaye ST, Schaus EE, Kutnink MA, Hawkes WC. Measurement of vitamin C in blood components by high-performance liquid chromatography. Implication in

assessing vitamin C status. *Ann N Y Acad Sci.* 1987;498:389–401.
43. Blackburn GL, Bistrian BR, Maini BS, et al. Nutritional and metabolic assessment of the hospitalised patient. *JPEN J Parenter Enteral Nutr.* 1977;1:11–22.
44. Chandra RK. Nutrition, immunity and infection: from basic knowledge of dietary manipulation of immune responses to practical application of ameliorating suffering and improving survival. *Proc Natl Acad Sci USA.* 1996;93(25);14304–14307.
45. Maggini S, Wintergerst ES, Beveridge S, Hornig DH. Selected vitamins and trace elements support immune function by strengthening epithelial barriers and cellular and humoral immune responses. *Br J Nutr*. 2007;98(S1);S29–S35.
46. Albers R, Antoine J-M, Bourdet-Sicard R, et al. Markers to measure immunomodulation in human nutrition intervention studies. *Br J Nutr*. 2005:94:452–481.
47. Hedrick VE, Dietrich AM, Estabrooks PA, et al. Dietary biomarkers: advances, limitations and future directions. *Nutr J.* 2012;11:109.
48. German JB, Zivkovic AM, Dallas DC, Smilowitz JT. Nutrigenomics and personalized diets: what will they mean for food? *Annu Rev Food Sci Technol.* 2011;2:97–123.
49. Ganesh V, Hettiarachchy NS. Nutriproteomics: a promising tool to link diet and diseases in nutritional research. *Biochim Biophys Acta.* 2012;1824(10):1107–1117.
50. Jones DP, Park Y, Ziegler TR. Nutritional metabolomics: progress in addressing complexity in diet and health. *Annu Rev Nutr.* 2012;32:183–202.

Body Composition and Growth

4

Kenneth J. Ellis, PhD

Traditionally, the most basic model of body composition partitions the body into two compartments: fat mass and all the other tissues, called the fat-free mass (FFM). Fat mass is based on the ether-extractable lipids in the body, whereas adipose tissue contains not only fat, but also includes its supporting cellular and extracellular structures. A glossary of other commonly used body composition terms is given in Table 4-1. Although FFM is treated as a single compartment, it consists of many different tissues. The three major components of FFM are water, proteins, and minerals, with water being the most abundant. In very obese individuals, however, the excess adipose tissue mass can equal or exceed the amount of body water. The concept of FFM was derived mainly to provide a way to estimate the body's fat mass using the two-compartment (2-C model). In general, some property of the FFM, such as body water, is measured and assumed to be directly proportional to the total FFM. Fat mass is then calculated as the difference between body weight and the estimate of FFM. The 2-C model continues to be widely used, partly because of the simplicity of its interpretation. The basic assumptions used with this model, however, can be problematic when the composition of FFM changes with any number of conditions.[1–4] To address this limitation, more advanced multicompartment models have been developed in which more than one component of the FFM is measured.[5]

TABLE 4-1 Glossary of Body Composition Terms

Term	Definition

Fat mass	All ether-extractable lipids in the body, including nonessential lipids (~90%), triglycerides, and essential lipids
Essential lipid	Lipids in bone marrow, cell membranes, phospholipids in neurons
Adipose tissue	Fat (80%–85%) and its supporting cellular and extracellular structures (2% protein and 13%–18% water)
Lean body mass (LBM)	Body mass free from adipose tissue
Fat-free mass	Nonfat mass, with four major constituents: water, protein, glycogen, and mineral; equals LBM plus nonfat components of adipose tissue
Total body water (TBW)	Total water amount in the body
Intracellular water (ICW)	Water content of the cellular compartment
Extracellular water (ECW)	Non metabolizing fluid surrounding cells; distributed primarily in the plasma and interstitial fluid compartments
Body cell mass	Cellular components of the body
Extracellular solids	Extracellular chemical compounds distributed in organic (collagen, reticular, and elastic fibers) and nonorganic (bone minerals, bicarbonate, and citrate) material
Bone mineral content (BMC)	Bone mineral content (g/cm), as measured by absorptiometric techniques
Bone mineral density (BMD)	Ratio of BMC to bone width (g/cm^2), as measured by absorptiometric techniques

MULTICOMPARTMENT MODELS

The advances in methodology allow for the measurement of several body compartments at the same time overcoming the limitations of the assumptions of the 2-C model. As a result, body composition analysis has evolved to three and four compartment models. These models have different levels of complexity as illustrated in Figure 4-1.[6–8] The five models are atomic, molecular, cellular, functional, and the whole body. The different equations and methods used to estimate components at the five levels are presented in Table 4-2.

Atomic Level

In the human body, there are more than 50 different elements, but six (oxygen, carbon, hydrogen, nitrogen, calcium, and phosphorus) account for more than 98% of body weight.[9] The remaining 44 elements make up less than 2% of body weight.[8,9] For years, elemental analysis of the human body was possible only in cadavers, which could be confounded by the potential alterations in composition caused by disease and death. Because of the availability of neutron activation analysis (NAA), it has been used for the in vivo measure of eight elements directly (hydrogen, carbon, nitrogen, calcium, phosphorus, sodium, chlorine, and magnesium). Whole-body counting can provide values for body potassium, and isotopic techniques can be used to measure body water. This information, determined at the atomic level, has been used to calculate more than ten body composition compartments in living humans.[10,11] The equations in Table 4-2 illustrate how atomic data can be used to calculate multiple levels of body composition.[8,11]

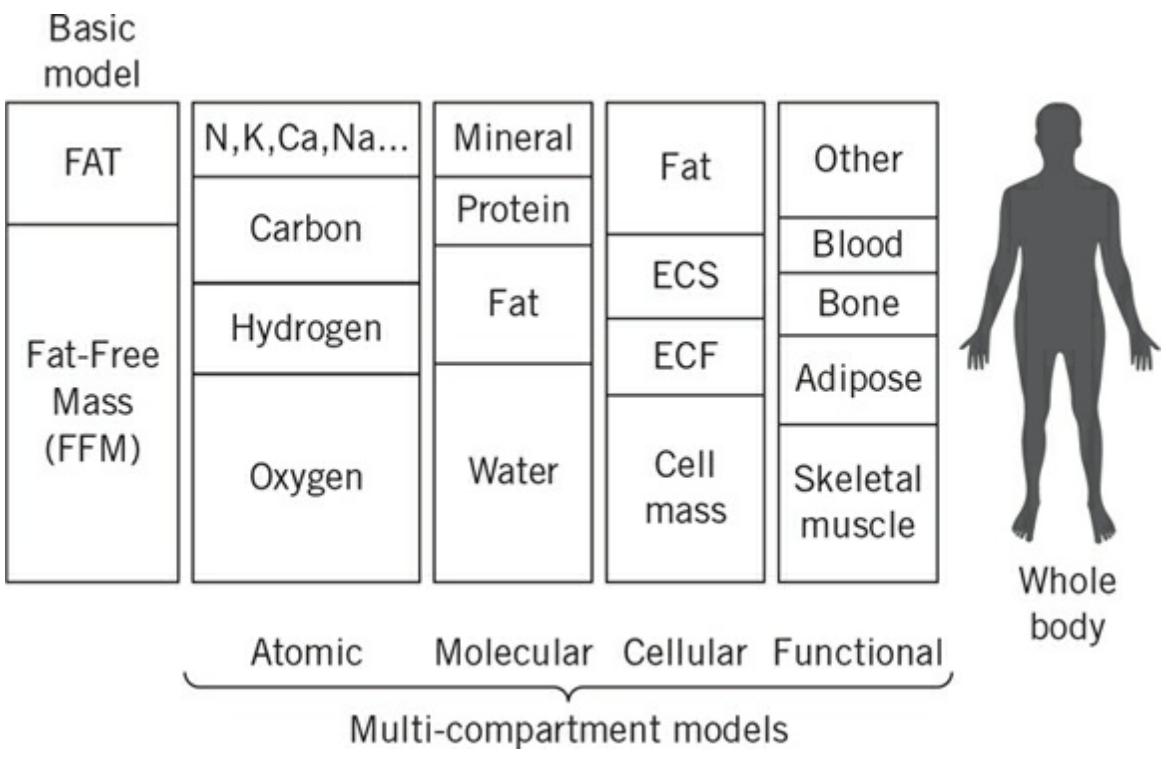


Figure 4-1 Body composition models (reproduced from

TABLE 4-2 Equations and Methods for Measuring

Body Composition by Level

Atomic level	Equation or Method
Na, Cl, Ca, N, C, P	Neutron activation analysis
K	^{40}K counting
H_2O	Isotope dilution
Body weight = TBO + TBC + TBH + TBN + TBCa + TBP + TBK + TBCl + TBNa + TBMg	
Molecular level	
Fat	Carbon in fat*/0.774
TBW	Isotope dilution
Total body protein	TBN × 6.25
Bone mineral	TBCa/0.364
	DXA
Soft tissue mineral	
K	TBK
Mg	TBK × 0.06
H_2PO_4-	TBK × 1.7
Na	TBNa
Cl	TBCl
HCO_3	TBCl × 0.43
Body weight = lipid mass + total body water + total body protein + bome mineral + soft tissue mineral	
Cellular level	
Body cell mass	0.00833 × TBK
ECW	Bromine isotope dilution
	Multi frequency bioelectrical impedance Analysis (BIS)
Extracellular solids	TBNa + TBCl + TBHCO3 + TBCa/0.177
Fat	Carbon in fat*/0.774
Body weight = body cell mass + extracellular water (ECW) + extracellular solids + fat	
Functional level	
Adipose tissue	Computed tomography (CT)
	Magnetic resonance imaging (MRI)
	Ultrasound
	Infrared interactance
	Anthropometry
Skeletal muscle	0.0196 × TBK – 0.0261 × TBN

Bone	TBCa
	DXA
	Quantitative CT
Other tissues	DXA
	CT
	MRI
	Ultrasound
Body weight = adipose tissue + skeletal muscle + bone + other tissues	
Whole body level	
	Anthropometry
	Densitometry
	Total body electrical conductivity
	Bioelectrical impedance

* Carbon in fat = TBC – [(3.37 + TBN) + (0.052 × TBCa) + (0.085 × TBCl)].
Data from Refs. 6,8,11.

Molecular Level

The molecular model of body composition is usually based on three main chemical groups: water, mineral, and organic.[5] The minerals are further separated into bone mineral and soft tissue components, and the organics are mainly defined by protein and fat components. Therefore, Body weight = fat (lipid mass) + total body protein + bone mineral + soft tissue mineral + TBW.[5,6] At this level, TBW can be determined directly by isotope dilution and bone mineral by whole-body dual-energy X-ray absorptiometry (DXA).[8] The remaining components must be estimated indirectly. For example, protein can be determined from TBN by making two assumptions: that all body nitrogen is in protein and that 16% of protein is nitrogen.[8,11] Total body fat can be calculated by subtracting the nonlipid carbon from total body carbon and then converting this into fat, assuming that triglycerides are 77.4% carbon.[12] Bone mineral can also be calculated from total body calcium.[11] An important aspect of these estimates is that there are no known age dependency on their relationships. That is, the proportions of carbon, nitrogen, and calcium are known and relatively constant for lipid, protein, and bone mineral, respectively.

Cellular Level

At the cellular level, the body is divided into four main compartments:

body cell mass (BCM), extracellular fluid, extracellular solids, and body fat. BCM is the work performing, oxygen consuming tissues of the body. The extracellular fluid is the fluid surrounding the cells, while extracellular solids consist of the organic and inorganic tissues, for example, collagen, reticular, elastic fibers and the mineral content of bone.[8] Extracellular fluid volume can be quantified directly by bromine dilution methods. Body cell mass can be estimated from the measurement of TBK or on the basis of the difference between TBW and ECW. Extracellular fluid and solids can also be estimated from the measurements of total body calcium, sodium, and chlorine by neutron activation analysis (NAA).[6,8,11]

Functional Level

The fourth level of complexity comprises tissues, organs, and systems. Three specific tissues are particularly important in body composition studies: bone, adipose tissue, and muscle, which together comprise about 75% of body weight.[8] The methods used to estimate these major compartments are usually the imaging techniques, such as computed tomography (CT), nuclear magnetic resonance imaging (MRI), and ultrasonography (US). These techniques are used to determine tissue volumes for regional and total body adipose tissues, skeletal muscle, liver, kidney, heart, spleen, and brain volumes. Alternately, DXA can be used to estimate the general compartments of fat mass, bone mineral, and skeletal muscle mass. Measurements of TBK, and body nitrogen and calcium by NAA allows the equal estimations of skeletal muscle mass, protein mass, and bone mass.[6]

Whole Body Level

The whole body level for body composition assessment is based on measurements of body size, shape, and physical characteristics.[8] The techniques used are mainly anthropometry, while body densitometry can also be considered part of this level. Anthropometric evaluations, although less accurate, are much simpler and easier to perform than are some of the techniques needed for the other four levels. Anthropometric techniques are often well suited for large-scale population studies or for field work. More recently, air displacement plethysmography (ADP), a densitometric method, has been used with children 5 years and older, as well as for infants until about 6 months of age.

MEASUREMENT METHODS

A number of methods have been developed for measurement of the composition of the human body.[5,6,8,11,12] Each of these methods provides information that relates, directly or indirectly, to one or more of the various components of the different body composition models described in the previous section. In some cases, these models are method-driven, meaning that to use that particular model, a specific measurement method must be used. Clearly, a multicompartment model would be the most informative, but these advanced models tend to require a combination of several different methods. Also, whether the information is to be used at the population level, such as in epidemiological or field studies, or for the assessment of an individual, such as in a clinical setting, can also strongly influence the choice of models and methods. Once a measurement is performed, the measured parameters have to be translated to their corresponding body composition value(s), which usually requires several assumptions.[6,13] For adults, the translation from the measured value to a body composition value often assumes relatively constant relationships, with occasional minor adjustments for age, sex, and ethnicity. For infants and children, these relationships are not always as stable, and can be easily perturbed during periods of rapid growth and sexual maturation. Of the various methods described in this section, only a few research centers can perform all of the assays. The information obtained from a combination of these assays, however, provides our best estimates of body composition and often serve as the basis for calibration of less sophisticated techniques. In most cases, only one of the methods is available to the investigator or clinician.

Anthropometry-Based Techniques

The measurement of body weight is probably the most basic of anthropometric measurements of the human body. This measurement requires a minimum of equipment, can be obtained relatively quickly, and as frequently as needed. Height or stature may be the second most measured parameter. These measurements are routinely used in anthropological and epidemiological studies, apart of public health surveys, and are commonly obtained as part of routine clinical exams. Often, the classification of subjects vulnerable to under- or overnutrition is based on these measurements alone. In recent years, BMI (body weight/stature^2) has been promoted as an international standard for the assessment of obesity in children[13,14] despite its limited relationship with

body fatness.[15]

Clearly auxology has contributed to our basic understanding of the general patterns of physical change of the body with growth and development. Many anthropometric-based measurements have been proposed for assessing body composition.[16] Whenever one of these indices has been directly compared with a more direct measure of body composition, the correlations have usually reached statistical significance only on a population basis. For the individual, however, these indices tend to be less reliable, except for extreme outlines in the population. Although, BMI appears to have the best validity of all the different weight-and-height indices,[17] its sensitivity and specificity for the individual is limited.[15] Standardized BMI curves for children (Appendix 1) are available from the US Centers for Disease Control and Prevention. Use of this index alone is quickly coming a routine part of health screening programs, including those in schools. The measurement of the subcutaneous fat layer using skinfold thickness is another anthropometric technique that has been used for many years. This technique is based on the assumption that the thicknesses of the fat layers below the skin are proportional to the total amount of fat in the body. In practice, skinfold thickness measurements need to be performed at standardized regions on the body and by experienced and well trained examiners The triceps has been the site most frequently selected, but no single body region appears to be consistently representative of the whole subcutaneous fat layer, much less the total fat mass. A number of different prediction equations have been published, some using the same measurement sites. To learn more on this topic, we would direct the reader to the literature for the use of skinfold measurements.[18,19] Alternate techniques, without the use of calipers, have been developed for the measurement of the subcutaneous fat layer. These techniques are based on the differences in the absorbance and reflectance properties of skin, fat, and lean tissue at different wavelengths.[20,21] These techniques are well tolerated by the subject being examined, and provide an alternative to skinfold measurement. Only a limited number of studies in different age groups have reported using this technology.[21–24] However, many of the same limitations for skinfold measurements equally apply, namely extrapolation to total body fat content. These techniques may be more useful for monitoring intrasubject changes than for intersubject comparisons. Also, there are no studies where it has been used in conjunction with a more direct method for measuring total body fat.

In Vivo Chemical Analyses

Historically, analytical chemical analyses of tissues are the quantitative basis for human body composition, and have contributed to our understanding the metabolic function of tissues. The advancement of modern physics lead to the development of nondestructive nuclear-based assays of tissue samples, which was later transferred to direct in vivo analyses in the 1970s.[25,26] The method, called neutron activation analysis (NAA), is based on the principle that when atoms are exposed to neutrons, there is a finite probability that some of the neutrons will interact with the nucleus of atoms, resulting in the production of gamma-rays that are characteristic of the interactions. These gamma-rays can be produced during the time of the neutron interaction, or observed for several minutes following the exposure to a low-intensity beam of neutrons. For body composition measurements, the gamma rays are detected external to the body.[25,26] Using this approach, the total body content of calcium, sodium, chlorine, phosphorus, nitrogen, carbon, oxygen, and hydrogen have been measured in humans.[26–28] This in vivo elemental information can be used for the chemical model, or converted to estimates of body protein, water, fat, and bone mass.

Body Potassium Measurement

A very small fraction (~0.012%) of all potassium atoms found in nature is radioactive. This isotope, called ^{40}K, decays emitting high-energy gamma-rays (1.46 MeV) about 11% of the time. At this energy, about half of the gamma-rays produced within the human body will escape and can be detected external to the body. To improve the precision of this measurement, the detectors must be shielded to reduce background interference from gamma rays originating from other radioactive sources in the environment. This type of instrument is called a whole-body counter.[26] The availability of stable electronics, high-efficiency detectors, and PC-based analyzers, has significantly decreased the cost of this technology. When properly calibrated, the whole body counting procedure can produce an accurate and precise measure of TBK, which is directly related to body cell mass, the actively metabolizing tissues in the body.[29] Since most of TBK is in the ICW volume at a relatively constant concentration, this measurement has also been used to estimate the ICW volume.[26] TBK is no longer recommended as a measure of the FFM because changes in the ECW compartment of FFM can occur without changes in body potassium. The most recent application of TBK is for

modeling of total body protein mass in adults and children.[30]

Body Water Measurements

Isotope Dilution Methods

Body water is the major component of the FFM, and is also the major contributor to body weight, with the exception of fat in extremely obese subjects. Hence the measurement of TBW may give one of the most reliable estimates of FFM, and when coupled with body weight, an estimate of body fat mass. The most common procedure used for the measurement of TBW is the tracer dilution technique, using nonradioactive isotopic labeled water. At least two body fluid samples are needed (blood, urine, or saliva), one before administration of the oral tracer dose and one at least several hours later. Both deuterium and ^{18}O-labeled water are used, with the quantitative assays based on mass spectroscopy or infrared absorbance.[31] The fluid samples are usually frozen for later analysis. In the clinical setting, this approach would not be practical when there is an immediate need for the results. To repeat the assay, there must be a sufficient time lapse in order for any earlier tracer to clear the body. Typically, this is 3–5 times the biological clearance of the tracer. Partly for these reasons, alternative methods such as bioelectrical impedance have been developed for the assessment of body water.

In a normally hydrated state, the ratio of TBW to FFM is assumed relatively constant for the individual. On average, for healthy adults and adolescents, this ratio is typically 0.732. At birth, the ratio is much higher (~0.80–83) and decreases during early infancy and childhood. Therefore, to estimate fat as the difference between body weight and FFM, age- and gender-specific adjustments for the hydration of FFM should be used.[1] The extracellular component of body water can also be determined using nonradioactive dilution methods. Bromide is the most commonly used tracer, and its concentration in blood or saliva is analyzed using either HPLC or X-ray fluorescence.[6] For infants, a sucrose tracer has also sometimes been used.[2] When these assays are obtained in conjunction with the TBW measurement, the difference between TBW and ECW be used to indirectly estimate ICW volume. Again, repeat measurements are limited by the amount of time needed for the tracer to clear the body from any earlier study. Partly for these reasons, multi frequency bioelectrical impedance methods have been developed for the assessment of extracellular body water volume in addition to TBW.

Bioelectrical Methods

In electrical terms, impedance is a measure of a conductor's frequency dependent opposition to the flow of an alternating electric current.[20] Because the body's fluids contain electrolytes, and the lean tissues in the body contain virtually all of body water, these tissues can conduct an electrical current. This is the core basis for the bioelectric impedance analysis (BIA) methods used to measure body composition in humans. How these tissues conduct a current is also dependent on the capacitance properties of the cells. At lower frequencies, the cell membranes are assumed to act like capacitors and not allow current to pass through the cells. At high frequencies, the capacitance properties of cells are significantly diminished, and the current is now assumed to also pass through the cells. That is, at low frequencies the current passes mainly through around the cells (e.g., the extracellular fluid volume), while at high frequencies the electric current passes both around and through cells (total body water). Since fat is a poor conductor of electricity at any frequency, a measurement of the body's impedance or resistance should be related to the body's water content. Most BIA instruments use a single frequency (50 kHz), while some instruments can make measurements at multiple frequencies.[33] The short-term precision of BIA measurements for an individual is good, but the accuracy of its translation of body composition values continues to be questioned in some studies.[34–36] Unfortunately, this has lead to a multitude of BIA-based prediction equations in the literature, most of which are incompatible with each other.[6,25] One factor that may account for some of these inconsistencies is the difference in body shapes among individuals of the same stature, as well as the placement of electrodes. In the simplest electrical model, the assumed shape (geometry) of the conductor affects the circuit's impedance value. In the case of the human body, where the electrodes are typically placed near the wrist and foot on one side of the body, the smaller cross-sectional areas of the arm and leg contribute the most to the resistance value. This is also the case when multiple segmental measurements are obtained. Altered states of hydration, such as edema or malnutrition can influence the resultant impedance values leading to inaccurate estimates of the true body composition. In diseased populations, the predicted body composition values for an individual should be independently verified with a more established method.[8,11,25] Recently, advanced models for the multifrequency method have been reported.[37,38] The single-frequency BIA method, however, continues to be the popular choice in many research

studies.

The BIA methods are not the only technique used for measurement of the body's resistance to the passage of an alternating electrical current through the body. If the body passes through an external uniform electromagnetic field, the body's tissues will interact with the external field. This method, developed in the 1980s is called.[39] The short term precision of the measurement was good and this approach showed promise.[6,35,36] However, the lower cost of the BIA method and that it could be performed relatively quickly, and offered portability, the total body electrical conductivity was not developed further.

Densitometry Methods

To measure the density of the whole body, one needs to accurately know the body's weight and volume, with density defined as weight/volume. Body weight can easily be measured with high precision with an accurate scale. A precise measurement of body volume, on the other hand, is more difficult to obtain. The classical method, used since the 1950s, has been to measure a subject while totally submerged underwater and with as much air as possible exhaled from the lungs.[29] By comparing the subject's weight in air and underwater, the volume of water displaced is the same as the subject's body volume. A person's weight is equal to the sum of its body composition compartments. Likewise, a person's volume is equal to the sum of the volumes of the same compartments. If the body is composed of two parts (e.g., fat and FFM), and the density of each is known, then the weight and volume equations can be rearranged to calculate the mass of either compartment.[31] This is the basic principle of all methods based on the measurement of body density. The underwater weighing technique has been considered by many as the "gold standard" measurement of body composition.[29,31] It is not feasible to use this method, however, in infants and young children, nor in most diseased populations or hospitalized patients. Hence, the development of air-displacement plethysmography has provided an alternative approach to the measurement of body volume.[40,41] For this approach, the body displaces a volume of air inside a closed chamber to determined body volume, replacing the need for total submersion in water. Corrections for lung volume are still needed, along with adjustments for the adiabatic effects of the skin.[42] However, once a body volume is obtained, the same calculations used in underwater weighting apply. At present, only one commercial instrument (BOD POD^{TM}, Cosmed, Concord, CA) is

available. Its performance has been shown to be in good agreement with underwater weighing for adults and children.[40,41] An infant-sized air-displacement plethysmography instrument (PEA POD™, Cosmed, Concord, CA) has also been developed.[43] Body composition results in healthy infants for this instrument were in good agreement with those obtained using the deuterium dilution method and four-compartment body composition model.[44,45] Further testing, however, is needed for preterm infants and in sick infants where there may be significant alteration of normal hydration.

Dual-Energy X-Ray Absorptiometry

This technology originated with the use of a collimated radioactive source passed over the distal end of the radius, and monitoring of the intensity of the transmitted photons on the opposite side of the arm.[6,25] Attenuation of the transmitted signal is related to the chemical makeup and thickness of the tissues in the beam path. In the mid-1990s, the radioactive sources were replaced with X-rays sources, different types of detectors were used, and measurements were made at two energies.[46] A low intensity X-rays are used and the exposure time is relatively brief resulting in a minimal exposure, comparable to natural background sources of radiation exposure one encounters daily.[6,25] DXA (sometimes denoted as DEXA) has emerged as a principal method for measuring bone mineral and body composition at all ages.[46–51] It has high precise for bone mineral measurements in adults and older children, and has become the de facto standard for assessing bone status. DXA measurements have also been obtained in a number of studies of infants and children.[46,48,52,53,] It should be noted that the adult-based software used with the initial pencil-beam instruments provided inaccurate body fat and lean tissue values for younger children.[49] The accuracy of DXA for use in infants has also been questioned.[51,59] The newer fan-beam DXA instruments, however, have improved software for body composition and bone mineral analysis specifically developed for use in pediatric populations. Another point to be noted is that DXA does not measure the true bone density (g/cm^3), but instead areal density (g/m^2) based on a reconstructed 2-D image.[6,25,46] To measure true bone density, a quantitative 3-D imaging method is needed. A technique, called peripheral quantitative computed tomography has been developed for this purpose. It has been used to measure the trabecular and cortical regions in the radius and tibia, and pediatric reference ranges are available.[54,55] Another feature of this technique is that the cross sectional

geometry of the bone can be measured.

Body Imaging Techniques

The most recent advances in measurement methods of body composition have been the use of imaging techniques.[56] Both CT and MRI have been used. Both methods can provide accurate reconstructed 3-D images of the body. An advantage for CT is that true density (g/cm^3) is obtained, while its major limitation is that a substantial high radiation dose is required. MRI, on the other hand, uses no ionizing radiation, and provides better images, but the information is more qualitative compared with CT. Furthermore, a CT scan typically takes only a few seconds to complete, whereas an MRI scan can take considerably longer, although the newer MRI instruments have reduced these differences. A strength of the CT and MRI techniques is that organ-specific information can be obtained which supports the Functional model (see Figure 4-1). To date, most of the emphasis using CT and MRI has been on the measurement of the subcutaneous and visceral components of abdominal adiposity.[56–58] Whole-body MRI has also been used to monitor changes in body composition during infancy, but only during early life.[59] An alternate magnetic-based technique has also been developed, but has had only limited use.[60]

Growth and Body Composition

Measurement of body composition, in both clinical and research settings, has advanced the understanding of human energy expenditure, metabolism, and physiologic changes seen during growth and disease. Growth is the maturation process characterized by change, compensation, and adaptation; it is affected by hormonal, nutritional, and environmental factors interacting with genetics. Changes in the components of body mass for an individual are a result of the dynamic events preceding the time of assessment. The importance of changes in body composition that can occur during acute and chronic diseases, and their treatment has led to the need for refinement of the techniques used in measuring these changes. Nutritional status can no longer be based on height and weight alone. In many diseases, such as protein-energy malnutrition, cancer, inflammatory bowel disease, chronic renal disease, acquired immune deficiency, and obesity, assessment of only body weight or weight-for-height can give an erroneous impression about the nutritional well-being of the subject. It is possible to achieve an acceptable weight while having excess body fat or

body water, thus confounding the benefits of traditional nutritional therapy. A closer look at body composition can illuminate the loss of lean tissues associated with disease, and may permit a more customized approach to nutritional management. The study of body composition helps identify the interactive effects of genetic, nutritional, and physical activity on changes in fat, muscle, and bone. It is important to monitor the amount of each component as well as their rates of growth and potential localized variations in an effort to better define possible relationships with current and future health risks.[61,62] Changes in body composition that occur during fetal life, infancy, childhood, adolescence, adulthood, and aging are presented as part of this chapter. An emphasis is placed on the changes in fluids, fat, muscle, and bone mass in the different stages of the life cycle. The measurement of body composition continues to become an important clinical tool in the monitoring of variations in growth of healthy subjects as well in the nutritional management of patients with acute or chronic illness.

The previous description of the various methods and models used for measurements of body composition is crucial in the interpretation of nutrition literature. Each method can provide a unique perspective, taking advantage of certain physical or chemical principles. On the whole, the use of different methods in the same set of subjects has allowed the cross-validation of the various techniques and has permitted a more detailed understanding of body composition.

BODY COMPOSITION CHANGES DURING GROWTH

The relative changes in body composition with age, as a percentage of body weight, are shown in Figure 4-2. More specific details of the compositional variation during typical growth are presented in Table 4-3. Figure 4-3 depicts the changes in body composition that occur during fetal development. These reference data can be helpful when assessing the growth of preterm infants.[6]

Changes in Body Fat Mass

Fat is the most variable of body components, ranging from as low as 5% to more than 50% of body weight. The normal range, however, is narrower and varies from about 10% to 25%–30%. During intrauterine life, the differentiation of fat starts in the second trimester of gestation, between the

fourteenth and twenty-fourth weeks of pregnancy. It becomes evident in the head and neck regions, followed by progressive increases for the trunk, arms, and, finally, the lower limbs. At the beginning of the third trimester, at approximately the twenty-eight weeks gestation, adipose tissue is already present in the principal fat deposit areas. This may indicate that the second trimester of pregnancy is a sensitive period for the later development of fat tissue.[66] Abnormalities in infant fat mass can be attributed to changes in maternal nutrition in late gestation, during the period of maximal fetal growth.[67] The possibility that nutritional alterations at critical periods of development influence nutritional status, such as for adult obesity, has been suggested by some authors.[67–69] Follow-up studies of adults with a history of maternal famine prenatally or during early in life and of infants of diabetic mothers strongly suggest that early under- or overnutrition influences the development of fatness and its related diseases in later life.[70,71] Altered maternal nutrition has been shown to change the expression of appetite-regulating neuropeptides in the fetal brain, enhancing the later risk for obesity.[69] Increased birth weight and postnatal growth are positively correlated with lean body mass, an effect that may persist throughout life.[63,72] Increased birth weight is positively correlated with body mass index (BMI) and inversely with central adiposity.[72,73] In general, low birth weight infants have higher percent body fat and less lean body mass, with an increased later risk for central obesity.[73,74] These data suggests that preventing growth failure in utero and in early life are important for body composition, and could have long term consequences.

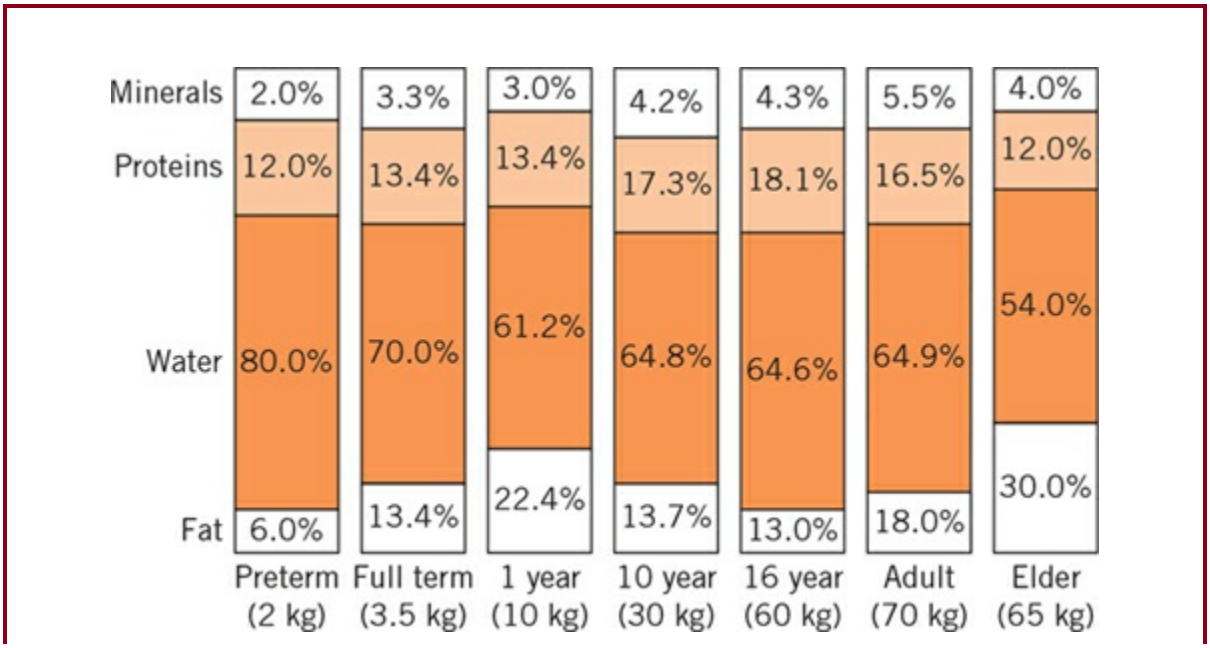


Figure 4-2 Relative changes in major body composition compartment with age.

On-going research also indicates that certain genes involved in embryonic development may play important roles in adipocyte development and fat distribution.[75] Before birth, there appear to be regional differences in fat accumulation that could have important significance for the future health. Cell characteristics and normal growth patterns differ among the fat depot sites. After birth, hormonal responsiveness of fat tissue also varies among these location.[66] It has also been suggested that the postnatal environment may equally influence the development of obesity in later life. Weight gain in the first weeks of life may be a critical determinant of adult obesity. In utero, the fetus increases its fat content from ~2.5% at 1 kg weight of a fat mass of 14%–15% at full term.[63] After birth, there is a progressive accumulation of fat mass peaking at 29%–32.0% at 6 months of age.[65] This effect is probably due to a high intake of energy during a period of relatively little activity. Breast-fed infants have a different accumulation of fat than formula-fed infants, with breast-fed infants having less fat, with the greatest differences evident at 9–15 months of age. Differences in body fat accumulation can also depend on the type of fat contained in certain formulas and the proportion of protein. However, some studies have reported that these differences do not persist after 1 year of life. Whether the choice of infant feeding mode affects body composition later in life is still uncertain.[76] Increasing with age in early life, the body fat mass decreases over the next several years to a minimum of ~13% at 7 years in boys and 16% at 6 years in girls. This follows a discrete increase to about 10 years of age, often referred to as "adiposity rebound."[63] Although there is evidence of some sexual differences in body composition at birth and during early life, the predominance of increased fatness in girls becomes notably evident during adolescence. For both sexes, puberty is characterized by substantial increases in total number of adipocyte and the fat content. There is also a shift of fat from peripheral sites to truncal storage sites, which is especially marked in males. In general, males attain their peak percentage fat during early puberty, and then decrease during adolescent growth.[76] Females show an initial decline in fatness, followed by a continuous increase in fat percentage thereafter. There are also significant gender differences in subcutaneous fat distribution, with a prevalence of an increase in adipose

tissue in the upper part of the body (above the umbilicus) in males and in the lower part in females. Despite this tendency for concentration of fat in the abdomen of males and the hips of females is clearly evident in adolescence, there are some indications it may be present as early as 4 years of age. Some of this variation can be attributed to the effects of hormones, with estrogen and progesterone inducing a slight increase in adipocyte volume and number, whereas in males, androgens appear to affect a decrease.[66] With aging in males, body fat slowly increases from about 18% in young adulthood to about 36% by 65 to 85 years of age. For females, there is a gradual increase from 33% to 45% of similar age groups. In a multiethnic longitudinal study of Americans, percentage fat in males increased from about 22% at age 18 years to about 26% at age 60–69 years. For the same ages in females, the increase was from about 32% to about 39%.[77] The increased fat mass in females of all ages, especially in the lower part of the body, could be related to their capacity for childbearing as an adaptive mechanism for the preservation of the species. There is a need for ample energy reserves for successful fetal and newborn growth.[78] At the same time, in a minority of normal adults, the adipose tissue distribution is reversed for the sexes.[78] In women with an android fat distribution, the risk factors for developing diabetes, hypertension, or hyperlipidemia are similar to those of men. In these women, there is no clear explanation for this different pattern of fat, although it has been suggested there might be an inherited increased sensitivity of adipocytes to androgens, and possibly for cortisol.[78] Ethnic differences in fat accumulation during growth have been observed. A study that included African-American and Hispanic girls showed their percentage of fat values to be higher than in the white group. Bone mineral content and lean tissue mass were also greater for the African-American girls than the white girls, whereas there were no differences in the Hispanic girls.[52] A similar multiethnic study of boys showed greater bone mineral content and lean tissue mass, with lower percentage fat for the African-American group. The Hispanic males had similar bone and lean tissues as the white group, but a higher percentage of body fat.[53] In another longitudinal study, no difference in total body fat was found across ethnic groups in males.[77] For females in this same study, total body fat was highest in the African-American group, followed by Mexican-Americans (Hispanic) and then white females.[77] It is to be noted, however, the BIA was used for the latter study, while DXA was used for the previous study. In general, white children have a more peripheral distribution of body fat than Latin

American, Japanese, or African-American children. African-American children tend to have a more centralized fat pattern. These ethnic differences are evident as early as the preschool period and continue throughout adolescence. The pattern of less peripheral fat has also been found in studies of children from several Latin American countries.[79] These differences could reflect genetic differences as well as different lifestyles and nutritional and health habits. Ethnic differences have also been found for Asian adolescents and adults. When combined, these studies suggest the need for ethnicity-specific references for body composition during growth. It appears that most overweight 1-year-olds exhibit normal levels of fatness as adolescents and adults. Overweight in early childhood is also not well correlated with adult obesity, whereas overweight in later childhood and adolescence is.[80] Long term follow-up studies in children have found that truncal fat predominance and increased BMI in childhood were associated with an increased risk of central obesity in adulthood.[81] However, in a longitudinal study from birth until age 43, it was found that for any given adult body size, those who had been lighter in childhood were more at risk for abdominal obesity, suggesting that insulin resistance may be linked to low weight in childhood.[82] The degree of body fatness as early as 7 years of age was shown to be correlated with risks for coronary heart disease and diabetes.[83] Overweight adolescent males also have an increased risk of mortality from all causes and from coronary heart disease after age 55 years.[84] Fat distribution also plays impacts the risks of developing cardiovascular disease, diabetes, hypertension, gallbladder disease, stroke, and overall mortality. It appears that at least three principal features of body fat are associated with health outcome: total amount of fat expressed as a percentage of body weight, amount of subcutaneous truncal or abdominal fat, and quantity of visceral fat located within the intraabdominal cavity.[85] Adult diabetogenic or atherogenic obesity tends to be more truncal than peripheral, That is more the abdominal “apple” shape than the gluteo femoral “pear” shape, more intra abdominal visceral than extra abdominal subcutaneous, and more in the upper body than in the lower body.[78] Risk factors for the development of obesity are both genetic and environmental, such as changes in diet, exercise, or lifestyle.[75] Correlation between body fat and the amount of time spent watching television have been reported by some, but not others.[86,87] Regular exercise can help decrease body fat as well as increase bone mineralization and muscle mass.[3] In general, athletes have a lower percentage of body fat and greater lean body mass than their nonathletic size-matched peers.

However, there is considerable variation among individuals and types of activities. Athletes participating in sports in which body weight is supported (e.g., swimming, cycling, kayaking) or in which there is minimum body movement (e.g., throwing, lifting) generally has a higher percentage fat than athletes who participate in sports that require horizontal or vertical body movement (e.g., running, jumping). In sports in which weight is a factor, such as gymnastics, ballet, and wrestling, the effort to decrease body fat can affect performance and induce eating disorders. This phenomenon occurs in athletes independent of gender.[88] In obese adults, intensive and extensive aerobic exercise can induce moderate losses of thigh fat and also central subcutaneous fat. This effect is more noticeable in males with excessively high truncal fat. The increased percentage of body fat seen in elderly populations may be attributable, in part, to decreased physical activity. With aging, there is a decline in muscle mass and strength that may be partly counteracted by regular physical exercise throughout life. The capacity for some increase muscle mass and strength is retained into old age. Physical training, especially resistance exercise of considerable intensity, frequency, and duration, may slow the loss of muscle mass and bone mineral, while decreasing fat mass. This, in turn, may reduce the associated abnormalities of diabetes, coronary heart disease, osteoporosis, and fractures.[89]

TABLE 4-3 Body Composition of Reference Children and Adolescences

Age/Sex (year)	Length (cm)	Weight (kg)	Percentage of Body Weight						
			Fat	Protein	Mineral	Carbohy-drate	TBW	ECW	ICW
Boys									
Birth	51.6	3.5	13.7	12.9	3.2	0.5	69.6	42.5	27.0
0.5	67.9	8.04	29.1	10.9	2.4	0.5	57.2	32.9	24.3
1	76.1	10.03	25.6	12.3	2.7	0.5	59.0	31.6	27.4
1.5	82.6	11.43	24.5	12.9	3.0	0.5	59.1	30.1	29.4
2	87.6	12.46	25.4	13.5	3.1	0.5	58.1	26.6	30.7
5	109.9	18.7	14.6	15.8	3.7	0.5	65.4	30.0	35.4
10	137.5	31.4	13.7	16.8	4.1	0.5	64.8	26.7	38.0
12.5	153.0	42.3	16.3	16.4	4.1	0.6	62.7	26.4	36.4
15.5	171.5	59.5	13.0	17.4	4.5	0.6	64.6	25.8	38.8
18.5	177.0	69.9	12.9	17.7	4.8	0.7	64.1	24.7	39.4
Girls									
Birth	50.5	3.3	14.9	12.8	3.2	0.5	68.6	42.0	26.7
0.5	66.5	7.6	32.0	10.4	2.3	0.5	54.9	31.7	23.2
1	75.3	9.5	27.6	12.2	2.7	0.5	56.9	29.7	27.4
1.5	82.0	10.94	26.3	12.7	2.9	0.5	57.8	29.0	28.5
2	87.7	12.02	25.4	13.1	3.0	0.5	57.7	29.0	29.4
5	108.4	17.7	16.7	15.0	3.1	0.5	64.6	31.0	33.6
10	138.3	32.6	19.4	15.0	3.1	0.5	62.0	28.1	33.9
12.5	154.6	43.8	21.5	15.4	4.2	0.5	58.5	25.6	32.9
15.5	162.1	55.0	24.7	14.9	4.5	0.5	55.5	23.7	31.8
18.5	164.0	57.0	25.0	14.9	4.4	0.7	55.2	23.5	31.7

ECW, extracellular water; ICW, intracellular water; TBW, total body water.
Adapted from Refs. 63–65.

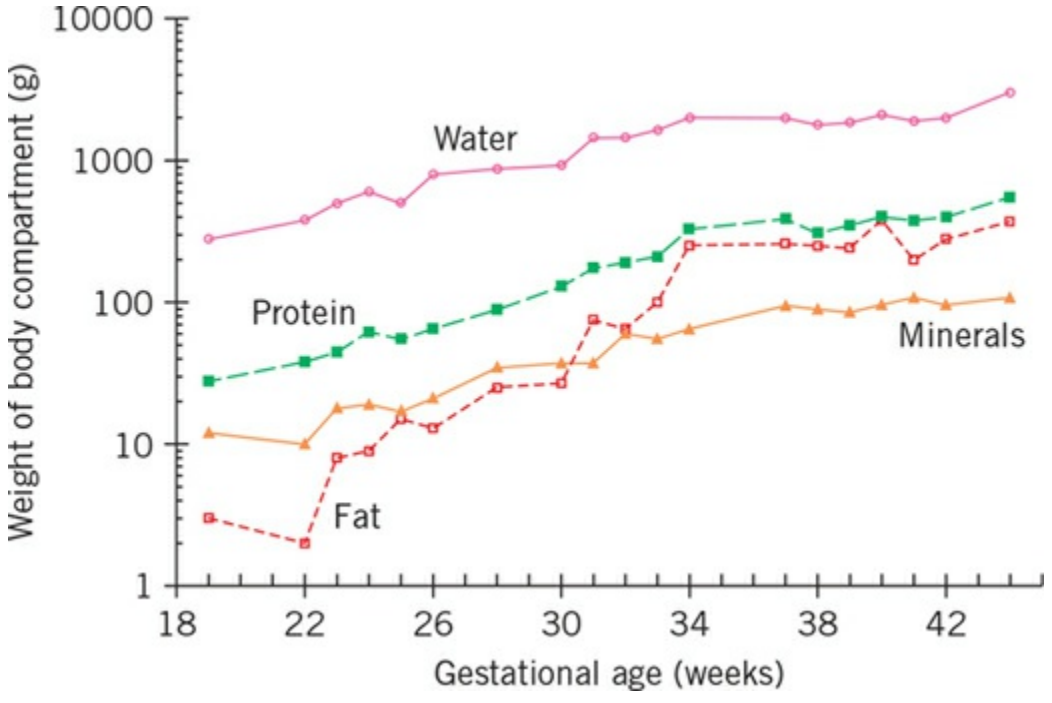


Figure 4-3 Changes in water, proteins, minerals, and fat during fetal development and early life. (Reproduced from Ref. 6).

Changes in Body Water

The body's content of water is greatest during fetal life—about 88.6% of weight at 24 weeks gestation, decreasing to about 75% at full term. After birth, the water content continues to decline rapidly, reaching about 60% of body weight at age 4 months. There is a continued, but slower, decline to adolescence, where the range between 60 and 65%.[90] During the first years of life, the extracellular water (ECW) volume exceeds the intracellular water (ICW) volume, while this ratio slowly decreases with growth and is reversed at older ages. Initially, ICW is about 25%%–30% of body weight, increasing to about 35%–40% by adolescence. This change is largely attributable to the increase in muscle mass. At the same time, ECW slowly drops from about 40% to about 20%–25% of body weight.[9,52,53,90] In the elderly there is a small decrease in total body water (TBW) and an increase in the ECW/ICW ratio, which can differ somewhat by race, with African Americans showing the greatest relative increase in ECW.[77,91] Preterm and small-for-gestational-age term infants have a greater proportion of body water at birth than do their appropriate peers, suggesting that fetal growth deficits in these groups are not attributable to abnormalities in body water regulation.[32,92] In disease states such as malnutrition, there is an expansion of ECW. Similar water distribution patterns have been observed in sepsis, after surgery, and in patients with cancer.[93] There is increasing evidence that changes in the ECW/ICW ratio may be an accurate predictor of clinical outcome and mortality.[93] A strong correlation between changes in the ECW/ICW ratio and protein and energy intakes has been observed. For this reason, measurements that can assess these two water compartments may aid the clinician in monitoring nutritional rehabilitation in the acute care of critically ill patients.[93]

Changes in Body Mineral Content

Throughout life, there are both accretion and resorption of bone. This process of bone modeling is characterized by changes in the shape, length, and strength of bones. During normal growth, accretion exceeds resorption. Thereafter, the bone tissues within the existing skeletal structure are being continuously formed and resorbed, with minimal change in bone volume.[94] Skeletal growth continues during infancy,

childhood, and adolescence. The rate of bone modeling varies greatly during these different biologic stages. By one year of age, skeletal mass has doubled and approaches about 37% of the total skeletal mass of an adult. The next major stage of bone growth is during puberty with over 25% accumulation of the final adult skeletal bone.[95,96] Although often ignored, there is a third phase during early adulthood where the skeletal mass continues to increase by about 7%–9%. This level of change is small, but it is important when considering the losses that occur at older ages, especially during menopause.

The annual increment in bone areal mineral density (BMD) is high during the first 3 years of life and decreases thereafter until puberty. This reflects an initial mineralization of bone that exceeds the increase in bone volume. The opposite tends to occur during puberty where there is accelerated height velocity followed by mineralization. The adult reference man has a calcium content of 1.2 kg.[9,29]

During the initial 10 weeks of gestation, the human skeleton is composed of cartilage, which is chemically and histologically distinctive from cortical bone. The next phase of fetal bone growth is ossified, starting with the shafts of long bones (e.g., femur, humerus). The ossification process gradually extends along the shaft, replacing the cartilage and also forming trabecular bone, such as the metaphyses.[97] Near the end of the second trimester, the fetal skeleton has a mass of about 500 g and about 42% cartilage. At full term, the total skeleton is about 33% cartilage and about 30 g of calcium, most of which was deposited during the last trimester.[9,94,98] Fetal calcium accretion increases dramatically during the last trimester, ranging from 130 mg/kg/d at 28 weeks gestation to 150 mg/kg/d at 36 to 38weeks gestation.[9,98] For a preterm infant, these rates are higher than what human milk alone can supply. Insufficient intake of calcium and phosphate is the primary cause of osteopenia and rickets of prematurity.[99,100] In premature infant studies, where supplementation of mother's milk or specialized preterm formula were used, bone mineralization increased significantly, leading to a more normal bone mass at expected term.[101,102] Intrauterine bone mineralization has been examined by measuring the BMD in appropriate for gestational age infants born at between 30 and 41weeks gestation and for infants born between 22 and 42 weeks gestation.[103,104] Bone width and areal BMD at birth were correlated with gestational age and birth weight and have provided reference curves for births at 30–41 weeks gestation.[105,106] BMD increases over the first year of life, initially at a lower rate during the first 6 months,

and can vary with the type of feeding.[107] Higher BMD values have been reported for supplemented breast-fed infants or cow's milk formula-fed infants, when compared with unsupplemented breast-fed infants.[101,108] In a study of preterm infants no difference in bone mineralization and total bone mass was found when fed either preterm formula or fortified breast milk.[105] Lower BMD values, however, have been observed in infants receiving soy-based formula compared with infants receiving cow's milk formula.[109] In later studies where the soy formula was modified to improve the bioavailability of minerals, no differences in BMD were found when compared with cow's milk formula.[110] The long-term consequences of the differences in BMD with various types of feedings in early infancy remains unknown. One study found that bone accretion is lower in infants fed human or low-mineral formula in the first 6 months of life, but those fed human milk had greater bone mass accretion in the second 6 months than those fed formula, such that was no difference in bone mass among the feeding groups at 1 year of age.[111] Similarly, no differences in BMD at 4 years of age were found in children who had been breast fed or formula fed.[112] Likewise, no gender or ethnic differences in BMD have been found in full-term or preterm newborns, while older males had higher BMD regardless of feeding type.[105,112] A wide variation in BMD values as a function of age, gender, and ethnicity have been reported, with gender is more important than race and boys having slightly higher BMD values than girls [113–115] children of African decent have higher values than whites.[52,53,116,117] Ethnic differences in BMD among blacks, whites, Lebanese, and Asians have also been reported in adolescents and adults.[113,117]

In the lumbar spine, there is a gradual, linear increase in BMD with age in both sexes before puberty, followed by accelerating increments during puberty. Until the age of 15 years, BMD values are generally higher in girls, possibly because of earlier pubertal development or Tanner Stage.[113,118] In girls, peak bone mass in the spine and femoral neck is usually achieved by 15 years of age, with the maximal peak mineralization rate occurring earlier at about 12.5 years.[96] In boys, the most rapid increase in lumbar BMD occurs at about 14years of age, with full bone mass achieved at about 17–18 years or Tanner Stage IV is reached.[96,118] By the end of puberty, skeletal bone mass reaches over 90% of its maximum, not peaking until age 25–30 years.[119] These studies show the relative importance in accumulating bone mass of early adolescence in girls as compared with late adolescence. This was confirmed in a study of

calcium kinetics in girls, ages 5–17 years old where calcium deposition and the size of the exchangeable bone pool reached a maximum during early puberty and decreased in late puberty,[120] Because peak bone mass accumulation is achieved during adolescence, current recommendations for calcium nutrition reflect a greater requirement for these age groups (See Appendix 2 for Dietary Reference Intakes). It has been suggested that prevention of osteoporosis consists, in part, of maximizing peak bone mass during growth, although there remains insufficient knowledge to verify this link.[121] Genetic factors, estimated to contribute 70–80% of the variance in bone mass, are perhaps the strongest predictors of peak bone density and loss with aging. A genetic influence has been observed in premenopausal women and premenopausal daughters of postmenopausal women with osteoporosis.[122,123] Additionally, BMD correlates more closely in monozygotic than in dizygotic twins.[122,124]

Within the modifiable environmental factors, calcium intakes can play a significant role in achieving peak bone mass. For the growing individual, there is a significant positive relationship between calcium intake and retention. Variations in calcium intakes early in life could account for as much as 5%–10% of the difference in peak bone mass. It is to be noted that older children and adolescents in the United States do not achieve the recommended intake of calcium, which may be important in reducing the risk of fractures and osteoporosis later in life.[125] However, a firm link between bone mineral status in childhood and the risk of future fractures has not been fully established.[121] Finally, physical exercise and weight-bearing activities throughout life have a positive impact on peak bone mass. From this prospective, prevention of osteoporosis has become a domain for the pediatrician as the relationship of genetics, nutrition, and activity is better understood. There is an increasing prevalence of obesity in childhood, and overweight children may have an increased risk of fractures owing to lower BMD.[126] Other studies, however, have not shown a difference in bone mineral content between obese and lean children.[122] In practical terms, the pediatrician should encourage the attainment of normal body weight for height and an adequate calcium intake at all stages of growth.

Changes in Skeletal Muscle Mass

There is no definitive in vivo method of measuring total muscle mass. Neither total body potassium (TBK) nor total body nitrogen (TBN) can specifically identify muscle cells. However, because about 60%–70% of

TBK is found in skeletal muscle, accurate models to predict skeletal muscle mass have been developed using these measurements. Although metabolic end products released from myocytes—creatinine and 3-methyl-histidine—have been proposed for estimation of whole body muscle mass, their application is limited by a number of constraints.[20,127] Although acceptable in certain well-controlled conditions (research lab), their general clinical use is restricted.[128] Approximate estimates of muscle mass can be obtained with anthropometric measurements of the mid-arm, mid-thigh, and mid-calf circumferences, which offers an inexpensive useful field method. This technique has been particularly useful in the clinical setting when subjects can be followed overtime, and more direct methods are not available. At full term birth, the skeletal muscle mass represents 22%–25% (~850 g) of the infant's body weight. As an adult, skeletal muscle mass is about 30%–45% (~28 kg) of body weight.[9] About 27% of TBN, 33% of TBK, and 28% of TBW is contained in the skeletal muscle of a newborn.[9,29,97] The chemical change in the composition of skeletal muscle during early development is a decrease in the percentage of water concurrent with an increase in the protein concentration. As the fibers increase in number and size, the extracellular material, which makes up a large part of fetal skeletal muscle, is replaced by muscle fibers.[97] The muscle mass constitutes the largest part of the soft tissue mass for normal individuals. For extremely obese individuals, the adipose tissue can dominate.[97] Depending on gender, age, and health status, between one-third and one-half of total body protein is within skeletal muscle.[9,127] During growth, the increase in total muscle mass is not uniform, developing slowly during childhood, and then a growth spurt in the adolescent (puberty) years, which continues longer in boys than girls.[29] The development of muscle mass is impacted by several factors, including age, sex, ethnicity, nutritional, hormonal and metabolic states, and exercise. Estimated muscle masses as a percentage of body weight in boys increases from about 42% at age 5 years to 54% by 17 years of age. In girls, the increase is from about 40 at age 5 years to 45% at 13 years of age, with a small decrease thereafter due to increasing body fat. From birth through adulthood, males tend to have higher muscle mass than females. There is a gradual decrease in muscle starting in the fourth decade of life with a more dramatic loss after 65 years of age.[113,129] This decrease is greater in men and in African-Americans. Low physical activity, tobacco smoking, and low testosterone are risk factors for sarcopenia in these populations.[129] For men and women over 64 years of age, sarcopenia was

reported at 22%–27% and increased further with advancing age.[130] It has been hypnotized that the sarcopenia that occurs in the elderly could be delayed by regular exercise, although numerous attempts have had minimum success.[89] Physical activity may need to be promoted during the early periods of life in order to create positive habits that favor the cardiovascular system, muscle, and bone.

Changes in Visceral Mass

Growth and development of the viscera parallel the changes in chemical compositions for muscle mass, in that there is a decrease in the proportion of water and an increase in the proportions of proteins and minerals.[9] Extracellular elements such as sodium and chloride decrease, whereas intracellular elements such as potassium, magnesium, and nitrogen either remain the same or increase. The brain grows rapidly from mid-gestation to 18 months after birth, when it reaches about 70% of its adult mass. Growth and development of the brain are characterized by a decrease in the percentage of water and an increase in protein, but differs from most other tissues in that lipids form an integral part of the structure. Myelination begins before birth, but most of it takes place between birth and 3 years of age, when the brain is about 80%–90% of its mass at maturity. Up to the age of 3 years there is a considerable increase in the concentration of lipid, with consequent decrease in the percentage of water. After 3 years, the changes in composition are relatively small.[97] During the myelination period there is a rapid accumulation in the brain of the omega-3 and omega-6 long chain polyunsaturated fatty acids (PUFAs)—docosahexaenoic acid (DHA; 22:6–3), arachidonic acid (AA; 20:4–6), and adrenic acid (22:4–6)—which are important structural components of the cell membrane phospholipids. The type of lipid that accumulates in the brain during these first few years is influenced by early nutrition and might have functional implications, especially related to visual maturation and cognitive development.[131] Preterm infants who receive formulas with a high linoleic to linolenic acid ratio and practically devoid of longer-chain PUFAs show a marked decrease in the DHA-to-AA ratio in brain and retina.[132] On the other hand, human milk contains enough precursors and long-chain PUFAs to fulfill the infant's requirements for brain growth. These factors have led to the study and development of infant formulas containing DHA and AA, which have been successful in approaching the fatty acid accretion pattern of breast-fed infants in babies who are fed with these formulas.[131]

The other major organs—liver, kidney, lung, heart, skin, and spleen—also change in chemical composition in a similar way, with a decrease in the proportion of water and an increase in proteins and minerals. At all ages, the kidney has more sodium and less potassium than muscle or liver.

Skin is an organ that changes in the composition. For the fetus, less than 20% by mass is collagen, the remaining being mainly elastin. By term, the collagen represents 63% and in the adult it is 90%.[97] The immune system is also an important component of the body's lean non muscle compartment, with lymphocytes accounting for about 2% of body weight and 8% of fat-free solids in the adult. The status of the immune system is intimately related to body composition via the known effects of cytokines on muscle protein metabolism.[2]

The relative proportions that major organs and tissues contribute to body weight are presented in Table 4-4. At any age, muscle, skeleton, skin, liver, and brain account for more than 70% of body mass. With increasing age, some organs, such as liver, spleen, and kidneys, appear to atrophy, whereas hypertrophy is common with others, such as prostate, lungs, and heart.[133]

General Changes in Fat-Free Mass

As noted earlier, fat-free mass (FFM) is not a true body composition compartment, but a concept developed in order to construct a two compartment model of body composition. FFM consists of multiple compartment, with water being the major component. Changes in FFM should not be over interpreted as necessarily reflecting similar changes in each of its subcomponents.

FFM increases rapidly during the late fetal and newborn periods, followed by a rapid slow down between the first and third years of life, and then a slow increase about 10 years of age, when the adolescent growth spurt is initiated. During the first years of life and childhood, there is a slightly higher FFM in males, which becomes more pronounced during the adolescent period. For boys, FFM increases rapidly between 12 and 15 years of age and continues until about to age 18 years. For girls, FFM increases steadily until about 15 years of age, then remains relatively constant.[134,135] The patterns of increase in FFM are correlated with changes in height.[29] At older adult ages there is a progressive loss of FFM, mainly reflecting loss of skeletal muscle and bone mass.[89]

Achieving Chemical Maturity

Healthy young adults have usually achieved chemical stability of their body composition by puberty.[1,9] The composition of FFM is relatively constant, with a water content of 72%–74%, a protein content of ~19%, and a mineral content of 6.8%.[1,29] The potassium content of the body is about 60–70 mmoles/kg for males and 50–60 mmoles/kg for females.[27] The completion of chemical maturation is also reflected in the stabilization of various constants relating one component of body composition to another, such as potassium to nitrogen and ECW to ICW, and intracellular potassium concentration.[90] However, in conditions such as pregnancy, advanced aging, disease, nutritional status, and genetics where an individual's FFM composition can be effected, the convenience of these constants is reduced. For example, the relative changes in the composition and density of FFM with age are shown in Figure 4-4. During growth, chemical maturity occurs at different stages for different constituents of FFM. Muscles, visceral tissues, and bone all reach chemical maturity at different rates. For example, bone mineral maturation appears to occur in the third decade of life. Muscle mass, reflected mainly by body potassium, increases gradually from birth to puberty for both sexes, then rapidly increases, reaching a plateau at about 20 years of age in males, and at about 17 years of age in females. Differences in peak muscle mass as it relates to timing of puberty are apparent between genders and among ethnicities.[135] By the end of the 3rd decade, there is a gradual decline in body potassium indicative of loss of muscle mass with aging.[27,29] (Figure 4-4A). In childhood, FFM contains relatively more water than that of adults. The hydration of FFM decreases from 80% to 83% in the newborn to about 75% in early puberty and then finally to about 73% in adults (Figure 4-4B). The body's protein content is about 15% at birth, increasing to about 19.5% in boys and 18.7% in girls by 10 years of age.[135] Approximately 0.6% of the FFM is carbohydrate or glycogen. Bone mineral content increases during early childhood and especially during adolescence. It is about 3.7% of FFM in infancy, increasing to about 5.02% in the prepubertal ages, then to about 6.2% in the late teen years, continuing to increase to about 6.8% in the adults.[1,27,52,53] A small gain in bone mass continues during the third decade of life and then it plateaus or has a slow decrease for the next several decades. Bone loss begins by the start of the 5th decade of life, with varying rates among skeletal sites and between sexes (Figure 4-4C). The density of the FFM (Figure 4-4D) also changes with age as it is dependent on its relative proportions of water, protein, and minerals. During growth, the water content decreases, while

the protein and mineral contents increase, resulting in an overall increase FFM density. With advanced aging, there are significant declines in muscle and bone mineral, such that FFM density declines at older ages (Figure 4-4D).[10] Consequently, estimates of body composition in children will be incorrect if adult constants for FFM hydration or density are used. For example, fat mass would be underestimated if calculated on the basis of adult TBW hydration and overestimated when calculated on the basis of body density.[136] Constants, adjusted for age and gender during childhood and adolescence, have been developed for FFM, making it possible to more accurately compare body composition by age, gender, and ethnic group.[1,135,136]

TABLE 4-4 Relative Contribution of Organs and Major Tissues to Body Weight

		Percent Age of Body Weight		
Organ or Tissue	**Fetus (20–24 weeks)**	**Full-Term Newborn**	**Adult Male**	**Adult Female**
Skeletal muscle	25.0	25.0	40.0	29.3
Skeleton	22.0	18.0	17.1	17.1
Skin	13.0	15.0	3.7	3.1
Liver	4.0	5.0	2.6	2.4
Brain	13.0	13.0	2.0	2.1
Lungs	3.3	1.5	1.4	1.4
Kidneys	3.1	1.6	0.4	0.5
Heart	0.6	0.5	0.5	0.4
Spleen	0.1	0.2	0.3	0.3

Adapted from Refs. 9 and 97.

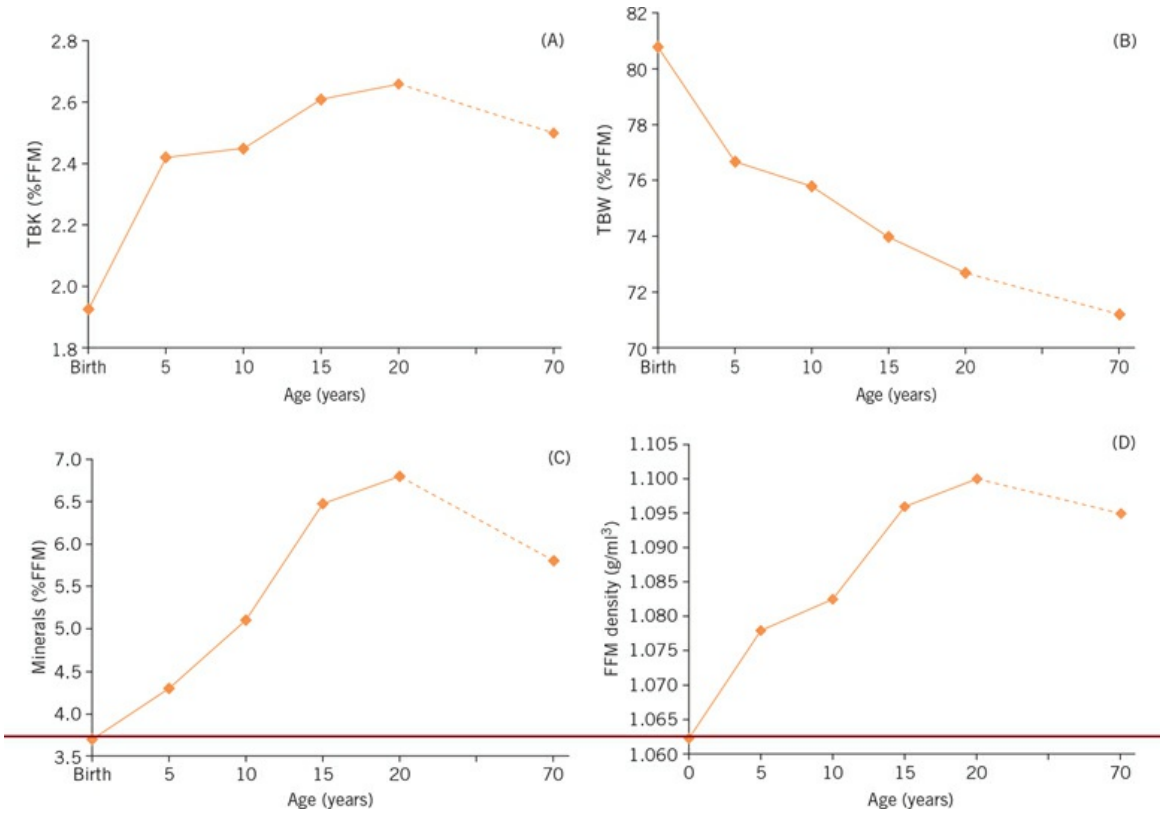


Figure 4-4 Changes in body composition components of the fat-free mass (FFM) over the human life span expressed as a percentage of FFM. (A) Total body potassium (TBK), (B) total body water (TBW), (C) Minerals, and (D) density of FFM (g/ml^3). (Adapted from Refs. 7,9,63,64.)

Changes in Linear Growth

During gestation, the fetus reaches its peak linear growth rate of 10–12 cm/month between the 10th and 30th weeks, reducing to about 3 cm/month (36 cm/year) at birth. Linear growth continues to decrease over the next three years: 15 cm/year at the end of the first year, 9 cm/year at the end of the second year, and 8 cm/year at the end of the third year. Linear growth continues to decrease over the next 2 years to about 6.5 cm/year, and is then relatively stable at about 5–6 cm/year until puberty. The start of puberty is both sex and ethnic dependent. Girls of European descent start their pubertal growth spurt at about 12 years of age, with a peak height velocity of 9 cm/year within 24 months. Boys of a similar ethnic decent start their pubertal growth spurt 2 years later, at about 14 years of age, with a higher peak velocity at 10 cm/year. Once peak height

velocity is achieved, annual growth increments decrease significantly.[137] In the Fels longitudinal study, adult stature was reached at 17.3 years for females and 21.2 years for males. Changes in body composition were related more to physiologic age than chronological age. Children who begin their adolescent growth spurt earlier tend to be taller and heavier than those who mature later.[134] Maximal height can also vary with ethnicity, where African-American males grow significantly taller than European-American males who are, in turn, taller than Mexican-American boys.[53,135] Similar ethnic differences are evident for females.[52,135] Tanner staging, an index of sexual maturity, is also an important component of growth assessment in children, particularly during the pubertal age period.

Changes with Weight Loss or Weight Gain

The range of weight gain, following full term birth, of healthy infants is 19–24 g/day. This decreases to roughly 9–10 g/day by the end of the first year, and to about 7 g/day by the end of the second year. Thereafter, weight gain is normally relatively stable at about 5.5 g/day until puberty. Weight change involves all the components of FFM and fat, with their contributions affected by the causative factors.[138] While BMI is considered a reliable indicator of excessive weight gain, changes in BMI often do not accurately reflect changes in body composition.[15,139] Malnourished children, for example, can continue to have abnormalities in body composition even after fully regaining weight.[140] The situation is similar for individuals who have a significant body weight increase in response to excessive intake. The composition of the weight gain is both fat and FFM. Though short-term weight gain is initially associated with proportionately greater increases in fat mass, both fat and FFM will be increased.[141] In obesity studies, the weight gained or lost is associated with about a 20%–30% changes in FFM. In subjects of low weight, such as malnourished children, premature infants, or patients with anorexia nervosa, the proportion of weight gained as FFM is larger, around 60%–75%, depending on the individual and type of diet. The excess weight of obese children is obviously composed of increased fat mass, but there are also increases in the protein and mineral masses. A propensity for greater fat accumulation in the trunk can promote greater mineral accumulation in the legs for older children.[142] An exception to this trend may be small for gestational age infants, who, following a catch up weight gain, tend to have greater adiposity and abdominal fat than appropriate for similar gestational age infants.[143] These features may be linked to long term risks

for central adiposity and insulin resistance later in life.

Changes in Body Composition during Malnutrition and Disease

Malnutrition and disease often results in body composition alterations beyond those expected from only underfeeding. Some of the methods used in the measurement body composition, and how it changes, may rely on assumptions derived from healthy children, which may have limited applicability in these cases. For example, electrolyte derangement and altered body water distribution related to a disease or clinical condition can invalidate the interpretation of the results of some techniques.[144,145] Disease-induced malnutrition can produce body compositional changes distinct from those caused by food deprivation alone. Significant changes in body composition may be masked by overall weight normalcy. Children with severe protein-energy malnutrition usually have increased TBW in relation to body weight, with ECW as an increased percentage of TBW. These changes are indicative of severe losses of cell mass, which may be detected by TBK measurements and loss of body fat. The cells retract and lose potassium, magnesium, and phosphorus. Body sodium is increased both extracellularly and intracellularly. The extracellular increase of sodium is generally less than the increase in ECW resulting in hyponatremia. Although serum potassium levels are usually normal, TBK is decreased. There are also deficiencies in magnesium, calcium, phosphorus, and trace elements (zinc, copper, selenium, and chromium). In severe cases, bone demineralization has been reported. The low TBK marked loss of muscle mass indicates depletion of body protein. If the energy imbalance continues, there can be slowing or cessation in linear growth, resulting in stunting. Therefore, alterations in ECW, sodium, and potassium should be taken into account early in the management of children with acute malnutrition. The process of recovery should start carefully and slowly, correcting for the water and electrolyte derangements, avoiding dangers associated with refeeding syndrome, particularly heart failure.[146] (See Chapter13, “Protein Energy Malnutrition: Pathophysiology, Clinical Consequences and Treatment”). In protein deprivation, there is an associated loss in non subcutaneous fat along with losses in muscle and visceral tissues. The ICW decreases, often with an expansion of the ECW as indicated by edema. These effects can obscure the severity of the malnutrition if body weight alone or a weight-for-height index alone is used to assess the child.

Changes in body composition are routinely observed in children with chronic diseases. These changes may be a direct consequence of the disease and caused by the treatment options (e.g., steroid associated osteopenia). Typically, lean mass is compromised during the most acute phase of illness, while the combination of lean and fat mass depletion is observed in less serious chronic conditions. Cancer, Crohn's disease, and cystic fibrosis are among the diseases where body composition abnormalities have been described. Children in remission from cancer have a greater percentage fat than healthy size-matched controls.[147–149] Similarly, children with Crohn's disease have reduced lean body mass, compared to controls, without significant concomitant reductions in fat mass. It has been shown that the degree of lean body mass loss is correlated with disease severity.[150] Cystic fibrosis is associated with deficits in lean body mass, fat mass, and bone BMD, with negative relationships with disease severity.[151] This suggests abnormalities in body composition are widespread in disease, and could be linked to patient outcome.[152]

CONCLUSIONS

Energy homeostasis is known to be important to human health, and the body's lean mass is the major contributor to metabolism and energy expenditure. Thus, assessment of body composition is an important aspect of understanding energy requirements during health and disease.[153] Body composition changes during normal growth, as well as in diseases and with their treatment. Growth includes chemical maturity of tissues, which occurs at different stages of development of the components of FFM. Fat mass also varies greatly during the life cycle and can be influenced by heredity, nutrition, lifestyle, and health status.

Body weight alone cannot be used as an acceptable measure of nutritional efficacy or status. The expansion of growth standards to include body composition reference ranges in childhood and adolescence could help in the recognition of abnormalities.[154] Body composition standards related to age, gender, body size, and ethnicity could also help in the recognition of differing patterns of distribution or accretion. In this sense, the beneficial role of exercise and the negative impact of a sedentary lifestyle could be emphasized. The possibility of critical periods for fat accretion and an understanding of the long term consequences of different body fat distributions might have important implications in the nutritional

management of children and adolescents.

As new and more accurate methods emerge, refinements for the reference infant, child, and adolescent models may be developed. The improvements will provide a measure of the biological diversity with age, gender, and ethnicity that can assist in the calibration of less expensive, surrogate methods more suitable for office practices and field studies. Although an ideal field method should be inexpensive, easy to use, reliable, noninvasive, and portable, it still needs to be validated and calibrated with one of the more advanced techniques. The clinician now has a variety of options in the body composition assessment armamentarium. The final interpretation of any results, however, should be assessed knowing the limitations and assumptions of the method selected.[155]

ACKNOWLEDGMENT

This chapter is a revised and updated version of the previous Body Composition and Growth chapter by Bechard L, Wroe E, and Ellis K in *Nutrition in Pediatrics: Basic Science and Clinical Applications*, 4th edition. Walker WA, Watkins JB, Duggan C, editors, 2006.

REFERENCES

1. Lohman TG. Applicability of body composition techniques and constants for children and youths. *Exerc Sport Sci Rev*. 1986;14:325–357.
2. Roubenoff R. Hormones, cytokines and body composition: can lessons from illness be applied to aging? *J Nutr*. 1993;123:469–473.
3. Hergenroeder AC, Klish WJ. Body composition in adolescent athletes. *Pediatr Clin North Am*. 1990;37:1057–1083.
4. Kuczmarski RJ. Need for body composition information in elderly subjects. *Am J Clin Nutr*. 1989;50:1150–1157; discussion 1231–1155.
5. Heymsfield SB, Waki M. Body composition in humans: advances in the development of multicompartment chemical models. *Nutr Rev*. 1991;49:97–108.
6. Ellis KJ. Human body composition: in vivo methods. *Physiol Rev*. 2000;80:649–680.
7. Heymsfi eld SB, Wang J, Lichtman S, et al. Body composition in elderly subjects: a critical appraisal of clinical methodology. *Am J Clin Nutr*. 1989;50:1167–1175; discussion1231–1165.
8. Wang ZM, Pierson RN, Jr, Heymsfi eld SB. The five-level model: A new approach to organizing body-composition research. *Am J Clin Nutr*. 1992;56:19–28.
9. Basic anatomical and physiological data for use in radiological protection: reference Values. A report of age- and gender-related differences in the anatomical and physiological characteristics of reference individuals. ICRP Publication 89. *Ann. ICRP*. 2002;32 (3–4):5–265.

10. Heymsfield SB, Wang Z, Baumgartner RN, et al. Body composition and aging: a study by in vivo neutron activation analysis. *J Nutr*. 1993;123:432–437.
11. Heymsfield SB, Waki M, Kehayias J, et al. Chemical and elemental analysis of humans in vivo using improved body composition models. *Am J Physiol*. 1991;261:E190–E198.
12. Kehayias JJ, Heymsfi eld SB, LoMonte AF, et al. In vivo determination of body fat by measuring total body carbon. *Am J Clin Nutr*. 1991;53:1339–1344.
13. Cole TJ, Faith MS, Pietrobelli A, Heo M. What is the best measure of adiposity change in growing children: BMI, BMI %, BMI z-score or BMI centile? *Eur J Clin Nutr*. 2005;59:419–425.
14. Dietz WH, Bellizzi MC. Introduction: the use of body mass index to assess obesity in children. *Am J Clin Nutr*. 1999;70:123S–125S.
15. Ellis KJ, Abrams SA, Wong WW. Monitoring childhood obesity: Assessment of the weight/height index. *Am J Epidemiol*. 1999;150:939–946.
16. Lohman TG, Roche AF, Martorell R. *Anthropometric Standardization Reference Manual*. Champaign, IL: Human Kinetics Books; 1988.
17. Mei Z, Grummer-Strawn LM, Pietrobelli A, et al. Validity of body mass index compared with other body-composition screening indexes for the assessment of body fatness in children and adolescents. *Am J Clin Nutr*. 2002;75:978–985.
18. Chumlea WC, Baumgartner RN. Status of anthropometry and body composition data in elderly subjects. *Am J Clin Nutr*. 1989;50:1158–1166; discussion 1231–1155.
19. Chumlea WC, Guo SS, Kuczmarski RJ, et al. Reliability for anthropometric measurements in the Hispanic Health and Nutrition Examination Survey (HHANES 1982–1984). *Am J Clin Nutr*. 1990;51:902S–907S.
20. Lukaski HC. Methods for the assessment of human body composition: Traditional and new. *Am J Clin Nutr*. 1987;46:537–556.
21. Moller R, Tafeit E, Smolle KH, et al. Estimating percentage total body fat and determining subcutaneous adipose tissue distribution with a new noninvasive optical device LIPOMETER. *Am J Human Biol*. 2000;12:221–230.
22. Fuller NJ, Dewit O, Wells JC. The potential of near infrared interactance for predicting body composition in children. *Eur J Clin Nutr*. 2001;55:967–972.
23. Jurimae T, Sudi K, Payerl D, et al. Relationships between bioelectric impedance and subcutaneous adipose tissue thickness measured by LIPOMETER and skinfold calipers in children. Eur J Appl Physiol. 2003;90:178–184.
24. Kabir N, Forsum E. Estimation of total body fat and subcutaneous adipose tissue in full-term infants less than 3 months old. *Pediatr Res*. 1993;34:448–454.
25. Mattsson S, Thomas BJ. Development of methods for body composition studies. *Phys Med Biol*. 2006;51:R203–R228.
26. Ellis KJ. Whole-body counting and neutron activation analysis. In: Heymsfield SB, Lohman TG, Wang ZM, Going SB, editors. *Human Body Composition*, 2nd ed. Champaign, IL: Human Kinetics; 2005:51–62.
27. Ellis KJ. Reference man and woman more fully characterized. Variations on the basis of body size, age, sex, and race. *Biol Trace Elem Res*. 1990;26–27:385–400.
28. Ellis KJ, Shypailo RJ, Hergenroeder A, et al. Total body calcium and bone mineral content: comparison of dual-energy X-ray absorptiometry with neutron activation analysis. *J Bone Miner Res*. 1996;11:843–848.
29. Forbes GB. *Human Body Composition. Growth, Aging, Nutrition and Activity*. New York, NY: Springer-Verlag; 1987.
30. Wang Z, Heshka S, Wang J, Heymsfield SB. Total body protein mass: validation of total body potassium prediction model in children and adolescents. *J Nutr*.

2006;136:1032–1036.
31. Schoeller DA. Hydrometry. In: Heymsfi eld SB, Lohman TG, Wang ZM, Going SB, editors. *Human Body Composition*, 2nd ed. Champaign, IL: Human Kinetics; 2005:35–50.
32. Bauer K, Bovermann G, Roithmaier A, et al. Body composition, nutrition, and fluid balance during the first two weeks of life in preterm neonates weighing less than 1500 grams. *J Pediatr*. 1991;118:615–620.
33. Segal KR, Burastero S, Chun A, et al. Estimation of extracellular and total body water by multiple-frequency bioelectrical-impedance measurement. *Am J Clin Nutr*. 1991;54:26–29.
34. Mast M, Sonnichsen A, Langnase K, et al. Inconsistencies in bioelectrical impedance and anthropometric measurements of fat mass in a field study of prepubertal children. *Br J Nutr*. 2002;87:163–175.
35. Coppini LZ, Waitzberg DL, Campos AC. Limitations and validation of bioelectrical impedance analysis in morbidly obese patients. *Curr Opin Clin Nutr Metab Care*. 2005;8:329–332.
36. Gelbrich G, Reich A, Muller G, Kiess W. Knowing more by fewer measurements: About the (In)ability of bioelectric impedance to enhance obesity research in children. *J Pediatr Endocrinol Metab*. 2005;18:265–273.
37. Kraemer M. A new model for the determination of fluid status and body composition from bioimpedance measurements. *Physiol Meas*. 2006;27:901–919.
38. Moissl UM, Wabel P, Chamney PW, et al. Body fluid volume determination via body composition spectroscopy in health and disease. *Physiol Meas*. 2006;27:921–933.
39. Fioritto M. Application of the TOBEC measurement for determining fat and fat-free mass of the human infant. In: Klish WJ, Kretchmer N, editors. *Body Composition Measurements in Infants and Children. Report of the 98th Ross Conference on Pediatric Research*. Columbus, OH: Ross Laboratories; 1989. pp. 57–64.
40. Fields DA, Goran MI, McCrory MA. Body-composition assessment via air-displacement plethysmography in adults and children: A review. *Am J Clin Nutr*. 2002;75:453–467.
41. Wells JC, Fuller NJ. Precision of measurement and body size in whole-body air-displacement plethysmography. *Int J Obes Relat Metab Disord*. 2001;25:1161–1167.
42. Dempster P, Aitkens S. A new air displacement method for the determination of Human Body Composition. *Med Sci Sports Exerc*. 1995;27:1692–1697.
43. Urlando A, Dempster P, Aitkens S. A new air displacement plethysmograph for the measurement of body composition in infants. *Pediatr Res*. 2003;53:486–492.
44. Ellis KJ, Yao M, Shypailo RJ, et al. Body composition assessment in infancy: Air-displacement plethysmography versus a four-compartment criterion model. *Am J Clin Nutr*. 2007;85:90–95.
45. Ma G, Yao M, Liu Y, et al. Validation of a new pediatric air-displacement plethysmograph for assessing body composition in infants. *Am J Clin Nutr*. 2004;79:653–660.
46. Mazess RB, Barden HS, Bisek JP, Hanson J. Dual-energy X-ray absorptiometry for total-body and regional bone mineral and soft-tissue composition. *Am J Clin Nutr*. 1990;51:1106–1112.
47. Venkataraman PS, Ahluwalia BW. Total bone mineral content and body composition by X-ray densitometry in newborns. *Pediatrics*. 1992;90:767–770.
48. Venkataraman PS, Duke JC. Bone mineral content of healthy, full-term neonates.

Effect of race, gender, and maternal cigarette smoking. *Am J Dis Child*. 1991;145:1310–1312.
49. Laskey MA, Prentice A. Comparison of adult and paediatric spine and whole body software for the Lunar dual energy X-ray absorptiometer. *Br J Radiol*. 1999;72:967–976.
50. Koo WW, Hammami M, Shypailo RJ, Ellis KJ. Bone and body composition measurements of small subjects: discrepancies from software for fan-beam dual energy X-ray absorptiometry. *J Am Coll Nutr*. 2004;23:647–650.
51. Hammami M, Walters JC, Hockman EM, Koo WW. Disproportionate alterations in body composition of large for gestational age neonates. *J Pediatr*. 2001;138:817–821.
52. Ellis KJ, Abrams SA, Wong WW. Body composition of a young, multiethnic female population. *Am J Clin Nutr*. 1997;65:724–731.
53. Ellis KJ. Body composition of a young, multiethnic, male population. *Am J Clin Nutr*. 1997;66:1323–1331.
54. Moyer-Mileur L, Xie B, Ball S, et al. Predictors of bone mass by peripheral quantitative computed tomography in early adolescent girls. *J Clin Densitom*. 2001;4:313–323.
55. Schoenau E, Neu CM, Rauch F, Manz F. Gender-specific pubertal changes in volumetric cortical bone mineral density at the proximal radius. *Bone* 2002;31:110–113.
56. Fuller MF, Fowler PA, McNeill G, Foster MA. Imaging techniques for the assessment of body composition. *J Nutr*. 1994;124:1546S–1550S.
57. Kvist H, Chowdhury B, Grangard U, et al. Total and visceral adipose-tissue volumes derived from measurements with computed tomography in adult men and women: Predictive equations. *Am J Clin Nutr*. 1988;48:1351–1361.
58. Seidell JC, Bakker CJ, van der Kooy K. Imaging techniques for measuring adipose-tissue distribution—a comparison between computed tomography and 1.5-T magnetic resonance. *Am J Clin Nutr*. 1990;51:953–957.
59. Olhager E, Flinke E, Hannerstad U, Forsum E. Studies on *Human Body Composition* during the first 4 months of life using magnetic resonance imaging and isotope dilution. *Pediatr Res*. 2003;54:906–912.
60. NapolitanoA, Miller SR, Murgatroyd PR, et al. Validation of a quantitative magneticresonance method for measuring Human Body Composition. *Obesity* 2008;16:191–198.
61. Rogers IS, Ness AR, Steer CD, et al. Associations of size at birth and dual-energy X-ray absorptiometry measures of lean and fat mass at 9 to 10 y of age. *Am J Clin Nutr*. 2006;84:739–747.
62. Wells JCK, Fewtrell MS. Measuring body composition. *Arch Dis Child*. 2006;91:612–617.
63. Fomon SJ, Haschke F, Ziegler EE, Nelson SE. Body composition of reference children from birth to age 10 years. *Am J Clin Nutr*. 1982;35:1169–1175.
64. Haschke F. Body composition of adolescent males. Part I. Total body water in normal adolescent males. Part II. Body composition of the male reference adolescent. *Acta Paediatr Scand Suppl*. 1983;307:1–23.
65. Butte NF, Hopkinson JM, Wong WW, et al. Body composition during the first 2 years of life: An updated reference. *Pediatr Res*. 2000;47:578–585.
66. Poissonnet CM, Burdi AR, Garn SM. The chronology of adipose tissue appearance and distribution in the human fetus. *Early Hum Dev*. 1984;10:1–11.
67. Budge H, Gnanalingham MG, Gardner DS, et al. Maternal nutritional programming of fetal adipose tissue development: Long-term consequences for

later obesity. *Birth Defects Res C Embryo Today*. 2005;75:193–199.
68. Euser AM, Finken MJ, Keijzer-Veen MG, et al. Associations between prenatal and infancy weight gain and BMI, fat mass, and fat distribution in young adulthood: A prospective cohort study in males and females born very preterm. *Am J Clin Nutr*. 2005;81:480–487.
69. McMillen IC, Muhlhausler BS, Duffield JA, Yuen BS. Prenatal programming of postnatal obesity: Fetal nutrition and the regulation of leptin synthesis and secretion before birth. *Proc Nutr Soc*. 2004;63:405–412.
70. Ravelli GP, Stein ZA, Susser MW. Obesity in young men after famine exposure in utero and early infancy. *N Engl J Med*. 1976;295:349–353.
71. Catalano PM, Thomas A, Huston-Presley L, Amini SB. Increased fetal adiposity: A very sensitive marker of abnormal in utero development. *Am J Obstet Gynecol*. 2003;189:1698–1704.
72. Rogers I. The influence of birth weight and intrauterine environment on adiposity and fat distribution in later life. *Int J Obes Relat Metab Disord*. 2003;27:755–777.
73. Oken E, Gillman MW. Fetal origins of obesity. *Obes Res*. 2003;11:496–506.
74. Rolland-Cachera MF, Deheeger M, Guilloud-Bataille M, et al. Tracking the development of adiposity from one month of age to adulthood. *Ann Hum Biol* 1987;14:219–229.
75. Gesta S, Bluher M, Yamamoto Y, et al. Evidence for a role of developmental genes in the origin of obesity and body fat distribution. *Proc Natl Acad Sci USA*. 2006;103:6676–6681.
76. Butte NF, Wong WW, Hopkinson JM, et al. Infant feeding mode affects early growth and body composition. *Pediatrics*. 2000;106:1355–1366.
77. Chumlea WC, Guo SS, Kuczmarski RJ, et al. Body composition estimates from NHANES III bioelectrical impedance data. *Int J Obes Relat Metab Disord*. 2002;26:1596–1609.
78. Vague J, Vague P, Jubelin J, Barre A. Fat distribution, obesities and health: Evolution of concepts. In: Bouchard C, Johnston FE, editors. *Fat Distribution During Growth and Later Health Outcomes. Current Topics in Nutrition and Disease*, Vol 17. New York, NY: Alan R. Liss; 1988:9–41.
79. Lopez-Blanco M, Hernandez-Valera Y, Landaeta-Jimenez M, Henriquez-Perez G. Crecimiento y nutricion en la region Latinoamericana. *An Venez Nutr*. 1993;6:47–90.
80. Whitaker RC, Wright JA, Pepe MS, et al. Predicting obesity in young adulthood from childhood and parental obesity. *N Engl J Med*. 1997;337:869–873.
81. Rolland-Cachera MF, Bellisle F, Deheeger M, et al. Influence of body fat distribution during childhood on body fat distribution in adulthood: A two-decade follow-up study. *Int J Obes*. 1990;14:473–481.
82. Kuh D, Hardy R, Chaturvedi N, Wadsworth ME. Birth weight, childhood growth and abdominal obesity in adult life. *Int J Obes Relat Metab Disord*. 2002;26:40–47.
83. Gutin B, Islam S, Manos T, et al. Relation of percentage of body fat and maximal aerobic capacity to risk factors for atherosclerosis and diabetes in black and white seven- to eleven-year-old children. *J Pediatr*. 1994;125:847–852.
84. Must A, Jacques PF, Dallal GE, et al. Long-term morbidity and mortality of overweight adolescents. A follow-up of the Harvard Growth Study of 1922 to 1935. *N Engl J Med*. 1992;327:1350–1355.
85. Despres JP. Is visceral obesity the cause of the metabolic syndrome? *Ann Med*. 2006;38:52–63.
86. Dennison BA, Erb TA, Jenkins PL. Television viewing and television in bedroom

associated with overweight risk among low-income preschool children. *Pediatrics*. 2002;109:1028–1035.
87. Burdette HL, Whitaker RC. A national study of neighborhood safety, outdoor play, television viewing, and obesity in preschool children. *Pediatrics*. 2005;116:657–662.
88. Sabatini S. The female athlete triad. *Am J Med Sci*. 2001;322:193–195.
89. Evans WJ, Campbell WW. Sarcopenia and age-related changes in body composition and functional capacity. *J Nutr*. 1993;123:465–468.
90. Spady DW. Normal body composition of infants and children. In: Klish WJ, Kretchmer N, editors. *Body Composition Measurements in Infants and Children. Report of the 98th Ross Conference on Pediatric Research*. Columbus, OH: Ross Laboratories; 1989:67–73.
91. Silva AM, Wang J, Pierson RN Jr, et al. Extracellular water: Greater expansion with age in African Americans. *J Appl Physiol*. 2005;99:261–267.
92. Hartnoll G, Betremieux P, Modi N. Body water content of extremely preterm infants at birth. *Arch Dis Child Fetal Neonatal Ed*. 2000;83:F56-F59.
93. Robert S, Zarowitz BJ, Hyzy R, et al. Bioelectrical impedance assessment of nutritional status in critically ill patients. *Am J Clin Nutr*. 1993;57:840–844.
94. Matkovic V, Ilich JZ. Calcium requirements for growth: Are current recommendations adequate? *Nutr Rev*. 1993;51:171–180.
95. Matkovic V. Calcium intake and peak bone mass. *N Engl J Med*. 1992;327:119–120.
96. Whiting SJ, Vatanparast H, Baxter-Jones A, et al. Factors that affect bone mineral accrual in the adolescent growth spurt. *J Nutr*. 2004;134:696S–700S.
97. White DR, Widdowson EM, Woodard HQ, Dickerson JW. The composition of body tissues (II). Fetus to young adult. *Br J Radiol*. 1991;64:149–159.
98. Chan GM, Mileur L, Hansen JW. Calcium and phosphorus requirements in bone mineralization of preterm infants. *J Pediatr*. 1988;113:225–229.
99. Forbes GB. Some remarks on bone mineralization. *J Pediatr*. 1988;113:167–171.
100. Steichen JJ, Gratton TL, Tsang RC. Osteopenia of prematurity: The cause and possible treatment. *J Pediatr*. 1980;96:528–534.
101. Greer FR, McCormick A. Improved bone mineralization and growth in premature infants fed fortified own mother's milk. *J Pediatr*. 1988;112:961–969.
102. Lapillonne A, Salle BL, Glorieux FH, Claris O. Bone mineralization and growth are enhanced in preterm infants fed an isocaloric, nutrient–enriched preterm formula through term. *Am J Clin Nutr*. 2004;80:1595–1603.
103. Minton SD, Steichen JJ, Tsang RC. Bone mineral content in term and preterm appropriate-for-gestational-age infants. *J Pediatr*. 1979;95:1037–1042.
104. Greer FR, Lane J, Weiner S, Mazess RB. An accurate and reproducible absorptiometric technique for determining bone mineral content in newborn infants. *Pediatr Res*. 1983;17:259–262.
105. De Schepper J, Cools F, Vandenplas Y, Louis O. Whole body bone mineral content is similar at discharge from the hospital in premature infants receiving fortified breast milk or preterm formula. *J Pediatr Gastroenterol Nutr*. 2005;41:230–234.
106. Mimouni F, Tsang RC. Bone mineral content: Data analysis. *J Pediatr*. 1988;113:178–180.
107. Hillman LS. Bone mineral content in term infants fed human milk, cow milk-based formula, or soy-based formula. *J Pediatr*. 1988;113:208–212.
108. Abrams SA, Schanler RJ, Garza C. Bone mineralization in former very low birth weight infants fed either human milk or commercial formula. *J Pediatr*.

1988;112:956–960.
109. Steichen JJ, Tsang RC. Bone mineralization and growth in term infants fed soy-based or cow milk-based formula. *J Pediatr*. 1987;110:687–692.
110. Mimouni F, Campaigne B, Neylan M, Tsang RC. Bone mineralization in the first year of life in infants fed human milk, cow-milk formula, or soy-based formula. *J Pediatr*. 1993;122:348–354.
111. Specker BL, Beck A, Kalkwarf H, Ho M. Randomized trial of varying mineral intake on total body bone mineral accretion during the first year of life. *Pediatrics* 1997;99:E12.
112. Young RJ, Antonson DL, Ferguson PW, et al. Neonatal and infant feeding: Effect on bone density at 4 years. *J Pediatr Gastroenterol Nutr*. 2005;41:88–93.
113. Arabi A, Nabulsi M, Maalouf J, et al. Bone mineral density by age, gender, pubertal stages, and socioeconomic status in healthy Lebanese children and adolescents. *Bone* 2004;35:1169–1179.
114. Taaffe DR, Cauley JA, Danielson M, et al. Race and sex effects on the association between muscle strength, soft tissue, and bone mineral density in healthy elders: the Health, Aging, and Body Composition Study. *J Bone Miner Res*. 2001;16:1343–1352.
115. Chan GM. Dietary calcium and bone mineral status of children and adolescents. *Am J Dis Child*. 1991;145:631–634.
116. Vidulich L, Norris SA, Cameron N, Pettifor JM. Differences in bone size and bone mass between black and white 10-year-old South African children. *Osteoporos Int*. 2006;17:433–440.
117. Bachrach LK, Hastie T, Wang MC, et al. Bone mineral acquisition in healthy Asian, Hispanic, black, and Caucasian youth: A longitudinal study. *J Clin Endocrinol Metab*. 1999;84:4702–4712.
118. Yilmaz D, Ersoy B, Bilgin E, et al. Bone mineral density in girls and boys at different pubertal stages: Relation with gonadal steroids, bone formation markers, and growth parameters. *J Bone Miner Metab*. 2005;23:476–482.
119. Abrams SA. Normal acquisition and loss of bone mass. *Horm Res*. 2003;60:71–76.
120. Abrams SA. Pubertal changes in calcium kinetics in girls assessed using 42Ca. *Pediatr Res*. 1993;34:455–459.
121. Prentice A. Diet, nutrition and the prevention of osteoporosis. *Public Health Nutr*. 2004;7:227–243.
122. Hunter DJ, de Lange M, Andrew T, et al. Genetic variation in bone mineral density and calcaneal ultrasound: A study of the influence of menopause using female twins. *Osteoporos Int*. 2001;12:406–411.
123. Seeman E, Hopper JL, Bach LA, et al. Reduced bone mass in daughters of women with osteoporosis. *N Engl J Med*. 1989;320:554–558.
124. Harris M, Nguyen TV, Howard GM, et al. Genetic and environmental correlations between bone formation and bone mineral density: a twin study. *Bone*. 1998;22:141–145.
125. Greer FR, Krebs NF. Optimizing bone health and calcium intakes of infants, children, and adolescents. *Pediatrics*. 2006;117:578–585.
126. Goulding A, Taylor RW, Jones IE, et al. Overweight and obese children have low bone mass and area for their weight. *Int J Obes Relat Metab Disord*. 2000;24:627–632.
127. Heymsfield SB, Smith R, Aulet M, et al. Appendicular skeletal muscle mass: Measurement by dual-photon absorptiometry. *Am J Clin Nutr*. 1990;52:214–218.
128. Poortmans JR, Boisseau N, Moraine JJ, et al. Estimation of total-body skeletal

muscle mass in children and adolescents. *Med Sci Sports Exerc*. 2005;37:316–322.
129. Szulc P, Duboeuf F, Marchand F, Delmas PD. Hormonal and lifestyle determinants of appendicular skeletal muscle mass in men: The MINOS study. *Am J Clin Nutr*. 2004;80:496–503.
130. Iannuzzi-Sucich M, Prestwood KM, Kenny AM. Prevalence of sarcopenia and predictors of skeletal muscle mass in healthy, older men and women. *J Gerontol A Biol Sci Med Sci*. 2002;57:M772–M777.
131. Fleith M, Clandinin MT. Dietary PUFA for preterm and term infants: Review of clinical studies. *Crit Rev Food Sci Nutr*. 2005;45:205–229.
132. Martinez M. Tissue levels of polyunsaturated fatty acids during early human development. *J Pediatr*. 1992;120:S129-S138.
133. St-Onge MP. Relationship between body composition changes and changes in physical function and metabolic risk factors in aging. *Curr Opin Clin Nutr Metab Care*. 2005;8:523–528.
134. Siervogel RM, Demerath EW, Schubert C, et al. Puberty and body composition. *Horm Res*. 2003;60:36–45.
135. Ellis KJ, Shypailo RJ, Abrams SA, Wong WW. The reference child and adolescent models of body composition. A contemporary comparison. *Ann N Y Acad Sci*. 2000;904:374–382.
136. Rolland-Cachera MF. Body composition during adolescence: Methods, limitations and determinants. *Horm Res*. 1993;39:25–40.
137. Roche AF. *Growth, Maturation and Body Composition. The Fels Longitudinal Study 1929–1991*. Cambridge, UK: Cambridge University Press; 1992.
138. Nicholls D, Wells JC, Singhal A, Stanhope R. Body composition in early onset eating disorders. *Eur J Clin Nutr*. 2002;56:857–865.
139. Demerath EW, Schubert CM, Maynard LM, et al. Do changes in body mass index percentile reflect changes in body composition in children? Data from the Fels Longitudinal Study. *Pediatrics* 2006;117:e487-e495.
140. Scalfi L, Polito A, Bianchi L, et al. Body composition changes in patients with anorexia nervosa after complete weight recovery. *Eur J Clin Nutr*. 2002;56:15–20.
141. Kyle UG, Zhang FF, Morabia A, Pichard C. Longitudinal study of body composition changes associated with weight change and physical activity. *Nutrition*. 2006;22:1103–1111.
142. Wells JC, Fewtrell MS, Williams JE, et al. Body composition in normal weight, overweight and obese children: Matched case–control analyses of total and regional tissue masses, and body composition trends in relation to relative weight. *Int J Obes (Lond)*. 2006;30:1506–1513.
143. Ibanez L, Ong K, Dunger DB, de Zegher F. Early development of adiposity and insulin resistance after catch-up weight gain in small-for-gestational-age children. *J Clin Endocrinol Metab*. 2006;91:2153–2158.
144. Warner JT, Evans WD, Webb DK, Gregory JW. Pitfalls in the assessment of body composition in survivors of acute lymphoblastic leukaemia. *Arch Dis Child*. 2004;89:64–68.
145. Wells JC. Body composition in childhood: Effects of normal growth and disease. *Proc Nutr Soc*. 2003;62:521–528.
146. Crook MA, Hally V, Panteli JV. The importance of the refeeding syndrome. *Nutrition*. 2001;17:632–637.
147. Murphy AJ, Wells JC, Williams JE, et al. Body composition in children in remission from acute lymphoblastic leukemia. *Am J Clin Nutr*. 2006;83:70–74.
148. Nysom K, Holm K, Michaelsen KF, et al. Degree of fatness after treatment for acute lymphoblastic leukemia in childhood. *J Clin Endocrinol Metab*.

1999;84:4591–4596; erratum appears in *J Clin Endocrinol Metab*. 2001;86:1758.
149. Nysom K, Holm K, Michaelsen KF, et al. Degree of fatness after allogeneic BMT for childhood leukaemia or lymphoma. *Bone Marrow Transplant*. 2001;27:817–820.
150. Burnham JM, Shults J, Semeao E, et al. Body-composition alterations consistent with cachexia in children and young adults with Crohn disease. *Am J Clin Nutr* 2005;82:413–420.
151. Bianchi ML, Romano G, Saraifoger S, et al. BMD and body composition in children and young patients affected by cystic fibrosis. *J Bone Miner Res*. 2006;21:388–396.
152. Kyle UG, Morabia A, Slosman DO, et al. Contribution of body composition to nutritional assessment at hospital admission in 995 patients: A controlled population study. *Br J Nutr*. 2001;86:725–731.
153. Zakeri I, Puyau MR, Adolph AL, et al. Normalization of energy expenditure data for differences in body mass or composition in children and adolescents. *J Nutr*. 2006;136:1371–1376.
154. Kelly TL, Wilson KE, Heymsfield SB. Dual Energy X-Ray Absorptiometry Body Composition Reference Values from NHANES. *PLoS ONE*. 2009;4(9):e7038. doi:10.1371/journal.pone.0007038.
155. Hill RJ, Davies PS. You look alright to me: Compromised nutritional status in paediatric patients with ulcerative colitis. *J Pediatr Gastroenterol Nutr*. 2012, Nov 27 (Epub ahead of print).

Carbohydrates

5

Clemens Kunz, PhD, Lena Geiger, MSc, and Christoph Fusch, MD, PhD

BACKGROUND AND OBJECTIVES

Carbohydrates (CHO) play an essential role with regard to the development of physiological functions of the entire gastrointestinal (GI) tract in neonates, infants, and children. In this chapter, we will not review the general aspects of digestion and absorption of CHO but select some special topics that are currently discussed. We focus on

i. the influence of CHO on intestinal microbial composition and activity;
ii. differences between prebiotics in human milk versus oligosaccharides added to milk and other food products;
iii. potential risks of a high fructose (Frc) intake;
iv. recommendations for CHO intake;
v. disorders involving CHO.

The nutritional status of pregnant women has been recognized to play a major role in fetal development, which may affect the manifestation of diseases later in life (metabolic programming).[1–4] From the time of birth, nutrition of the infant becomes most important. Here, the intestinal microbiota is of particular relevance owing to its strong impact on the development of the unspecific defense and on the immune system.[5–11] Currently, the question is intensely discussed how composition and activity of the microbiota can be modulated by the intake of dietary components such as CHO.[12,13] Products enriched with special components or mixtures, such as prebiotic oligosaccharides (PBOs), play a major role in the nutrition of infants and children.

Amongst CHO, Frc has received special attention in recent years not only because its intake increased markedly during the last 10 years but also the metabolic processes of Frc is also considered to be potentially linked to

various health risks. Therefore, the controversy about Frc and its health effects, for example, the potential contribution to the obesity pandemic, will be covered as well.

The principle how to establish recommendations for digestible CHO intake as well as for distinct CHO members are reviewed; actual figures are given and the role of body growth is critically discussed. Second, the basis of recommendations for the intake of nondigestible CHO, that is, dietary fibers (DFs) is delineated. Finally, an overview about diseases that lead to or involve CHO metabolism is given.

In the following, we will first give a short introduction into some basic aspects of CHO followed by their functional roles in human nutrition.

DEFINITION, ORIGIN, AND CLASSIFICATION OF CARBOHYDRATES

CHO consist of carbon, hydrogen, and oxygen in the ratio of 1:2:1 (empirical formula $C_nH_{2n}0_n$). They are found in a variety of foods in the human diet and are largely derived from plant sources. However, sugars are also widely used in many products that are not of plant origin.

In humans, CHO occur as a product of the intermediary metabolism (gluconeogenesis, glycogenesis). Under starvation, the body can produce CHO from glycoplastic amino acids; synthesis from glycerol of triacylglycerols (fat) and fatty acids is also possible.

Classification

Like other macronutrients, CHO are classified into four main groups, based on their molecular size, which is determined by the degree of polymerization:

- Monosaccharides
- Disaccharides (2 monosaccharides)
- Oligosaccharides (3–9 monosaccharides)
- Polysaccharides (>9 monosaccharides)

The term “sugars” comprises monosaccharides and disaccharides. They occur as single and small molecules and are the building blocks of the more complex CHO. These can be stored or can be used as structural components.

Monosaccharides and Disaccharides (Sugars)

Monosaccharides are the smallest building blocks of CHO. In contrast to disaccharides, oligosaccharides, and polysaccharides, monosaccharides cannot be further degraded by acid hydrolysis. They are composed of a chain of three to seven carbons. Based on the number of carbons, the monosaccharides are classified into trioses, tetroses, pentoses, hexoses, and heptoses. Only 3 of the 12 naturally occurring hexoses can be absorbed and metabolized by the human body. These are glucose, galactose, and Frc.

Disaccharides are composed of two equal or different monosaccharides. The common disaccharides are lactose, sucrose, and maltose. Lactose is found in very high amounts in milk and dairy products and is the major source of CHO in the nutrition of milk-fed infants.

Oligosaccharides (3–9 Monosaccharides)

Oligosaccharides are found in fermented products as well as in vegetables. Various oligosaccharides are also highly abundant in breast milk.

Oligosaccharides are a diverse group of soluble CHO, including trisaccharides and tetrasaccharides such as raffinose and stachyose, maltodextrins from partial degradation of starch, fructosaccharides and galactooligosaccharides, and inulin. Many of the oligosaccharides are resistant against the enzymes of the intestinal tract and are fermented in the colon. Therefore, they are called nondigestible oligosaccharides.

Polysaccharides (>9 Monosaccharides)

The most important building block of natural polysaccharides is glucose. In nature, many polysaccharides are used for energetic purposes as building blocks for structural components. Polysaccharides may be divided into starch and nonstarch polysaccharides (NSP). They are distinguished by the presence of α-glucosidic bonds.

Starch

The most common storage form in plants is starch, which is composed of α-glucan and a mixture of the two polymers amylose and amylopectin. Amylose, which usually comprises 20%–30%, is a polymer of α-1,4 linked D-glucose. The molecule consists of linear chains that are turned into a tight helix. The second and quantitatively major component of naturally occurring starch (70%–80%), amylopectin, contains additional α-1,6 glycosidic linkages, resulting in the highly branched structure of this polymer.

NSPs are mostly components of plant cell walls. Other NSPs are also present in plant food as gums or mucilages. Cell wall NSPs are quantitatively more significant and are found in all fruits, vegetables, cereals, and nuts. They escape digestion in the upper gut and pass into the large intestine where they are fermented by the microbiota.

The classification of CHO and their food source is given in Table 5-1.

FUNCTIONAL ASPECTS OF CARBOHYDRATES

Carbohydrates and Intestinal Microbiota

As summarized in Figure 5-1, CHO is either a source of energy or essential for various systemic effects after being absorbed. Local effects within the GI tract include being a substrate for the microbiota or by directly influencing and thereby modulating epithelial cell functions.

Interactions between microorganisms and the human host are strongly influenced by the fermentation of CHO with a subsequent effect on, for example, (a) growth promotion of bacteria that can utilize CHO, (b) formation of short-chained fatty acids (SCFAs), which are an important supplier of energy for epithelial cells, or (c) systemic effects of SCFA, which eventually may affect CHO and lipid metabolism.[14]

The use of CHO for fermentation strongly depends on the microbial composition of the GI tract, which changes from infancy to adulthood. Current studies aim at defining the dominant forces, which shape the microbiota.[15] As microorganisms differ in their preference for CHO, the dietary supply with various CHO as well as glycans from mucosal secretions has a direct influence on the microbial balance in the gut. CHO are of particular importance in situations when the normal gut microbiota has been destroyed and needs to be restored or when the microbiota has to be established, as is the case in the postnatal period.

TABLE 5-1 Classification of Carbohydrates and Sources

Class (DP)	Subgroup	Principal Components	Source(s)
Sugars (1–2)	Monosaccharides	Glucose Galactose Fructose	Fruits and vegetables
	Disaccharides	Lactose Sucrose	Milk and milk products

			Fruits and vegetables
Oligosaccharides (3–9)	Malto-oligosaccharides Nondigestible oligosaccharides	Maltose Stachyose, raffinose Fructo-oligosaccharides (FOS) Galacto-oligosaccharides (GOS)	Glucose syrups Sugar-beet root, legume, cucurbits Vegetables Milk and milk products
Polysaccharides (≥9)	Starch	Amylose Amylopectin Modified starches	Cereals, potatoes
	Nonstarch polysaccharides	Cellulose Hemicellulose Pectin	Vegetables, cereals, fruits

Abbreviations: DP, degree of polymerization.

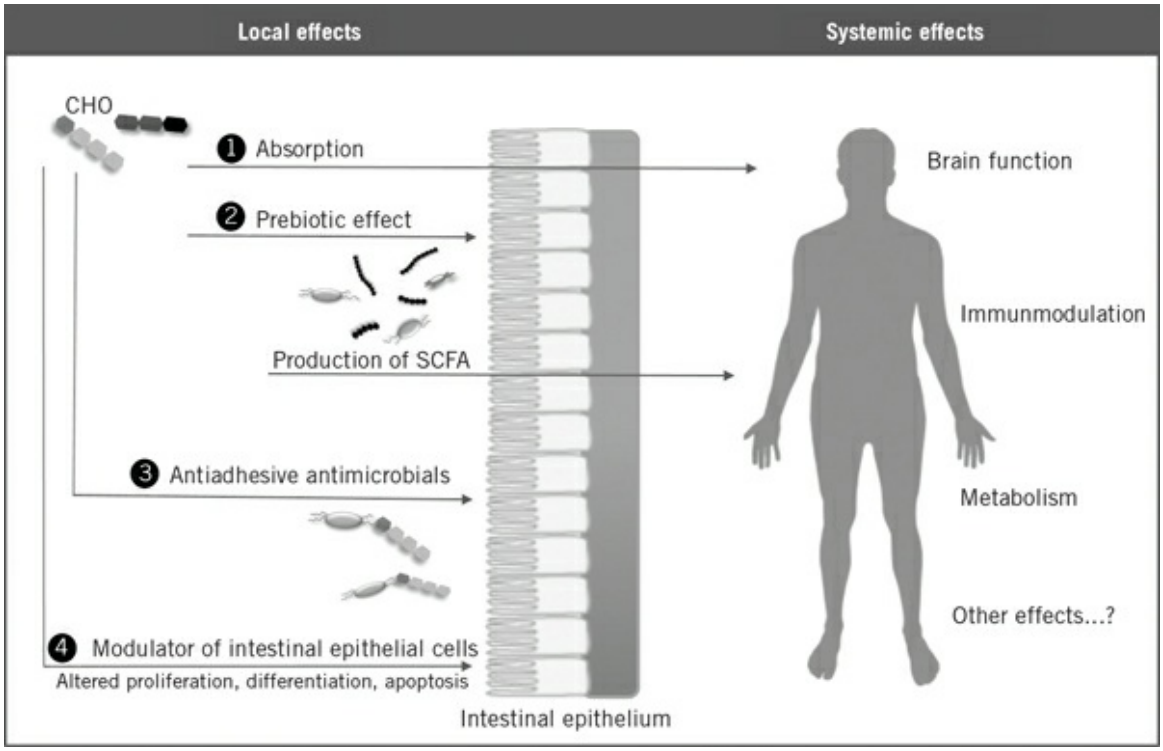


Figure 5-1 Generalized local and systemic effects of carbohydrates. The numbers 1–4 indicate specific functions, i.e., (1) absorption of carbohydrates, (2) influence on the microbiota composition and/or activity, (3) prevention of pathogen adhesion, and (4) direct effects on epithelial cells and systemic effects.

A schematic overview about changes in the microbial composition from birth to adulthood is shown in Figure 5-2.

The figure demonstrates the importance of bifidobacteria soon after birth. As anaerobes, they grow well in an oxygen-depleted environment, such as the infant's colon. The lactic acid produced keeps the surrounding pH low, thereby preventing the growth of potential pathogens. Although many factors, such as the mode of delivery or the environment, influence the development of the microbiota, human milk is considered to have a major impact as well.[11,17] Obviously, human milk contains components that favor the growth of bifidobacteria, which is discussed later. After exclusive breastfeeding, the introduction of solid food marks an important change in the microbial composition, which slowly leads to an adult microbiome. The first years after birth are a sensitive time frame where, among others, nutritional factors are of particular importance for the establishment of the microbiota. Our knowledge of the development of the microbiota and factors influencing the changes will certainly be modified due to the vast number of publications in recent years based on the availability of methods such as 16sRNA or pyrosequencing, which allow a much better characterization of microbial communities.[6] Here, recent observations of a specific "enterotype" of an individual are of particular interest.[18] It means that single persons can be grouped to a certain gut microbiome within a population, although the microbial composition varies substantially from person to person. Current data indicate that the human population can be associated with at least three enterotypes, and within one group, only few species dominate. *Enterotype 1* is characterized by bacteroides with a broad spectrum of saccharolytical (i.e., galactosidases, hexosaminidases, etc.) and proteases activities, as well as the enzymatic equipment for glycolysis and pentose phosphate pathway. *Enterotype 2* is comparable to type 1 with additional sulfate-reducing bacteria (*desulfovibrio*). This type is possibly important for mucin-degrading pathways with the simultaneous existence of *Prevotella*, which releases sulfate during the degradation of mucins. Sulfate is removed by *desulfovibrio* in order not to inhibit the degradation of mucins by the rising sulfate concentration. *Enterotype 3* is characterized by the family of Lachnospiraceae, which produces butyrate. Besides, they contain mucin-degrading and sugar-metabolizing enzymes that allow them to ferment a wide variety of CHO. This ensures that otherwise indigestible CHO are actually metabolized.[18] The products of hydrolysis can then be fermented, resulting in the production of SCFA that provide about 10% of the daily energy.

Interestingly, those enterotypes seem to be influenced by the nutrition pattern of the respective individual: a diet rich in lipids will lead to a cluster being dominated by bacteroides, whereas a diet rich in CHO will lead to a dominance of *Prevotella* (firmicutes). This also confirms observations made in other studies, which show that diet substantially affects the microbial composition of the intestine and thus the overall health. The question which mechanisms are underlying these observations has not been answered yet. In the case of preventive and therapeutic measures, it is important that the microbial composition of the intestine can be changed within one day, but it is unclear whether the predominant pattern of enterotypes can actually be changed in long-term studies.

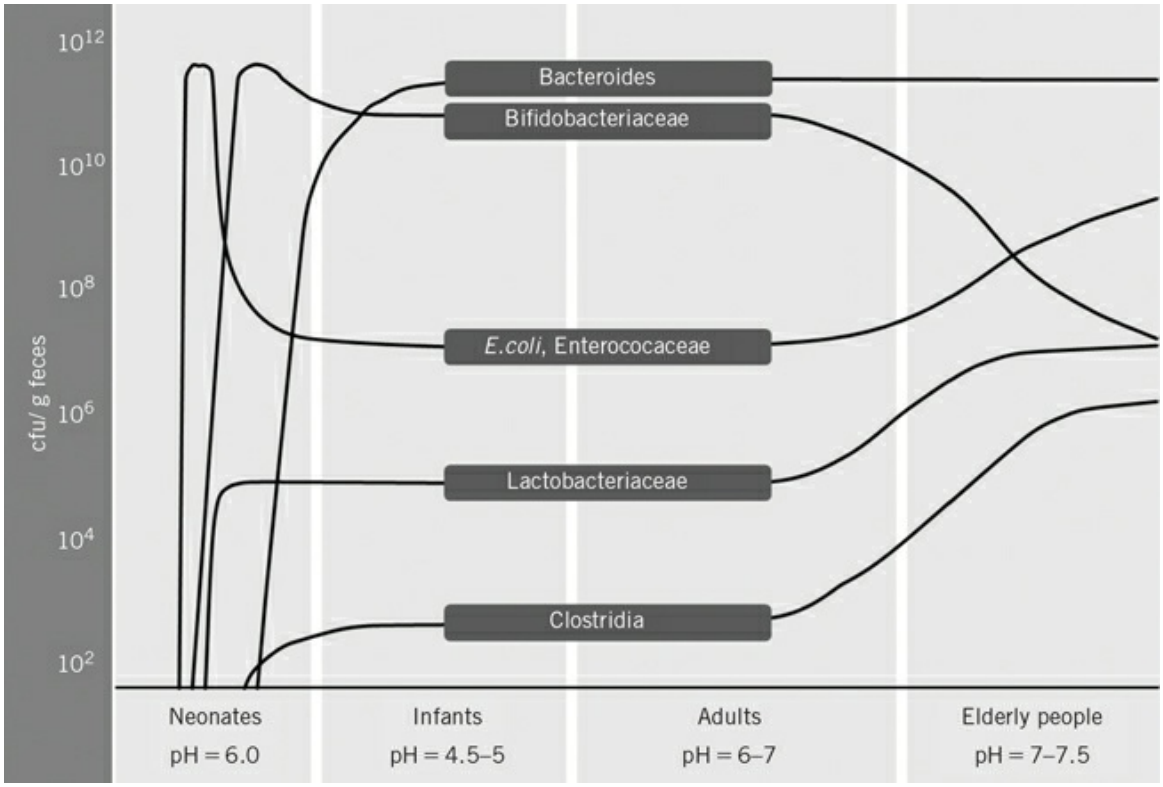


Figure 5-2 Changes in microbial composition from birth to advanced age (according to Walker et al. [2000]). CFU, colony-forming units.

Importance of Milk Oligosaccharides and Gut Microbiota Development

Intestinal colonization with a balanced microbiota is of major importance for an appropriate development of the immune system, and there is an

enormous scientific and commercial interest in modifying the microbiota for health promotion.[19] As the gut is sterile at birth, it is an organ sensitive to environmental influences. Furthermore, there is an intensive cross talk between gut microbes and the intestinal epithelium throughout life.[20–22] The mechanisms by which the intestinal mucosa perceives and responds to microbes, both pathogenic and commensal, are not completely known yet. Here, it is intriguing to investigate the role of human milk oligosaccharides (HMOs) or specific single monosaccharides and their effects on the selective growth of microorganisms in the gut.

As there are major differences in the CHO composition of human milk and bovine milk, and hence in the composition of infant formula, the effects of these types of feeding within the GI tract are different. For comparison, the overall composition of human milk and bovine milk is shown in Table 5-2.

Since the pioneering work of György et al. in the middle of the last century demonstrating the effect of *N*-acetylglucosamine (GlcNAc) containing oligosaccharides on the growth of *Bifidobacteria bifidum* subsp. *Pensylvanicum*, a strain György et al. isolated from infant feces, this topic is still of great scientific interest today.[26–28]

TABLE 5-2 Macronutrients and HMO in Mature Human Milk and Cow Milk

	Human Milk	Bovine Milk
Protein (g/L)	10	35
Fat (g/L)	41	37
Lactose (g/L)	65	48
Oligosaccharides (g/L)	5–15	0,05
Number of identified oligosaccharides	>150	~40
% Neutral oligosaccharides	~70%	~1%
% Sialylated oligosaccharides	~30%	~90%

Refs. 23–25 and cited references in these reviews.

Recently, Marcobal et al. showed that *Bifidobacterium longum* subsp. *infantis* can use HMO as the sole carbon source, whereas most of the other intestinal bacteria they studied were unable to assimilate HMO.[29] The genome of *B. longum* subsp. *infantis* (ATCC15697) contains a large gene cluster that comprises several glycosidases and specific transporters

suggested to be involved in the metabolism of HMO.[30] From the results of a comparative genomic survey, the conclusion was that the occurrence of the cluster correlates with the survival of this subspecies on HMO.[31] Therefore, David Mills and coworkers suggested that a HMO-consuming phenotype (HMO^+) of the subspecies can be attributed to the presence of this cluster (designated as the HMO cluster-1).[31]

Using a different methodological approach, Asakuma et al. reported the occurrence and localization of HMO-degrading enzymes in different bifidobacterial strains (Table 5-3).[32] The presence of external enzymes such as specific fucosidases and lacto-*N*-biosidases, which are secreted by bacterial cells and specific transporters for lacto-*N*-biose (LNB; Gal-β-(1-3)-Glc*N*Ac) and galacto-*N*-biose (GNB; Gal-β-(1-3)-Gal*N*Ac) explains why specific HMO can be used differently by bifidobacteria. Kitaoka et al. have recently elucidated that *B. bifidum*, which is another consumer of HMO, has a special pathway for degrading type-1 HMO.[33,34] The authors reported that *B. bifidum* uses a secretory lacto-*N*-biosidase to hydrolyze lacto-*N*-tetraose (LNT) to LNB and lactose. The liberated LNB is then incorporated into the cells by an ABC transporter specific for LNB and GNB.[35] LNB and GNB are converted to galactose and Glc*N*Ac-1-phosphate or Gal*N*Ac-1-phosphate by the action of GLB/LNB phosphorylase.[33]

This pathway, the GNB/LNB pathway, which involves LNB as a key intermediate, occurs in *B. bifidum* and some strains of *B. longum* subsp. *longum*, but apparently does not work in *B. longum* subsp. *infantis* because all of the genomes having been determined so far do not contain lacto-*N*-biosidase homologs.[31,34] *B. longum* subsp. *infantis* possesses a GNB/LNB phosphorylase, but this enzyme exclusively acts on the disaccharide and not on LNT.[33,36]

In a study on *B. longum* subsp. *Infantis*, Yoshida et al. demonstrated that this strain directly incorporates LNT and hydrolyses it inside the cell by a specific β-galactosidase. This β-galactosidase seems to have the substrate specificity for the type-1 chain (Galβ1-3Glc*N*Ac), but this strain also has another β-galactosidase that is specific for lactose and type-2 chain (Galβ1-4Glc*N*Ac).[37,38] This new data indicate that the organism uses two different β-galactosidases to selectively degrade type-1 and type-2 HMO. It also supports the view that the HMO^+ phenotype of this subspecies should not be attributed solely to the presence of the HMO cluster-1.

So far, investigations on growth-promoting factors have primarily been

focused on neutral HMO, although a few studies on the effects of sialylated oligosaccharides have been published. Sialidases have been found in *B. longum* subsp. *infantis* and *B. bifidum*.[39,40]

Prebiotic Oligosaccharides from Different Matrices

CHO with prebiotic effects belong to a functional food concept claiming to positively influence GI health through various mechanisms. The concept has been introduced in 1995 by Gibson and Roberfroid who defined prebiotics as a nondigestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon, thus improving host health.[41]

Prebiotics are considered to have specific effects, especially in the GI tract, such as influencing the microbial composition, digestive processes, regulatory mechanisms, hormone activities, and nutrient availability. This directly or indirectly also affects the immune system (Figure 5-3).

TABLE 5-3 HMO Metabolism by Bifidobacteria—Involved Enzymes and Their Location

Activities	*Bifidobacteria* bifidum JCM 1254	*Bifidobacteria* longum subsp. *infantis* JCM1222	B. *longum* subsp. *longum* JCM1217	*Bifido breve* JCM1
1,2-α-L-Fucosidase	Cell bound	Cytosol	–	Gene n
1,3-1,4-α-L-Fucosidase	Cell bound	Cytosol	–	–
1,4-β-Galactosidase	Cell bound	Cytosol	Cytosol	Cytosol
β-*N*-Acetylhexosaminidase	Cell bound	Cytosol	Cytosol	Cytosol
Lacto-*N*-biosidase	Cell bound	–	Cell bound[b]	–
GLBP (GNB/LNB transporter)	+	+	+	+
Transporters in HMO cluster	–	Transmembrane	–	–
LNT β-galactosidase	Cytosol	Cytosol	Cytosol	Cytosol

According to Refs.[27,32,37]

+, exists; –, does not exist.

M. Kitaoka and T. Katayama, personal communication.

[a]1,2-α-*L*-Fucosidase activity not detected; this strain does not utilize 2′fucosyllactose.

[b]No gene encoding GH20 LNBase.

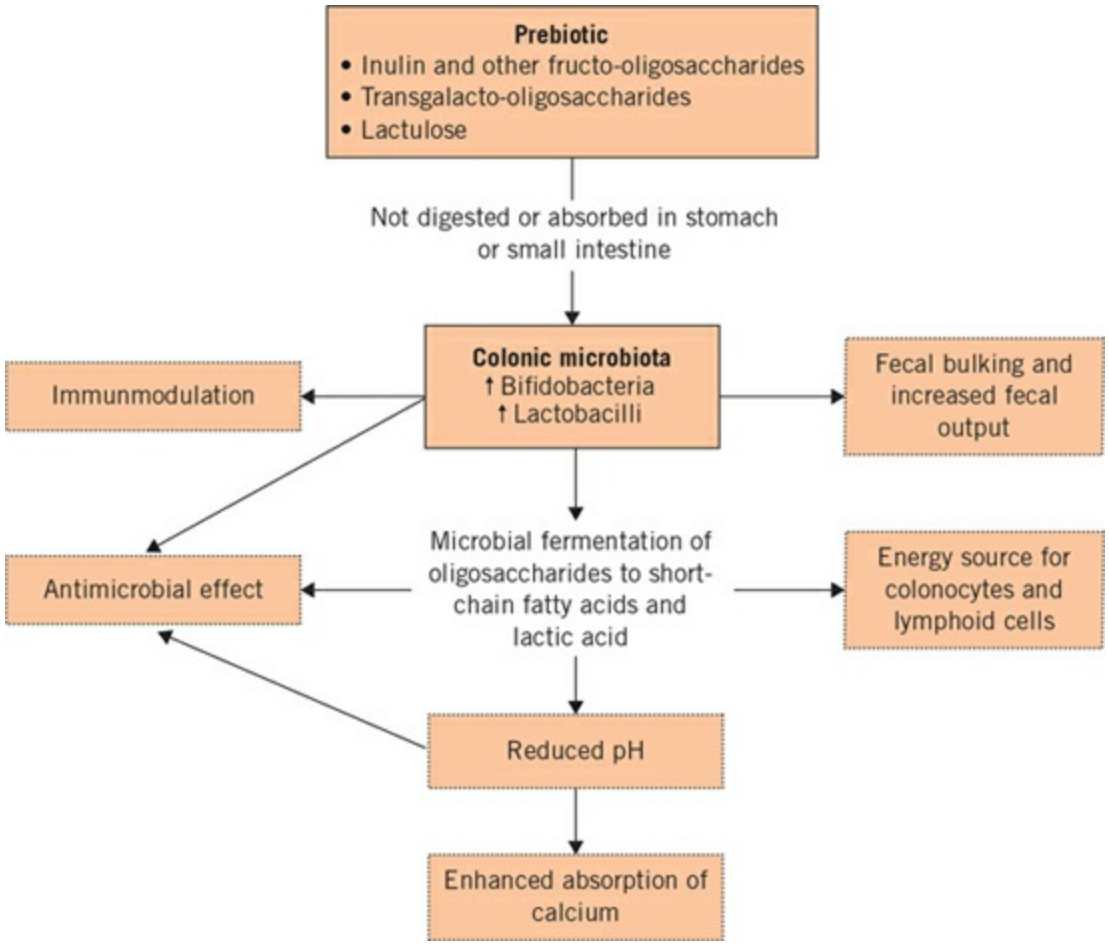


Figure 5-3 Representation of potential effects of prebiotics in the intestinal tract (Sherman et al.[43]).

The bifidogenic effects of oligosaccharides, especially of inulin and oligofructose, have been well proven even in humans. In one of their first experiments, Gibson and Roberfroid have shown that within a few days after the oral administration of oligofructose (15 g for 14 days) to healthy volunteers, a marked change of the microbiota toward the predominance of bifidobacteria occurred.[42] Switching back to the original diet restores the microbiota within a few days.

Currently, research concentrates on searching for ways how to improve the growth of bifidobacteria as these microorganisms dominate in the infants' GI tract. A direct influence of bifidobacteria on the growth of colon cells by differentiation, proliferation, and apoptosis is considered to

be the mechanism underlying the postulated effects of prebiotic products. In this context, the SCFA butyrate, propionate, and acetate, which are formed during fermentation, play a special role.

Galacto-oligosaccharides (GOS) and fructo-oligosaccharides (FOS) belong to the best-known prebiotics, but moreover, numerous other CHO can be found as prebiotics on the global market.[43,44] There is a huge number of publications of studies using prebiotic components in different clinical settings or for various reasons (for review see Refs. 43 and 45). Most often, the overall aim of these studies is to improve the GI health by investigating the effects on (a) the bifidobacterial composition, (b) on the production of SCFA, or (c) on changes in the fecal pH. Each of these effects would be beneficial to the infant. However, often, the data are rather difficult to interpret, and the impression is being created that if an effect on the microbial composition cannot be found, although it was the major aim of the study, the authors focus on the difference in SCFA production or, if this fails as well, they concentrate on differences in the pH in various groups. In agreement with pediatric authorities, there is still a lack of RCTs in this area to recommend a special prebiotic to be used.[46]

Figure 5-4 shows a scheme of the postulated effects of milk oligosaccharides and prebiotic plant-derived oligosaccharides. It needs to be shown which specific process and underlying mechanism is induced by which substrate and whether there are similarities between different prebiotics. Like with other functional food components, special attention needs to be addressed as to whether prebiotics are recommended for preventive purpose in healthy persons or for therapeutic considerations in certain diseases.

However, there are very interesting data worthwhile for further investigations. Besides the important observation that the amount of calcium absorbed can be influenced by prebiotics, it has been shown for the first time that applying prebiotics implies the need of differentiating between responder and nonresponder.[47] This observation may explain that in many studies, the interpretation of data is rather difficult as no distinction between a different individual responsiveness has been made.

Another difficulty is that most studies do not allow unambiguous statements regarding structural interactions. CHO, in general, have a high potential to exert health effects besides the delivery of glucose as energy source. However, much remains speculative in their prebiotics functions. Most studies in recent years concentrated on the impact on the growth of bifidobacteria themselves, where as the effects of bacterial metabolites and their impact on health have been neglected.[48] Important questions still

cannot be answered, such as (i) should the prevailing microbial composition throughout the whole GI tract be modified in healthy humans and (ii) is it actually of importance to have an intestinal microbiota dominated by bifidobacteria? Apart from promoting a microbiota dominated by bifidobacteria in neonates, further research should consider possibilities how to influence the metabolic activity of colon cells, the increase of mineral bioavailability, or systemic effects regarding CHO and lipid metabolism.

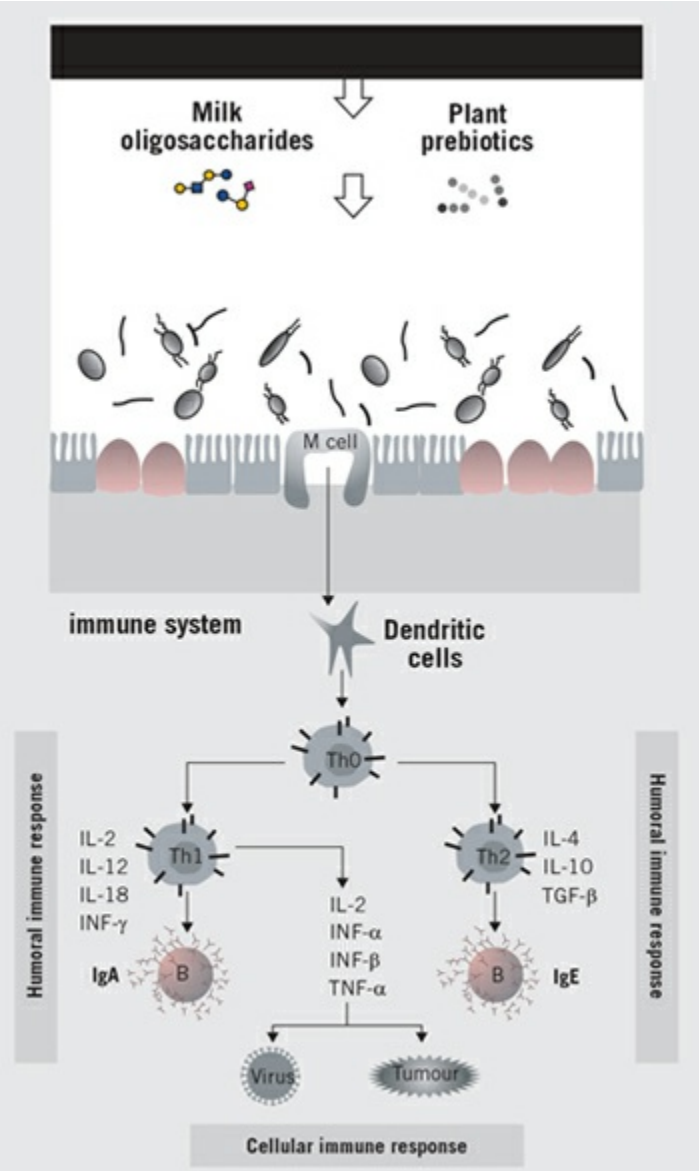


Figure 5-4 Metabolic fate of an oral fructose load in healthy subjects.

In our view, discussion needs to take into account whether prebiotics are useful for primary prevention in healthy people, for secondary prevention in case of a given risk (e.g., genetic predisposition), or for therapy in case of certain diseases.[46]

Human Milk Oligosaccharides Versus Prebiotic Oligosaccharides

In many scientific publications, including reviews on functional aspects of PBOs, these components are often compared with HMOs as those CHO are believed to also exert a bifidogenic effect on the infant. Some formula producing companies go as far as claiming in consumer information that both groups of CHO share great functional as well as structural aspects.

However, PBOs and HMOs do not share any structural similarity that is required to speculate about similar biological functions. A brief summary is given regarding the structural diversity of PBOs and HMOs.

TABLE 5-4 Monosaccharide Composition of Human Milk Oligosaccharides and of Representative Prebiotics

Compound	Human Milk Oligosaccharides	Prebiotics
Glucose	Traces	+
Galactose	+	+
N-acetylglucosamine	+	–
Fucose	+	–
N-acetylneuraminic acid	+	–
Fructose	–	+
Xylose	–	+
Arabinose	–	+
Rhamnose	–	+
Galacturonic acid	–	+

TABLE 5-5A Specific Monosaccharide Linkages in Lactose-Derived Oligosaccharides

Lactose-Derived Oligosaccharides	Monosaccharide Linkage
Fucosyl-lactose	α1-3, β1-4,
Lacto-*N*-tetraose	β1-3, β1-4

Sialyl-lactose	α 2-3, α 2-6, β1-4,
Sialyl-lacto-*N*-tetraose	α 2-3, α 2-6, β1-4, β1-3

Definition of HMOs and of PBOs

HMOs are solely produced in the lactating mammary gland. Their core structures are based on lactose, which is then modified by the addition of specific monosaccharides like the amino sugar Glc*N*Ac and galactose that are present as lactosamine units and/or fucose, or acidic components (*N*-acetylneuraminic acid, also called sialic acid) (Table 5-4).[49] An important core structure is LNT and its fucosylated derivatives (Table 5-5a). Hence, there is a large variability of HMOs with a total number of more than 150 compounds characterized so far. The total concentration is rather high (5–10 g/L) compared to the protein content of human milk.[50]

For supplementation of infant formula, galactosylated and/or fructosylated oligosaccharides are often used. They may be derived from plants or produced by technological means, for example, by transgalactosylation using the galactose moiety of hydrolyzed lactose. Such components are not present in human milk.

So far, the main monosaccharides in PBOs are galactose, glucose, Frc, xylose, and arabinose with a great potential for new components to be available soon (Table 5-4).[48] With the exception of glucose and galactose, these monosaccharides are not present in human milk.

Linkage Specificity Between Monosaccharidesin HMOs and PBOs

FOS like inulin or oligofructose are well-characterized components. They consist of a number of β1-6-linked galactosyl residues bound to a terminal glucose unit through an α1-4-linkage. A thorough review on this topic has been given by Rastall.[51] In human milk, however, small oligosaccharides are fucosylated or sialylated, but solely galactosylated components do not occur. In addition, the linkage between monosaccharides is important for biological effects as well. Therefore, examples for some specific linkages in HMOs and PBOs are shown in Table 5-5b.

In recent clinical studies, it was shown that the addition of GOS and FOS to a formula may increase the number of bifidobacteria. However, it has not been convincingly demonstrated yet that, at the same time as the number of bifidobacteria increases, the number of potentially pathogenic microorganisms in the infants' colon decreases, which then leads to the bifidus-predominant gut flora in breastfed infants. Milk oligosaccharides

are thus thought to exert direct effects on the adhesion of microorganisms, and even their toxins, by competitive inhibition of their adhesion to the intestinal wall. For detailed information, see reviews by Bode[52] and Kunz et al.[53]

Potential Risks of a High Fructose Intake

A typical westernized diet is characterized by a high intake of refined CHO and fats combined with a reduced intake of plant polysaccharides. Often, this pattern involves an increasing consumption of Frc, sugar substitutes, or artificial sweetener, which is controversially discussed as one major factor for the obesity pandemic.[54,55]

TABLE 5-5B Specific Monosaccharide Linkages in Prebiotics

Oligosaccharide	Monosaccharide Linkage
Fructo-	β1-2,
Galacto-	α 1-2, α 1-4, α 1-6,
Transgalacto-	and
Soybean-	other linkages

Frc is a monosaccharide naturally occurring in many fruits and vegetables. Its contribution to the overall energy intake is estimated to be about 10%.[56] In comparison to the amount of Frc, for example, in high corn fructose syrup (HCFS), the amounts in fruits and vegetables are relatively low. HCFS consists of free Frc and glucose. The ratio may be very different often being 55–45. Currently, the question as to whether the marked increase of Frc intake is a particular high risk for obesity is highly controversially discussed.[57–62]

Negative effects might be due to different satiety effects compared to glucose with a subsequently higher intake of energy. It is conceivable that a lower increase in blood glucose results in a lower secretion of insulin followed by a diminished increase in leptin and, subsequently, by a lower ghrelin suppression.

Bray and Popkin were the first to describe the link between Frc and obesity. Since then, Frc has often been associated with the negative effects, especially concerning the promotion of cardiovascular disease. Frc is found to have negative effects on blood lipids, uric acid, blood pressure, body weight, glucose, and insulin regulation, which is schematically

shown in Figure 5-5b.

Metabolic Aspects of Fructose

Special effects of Frc metabolism in the liver result in the preference of the lipogenic pathway with an influence on the de novo lipogenesis (DNL).[63] In contrast to glucose dietary, Frc does not stimulate insulin or leptin, which are important regulators of energy intake and body adiposity. Stimulated triglyceride synthesis most likely leads to the hepatic accumulation of triglyceride, which has been shown to reduce hepatic insulin sensitivity, as well as an increased formation of very low-density lipoprotein (VLDL) particles due to higher substrate availability, increased apo B stability, and higher microsomal triglyceride transfer protein, a critical factor in VLDL assembly.[63]

The intestinal absorption of glucose and Frc is distinctly different. Frc is primarily absorbed through the GLUT-5 transporter. After the transport through the portal vein, a complete uptake of Frc by the liver through GLUT-2 follows, where Frc can be converted to glucose and/or lactate or stored as glycogen (Figure 5-6). There is the decisive point, where one part of the absorbed Frc can be used for the DNL.

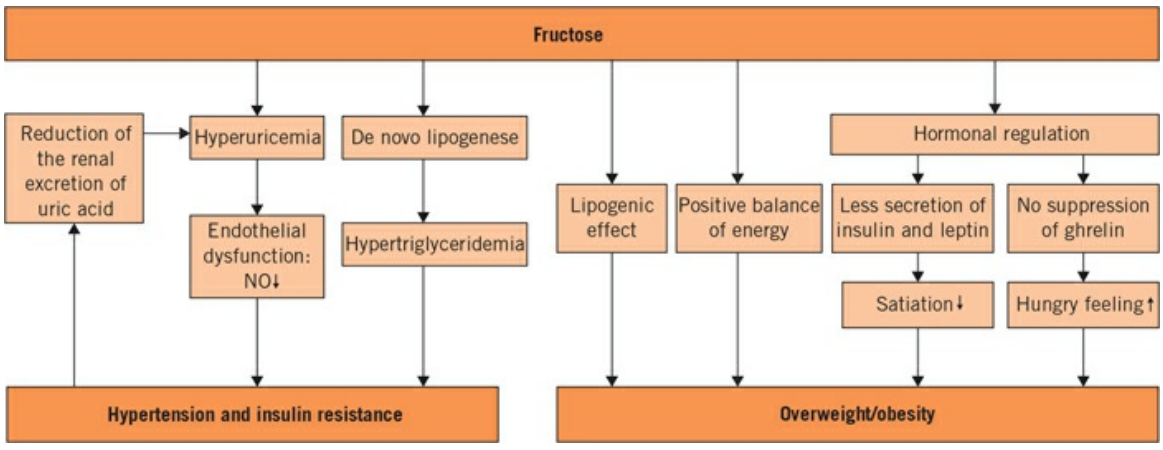


Figure 5-5 Fructose and the potential link to hypertension, insulin resistance, and obesity.

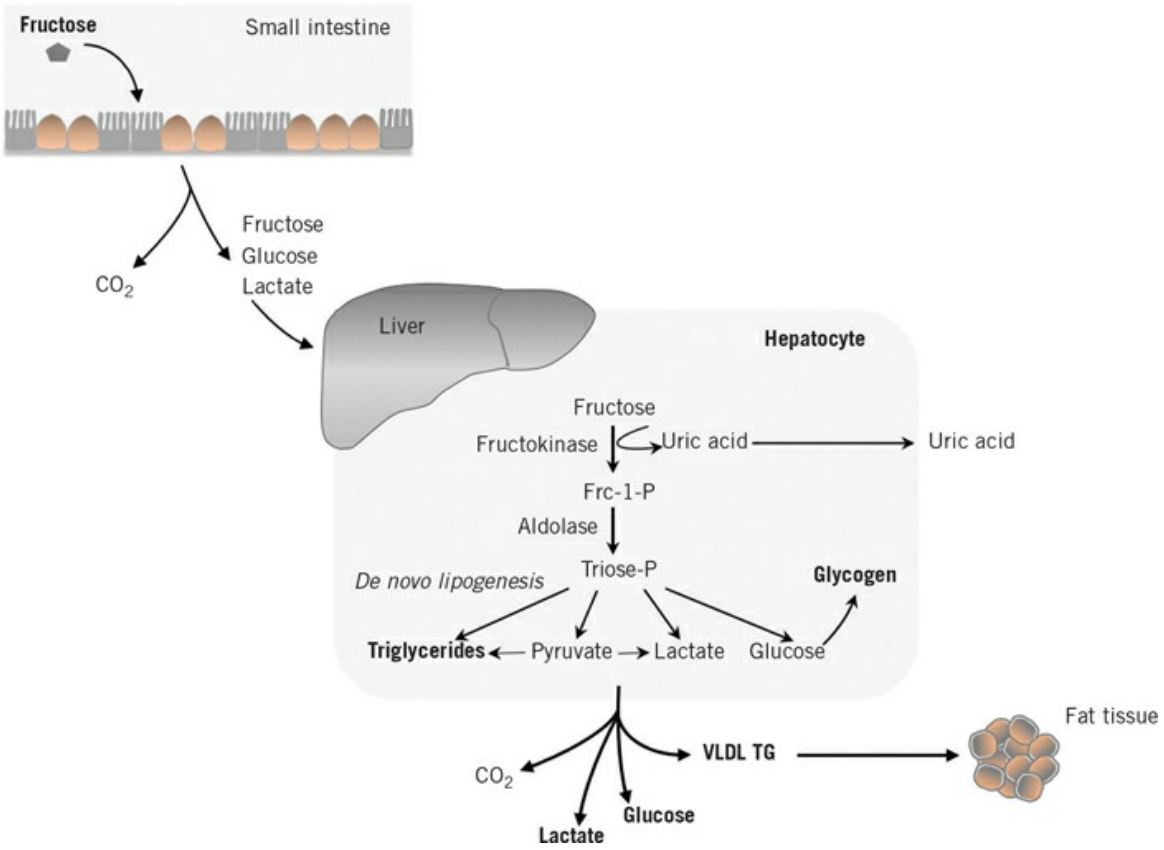


Figure 5-6 Potential of milk oligosaccharides and plant-derived prebiotics to influence the intestinal immune system.

On the extent of DNL after the intake of Frc, the opinions, however, differ greatly (see below). The assumption is that with Frc, in contrast to glucose, the regulation through phosphofructokinase as the most effective controlling enzyme is avoided so that the whole Frc is available as a substrate for DNL.

Controversial Opinions

Intervention studies with high amounts of Frc (e.g., with a 25% energy excess) are regarded as evidence for the negative effects. But a negative metabolic implication has not been shown in investigations with isocaloric diets, in which Frc was exchanged by other CHO sources under energy-matched conditions. In contrast, studies with hypercaloric diets lead to strongly and permanently increased triglycerides and apo-B-values.

However, this mechanism of DNL has been proven so far only in animal and not in human studies. In animal models, however, the

proportion of CHO being available for DNL is much higher than in humans (up to 96% in mice, >50% in rats, and only 5% in man).[64] In addition, there is even evidence that smaller amounts of Frc (<10g) positively influence the regulation of blood glucose through an increase of glycogen synthesis in the liver and a decreased glucose production. Apart from that, 60% of the energy intake consists of Frc in animal studies, whereas it is only 10% in man.

Despite those serious doubts concerning the negative interpretation of many studies, nutrition guidelines are very restrictive regarding the Frc intake.

In various studies, no indication for negative effects of Frc on blood lipids, uric acid, blood pressure, body mass index (BMI), or insulin has been found as long as low to moderate amounts of Frc (<60 g/d or <10% of energy) are consumed together with an isocaloric exchange with other CHO. It seems that the risks of a too high Frc supply will rise, if intake exceeds 100g/d (relevant for all population groups), intake exceeds 60 g/d in case of diabetes mellitus (DM) type 2, and in case of hypercaloric nutrition (>20% excess energy).

Dyslipidemia, increased uric acid levels, and a gain of weight are observed. In case of hypercaloric nutrition, however, it may be that the surplus of calories may be causing the negative effects and not Frc itself.[64]

The situation regarding the potential harm of a Frc intake is still a matter of most controversial discussion. For a detailed overview, see Refs. 58–62.

In general, it should be kept in mind, irrespective of whether studies found negative impacts or not, that all studies so far have shown more or less striking shortcomings with respect to study design, methods applied, duration, and so forth.

To finally evaluate the current discussion, one may conclude that free Frc is to be evaluated similar to bound Frc; Frc as HFCS or sugar in soft drinks differs from Frc consumed as part of a diet rich in fruits; and transferability of results from animal studies to humans is not always given due to (i) unphysiologically high doses of Frc, (ii) different metabolic situation (e.g., rats have uricase), and (iii) the fact that not all effects observed in animals can be found in human studies.

The controversial topic of Frc and its negative effects is, in our opinion, best summarized by Tappy and Mittendorfer (2012)[65]:

- There is some evidence suggesting excessive Frc consumption, especially in combination with excess energy intake may have

adverse effects on metabolic health.

- The results from clinical trials do not support a significant detrimental effect of Frc on metabolic health when consumed as part of a weight-maintaining diet in amounts consistent with the average estimated Frc consumption in Western countries.
- Public health policies to eliminate or limit Frc in the diet should be considered to be premature.
- Crucial studies providing definitive answers to the concerns of Frc toxicity and others, which should guide decisions of public policy makers, are still missing.

RECOMMENDATIONS

Preamble

The following paragraph will deal with recommendations for CHO intake: its first part will refer to the "digestible" fraction of the CHO family, and the second part will deal with the "nondigestible" fraction, also called dietary fibers (DF).

Due to the fact that child physiology is influenced by physical growth, pediatric recommendations for nutrient intakes usually vary with age and body weight; it is also a fact that recommendations have not been established for all age groups by sound research, but were delineated for some age groups by extrapolation, thereby applying known data from other age groups. For example, frequent recommendations for toddlers and school kids are based on extrapolating data established for newborns and adolescents. These different approaches lead to different types of recommendations and are summarized as follows[66]:

Estimated average requirement (EAR) meets the estimated nutrient needs in half of the population.

Recommended dietary allowance (RDA) covers the needs of 97%–98% of the population and is, therefore, equal to EAR plus twice the coefficient of variation (CV).

Acceptable macronutrient distribution range (AMDR) is the range of adequate macronutrient intakes that are associated with reduced risks of chronic disease while providing recommended intakes of other essential nutrients. AMDRs are especially useful in subjects with special energy needs and are therefore given as percentage of total energy intake.

Adequate intake (AI) is the observed average or experimentally determined intake by a defined population or subgroup that appears to

sustain a defined nutritional status such as growth rate, normal circulation nutrient levels, or other functional indicators of health. AI is used if sufficient scientific evidence is not available to derive an EAR. AI is not equivalent with RDA. For infants, AI of CHO intake is derived using what is consumed from human milk and complementary foods.

Composition of Carbohydrates During Childhood

The composition of ingested CHO varies during childhood and—as gut matures—becomes more complex. During fetal life, the sole source of CHO is provided as a monosaccharide, that is, glucose. Postnatally, glucose is still the only CHO of choice when a newborn cannot be enterally fed and is in need for intravenous nutrition. In healthy newborns who can be enterally fed, the sole source of CHO becomes a disaccharide (lactose); with introduction of solid food and later family meals, the CHO source expands to starch and other disaccharides like sucrose, maltose, and maltodextrin, a glucose-based oligosaccharide (chain length of n = 3–20, also known as glucose syrup). Comparative data from international studies indicate that lactose intake drops from 20% to 25% (of total energy intake) during the first year of life toward <10% after the age of two years.[67] However, currently, no recommendations exist that address the composition of CHO during childhood.

Recommendations Based on the Intake of Digestible Carbohydrates

As mentioned earlier, digestible CHO consist mainly of sugars and starches, and their primary role is that of an energy source to the tissue. Muscle, heart, kidneys, and especially the brain rely mainly on CHO as an energy substrate. The recommended dietary intake (RDI) is therefore based to provide sufficient CHO as primary energy source for the brain and to avoid the formation of keto-bodies. Besides the intake that is needed to support brain metabolism, there is an AMDR, which takes into account the role of CHO in maintaining a constant body weight/composition or—as it is the case in children—in achieving adequate growth.[66] AMDR also takes into account specific circumstances like the physical activity level (PAL).

As a general rule, it is said that for children beyond 2 years of age, more than 50% of energy intake shall be provided by CHO.[68–70] Thus, the dietary recommendations for CHO—on one hand—are strongly related to energy requirements and intake. On the other hand, there is strong

evidence that the brain of infants consumes up to 60% of all CHO; thus, in childhood, RDI for CHO is also closely correlated to brain size and growth. The brain of a term baby weighs 380–400 g; at 12 months of age, it grows up to about 1000 g; at the age of 5 years, the brain weight is about 1300 g; and that of a young adult is between 1350 and 1450 g.[66] Brain glucose consumption has been found to be approximately 8 g/100 g brain tissue/d. Term infants will therefore need 32 g/d as adequate cerebral CHO supply. Infants 1 year of age will require 80 g of CHO per day and young adults 100–110 g/d. In addition to cover the energetic needs of the brain, glucose must also be provided for those body cells that rely on anaerobic glycolysis like red and white blood cells (approximately 35 g/d in adults). On the other hand, endogenous gluconeogenesis from proteins and fatty acid oxidation contributes to about 30 g of glucose production per day in young adults. Beyond the first year of age, the estimated average of requirements (EAR) for CHO intake seems therefore to be 100 g/d. Assuming an adequate energy intake contributed by the other macronutrients, this amount should be appropriate to provide sufficient energy to the brain, thereby preventing the formation of keto-bodies. The RDA (calculated as EAR plus twice the CV of 15%) is thus 130 g/d.

In infants below 12 months of age, the EAR/RDA approach to calculate CHO intake solely based on its role to provide sufficient cell fuel is not valid. In this age range, the effect of growth on energy and CHO requirements needs to be taken into account more specifically. Growth rates are highest around birth. From 40 weeks of gestation until 3 months of age, yearly growth rates are 70%, decreasing afterward to about 10% until the age of 6 months and to 3% per year at 12 months. On the other end, premature born infants experience even higher growth rates of up to 200% during their period from 26 to 34 weeks of gestation. Adequate growth requires sufficient intake of macronutrients, so that they can be deposited as new body cell mass, but also the synthesis of new cell mass itself is an energy-consuming process.

The dietary requirements of infants below 1 year of age can basically be divided into those needed for maintenance (like in young adults) and those needed for growth. Although figures for maintenance (expressed as kilocalories per kilogram per day) do not vary significantly during the first 12 months of life, the needs for growth are considerably higher during the first 3 to 5 months of life because growth rates are highest during that period; thus, at 1 month of age, requirements for growth account for 40% of total intake, at 3 months for 22%, at 6 months for 7%, and at 12 months for 3.5%. Infants born very premature may even need up to 70% of their

intake for growth. As a consequence, there is a fundamental difference in the composition of those macronutrients that are either needed for growth or to provide energy for maintenance. Appropriate growth from –3 until + 12 months after term can only occur by providing excess energy as well as sufficient protein, that is, the amount purely needed for growth exceeds that one needed for maintenance by a factor of 2–4.

As a consequence, recommendations about a safe intake of CHO can therefore not be made for this young age group by focusing only on the role of CHO for brain metabolism. The picture will become more complete by taking into account (i) theoretical considerations about the composition of growth (i.e., factorial approach) as well as (ii) the CHO intake achieved from breast milk, which is assumed to be the reference nutrition for healthy term infants. Assuming an average digestible CHO content of 6.2–7.5 g/100mL in mature breast milk,[71–74] and a total fluid intake (TFI) of 150–160 ml/kg/d, a term infant will thus achieve a CHO intake of 10.5–12 g/kg/d or 32–50 g/d. At 3 months of age, the intake increases to 50–65 g/d and to 90–120 g/d at the age of 9–12 months.

In premature born infants, the previous approach is not valid because breast milk has not been designed by nature to meet the enhanced nutritional needs for growth in very low-birth-weight (VLBW) infants. Applying the factorial approach leads to an overall requirement of 100–135 kcal/kg/d.[75] Approximately one half of these calories should be provided in the form of CHO (i.e., 50–65 kcal/kg/d), which translates into an enteral CHO intake of 12–17 g/kg/d. The upper value of this range comes closer to the upper limit of 16 g/kg/d needed for CHO oxidation.[74,75] It has also been shown that it is beneficial for protein accretion and longitudinal growth to provide nonprotein calories preferentially as CHO. Providing >50% of non proteincalories as CHO reduces amino acid breakdown, protein oxidation, and urea formation.[74,75] Applying standard fortification (3 g lactose per 100 mL) of human breast milk provides an intake of 15–17 g CHO/kg/d at a standard TFI of 150–160 mL/kg/d. It has been shown that growth rates above 17 g/kg/d can be achieved and lead to postnatal growth that is parallel to intrauterine growth curves.[76]

Table 5-6 summarizes data about RDA, EAR, and AIs for CHO in different age groups. It also shows data of a recent review paper about total intake of CHO, sugar, and starch in infants and toddlers.[67]

Recommendations Based on the Intake of Dietary Fibers

DFs consist of complex, partially branched, chained polysaccharides originating mainly from plants (see Chapter 2). The spectrum of enzymes in the human intestine is unable to cleave and digest these molecules. However, some intestinal bacterial strains may use them as a substrate. DFs have an impact on gastric emptying, absorption of lipid components, blood glucose response, the gut microbiome, laxation, and stool weight. A recent commentary by the ESPGHAN reviews the basics of non digestible CHO in childhood.[77]

DFs seem to have no systemic effects as they are not being absorbed. For the same reason, serum concentrations of DFs cannot be measured and —unlike for other nutrients—can therefore not provide a basis for recommendations. Current recommendations are derived from adult epidemiological studies that investigate DF intake and adult risk for hyperlipidemia, hypertension, and coronary heart disease. Different studies in adults have shown that a DF intake (particularly from cereals) of 12.9–14.4 g per 1000 kcal significantly reduces the risk for coronary heart disease.[78–80] There are no further studies available that could provide the basis for specific recommendations for DF intake during childhood. Current recommendations of DF intake for the period beyond 1 year of age are therefore mainly based on adult data. It might be questioned whether endpoints like adult CHD risk are appropriate to guide recommendations in infants older than 1 year of age.

For term and preterm newborns, AI can be calculated from the composition of breast milk assuming standard TFIs. Assuming that breast milk contains 1.3–2.1 g of DF per 100 ml, newborns will ingest between 2.1 and 3.5 g DF/kg BW/d. For infants up to 1 year of age, the additional intake of complementary solid foods needs to be taken into account. There are actually no published data available about the DF intake in that age group. However, several manufacturers of commercially available solid food products give ranges from 0.3 to 2.5 g DF/100g of product. Although this content is not very different from that of breast milk, AI of DFs may be assumed to be in the order of 1.8–3.0 g/kg BW/d.

Table 5-7 gives a summary of recommended intakes of DFs during childhood.

There are currently no recommendations available that address from which source DFs preferably should be taken from. For infants on human milk or formula, there is an ongoing debate whether artificial DFs like GOS and FOS added to formula products have the same biological properties as the natural spectrum of oligosaccharides in human milk (see also Chapter 3).[81,82] Results of this debate could have an influence on

whether there is a justification for current practice of supplementing formula or solid foods with artificial DFs.

TABLE 5-6 Recommendations for Intake of Digestible Carbohydrates

Age		Criterion	EAR[66](g/d)	RDA[66](g/d)	AI(g/d)	Energy Requirements Boys/Girls[66]	Reported Intakes[67] Total/Sugars/ Starch
Newborn	Preterm	Factorial approach			10.5–12.0g/100 kcal[71,75]	100–135 kcal/kg/d[71,75]	
	Term	Composition of breast milk			35	90–110 kcal/ kg/d	
Infants	0–6 mo	Composition of breast milk			60[66]	570/520	90–115
	7–12 mo	Composition of breast milk and solid food			95[66]	740/680	90–115/ 60–75/35–40
Children	1–3 y		100	130		1050/1000	130–190/ 90–115/60–80
	4–8 y		100	130		1740/1640	
	9–13 y		100	130		2300/2100	
	14–18 y	Brain glucose utilization	100	130		3200/2400	

General: For children older than 2 years of age 50% of caloric intake should be provided as carbohydrates.[2–4]

TABLE 5-7 Recommendations for the Intake of Dietary Fibers (DF)

Age		Fibers[66](g/d) Boys/Girls	Energy Requirements[66](Kcal/d) Boys/Girls
Newborn	Preterm	1.3–2.1 g/100 mL BM[72]	100–135 kcal/kg/d[71,75]
	Term	1.3–2.1 g/100 mL BM[72]	90–110 kcal/kg/d
Infants	0–6 mo	2.1–3.5 g/kg BW/d	570/520
	7–12 mo	1.8–3.0 g/kg BW/d	740/680
Children	1–3 y	19	1050/1000
	4–8 y	25	1740/1640

9–13 y	31/26	2300/2100
14–18 y	38/26	3200/2400

General: Data for newborn and infants are based on composition of ingested breast milk (and complementary solid food) in these age groups. Breast milk contains 1.3–2.1 g/100 g, and commercially available solid foods contain 0.3–2.5 g/100 g.

For all other age groups: A dietary intake of 14 g per 1000 kcal has been identified as most protective for coronary heart disease.[66]

DISORDERS INVOLVING CARBOHYDRATES

A variety of acute and chronic disorders can affect different steps of CHO metabolism. All of these lead usually to a more severe clinical picture. Due to space restrictions, we will not be able to discuss these diseases in detail, but will aim to give rather a high-level overview.

Only monosaccharides can be absorbed by the intestinal border. *Malabsorption* describes various acute and chronic conditions where CHO escape intestinal digestion and absorption. Different from lipid and protein digestion, pancreatic exocrine insufficiency does not play a major role in CHO malabsorption. It is only α-amylase that is provided by the pancreas and it cleaves the α-1,4 glucose–glucose bonds of starch. CHO malabsorption is usually caused by an insufficient activity of brush border located enzymes that cleave disaccharides. Such enzymes are lactase (cleaving galactose–glucose bonds), sucrase isomaltase (glucose–glucose and glucose–Frc bonds), and gluco amylase (cleaving glucose from glucose polymers). The resulting monosaccharides are transported into the blood through specific transporters (like GLUT-2 and GLUT-5). Activity of disaccharide-cleaving enzymes can be decreased due to individual genetic conditions (rare), or can secondarily develop as “adult-onset” type (common in Asians, African Americans, and Mediterranians), or as a consequence of mucosal injury (i.e., gastroenteritis, drugs, radiation). Rarely, activity of transluminal monosaccharide transporters can be decreased due to genetic defects. Clinical symptoms of malabsorption are chronic diarrhea, abdominal distension, and failure to thrive. H-breath tests are helpful in making the diagnosis. A comprehensive review of malabsorption is found in Ref. 83.

Fructose intolerance: Frc is absorbed by two major transporters: GLUT-2 and GLUT-5, although, currently, GLUT-8 and GLUT-12 have also been identified to play a role.[84] Deficiency of these transluminal transporters leads to bacterial fermentation of Frc with increased intraluminal gas production and distension and also to osmotic influx of

water, resulting in severe abdominal pain and diarrhea. Secondary effects in skin integrity, chronic respiratory, and urogenital infections have been described.[85] Diagnosis may be made with H-breath tests. Dietary restrictions of Frc intake as well as novel enteral enzyme replacement therapies help to cure the symptoms.[86] A comprehensive review of Frc intolerance is given in Ref. 87.

Glycogen is a multi branched glucose polymer ("animal starch") and serves as a short-term energy storage for excess glucose. *Glycogen storage diseases* comprises a group of 11 inherited enzymatic defects that either impair the release of glucose molecules from the glycogen polymer or lead to incorrect phosphorylation of released molecules or build up poorly branched glycogen with altered solubility in the cellular and extracellular matrix. Symptoms arise from hypoglycemia due to poor glucose release in periods of hunger and accumulation of glycogen in a variety of organs, leading to impairment of function. Depending on the nature of the underlying enzyme defect, life expectancy can be severely reduced and also mental function might be severely impaired. There is no causative cure, some forms benefit from dietary regimes, high-dose vitamin supplementation, or enzyme replacement therapy. Gene therapy or stem cell applications have not yet reached a stage of maturity to be tested in humans. A comprehensive review of glycogen storage diseases is given in Ref. 88.

Galactosemia is caused by a group of three inherited enzyme defects that are needed for the regular metabolism of galactose into glucose. The enzyme block leads to an accumulation of galactose that leads to the formation of galactitol, an alcohol of galactose. Galactitol itself is an end product that accumulates in the body and leads to hepatomegaly and cirrhosis, renal failure, cataracts, and brain damage. Undiagnosed, lethality is high (>75%). Therapy consists of eliminating lactose and galactose from the diet. Early detection of affected newborns by determining enzyme activity in dried spot blood is now part of many newborn screening programs. A comprehensive review of galactosemia is given in Ref.[89].

Hyperinsulinism describes a series of disorders with hypoglycemic episodes caused by an overproduction and secretion of insulin. This can be the effect of acute pancreatic overstimulation like in newborns from diabetic mothers or of chronic overproduction by autonomously producing islet cells (focal or diffuse form). Hyperinsulinism is due to peripheral resistance of insulin, which usually leads to hyperglycemia and will be described in the following paragraph. The main risk rising from unregulated insulin hypersecretion in the presence of a disturbed feedback

loop are repeated episodes of hypoglycemia. These pose a significant threat to neuronal integrity and function. A comprehensive review about hyperinsulinism is given in Ref. 90.

DM is the insufficient secretion of insulin, or peripheral resistance to insulin impairs uptake of glucose into body cells. This lack results in elevated glucose levels in the extracellular compartment as well as to disturbed intracellular energy supply. The latter leads to an acute metabolic crisis either as a ketoacidosis or as a nonketotic hyperosmolar state, both are potentially life-threatening disorders. Elevated blood glucose levels induce osmotic polyuria and can acutely culminate in shock due to dehydration. On the other hand, chronically elevated glucose levels induce glycation of structural proteins in the vessel walls. The silent denaturation of these proteins advances aging of vessels, induces chronic endothelial inflammation, and promotes activation of different metabolic cascades because blood cells interact with the altered vessel walls and get stimulated. Clinically, this leads to an increased risk of hypertension, cardiovascular events, and stroke (macroangiopathy). Early signs of microangiopathy can be best assessed in the eye (cataract, retinal vessels).

Insufficient secretion of insulin is more frequently observed with type-I DM as a result of an acute injury to pancreatic islet cells (infection, autoimmune disorder, for example) and affects younger individuals. In contrast, peripheral insulin resistance (type-II DM) is more frequently observed during ageing; or because a subtle, but chronic increase in fat mass during lifetime (obesity) induces secretion of adipokines like leptin that directly causes insulin resistance. Treatment strategies for type-II DM are normalization of body weight and fat mass, increasing physical activity, changing the dietary regime, and/or oral antidiabetic drugs that stimulate pancreatic insulin secretion or—in severe cases—subcutaneous insulin injections. The latter one is also usually required to treat type-I DM.

Obesity describes conditions with increased body fat. On one hand, obesity leads to disturbed metabolism with high serum levels of glucose and fatty acids because there is an imbalance between energy-consuming body mass (lean and muscle mass, respectively) and energy reserves (fat mass). On the other hand, recent research confirmed that adipocytes are metabolically highly active and secrete adipokines as feedback signals to the body. For example, the release of leptin is proportional to the amount of body fat and thus provides information of how much energy is stored within the body. As a consequence, distinct metabolic or signaling pathways will be switched on or off: like a certain level of leptin is needed

to switch on the LH-RH axis to allow menstrual cycle and ovulation. An excess of fat mass leads to higher levels of adipokines, some of them induce inflammation (like C-reactive protein). In addition to the known negative effects of chronically elevated substrate levels on vascular integrity (see paragraph about “Diabetes”), obesity needs to be considered as a condition with chronic inflammation. Obesity reduces life expectancy and increases the risk for preceding morbidity in all body organs. Worldwide, the incidence of obesity is dramatically increasing and is affecting more frequently young adults and children. Obesity seems to be the result of an imbalance between nutrient intake and energy expenditure, with a higher incidence in populations with a lower socioeconomic status. Lifestyle in modern societies leads to permanent availability of highly palatable food and to reduced physical activity. Education and change in lifestyle can reduce the incidence of obesity, and recent data suggest that some populations have managed to stop the increasing incidence. BMI is a parameter that tries to reflect nutritional status and body fat. For clinical use, cutoff values have been established that define overweight and obesity; however, genetic disposition or lifestyle-induced changes of muscle mass (e.g., weight lifting) are not taken into account. In contrast to adults, the amount and distribution of body fat changes during childhood. Age and gender-specific percentile curves are available for BMI during childhood.

REFERENCES

1. Salisbury C, Robertson C. Maternal nutrition: building foundations of long-term good health. *Nutr Bull.* 2013;38:249–253.
2. Gluckman PD, Hanson MA, Beedle AS. Early life events and their consequences for later disease: a life history and evolutionary perspective. *Am J Hum Biol.* 2007;19:1–19.
3. Barker DJP, Godfrey KM, Gluckman PD, et al. Fetal nutrition and cardiovascular disease in adult life. *Lancet.* 1993;341:938–941.
4. Bloomfield FH. How is maternal nutrition related to preterm birth? *Ann Rev Nutr.* 2011;31:235–261.
5. Sommer F, Bäckhed F. The gut microbiota—masters of host development and physiology. *Nat Rev Microbiol.* 2013;11(4):227–238.
6. Roger LC, McCartney AL. Longitudinal investigation of the faecal microbiota of healthy full-term infants using fluorescence *in situ* hybridization and denaturing gradient gel electrophoresis. *Microbiology.* 2010;156:3317–3328.
7. Ouwehand A, Isolauri E, Salminen S. The role of the intestinal microflora for the development of the immune system in early childhood. *Eur J ClinNutr.* 2002;41:132–137.
8. Maslowski KM, Mackay CR. Diet, gut microbiota and immune responses. *Nat Immunol.* 2011;12(1):5–9.

9. Kuo SM. The interplay between fiber and the intestinal microbiome in the inflammatory response. *AdvNutr*. 2013;4:16–28.
10. Friedrich MJ. Genomes of microbes inhabiting the body offer clues to human health and disease. *JAMA.* 2013;309(14):1447–1449.
11. Adlerberth I, Wold AE. Establishment of the gut microbiota in Western infants. *Acta Paediatr.* 2009;98:229–238.
12. Flint HJ. The impact of nutrition on the human microbiome. *Nutr Rev*. 2012;70(suppl 1):S10–S13.
13. Brownawell AM, Caers W, Gibson GR, et al. Prebiotics and the health benefits of fiber: current regulatory status, future research, and goals. *J Nutr*. 2012;142(5):962–974.
14. Delzenne NM, Neyrinck AM, Cani PD. Modulation of the gut microbiota by nutrients with prebiotic properties: consequences for host health in the context of obesity and metabolic syndrome. *Microb Cell Fact.* 2011;10(suppl 1):S10–S20.
15. Koropatkin NM, Cameron EA, Martens EC. How glycan metabolism shapes the human gut microbiota. *Nat Rev Microbiol.* 2012;10:323–335.
16. Harmsen HJ, Wildeboer-Veloo AC, Raangs GC, et al. Analysis of intestinal flora development in breast-fed and formula-fed infants by using molecular identification and detection methods. *J Pediatr Gastroenterol Nutr*. 2000;30:61–67.
17. Isolauri E. Development of healthy gut microbiota early in life. *J Paediatr Child Health*. 2012;48(suppl 3):1–6.
18. Arumugam M, Raes J, Pelletier E, et al. Enterotypes of the human gut microbiome. *Nature*. 2011;473(7346):174–180.
19. Walker AW, Ince J, Duncan SH, et al. Dominant and diet-responsive groups of bacteria within the human colonic microbiota. *The ISME J*. 2010;5:220–230.
20. Kau AL, Ahern PP, Griffin NW, et al. Human nutrition, the gut microbiome and the immune system. *Nature.* 2011;474:327–336.
21. Lozupone CA, Stombaugh JI, Gordon JI, et al. Diversity, stability and resilience of the human gut microbiota. *Nature.* 2011;489:220–230.
22. Maynard CL, Elson CO, Hatton RD, Weaver CT. Reciprocal interactions of the intestinal microbiota and immune system. *Nature.* 2012;489:231–241.
23. RudloffS, Kunz C. Proteins and non-protein nitrogen in human milk, bovine milk and infant formula. Quantitative and qualitative aspects in infant nutrition. *J Paediatr Gastroenterol Nutr.* 1997;24:328–344.
24. Jensen RG. *Handbook of Milk Composition*. San Diego, CA: Academic Press; 1995.
25. Urashima T, Taufik E, Fukuda K, Asakuma S. Recent advances in studies on milk oligosaccharides of cows and other domestic farm animals. *Biosci Biotechnol Biochem*. 2013;77(3):455–466.
26. György P, Norris RF, Rose CS. Bifidus factor I. A variant of *Lactobacillus bifidus* requiring a special growth factor. *Arch Biochem Biophys.* 1954;48:193–201.
27. Kitaoka M. Bifidobacteria lenzymes involved in the metabolism of human milk oligosaccharides. *AdvNutr.* 2012;3:422S–429S.
28. Sela DA, Mills DA. Nursing our microbiota: molecular linkages between bifidobacteria and milk oligosaccharides. *Trends Microbiol.* 2010;18:298–307.
29. Marcobal A, Barboza M, Sonnenburg ED, et al. Bacteroides in the infant gut consume milk oligosaccharides via mucus-utilization pathways. *Cell Host Microbe.* 2011;10:507–514.
30. Sela DA, Chapman J, Adeuya A, et al. The genome sequence of *Bifidobacterium longum* subsp. *infantis* reveals adaptations for milk utilization within the infant

microbiome. *Proc Natl Acad Sci USA.* 2008;105:18964–18969.
31. LoCascio RG, Desai P, Sela DA, et al. Broad conservation of milk utilization genes in *Bifidobacterium longum* subsp. *infant i*sas revealed by comparative genomic hybridization. *Appl Environ Microbiol.* 2010;76(22):7373–7381.
32. Asakuma S, Hatakeyama E, Urashima T, et al. Physiology of consumption of human milk oligosaccharides by infant gut-associated bifidobacteria. *J Biol Chem.* 2011;286:34583–34592.
33. Kitaoka M, Tian J, Nishimoto M. Novel putative galactose operon involving lacto-*N*-biosephoshorylase in *Bifidobacterium longum*. *Appl Environ Microbiol.* 2005;71:3158–3162.
34. Wada J, Ando T, Kiyohara M, et al. *Bifidobacterium bifidum*lacto-*N*- biosidase, a critical enzyme for the degradation of human milk oligosaccharides with a type 1 structure. *Appl Environ Microbiol.* 2008;74:3996–4004.
35. Suzuki R, Wada J, Katayama T, et al. Structural and thermodynamic analyses of solute-binding protein from *Bifidobacterium longum* specific for core 1 disaccharide and lacto-*N*-biose I. *J Biol Chem.* 2008;283:13165–13173.
36. Hidaka M, Nishimoto M, Kitaoka M, et al. The crystal structure of galacto-*N*-biose/lacto-*N*-biose I phosphorylase: a large deformation of a TIM barrel scaffold. *J Biol Chem.* 2009;284:7273–7283.
37. Yoshida E, Sakurama H, Kiyohara M, et al. *Bifidobacterium longum* subsp. *infantis* uses two different galactosidases for selectively degrading type-1 and type-2 human milk oligosaccharides. *Glycobiology.* 2012;22:361–368.
38. Urashima T, Asakuma S, Leo F, et al. The predominance of type I oligosaccharides is a feature specific to human breast milk. *Adv Nutr.* 2012;3:473S–482S.
39. Sela DA, Li Y, Lerno L, et al. An infant-associated bacterial commensal utilizes breast milk sialyloligosaccharides. *J Biol Chem.* 2011;286:11909–11918.
40. Kiyohara M, Tanigawa K, Chaiwangsri T, et al. An exo-α-sialidase from bifidobacteria involved in the degradation of sialyloligosaccharides in human milk and intestinal glycoconjugates. *Glycobiology.* 2011;21:437–447.
41. Salminen S, Bouley C, Boutron-Ruault MC, et al. Functional food science and gastrointestinal physiology and function. *Br J Nutr*. 1998;80(suppl 1):S147–S171.
42. Gibson GR, Beatty ER, Wang X, Cummings JH. Selective stimulation of bifidobacteria in the human colon by oligofructose and inulin. *Gastroenterology*. 1995;108(4):975–982.
43. Sherman PM, Cabana M, Gibson GR, et al. Potential roles and clinical utility of prebiotics in newborns, infants, and children: proceedings from a global prebiotic summit meeting, New York city, June 27–28, 2008. *J Paediatr*. 2009;155:S61–S70.
44. Meyer D, Stasse-Wolthuis M. The bifidogenic effect of inulin and oligofructose and its consequences for gut health. *Eur J Clin Nutr*. 2009;63(11):1277–12789.
45. Thomas DW, Greer FR, Committee on Nutrition, et al. Clinical report—probiotics and prebiotics in pediatrics. *Pediatrics.* 2010;126(6):1217–1231.
46. Vandenplas Y, De Greef E, Hauser B, et al. Probiotics and prebiotics in pediatric diarrheal disorders. *Expert Opin Pharmacother*. 2013;14(4):397–409.
47. Abrams SA, Griffin IJ, Hawthorne KM, et al. A combination of prebiotic short- and long-chain inulin-type fructans enhances calcium absorption and bone mineralization in young adolescents. *Am J Clin Nutr.* 2005;82(2):471–476.
48. Latulippe ME, Meheust A, Augustin L, et al. ILSI Brazil International Workshop on Functional Foods: a narrative review of the scientific evidence in the area of carbohydrates, microbiome, and health. *Food Nutr Res.* 2013;57[EpubFebruary 7,

2013].
49. Kunz C, Rudloff S, Baier W, et al. Oligosaccharides in human milk. Structural, functional, and metabolic aspects. *Annu Rev Nutr.* 2000;20:699–722.
50. Rudloff S, Kunz C. Milk oligosaccharides and metabolism in infants. *Adv Nutr.* 2012;3:398–405.
51. Rastall RA. Functional oligosaccharides: application and manufacture. *Annu Rev Food Sci Technol*. 2010;1:305–339.
52. Bode L. Human milk oligosaccharides: every baby needs a sugar mama. *Glycobiology.* 2012;22:1147–1162.
53. Kunz C, Kuntz S, Rudloff S. Bioactivity of human milk oligosaccharides. In: Javier MF, Luz SM, eds. *Food Oligosaccharides: Production, Analysis and Bioactivity*. Oxford: Wiley-Blackwell. In press.
54. Bray GA. How bad is fructose? *Am J Clin Nutr.* 2007;86:895–896.
55. Payne AN, Chassard C, Lacroix C. Gut microbial adaptation to dietary consumption of fructose, artificial sweeteners and sugar alcohols: implications for host-microbe interactions contributing to obesity. *Obes Res.*2012;13(9):799–809.
56. Hellerstein MK. Mitigating factors and metabolic mechanisms in fructose-induced nonalcoholic fatty liver disease: the next challenge. *Am J Clin Nutr*. 2012;96(5):951–952.
57. Klurfeld DM. What do government agencies consider in the debate over added sugars? *Adv Nutr*. 2013;4(2):257–261.
58. White JS. Challenging the fructose hypothesis: new perspectives on fructose consumption and metabolism. *Adv Nutr.* 2013;4(2):246–256.
59. Rippe JM, Angelopoulos TJ. Sucrose, high-fructose corn syrup, and fructose, their metabolism and potential health effects: what do we really know? *Adv Nutr.* 2013;4(2):236–245.
60. Rippe JM, Saltzman E. Sweetened beverages and health: current state of scientific understandings. *Adv Nutr.* 2013;4(5):527–529.
61. Bray GA. Energy and fructose from beverages sweetened with sugar or high-fructose corn syrup pose a health risk for some people. *Adv Nutr.* 2013;4(2):220–225.
62. Lustig RH. Fructose: it's "alcohol without the buzz". *Adv Nutr.* 2013;4(2):226–235.
63. Basciano H, Federico L, Adeli K. Fructose, insulin resistance, and metabolic dyslipidemia. *Nutr Metab.* 2005;2:5.
64. Sievenpiper JL, de Souza RJ, Kendall CW, Jenkins DJ. Is fructose a story of mice but not men? *J Am Diet Assoc*. 2011;111(2):219–220.
65. Tappy L, Mittendorfer B. Fructose toxicity: is the science ready for public health action? *Curr Opin Clin Nutr.* 2012;15(4):357–361.
66. Panel on Macronutrients, Panel on the Definition of Dietary Fiber, Subcommittee on Upper Reference Levels of Nutrients, Subcommittee on Interpretation and Uses of Dietary Reference Intakes, and the Standing Committee on the Scientific Evaluation of Dietary Reference Intakes. Food and Nutrition Board. Institute of Medicine of the National Academics. *Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids*. National Academy of Sciences;28–29, ISBN 978-0-309-08525-0.
67. Stephen A, Alles M, de Graaf C, et al. The role and requirements of digestible dietary carbohydrates in infants and toddlers. *Eur J Clin Nutr.* 2012;66:765–779.
68. Food Agriculture Organisation. *Carbohydrates in Human Nutrition. Report of a Joint FAO/WHO Expert Consultation*. Rome: Food and Agriculture Organisation of the United Nations; 1998.

69. World Health Organisation. *Diet, Nutrition and the Prevention of Chronic Diseases. Report of a Joint WHO/FAO Expert Consultation*. Geneva: World Health Organisation; 2003.
70. EFSA (European Food safety Authority) Panel on Dietetic Products, Nutrition and Allergies. Scientific opinion on dietary reference values for carbohydrates and dietary fibre. *EFSA J.* 2010;8:14–62.
71. Agostini C, Buonocore G, Carnielli VP, et al. Enteral nutrient supply for preterm infants: commentary from the European Society for Paediatric Gastroenterology, Hepatology, and Nutrition. Committee on Nutrition. *JPGN.*2010;50:89–95.
72. Coppa GV, Gabrielli O, Pierani P, et al. Variations of carbohydrates in human milk during the first 4 months of lactation. *Pediatrics.*1993;91:637–641.
73. Loennerdal B, Smith C, Keen CL. Analysis of breast milk: current methodologies and future needs. *JPGN.* 1984;3:290–295.
74. Loennerdal B, Forsum E, Hambraeus L. A longitudinal study of the protein, nitrogen and lactose contents of human milk from Swedish well-nourished mothers. *AJCN.* 1979;29:1127–1132.
75. Ziegler EE, Thureen PJ, Carlson SJ. Aggressive nutrition of the very low birth weight infant. *Clin Perinatol.* 2002;29:225–244.
76. Kashyap S, Towers HM, Sahni R, et al. Effects of quality of energy on substrate oxidation in enterally fed, low-birth-weight infants. *Am J ClinNutr*. 2001;74:374–380.
77. Aggett PJ, Agostoni C, Axelsson I, et al. Nondigestible carbohydrates in the diets of infants and young children: a commentary by the ESPGHAN Committee on Nutrition. *JPGN*. 2003;36:329–337.
78. Rimm EB, Ascherio A, Giovannucci E, et al. Vegetable, fruit, and cereal fiber intake and risk of coronary heart disease among men. *JAMA.*1996;275:447–451.
79. Wolk A, Manson JE, Stampfer MJ, et al. Long-term intake of dietary fibre and decreased risk of coronary heart disease among women. *Am J Clin Nutr.* 1999;49:1179–1183.
80. Pietinnen P, Rimm EB, Korhonen P, et al. Intake of dietary fibre and risk of coronary heart disease in a cohort of Finnish men. The Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study. *Circulation.* 1996;94:2720–2727.
81. Mugambi MN, Musekiwa A, Lombard M, et al. Probiotics, prebiotics infant formula use in preterm or low birth weight infants: a systematic review. *Nutr J.*2012;11:58.
82. Kunz C, Rudloff S. Health promoting aspects of milk oligosaccharides. *Int Dairy J.* 2006;16(11):1341–1346.
83. Ammoury RF, Croffie JM. Malabsorptive disorders in childhood. *Pediatr Rev.* 2010;31:407–415.
84. DeBosch BJ, Chi M, Moley KH. Glucose transporter 8 (GLUT8) regulates enterocyte fructose transport and global mammalian fructose utilization. *Endocrinology*. 2012;153:4181–4191.
85. Sui Y, Juras G, Kostner GM. Fructose malabsorption influences chronic and recurrent infectious diseases, dyspepsia and heartburn. *Open Gastroenterol J.* 2012;6:1–7.
86. Komericki P, Akkilic-Materna M, Strimitzer T, et al. Oral xylose isomerase decreases breath hydrogen excretion and improves gastrointestinal symptoms in fructose malabsorption—a double-blind, placebo-controlled study. *Aliment Pharmacol Ther*. 2012;36:980–987.
87. Putkonena L, Yaob CK, Strimitzer T, et al. Fructose malabsorption syndrome. *Curr Opin Clin Nutr Metab Care.* 2013;16:473–477.

88. Hicks J, Wartchow E, Mierau G. Glycogen storage diseases: a brief review and update on clinical features, genetic abnormalities, pathologic features, and treatment. *Ultrastruct Pathol.* 2011;35:183–196.
89. Bosch AM. Classical galactosaemia revisited. *J Inherit Metab Dis.* 2006;29:516–525.
90. Arnoux JB, Verkarre V, Saint-Martin C, et al. Congenital hyperinsulinism: current trends in diagnosis and therapy. *Orphanet J Rare Dis.* 2011;6:63.

Proteins and Amino Acids

6

Johannes B. van Goudoever, MD, PhD

Protein is the major functional and structural component of all the cells of the body. The defining characteristic of protein or amino acid is its requisite amino nitrogen group. This nitrogen group distinguishes an amino acid from, for example a sugar. Proteins are macromolecules consisting of long chain of amino acid subunits. In the protein molecule, the amino acids are joined together by peptide bonds. In biological systems, the chains formed might be anything from a few amino acids (di-, tri-, or oligopeptide) to thousands of units (polypeptide). The sequence of amino acids in the chain is known as the primary structure. A critical feature of proteins is the complexity of their physical structures. Polypeptide chains do not exist as long chains, but they fold into a three-dimensional structure. The chains of amino acids tend to coil into helices (secondary structure). Sections of the helices may fold on each other due to hydrophobic interactions between nonpolar side chains and, in some proteins, to disulphide bonds so that the overall molecule might be globular or rod-like (tertiary structure). Their exact shape depends on their function and for some proteins, their interaction with other molecules (quaternary structure).[1]

AMINO ACIDS

The amino acids that are incorporated into mammalian protein are α–amino acids. This means that amino acids have a carboxyl group, an amino, nitrogen group, and a side chain attached to a central α-carbon (Figure 6-1).

Proline is the only exception, being unique among the 20 protein-

forming amino acids in that the amine nitrogen is bound to not one, but two alkyl groups, thereby making it an α-iminoacid. Functional differences among the amino acids lie in the structure of their side chains (Table 6-1).

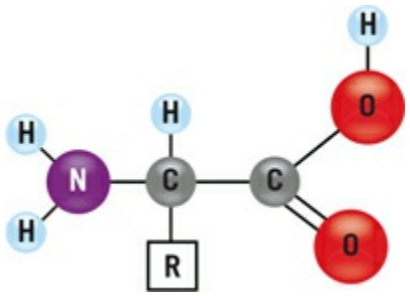


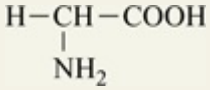
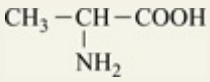
Figure 6-1 An amino acid: an amino (NH_2) group, a carboxyl group (COOH) and a side chain attached to a central α-carbon (R).

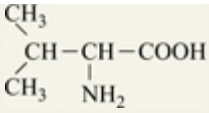
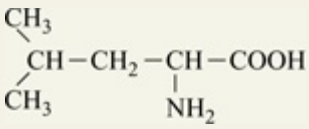
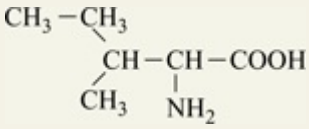
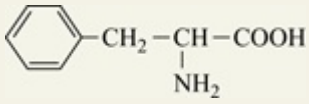
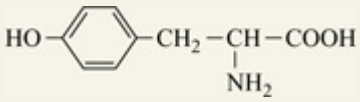
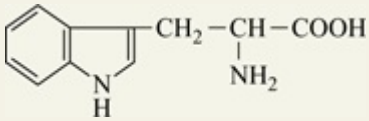
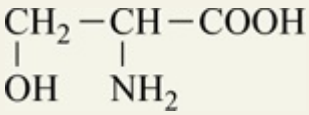
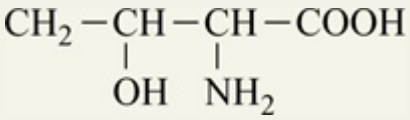
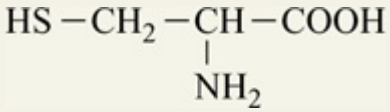
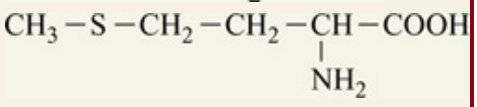
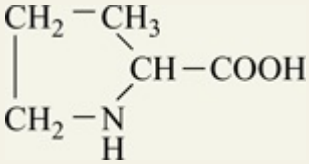
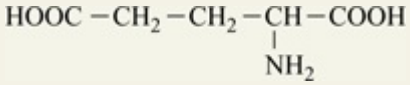
Classification of amino acids was initially based on the classic studies of Rose and co-workers.[2,3] These studies were based on the qualitative significance of each amino acid in human nutrition. Rose defined amino acids as essential or nonessential based on their capacity to maintain nitrogen balance in young men (Table 6-2). Hol[4], Snyderman,[5–7] and Fomon[8] extended Rose's studies to the pediatric population. In their studies, of which only a few are referenced here) they demonstrated that only the "essential" amino acids were needed to maintain growth in young infants and therefore conformed according to the original definition of "essentiality of amino acids." Through the following decades it became evident that Rose's classification was limited and that a distinction between essential and nonessential amino acids was complex and could not be defined based solely on the nutritional criteria of maintenance of nitrogen balance or growth.[9] For example, histidine, an nonessential amino acid in the rat that was considered nonessential in humans in the original Rose classification, has been proven to be essential in health and disease, and was included as an essential amino acid by the Food Agricultural Organization/World Health Organization/United Nations University (FAO/WHO/UNU) expert report in 1985 and subsequent reports.[10]

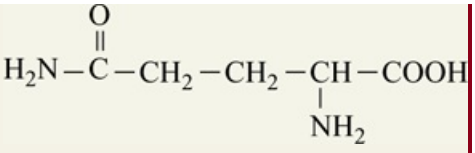
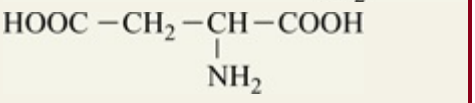
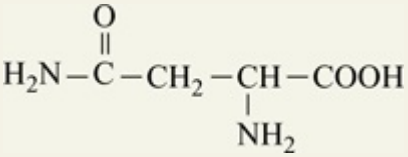
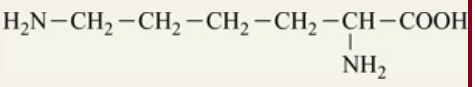
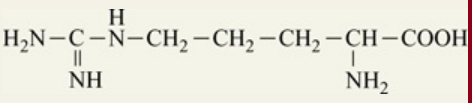
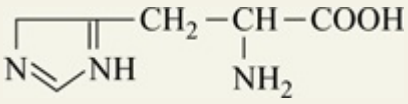
Over the past decades the concept of essentiality based on maintenance of growth or of nitrogen equilibrium has been revisited and, with contributions by many[11–15] the concept of amino acid essentially has changed. Mitchell[16] considered a more dynamic concept of "essentiality"

based on the ratio of supply to demand, rather than on the maintenance of nitrogen balance or growth. The term “conditionally indispensable” has been used to refer to those nonessential or dispensable amino acids that become essential or indispensable under pathophysiologic conditions. Because the carbon skeleton of the amino acid is the moiety that determines its nutritional indispensability, some classification of amino acids has been based on the ability to synthesize and/or aminate the carbon skeleton.[13] In metabolic terms, the amino acids that are generally considered to be nutritionally essential contain specific chemical structures, the synthesis of which cannot be catalyzed by mammalian enzymes, such as the branch aliphatic side chain of leucine, isoleucine, or valine; the primary amine of lysine; the secondary alcohol of threonine; the secondary thiol of methionine; the indol ring of tryptophan; the aromatic ring of phenylalanine; or the imidazole ring of histidine. Table 6-3 summarizes the classic nutritional classification of amino acids and Table 6-4 shows an updated classification of amino acids It is important, however, to remember that an amino acid classification should not be based solely on nutritional (growth and maintenance), metabolic (de novo synthesis, tissue utilization, and catabolism), or chemical aspects of the amino acids, but also, and perhaps most importantly, on the maintenance of amino acid function. Some amino acids that would be considered truly dispensable in nutritional, metabolic, and chemical terms are utilized as precursors of other amino acids that maintain important physiologic functions, such as glutamate and serine, which serve, respectively, as precursors of glutamine and cystathionine. Other dispensable amino acids like alanine and aspartate contribute to physiological and pathophysiologic events.[17,18] The involvement of amino acids in physiological and metabolic functions is extensive and a partial description is shown in Table 6-5.

TABLE 6-1 Amino Acid Structure

Name	Abbreviation	Formula
Aliphatic side chains		
Glycine	Gly	
Alanine	Ala	
Valine[a]	Val	

		
Leucine[a]	Leu	
Isoleucine[a]	Ile	
Aromatic side chains		
Phenylalanine	Phe	
Tyrosine	Tyr	
Tryptophan	Trp	
Hydroxyl groups in side chains		
Serine	Ser	
Threonine	Thr	
Sulfur-containing side chains		
Cysteineb	Cys	
Methionine	Met	
Imino Acids		
Proline[†]	Pro	
Acidic side chains and their amides		
Glutamic acid	Glu	
Glutamine	Gln	

		
Aspartic acid	Asp	
Asparagine	Asn	
Basic side chains		
Lysine	Lys	
Arginine	Arg	
Histidine	His	

[a] Leucine, valine, and isoleucine are known as the branch chain amino acids.

[b] Cysteine is found as a dimer, linked through the sulfur atoms by oxidation

[†] Proline is strictly an imino acid rather than amino acid.

From Institute of Medicine, Dietary Reference Intakes for Energy Carbohydrate Fiber Fat Fatty Acids Cholesterol Protein and Amino Acids. Washington, DC: National Academy of Sciences; 2002.

TABLE 6-2 Amino Acid Classification by the FAO/WHO/UNU Expert Report, 1985

Essential	Non-Essential
Lysine	Glycine
Tryptophan	Alanine
Threonine	Serine
Isoleucine	Cystine
Leucine	Tyrosine
Methionine	Aspartic acid
Phenylalanine	Glutamic acid
Valine	Proline
Histidine	Hydroxyproline
	Arginine

TABLE 6-3 Nutritional Classification of Amino Acids

Indispensable

(a) Carbon skeleton cannot be synthesized.

(b) Rate limiting enzymes of catabolism regulated in relation to adequacy of intake and tissue supply.

Conditionally Indispensable

(a) Indispensable amino acid is a precursor (i.e., phenylalanine-tyrosine).

(b) Synthesis and degradation are modulated by dietary supply (i.e., arginine, glycine).

(c) Stressful states cause tissue depletion where rates of synthesis are insufficient to meet the increased rate of metabolic demand (proline, glutamine, arginine, glycine, taurine) or match the catabolic rate.

Dispensable

(a) Rate of synthesis is not down regulated by intake of amino acid; metabolism largely a function of metabolic status of major energy-yielding substrates and/or overall nutritional status (alanine, glutamate, aspartate).

From Ref.[31]

TABLE 6-4 Involvement of Amino Acids in Physiologic and Metabolic Function

System	Function	Product	Precursor
Intestine	Energy generation	ATP	Glu, Asp, Glutamine
	Proliferation	Nucleic acids	Glutamine, Gly, Asp
	Protection	Glutathione	Cys, Glu, Gly
		Nitric oxide	Arg
		Mucins	Thr, Cys, Ser, Pro
Skeletal muscle	Energy generation	Creatine	Gly, Arg, Met
	Peroxidative protection	Taurine	Cys
Nervous system	Transmitter synthesis	Adrenergic	Phe
		Serotinergic	Try
		Glutaminergic	Glu
		Glycinergic	Gly
		Nitric oxide	Arg
	Peroxidative protection	Taurine (?)	Cys
Immune system	Lymphocyte proliferation	(?)	Glutamine, Arg, Asp

	Peroxidative protection	Glutathione	Cys, Glu, Gly
Cardiovascular	Blood pressure regulation	Nitric oxide	Arg
	Peroxidative protection (?)	Red cell glutathione	Cys, Glu, Gly

Source: Adapter from the previous version of this chapter.
From Ref.[33]

TABLE 6-5 1996 IDECG Revised Estimates on Average Protein Intake in Breast Fed Infants 0–6 Months of Age and the Intakes Estimated by the 1985 FAO/WHO/UNU Expert Report

Age (Months)	1985 FAO/WHO/UNU[a] "Crude Protein" (g kg^{-1} d^{-1})	1996 IDECG[b] "Adjusted Protein" (g kg^{-1} d^{-1})
0–1	2.46	1.95–2.04
2	1.93	1.41–1.48
3	1.74	1.19–1.25
4	1.49	1.11–1.16
6		1.05–1.11

[a] From 1985 FAO/WHO/UNU 1985.[28]

[b] From 1996 IDECG.[41] Exclusively breastfed infants. Nitrogen intake includes non-protein nitrogen (NPN). Adjusted protein intake is based on milk protein concentration plus 41%–61% of the NPN (protein = 6.25 × nitrogen).

Source: Adapted from the previous version of this chapter.

PROTEIN METABOLISM AND DISTRIBUTION

Most proteins in the body are constantly being synthesized from amino acids and degraded back to them. The turnover rate of proteins in neonates and children is twice to three times higher than in adults, resulting in a higher energy demand for children per kilogram body weight than for adults.

The "pool" of amino acids is in dynamic equilibrium with tissue protein as depicted in Figure 6-2. Amino acids are continually taken from the pool for protein synthesis and replaced by the hydrolysis of dietary and tissue protein.

There are three main reactions essential for amino acid metabolism: transamination, oxidative deamination and hydroxylation. Transamination

converts one amino acid into another by catalyzing the transfer of the α-amino (NH_3^+) group from an amino acid to an α-keto-acid. All transamination reactions are fully reversible and therefore this pathway is. Lysine and threonine do not transaminate. The second reaction, which is irreversible, is the oxidative deamination. It removes the amino group, leaving behind the carbon skeleton. The ammonia (NH_4^+) formed enters the urea cycle and the carbon skeletons are all glycolytic and TCA intermediates. These products can, depending on the energy status of the cell, either be oxidized in the citric acid cycle to generate energy or used to synthetize glycogen or fat. Phenylalanine, tyrosine and tryptophan can be hydroxylated. Tryptophan hydroxylase controls levels of serotonin in the brain and the gastrointestinal tract, while tyrosine hydroxylase controls levels of dopamine, epinephrine, and norepinephrine in the brain and the adrenal medulla. Deficiency in phenylalanine hydroxylase causes hyperphenylalaninaemia, and when blood phenylalanine levels increase above 20 times the normal concentration, the metabolic disease phenylketonuria results.

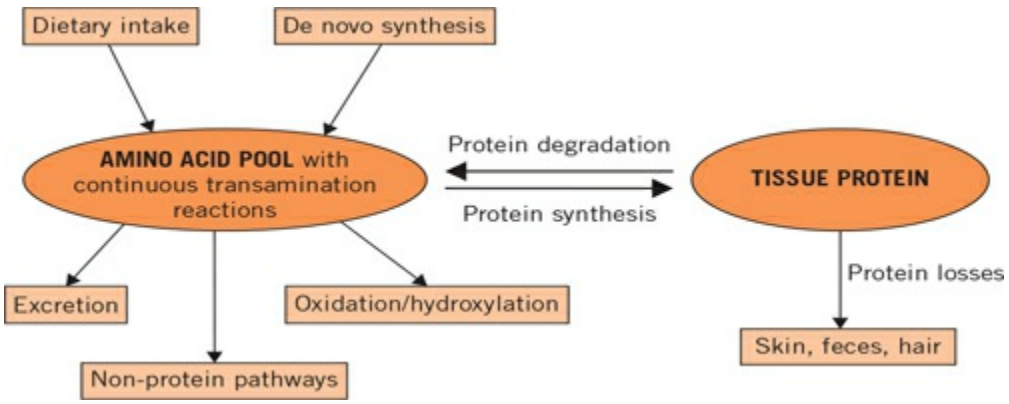


Figure 6-2 Exchange between body protein and free amino acid pools (adapted from Ref.1).

The distribution of protein among the different organs varies with developmental age. Children have, proportionally, a larger amount of visceral (liver, kidney, brain, heart, and lung) protein than in the skeletal muscle, while in adults protein constitutes about 15% of their weight. During starvation or low protein intake, visceral protein is mobilized. This is called the "labile protein content." However, these "reserves" account only for 1% of total body protein.[19] During disease states muscle becomes

the main contributor to protein loss.[20]

Not all proteins in the body have a similar turnover rate. Figure 6-3 shows the protein synthesis rate of different tissues in pigs and rodents, expressed as % of the protein pool of a specific organ that is synthesized per day. Despite the high fractional turnover rate of gut proteins, most protein synthesis and degradation occurs in the muscle because of the large pool size. Nevertheless, the relatively high turnover rate of the gut indicates that the intestines are metabolically a very active organ. This is illustrated by the fact that first pass uptake or splanchnic utilization is high.

DIGESTION AND ABSORPTION OF PROTEINS

Proteins are digested and absorbed along the upper part of the intestinal tract, with for human milk proteins an almost complete removal from the lumen. Proteins are broken down in the stomach to smaller polypeptide chains via hydrochloric acid and proteases in the duodenum and jejunum. Peptides and free amino acids can be absorbed by the enterocyte. However the systemic availability of amino acids is not 100% because 30%–50% of the amino acids are utilized within the intestine.

These amino acids are used as substrate for (glyco-)protein synthesis and as an energy source during high protein intakes.[26–30] The rate of first pass utilization is different for each individual amino acid as is shown in Figure 6-3. However, also dietary amino acids that are used within the gut to synthesize constitutive proteins or proteins that are being secreted in the lumen may become available to the systemic circulation in a later phase due to recycling of these proteins further on in the gastro-intestinal tract.[28] The availability depends on the digestibility of the (glyco-)protein. The structural component of the mucus layer, which forms a physical barrier between the underlying epithelium and the lumen of the gastrointestinal tract is secretory mucin 2 (MUC2). MUC2 is secreted by goblet cells of the intestine and gives intestinal mucus a high density and viscoelasticity. MUC2 is a glycoprotein rich in oligosaccharides, O-linked to threonine and serine residues in the peptide backbone. This glycoprotein with its high threonine content reaches almost completely intact the colon, where proteins amino acids are not absorbed, with subsequently a low systemic availability of threonine.[31]

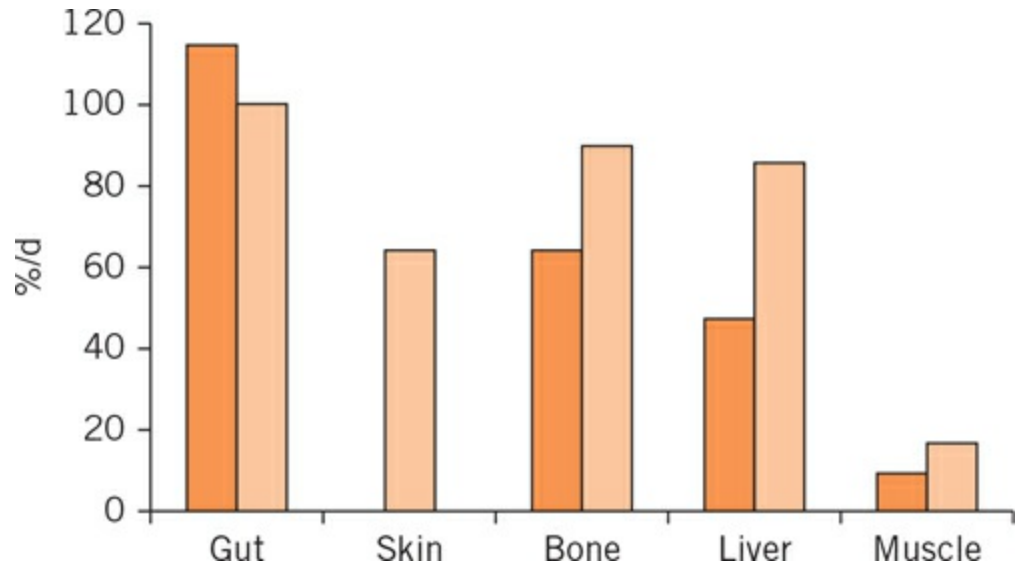


Figure 6-3 Fractional protein synthesis rates of different organs expressed as percent per day in pigs (dark orange bars) and rats (light orange bars).[21–24]

REQUIREMENTS FOR PROTEINS

Adequate nitrogen (protein) intake is essential for growth, development, and function. However, estimating "adequate" protein or specific amino acid requirement for the pediatric population is a complex and challenging endeavor.[32] First, the pediatric population constitutes a dynamic group, involving different clusters of individuals from newborn to adolescence. The pediatric population shows marked differences among the age groups regarding weight, rate of growth, hormonal milieu, activity, and other factors that may affect nutritional and metabolic status. Second, dietary recommendations for macro- and micronutrients among the different pediatric groups have been largely based on limited and inadequate data.[33–38] Because of ethical constraints that prevent the use of healthy children for more invasive research studies, data in the pediatric population are based on nitrogen balance studies, many times obtained from children recovering from malnutrition.[38,39] Hence, pediatric data are rather limited. Third, with new knowledge emerging on nonnutritional function of nutrient hyperhomocysteinaemia with folic acid[40] as well as the physiological and pharmacological properties of amino acids, the concept of amino acid requirement has changed from the traditional

nutritional criteria based on growth or weight maintenance, to a broader functional outcome. Therefore, "adequacy" of nutritional support is ill defined and conditional to the end goals to be achieved. Based on this new knowledge about nonnutritional functions of amino acids,[11,13] a key question regarding nutritional adequacy is "what is the objective to be achieved?" Is the objective to promote growth and weight maintenance? To maintain nonnutritional functions? To prevent pathophysiologic events? To act as pharmacological agent(s)? To induce gene expression? Or for nutritional rescue? These important questions are just beginning to be explored and will be better answered in the future.

TABLE 6-6 Protein Needs for Growth and Maintenance in Children of Different Ages

Age	Protein Gain (g/kg/d)	Maintenance (g/kg/d)	Maintenance/Total (%)	Growth/Total (%)
0–1 mo	1.00	0.56	36	64
1–3 mo	0.57	0.56	50	50
3–6 mo	0.30	0.56	65	35
6–12 mo	0.18	0.56	76	24
1–2 y	0.11	0.56	89	11
1–2 y	0.11	0.56	89	11
2–5 y	0.07	0.56	89	11

Maintenance assumed to be 90 mg kg^{-1} d^{-1}.

Source: Modified from Ref.41.

TABLE 6-7 Mean Daily Rates of Protein Deposition and Factorial Model Calculations of Mean Requirements for Protein

	Girls		Boys	
	Protein,	Mean,	Protein,	Mean,

Age (Years)	Deposition (mg/kg/d)	Requirement (g/kg/d)	Deposition (mg/kg/d)	Requirement (g/kg/d)
0.75	232	1.09	252	1.12
1	189	1.01	160	0.96
1.5	119	0.89	114	0.88
2	83	0.83	88	0.84
3	54	0.78	57	0.78
1–3	111	0.88	105	0.87
4	48	0.77	44	0.76
5	44	0.76	40	0.76
6	48	0.77	42	0.76
7	46	0.76	46	0.76
8	42	0.76	51	0.77
4–8	46	0.76	45	0.76
9	48	0.77	55	0.78
10	36	0.74	51	0.77
11	35	0.75	48	0.77
12	39	0.75	48	0.77
13	29	0.74	41	0.76
9–13	37	0.75	49	0.77
14	23	0.73	38	0.75
15	19	0.72	34	0.74
16	8	0.70	28	0.73
17	0	0.69	19	0.72
18	0	0.69	6	0.70
14–18	10	0.71	25	0.73

From Ref.38.

TABLE 6-8 Estimated Average Requirement (EAR) and Recommended Dietary Allowance (RDA) of Amino Acids at Different Ages

Age and Gender	Maintenance Amino Acid Deposition (mg/kg/day)	Total EAR (mg/kg/day)	RDA (mg/kg/day)	Amino Acid (mg/kg/day)
7–12 Months, Boys, Girls				
Histidine	11	7	22	32

Isoleucine	15	9	30	43
Leucine	34	18	65	93
Lysine	31	18	62	89
Methionine + cysteine	15	9	30	43
Phenylalanine + tyrosine	27	18	58	84
Threonine	16	10	34	49
Tryptophan4	3	9	13	
Valine	19	12	39	58
1–3 y, Boys, Girls				
Histidine	11	3	16	21
Isoleucine	15	4	12	28
Leucine	34	8	48	63
Lysine	31	8	45	58
Methionine + cysteine	15	4	22	28
Phenylalanine + tyrosine	27	8	41	54
Threonine	16	5	24	32
Tryptophan	4	1	6	8
Valine	19	5	28	37
4–8 y, Boys, Girls				
Histidine	11	1	13	16
Isoleucine	15	2	18	22
Leucine	34	4	40	49
Lysine	31	3	37	46
Methionine + cysteine	15	2	18	22
Phenylalanine + tyrosine	27	3	33	41
Threonine	16	2	19	24
Tryptophan	4	1	5	6
Valine	19	2	23	28
9–13 y, Boys				
Histidine	11	1	13	17
Isoleucine	15	2	18	22
Leucine	34	4	40	49

Lysine	31	4	37	46
Methionine + cysteine	15	2	18	22
Phenylalanine + tyrosine	27	4	33	41
Threonine	16	2	19	24
Tryptophan	4	1	5	6
Valine	19	2	23	28
9–13 y, Girls				
Histidine	11	1	12	15
Isoleucine	15	1	17	21
Leucine	34	2	38	47
Lysine	31	2	35	43
Methionine + cysteine	15	1	17	21
Phenylalanine + tyrosine	27	2	31	38
Threonine	16	1	18	22
Tryptophan	4	<0.5	5	6
Valine	19	2	22	27
14–18 y, Boys				
Histidine	11	1	12	15
Isoleucine	15	1	17	21
Leucine	34	2	38	47
Lysine	31	2	35	43
Methionine + cysteine	15	1	17	21
Phenylalanine + tyrosine	27	2	31	38
Threonine	16	1	18	22
Tryptophan	4	<0.5	5	6
Valine	19	1	22	27
9–13 y, Girls				
Histidine	11	<0.5	12	14
Isoleucine	15	<0.5	16	19
Leucine	34	1	35	44
Lysine	31	1	32	40
Methionine +	15	<0.5	16	19

cysteine				
Phenylalanine + tyrosine	27	1	28	35
Threonine	16	<0.5	17	21
Tryptophan	4	<0.5	4	5
Valine	19	1	20	24

Source: Modified from Ref.38.

TABLE 6-9 Food and Nutrition Board Amino Acid Scoring Pattern in Infants and in Children >1 Year of Age and in All Other Older Age Groups

	Amino Acid (mg/g protein)	Infants[a] (mg/g protein)	All other groups[b] (mg/g N)
Histidine	23	18	114
Isoleucine	57	25	156
Leucine	101	55	341
Lysine	69	51	320
Methionine + cysteine	38	25	156
Phenylalanine + tyrosine	87	47	291
Threonine	47	27	170
Tryptophan	18	7	43
Valine	56	32	199

[a] Pattern based on amino acid composition of breast milk.

[b] Pattern derived from estimated average requirement for amino acid divided by estimated amino acid requirement for protein according to age.

Source: Modified from Ref. 38.

METHODS TO DETERMINE PROTEIN OR AMINO ACID REQUIREMENTS

Dietary protein requirements can be estimated by essentially two different methods. The first method calculates the daily requirement theoretically, by a factorial approach which is based on estimating the nitrogen (obligatory) losses that occur when a person is fed a diet that meets energy needs but is essentially protein free and, when appropriate, also relies on

estimates of the amount of nitrogen that is accreted during periods of growth or lost to mothers during lactation. The second, the empirical approach, measures biochemical or physiological responses to graded intakes. It includes anthropometry, chemical indices like serum albumin, total protein, immunoglobulin, retinol-binding protein and transthyretin and biochemical markers of protein excess such as blood urea nitrogen (BUN), pH, pCO_2, bicarbonate. They also include nitrogen balance and isotopic studies of whole-body nitrogen kinetics.

FACTORIAL APPROACH

The factorial approach is based on the assumption that the basal requirements of a component are the same throughout the life cycle and requirements in infants and children are higher than those in adults due to growth. The factorial approach considers the total requirement for a nutrient as the sum of obligatory losses (e.g., urine, faeces, skin), plus the amount incorporated into newly formed tissues. This method, originally described for use in humans by Hegsted[41] has been useful for estimating requirements and designing experiments for obtaining definitive empirical data.[42] In this method the obligatory losses (maintenance requirement) are added to the nutrients needed for protein synthesis (growth requirement).

NITROGEN BALANCE

Historically, descriptive or gross measures like growth and nitrogen balance have been used. In this method an increasing intake of the test amino acid results in an increase in growth rate (usually weight gain) or nitrogen balance until the required level is met for the test amino acid, after which there is no further increase in either growth rate or nitrogen balance. Nitrogen balance is the difference between nitrogen intake and the amount excreted in urine, faeces, skin, and miscellaneous losses such as breath and sweat. A limitation of the method is the need for a 7-day adaptation time to the diet since the body urea pool needs 7–10 days to adapt. In addition, intakes of nitrogen are often being overestimated and excretion is often underestimated.[43] Since it is considered unacceptable to maintain children or infants on either deficient of excessive intakes for longer periods nowadays, alternative methods were needed. The availability of isotopically labeled tracers made it possible to determine the metabolic fate of a labeled amino acid at varying dietary intakes as will be

discussed next.

ISOTOPIC TRACER METHODS

Isotopic tracer methods measure amino acid oxidation, and are based on the principle that the oxidative losses are reversible and need daily replenishment, and that the amino acids provided in excess of the needs of protein synthesis, are preferentially oxidized. A ^{15} N- or a ^{13} C-labeled amino acid is administered enterally or parentally and is distributed throughout the body's free nitrogen pool. Stable isotopes are used: atoms with an extra neutron in the nucleus which slightly increases the mass which can be detected by mass spectrometry techniques. Different techniques using stable isotopes have been developed to determine the requirements of the indispensable amino acids like the direct and indirect amino acid oxidation methods, in both healthy and disease state.[44–46] Isotopically labeled tracers are used to determine the metabolic rate of a labeled amino acid at varying dietary intakes. Amino acids consumed in excess of requirements for protein synthesis are oxidized. Deficient intake of an indispensable amino acid will result in only minimal obligatory losses. As intake increases above the required level, the oxidation of the amino acid will also increase linearly. In the direct amino acid oxidation (DAAO) technique a known dose of labeled indispensable amino acid, for example ^{13} C-lysine is provided to subjects fed graded dietary intakes of lysine, with all other dietary factors being constant. The inflection point at which oxidation begins to increase rapidly over the obligatory minimum is referred to as the breakpoint and represents the dietary requirement for that individual (Figure 6-4).[47] A refinement of the direct oxidation technique is the 24h tracer balance (24-h DAAB).[48,49] This method uses a daily balance of the amino acid that is measured and determines the requirement level of the test amino acid as the minimum intake at which the estimate of daily balance is zero.[50] El-Khoury et al. showed that the short (3–6 h) direct oxidation method was within 5% of the 24 h estimate and thus is a valid representation of daily amino acid requirements.[49] Since a 6-day adaptation to the study diet is needed, this method is not applicable to study vulnerable populations like infants.

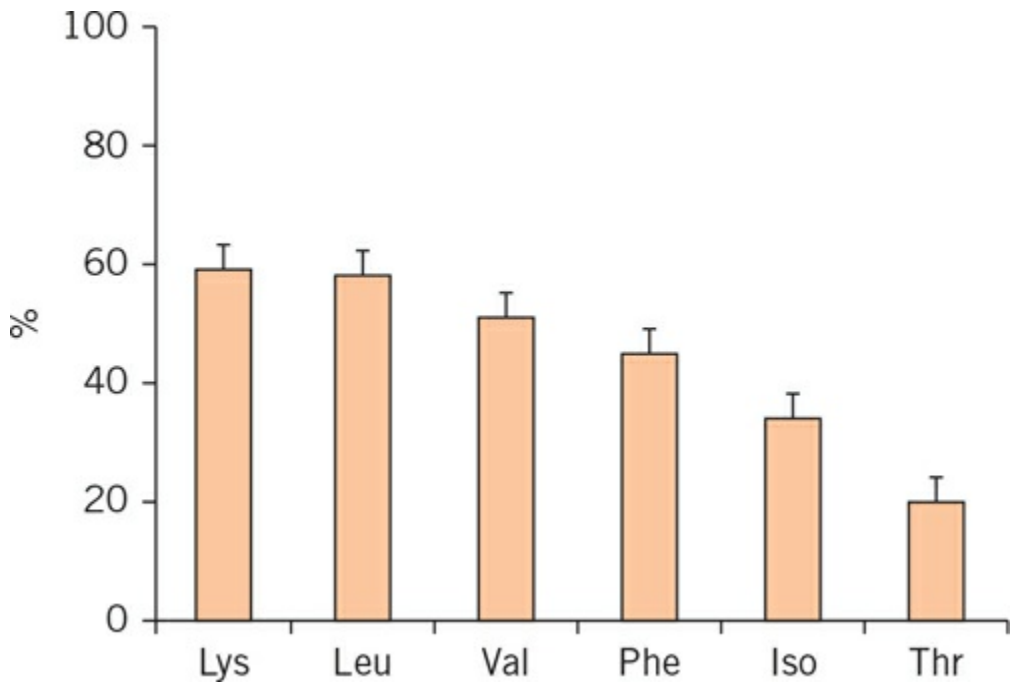


Figure 6-4 Systemic availability (expressed as percentage of intake) of enterally ingested amino acids in newborn pigs.[25] *Adapted from the previous version of this chapter.*

An alternative approach of estimating indispensable amino acid requirements using amino acid oxidation is the indirect amino acid oxidation (IAAO) technique (Figure 6-4). The main principle of this method is that because there is no storage of free amino acids, deficiency of one essential amino acid will limit protein synthesis. If the tested amino acid is deficient in the diet, this will limit protein synthesis and the indicator amino acid will be oxidized. If the dietary intake of the test amino acid increases, the oxidation of the indicator will decrease until requirement of the test amino acid is met. When intake meets the requirement, then protein synthesis occurs at an optimum capacity and the oxidative degradation of all other essential amino acids plateau. The mean requirement or estimated average requirement (EAR) of the test amino acid is identified by this breakpoint (Figure 6-4).[51–53]

The advantage of the DAAO and IAAO method are the short adaptation time needed (only one day) to the study diet which makes them applicable to study vulnerable groups such as infants and children.[54] A limitation of the DAAO method is that it is not applicable to all indispensable amino acids, whereas with the IAAO method the

requirements of any essential amino acid or conditionally indispensable amino acid can be studied. Thereby there is no dietary restriction on the test amino acid intake in the IAAO technique as a result of the fact that requirement of one amino acid is determined by the oxidation of another amino acid. This makes it possible to study all possible dietary levels of essential amino acids. Recent studies have used these techniques to determine amino acid and protein requirements, both in healthy and sick children.[55–60]

CURRENT PROTEIN AND AMINO ACID REQUIREMENT RECOMMENDATIONS

Chapter 11 describes in detail the different definitions and ranges, including the upper safe level and how the requirements are established. The requirements for the different age ranges are provided in Appendix B. The factorial method and nitrogen balance technique have been the methods of choice most frequently since not enough data are available using the amino acid oxidation method yet. However, this method will be used in the future as more data are emerging, since it has been shown that current recommendations using older methods might be an underestimation.[59,61]

AMINO ACID TOXICITY

The risk of harmful interactions elicited by pharmacological doses of single amino acid supplements may be significant. In addition, supplement purity is a major concern. Hence, to date, there is controversy about single amino acid supplementation. The safe upper level of intake is defined as the maximum level of habitual intake from all sources of a nutrient or related substance, which is judged to be unlikely to lead to adverse effects in humans (Chapter 11). Because of the higher risk of adverse effects, the safe upper level or tolerable upper level of intake is not desirable. There are limited data on amino acid toxicity in humans, and more extensive data in animal models. In regard to branched chain amino acids, and despite the role of these amino acids in protein synthesis, there is no consensus on its use in pathophysiological conditions.[62,63] The tolerable upper level for leucine in adults was determined using indicator amino acid techniques as well recently,[64] no data are available for infants or children. Sulfur amino acids exert beneficial effects through their role as precursor of glutathione,

DNA methylation reactions, and synthesis of important substrates, such as creatine, choline, etc. Sulfur amino acids also increase high-density lipoprotein and decrease very low density lipoprotein.[65] However, methionine supplementation can be associated with increased homocysteine, a marker of cardiovascular risk and promoter of oxidative injury.[66] A large intake of methionine has been associated with fatalities in the pediatric population.[67] Toxicity of sulfur amino acids can be significant and supplementation is not indicated. However, supplementation with N-acetyl cysteine appeared promising in conditions of oxidative injury[68] but a large recent RCT did not confirm a beneficial effect.[69] N-acetyl cysteine may be of some benefit in acute airway infections.[70]

CONCLUSION

In summary, the current recommendations for amino acid and protein intakes are based on limited data, although recently great progress has been made using stable isotope techniques. There is great need for further investigation on nutritional and functional requirements of amino acids and their safe upper levels of intake. Amino acid metabolism and requirements under pathophysiological conditions of the enteral and parenteral routes need to be further studied. The effect of developmental influences on the interactions between amino acids and energy (glucose and fat) metabolism needs to be closely explored.

REFERENCES

1. Institute of Medicine FaNB. Dietary reference intakes for macronutrients. In: Academies UN, editor. Washington, DC: National Academy Press; 2005.
2. Rose WC. Amino acid requirements of man. *Fed Proc*. 1949;**8** (2):546–552.
3. Rose WC. The amino acid requirements of adult man. *Nutr Abstr Rev Ser Hum Exp*. 1957;**27**(3): 631–647.
4. Holt LE Jr, Snyderman SE. Protein and amino acid requirements of infants and children. *Nutr Abstr Rev*. 1965;**35**:1–13.
5. Snyderman SE, Pratt EL, Cheung MW, et al. The phenylalanine requirement of the normal infant. *J Nutr*. 1955;**56**(2):253–263.
6. Snyderman SE, Holt LE Jr., Smellie F, et al. The essential amino acid requirements of infants: valine. *AMA J Dis Child*. 1959; **97**(2): 186–191.
7. Snyderman SE, Boyer A, Roitman E, et al. The histidine requirement of the infant. *Pediatrics*. 1963;**31**:786–801.
8. Fomon SJ. Requirements and recommended dietary intakes of protein during infancy. *Pediatr Res*. 1991; **30**(5): 391–395.
9. FAO/WHO. *Energy and Protein Requirements: Report of a Joint FAO/WHO Add*

Hoc Expert Committee. Geneva, Switzerland: WHO; 1971.
10. WHO/FAO/UNU. Protein and amino acid requirements in human nutrition. *Public Health Nutr*. 2005; **8: No. 7(A). 7. 92-4-120935-6_CH01_7**
11. Reeds PJ. Dispensable and indispensable amino acids for humans. *J Nut*. 2000;**130**(7):1835S–1840S.
12. Chipponi JX, Bleier JC, Santi MT, Rudman D. Deficiencies of essential and conditionally essential nutrients. *Am J Clin Nutr*. 1982;**35**(5 Suppl):1112–1116.
13. Reeds PJ, Biolo G. Non-protein roles of amino acids: an emerging aspect of nutrient requirements. *Curr Opin Clin Nutr Metab Care*. 2002;**5**(1):43–45.
14. Jackson AA. Aminoacids: essential and non-essential? *Lancet*. 1983;**1**(8332):1034–1037.
15. Laidlaw SA, Kopple JD. Newer concepts of the indispensable amino acids. *Am J Clin Nutr*. 1987;**46**(4):593–605.
16. Mitchell H. *Comparative Nutrition of Man and Domestic Animals*. New York: Academic Press; 1962. pp. 166–172.
17. Skolnick P. Modulation of glutamate receptors: strategies for the development of novel antidepressants. *Amino Acids*. 2002;**23**(1-3):153–159.
18. Tsai G, Coyle JT. N-acetylaspartate in neuropsychiatric disorders. *Prog Neurobiol*. 1995;**46**(5):531–540.
19. Young VR, Hussein MA, Scrimshaw NS. Estimate of loss of labile body nitrogen during acute protein deprivation in young adults. *Nature*. 1968; **218**(5141): 568–569.
20. Hansen RD, Raja C, Allen BJ. Total body protein in chronic diseases and in aging. *Ann NY Acad Sci*. 2000;**904**:345–352.
21. Garlick PJ, Fern M, Preedy VR. The effect of insulin infusion and food intake on muscle protein synthesis in postabsorptive rats. *Biochem J*. 1983;**210**(3):669–676.
22. Seve B, Reeds PJ, Fuller MF, et al. Protein synthesis and retention in some tissues of the young pig as influenced by dietary protein intake after early-weaning. Possible connection to the energy metabolism. *Reprod Nutr Dev*. 1986;**26**(3):849–861.
23. Burrin DG, Davis TA, Fiorotto ML, Reeds PJ. Stage of development and fasting affect protein synthetic activity in the gastrointestinal tissues of suckling rats. *J Nutr*. 1991;**121**(7):1099–1108.
24. McNurlan MA, Garlick PJ. Rates of protein synthesis in rat liver and small intestine in protein deprivation and diabetes. *Proc Nutr Soc*. 1979;**38**(3):133A.
25. van Goudoever JB, van der Schoor SR, Stoll B, et al. Intestinal amino acid metabolism in neonates. *Nestle Nutr Workshop Ser Paediatr Program*. 2006; **58**: 95–102; discussion 108.
26. van Goudoever JB, Stoll B, Henry JF, et al. Adaptive regulation of intestinal lysine metabolism. *Proc Natl Acad Sci USA*. 2000;**97**(21):11620–11625.
27. van der Schoor SR, van Goudoever JB, Stoll B, et al. The pattern of intestinal substrate oxidation is altered by protein restriction in pigs. *Gastroenterology*. 2001;**121**(5):1167–1175.
28. Van Der Schoor SR, Reeds PJ, Stoll B, et al. The high metabolic cost of a functional gut. *Gastroenterology*. 2002;**123**(6):1931–1940.
29. van der Schoor SR, Reeds PJ, Stellaard F, et al. Lysine kinetics in preterm infants: the importance of enteral feeding. *Gut*. 2004;**53**(1):38–43.
30. Schaart MW, Schierbeek H, van der Schoor SR, et al. Threonine utilization is high in the intestine of piglets. *J Nutr*. 2005;**135**(4):765–770.
31. Schaart MW, Schierbeek H, de Bruijn AC, et al. A novel method to determine small intestinal barrier function in human neonates in vivo. *Gut*. 2006;**55**(9):1366–

1367.
32. Dewey KG, Beaton G, Fjeld C, et al. Protein requirements of infants and children. *Eur J Clin Nutr*. 1996;**50 (Suppl 1)**:S119–S147; discussion S47-S50.
33. Dewey KG. Protein and amino acids. *Pediatrics*. 2000;**106**(5):1292.
34. Fomon SJ, Ziegler EE, Nelson SE, Edwards BB. Requirement for sulfur-containing amino acids in infancy. *J Nutr*. 1986; **116**(8): 1405–1422.
35. Huang PC, Lin CP, Hsu JY. Protein requirements of normal infants at the age of about 1 year: maintenance nitrogen requirements and obligatory nitrogen losses. *J Nutr*. 1980; **110**(9): 1727–1735.
36. Torum BYV, Rand WM. Obligatory nitrogen losses and factorial calculations of protein requirements of pre-school children. In: Torum BYV, Rand, WM, editors. *Protein-Energy Requirements of Developing Countries: Evaluation of New Data*. Tokyo, Japan: United Nations University; 2007: 159–163.
37. Nakagawa I, Takahashi T, Suzuki T, Kobayashi K. Amino acid requirements of children: nitrogen balance at the minimal level of essential amino acids. *J Nutr*. 1964;**83**:115–118.
38. Young VR, Borgonha S. Nitrogen and amino acid requirements: : the Massachusetts Institute of Technology amino acid requirement pattern. *J Nutr*. 2000; **130**(7): 1841S–1849S.
39. Intengan CL RB, Loyola A, Carlos E. Protein requirements of Philipino children 20 to 29 months old consuming local diets. In: Torum EBYV, Rand WM, editors. *Protein-Energy Requirements of Developing Countries: Evaluation of New Data*. Tokyo, Japan: United Nations University; 2007. pp. 249–257.
40. Mayer O, Filipovsky J, Hromadka M, et al. Treatment of hyperhomocysteinemia with folic acid: effects on homocysteine levels, coagulation status, and oxidative stress markers. *J Cardiovasc Pharmacol*. 2002;**39**(6):851–857.
41. Hegsted DM. Theoretical estimates of the protein requirements of children. *J Am Diet Assoc*. 1957;**33**(3):225–232.
42. el Lozy M, Hegsted DM. Calculation of the amino acid requirements of children at different ages by the factorial method. *Am J Clin Nutr*. 1975;**28**(9):1052–1054.
43. Duncan DL. The interpretation of studies of calcium and phosphorus balance in ruminants. *Nutr Abstr Rev Ser Hum Exp*. 1958;**28(3):695–715.**
44. Kurpad AV, El-Khoury AE, Beaumier L, et al. An initial assessment, using 24-h [13C]leucine kinetics, of the lysine requirement of healthy adult Indian subjects. *Am J Clin Nutr*. 1998;**67**(1):58–66.
45. Kurpad AV, Raj T, El-Khoury A, et al. Daily requirement for and splanchnic uptake of leucine in healthy adult Indians. *Am J Clin Nutr*. 2001;**74**(6):747–755.
46. Zello GA, Wykes LJ, Ball RO, Pencharz PB. Recent advances in methods of assessing dietary amino acid requirements for adult humans. *J Nutr*. 1995; **125**(12):2907–2915.
47. Zello GA, Pencharz PB, Ball RO. Phenylalanine flux, oxidation, and conversion to tyrosine in humans studied with L-[1-13C]phenylalanine. *Am J Physiol*. 1990;**259**(6 Pt 1):E835–E843.
48. Basile-Filho A, el-Khoury AE, Beaumier L, et al. Continuous 24-h L-[1-13C]phenylalanine and L-[3,3-2H2]tyrosine oral-tracer studies at an intermediate phenylalanine intake to estimate requirements in adults. *Am J Clin Nutr*. 1997;**65**(2):473–488.
49. el-Khoury AE, Fukagawa NK, Sanchez M, et al. Validation of the tracer-balance concept with reference to leucine: 24-h intravenous tracer studies with L-[1-13C]leucine and [15N-15N]urea. *Am J Clin Nutr*. 1994;**59**(5):1000–1011.
50. Kurpad AV, Regan MM, Raj T, Gnanou JV. Branched-chain amino acid

requirements in healthy adult human subjects. *J Nutr*. 2006;**136**(1 Suppl):256S–263S.
51. Pencharz PB, Ball RO. Different approaches to define individual amino acid requirements. *Annu Rev Nutr*. 2003; **23**:101–116.
52. Elango R, Ball RO, Pencharz PB. Individual amino acid requirements in humans: an update. *Curr Opin Clin Nutr Metab Care*. 2008;**11**(1):34–39.
53. Elango R, Ball RO, Pencharz PB. Indicator amino acid oxidation: concept and application. *J Nutr*. 2008;**138**(2):243–246.
54. de Groof F, Huang L, Twisk JW, et al. New insights into the methodological issues of the indicator amino acid oxidation method in preterm neonates. *Pediatr Res*. 2013;**73**(5):679–684.
55. Huang L, Hogewind-Schoonenboom JE, de Groof F, et al. Lysine requirement of the enterally fed term infant in the first month of life. *Am J Clin Nutr*. 2011;**94**(6):1496–1503.
56. Huang L, Hogewind-Schoonenboom JE, van Dongen MJ, et al. Methionine requirement of the enterally fed term infant in the first month of life in the presence of cysteine. *Am J Clin Nutr*. 2012; **95**(5):1048–1054.
57. Bross R, Ball RO, Clarke JT, Pencharz PB. Tyrosine requirements in children with classical PKU determined by indicator amino acid oxidation. *Am J Physiol Endocrinol Metab*. 2000; **278**(2): E195–E201.
58. Elango R, Ball RO, Pencharz PB. Recent advances in determining protein and amino acid requirements in humans. *Br J Nutr*. 2012;**108(Suppl 2)**:S22–S30.
59. Elango R, Humayun MA, Ball RO, Pencharz PB. Evidence that protein requirements have been significantly underestimated. *Curr Opin Clin Nutr Metab Care*. 2010;**13**(1):52–57.
60. Elango R, Ball RO, Pencharz PB. Amino acid requirements in humans: with a special emphasis on the metabolic availability of amino acids. *Amino Acids*. 2009;**37**(1):19–27.
61. Elango R, Humayun MA, Ball RO, Pencharz PB. Protein requirement of healthy school-age children determined by the indicator amino acid oxidation method. *Am J Clin Nutr*. 2011; **94**(6): 1545–1552.
62. Cynober L, Harris RA. Symposium on branched-chain amino acids: conference summary. *J Nutr*. 2006;**136**(1 Suppl):333S–336S.
63. De Bandt JP, Cynober L. Therapeutic use of branched-chain amino acids in burn, trauma, and sepsis. *J Nutr*. 2006;**136(1 Suppl): 308S–3013S.**
64. Elango R, Chapman K, Rafii M, et al. Determination of the tolerable upper intake level of leucine in acute dietary studies in young men. *Am J Clin Nutr*. 2012; **96**(4):759–767.
65. Oda H. Functions of sulfur-containing amino acids in lipid metabolism. *J Nutr*. 2006;**136**(6 Suppl): 1666S–1669S.
66. Cattaneo M. Hyperhomocysteinemia: an important risk factor for cardiovascular disease? Potentially, yes. Journal of thrombosis and haemostasis. *J Thromb Haemost*. 2003;**1**(9):1878–1879.
67. Harvey Mudd S, Braverman N, Pomper M, et al. Infantile hypermethioninemia and hyperhomocysteinemia due to high methionine intake: a diagnostic trap. *Mol Genet Metab*. 2003;**79(1):6–16.**
68. Marzullo L. An update of N-acetylcysteine treatment for acute acetaminophen toxicity in children. *Curr Opin Pediatr*. 2005;**17(2):239–245.**
69. Squires RH, Dhawan A, Alonso E, et al. Intravenous N-acetylcysteine in pediatric patients with nonacetaminophen acute liver failure: a placebo-controlled clinical trial. *Hepatology*. 2013;**57(4):1542–1549.**

70. Chalumeau M, Duijvestijn YC. Acetylcysteine and carbocysteine for acute upper and lower respiratory tract infections in paediatric patients without chronic broncho-pulmonary disease. *Cochrane Database Syst Rev*. 2013;**5**:CD003124.

Macronutrient Requirements for Growth: Fats and Fatty Acids

7

Kathrin Krohn, MD, Hans Demmelmair, PhD, and Berthold Koletzko, MD, PhD

INTRODUCTION

An adequate fat intake is essential for growth and development of infants and children. Dietary fat provides a large portion of energy and is the source of lipid soluble vitamins and essential polyunsaturated fatty acids. The composition of dietary fat influences lipid absorption, metabolism and tissue deposition, and thereby growth and body composition.

Fats or lipids can be defined as substances that have no or very limited solubility in water, but are soluble in organic solvents (e.g., chloroform, hexane). Most of our dietary fats are triglycerides containing three fatty acids esterified to a glycerol backbone. Vegetable oils, for example, consist almost entirely of triacylglycerols. Physicochemical properties of triacylglycerols (e.g., the melting point) are determined by the composition of its fatty acids. Watery foods may contain triacylglycerols either as a separate lipid layer (e.g., lipid droplets on a clear soup) or as an emulsion (e.g., milk or intravenous lipid emulsion). Emulsification is facilitated by amphipathic lipids that contain both hydrophobic parts, which associate with neutral lipids, as well as hydrophilic components, which interact with polar molecules such as water. Amphipathic lipids include phospholipids, monoglycerides and unesterified cholesterol and plant sterols, and lipid soluble vitamins bound to hydrophilic groups such as vitamin E acetate (tocopherol acetate). In contrast, cholesterol esters (esterified with a long-

chain fatty acid) and vitamin A esters such as retinol palmitate are non polar.

FAT DIGESTION

Fat digestion and absorption involve

i. intraluminal solubilization and hydrolysis of lipids,
ii. the subsequent intestinal absorption of hydrolyzed fatty acids and monoacylglycerols, re-esterification of fatty acids and secretion into the lymphatics in chylomicrons, or release of unesterified fatty acids into the portal venous system, and
iii. uptake of unesterified fatty acids or chylomicron triacylglycerols into the tissues.

Triacylglycerols represent the major portion of dietary lipids and are hydrolyzed in the gut by intestinal lipases. The digestion process begins in the stomach and continues throughout the intestinal tract. The major proportion of fat is hydrolyzed in the duodenum, but initial partial hydrolysis in the stomach may be critically important to increase efficiency of pancreatic lipase.[1,2] About 10%–30% of dietary fat is hydrolyzed in the stomach.[3] The rate of hydrolysis may be influenced by the initial droplet size in the stomach. A fine emulsion results in a higher degree of lipolysis compared to a coarse mixed emulsion.[4] This may not have major consequences for normal digestion since the capacity for hydrolysis is high, but it may become important in fat malassimilation, for example, due to exocrine pancreatic insufficiency and with tube feeding.[3]

Gastric lipase has an acidic pH optimum. When exposed to the pancreatic proteases (trypsin, chymotrypsin) it is rapidly inactivated. In healthy individuals, gastric lipase may not be important for intraduodenal fat digestion. However, in patients with pancreatic insufficiency, such as cystic fibrosis patients, the low intraluminal concentrations of pancreatic proteases and acidic pH may favor its function.[5]

Gastric lipase has a functional specificity for the sn-3 ester bond of triacylglycerols and gives rise to free fatty acids and sn-1,2 diglycerides. Monoglycerides and free fatty acids enter the duodenum and stimulate the release of cholecystokinin in the duodenum. This in turn stimulates the release of biliary secretions and pancreatic enzymes. Digestion products are emulsified by bile salts, thereby increasing the available surface. Free fatty acids, which are released by gastric lipase, induce the binding between the colipase-lipase complex and fat globules.[6] Colipase-

dependent lipase has a specificity for sn-1,3 ester bonds to give rise to sn-2 monoglycerides and free fatty acids. The level of enzyme secreted into the intestinal lumen changes with age. The level of colipase-dependent lipase secreted in adults is estimated to be 1000-fold in excess of what would be necessary to hydrolyze the dietary fat ingested daily. However, in neonates postprandial luminal levels are much lower, since the pancreas is not fully developed after birth.[5] In addition to a low activity of colipase-dependant pancreatic lipase, neonates have a limited bile salt pool, and low intraluminal bile salt concentrations.

In infants fed human milk, hydrolysis of triacylglycerols results from the combined action of bile salt stimulated lipase (BSSL), which is produced in the mammary gland and secreted into the milk and the endogenous intestinal lipases. BSSL hydrolysis triacylglycerols at all three ester bonds resulting in the formation of free fatty acids and glycerol[3] and is present in milk from about 26 weeks of gestation.[6,7] The considerable contribution of BSSL to the lipolytic activity in breast milk is eliminated by pasteurization which inactivates BSSL.[8] This effect on fat absorption is clinically relevant in preterm infants. A study in preterm infants born with a gestational age below 32 weeks showed significantly higher weight gain with raw human milk as compared to pasteurized milk.[9] The addition of recombinant BSSL to pasteurized human milk is currently evaluated in preterm infants.[8]

FAT ABSORPTION

Unsaturated fatty acids are more readily absorbed than saturated fatty acids.[10] The preferential absorption of unsaturated fatty acids may be facilitated through better solubilization by bile salts, greater gastric lipolysis, preferential binding to intracellular fatty acid binding protein and subsequent re-esterification or a decreased tendency to form soaps.[10,11]

Fat absorption is modulated by the chain length and degree of unsaturation of the fatty acids. High proportions of monounsaturated oleic acid (18:1) and polyunsaturated linoleic acid (18:2n-6) from vegetable oil triacylglycerols are absorbed (>90%).[12] In contrast, absorption of the saturated fatty acids myristic acid (14:0), palmitic acid (16:0), and stearic acid (18:0), is lower and decreases with increasing fatty acid chain length.

Absorption of saturated fatty acids depends on the position of the fatty acid in the triacylglycerol molecule.[13] In human milk, palmitic acid is the predominating saturated fatty acid. Most human milk palmitic acid is

present at the sn-2 position of the triacylglycerol molecule.[14] Formula milks contain smaller quantities of palmitic acid compared to human milk, but the saturated fatty acids from the originating vegetable oils are largely esterified at the outer sn-1,3 positions of the triacylglycerol molecule.[15] The lipolytic enzymes gastric and colipase-dependent lipases cleave fatty acids preferentially in sn-1- and sn-3-position (Figure 7-1). Whereas 2-monoglyceride of palmitic acid is readily absorbed, free palmitic acid and palmitic acid in the sn-1 and sn-3 positions of monoglycerides are not.[16] Therefore, infants fed formula milks with palmitic acid in these positions may be more likely to excrete more soaps and pass firmer stools than breast fed infants.[17] Soaps result from binding of free fatty acids released by lipolysis of triacylglycerols from the sn-1 and sn-3 positions and calcium, resulting in reduced fat and mineral absorption.[18] However, the difference in fat absorption may easily be compensated for by the infants' ability to adapt the amount of milk consumed daily.[19] Structured lipids with palmitic acid enzymatically esterified into the beta (sn-2) position have been used in formulas for term and particularly preterm infants with the key aim to soften stools.

Monoacylglycerols with a fatty acid in the sn-2 position are well absorbed, and fatty acids are preserved in this position during the absorption process.[20] In contrast, fatty acids originating from the sn-1,3 positions are randomly re-esterified during triacylglycerol re-synthesis via the monoacylglycerol pathway. Following reassembly, the triacylglycerols are secreted as components of chylomicrons.

Medium chain triacylglycerols (MCTs) contain predominantly saturated fatty acids with 8 and 10 carbon atoms. MCTs have high water solubility, are rapidly cleaved by lipases and are readily absorbed even in the presence of low amounts of intraluminal bile salts and pancreatic lipases.[21] MCTs are used in low birth weight infant (LBWI) formulas to increase fat absorption. Preterm infant formulas in the USA contain up to 50% and in Europe up to 40% of total fat as MCT. Proposed advantages of MCT when compared to long chain triacylglycerols (LCT) with saturated fatty acids in LBWI formulas include rapid, portal venous transport of the albumin-bound medium chain fatty acids (MCFAs) to the liver, the carnitine-independent transport into the mitochondria, and subsequent oxidation that is more rapid than for longer chain fatty acids. However, the energy content of MCT is about 16% lower than that of LCT (Figure 7-2). Therefore, a benefit of providing MCT rather than LCT with respect to increased energy absorption and supply can only be expected in subjects

with a very severe degree of fat malassimilation, such as children with severe cholestasis, whereas most patients will not achieve enhanced energy absorption with the use of MCT. Indeed, a Cochrane review did not demonstrate an advantage of preterm infant formulas containing high amounts of MCT (40% or more) versus formulas with low amounts of MCT (20% or less) with respect to weight gain achieved.[22] In patients with short bowel syndrome, MCTs are used to improve fat absorption.[23] In adult short bowel patients the colon as been suggested as a digestive organ for medium chain fat, probably absorbed as MCFA.[24] However, the available data on clinical effects achieved are limited. Since MCFAs are preferentially oxidized, their provision spares polyunsaturated fatty acids (PUFAs) from oxidation. Randomized trials in preterm infants demonstrated that enteral and parenteral MCFA supply enhanced PUFA availability.[25,26]

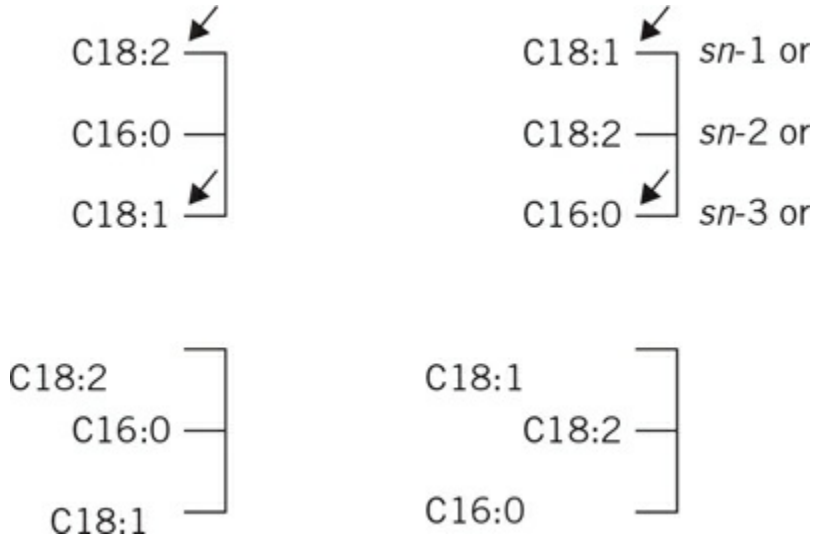


Figure 7-1 Intestinal lipases preferentailly hydrolyse triglyceride fatty acids in the sn-1 and sn-3 positions. Human milk lipids, lard, and some specifically structured lipids contain the saturated palmitic acid (C16:0) predominantly in the sn-2 position (left panel). Hydrolysis of these substrates produces most C16:0 as a monoglyceride, which is highly water soluble and absorbed to a large extent. In contrast, saturated fatty acids in vegetable oils are more or less randomly esterified to the different positions of the molecule, and a greater proportion of C16:0 is liberated as a hydrophobic free fatty acid, which tends to bind to cations (soap formation), resulting in poor absorption of both fat and calcium. Benficial effects of structured lipids with saturated fatty acids in the sn-2

position for both fat and mineral absorption have been shown in infants.

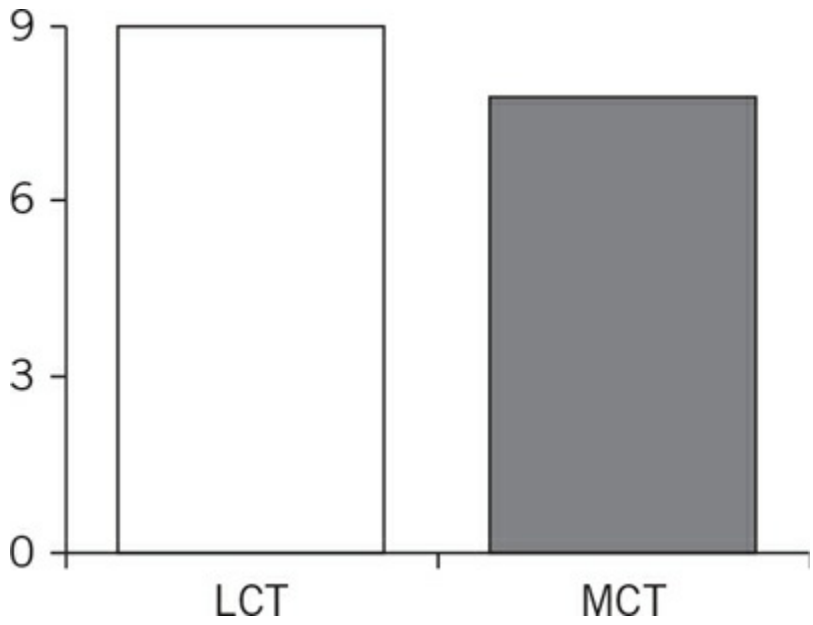


Figure 7-2 The energy content (kcal/g fat) of medium chain triacylglycerols (MCTs) is about 16% lower than that of long-chain triacylglycerols (LCTs). Therefore, an increased energy absorption by the use of MCT can only be expected in subjects with very severe fat malassimilation, such as children with severe cholestasis.

TRANSPORT OF LIPIDS

Some lipids are transported predominantly via the portal vein, whereas others are preferentially transported via the lymphatic system. The route of transport is generally related to chain length, the level of unsaturation and the structure of the lipid. The chain length is the most important factor affecting portal transport of fatty acids.[3]

MCT may be incorporated directly into mucosal cells and hydrolyzed. Liberated MCFA are then transported bound to albumin predominantly via the portal system. However, the separation of MCFA to be transported as unesterified, albumin-bound fatty acids via the portal venous route, and of fatty acids of carbon chain length 14 and longer to be transported in chylomicron triacylglycerols via the lymphatic system, is not complete. Physiologically significant amounts of saturated and unsaturated fatty acids, including linoleic acid (18:2n-6) and alpha-linolenic acid (18:3n-3),

are transported via the portal pathway,[27] while MCFAs are found in chylomicron triacylglycerols.

ASSEMBLY OF LIPOPROTEINS

In the enterocyte, absorbed lipids are packed into lipoproteins, which enable transport in the aqueous environment of blood plasma. The major lipoproteins secreted by the intestine are chylomicrons and very low-density lipoprotein (VLDL). Chylomicrons contain triacylglycerols, cholesterol ester, free cholesterol and phospholipids.[3] They are secreted in the small intestinal epithelium after a meal to transport absorbed lipids.[28] VLDL-sized particles are formed in intestinal cells during fasting.[3] The intestinal lipoproteins are secreted into the lymphatic system and enter the bloodstream via the thoracic duct.

Triacylglycerol constituents of intestinal chylomicrons and hepatic VLDL are hydrolyzed by lipoprotein lipase released from adipose tissue, muscle, and possibly other tissues to the capillary endothelium.

TOTAL DIETARY FAT INTAKE IN INFANTS

Dietary fat is essential for growth and development. It provides a source of energy that covers the needs for energy expenditure, fat oxidation and fat deposition during growth. However, there is limited evidence on acceptable ranges of dietary fat intake in infants and current intake recommendations are guided be the fat intake with human milk.[29,30]

With an energy content of about 9 kcal/kg, LCT have more than twice the energy content per unit weight than carbohydrates or protein. Growing infants have a very high energy requirement estimated to be about 120 kcal/kg per day during the first month of life and close to 100 kcal/kg per day during the rest of the first year of life. The tissue deposition of fat in healthy infants is very high: about 90% of the total energy deposited in growing tissues during the first six months of life consists of fat.[31] To achieve this high fat deposition, about 25% of dietary energy supply is deposited as fat during the first two months of life, but the proportion rapidly decreases with the slower growth rate to less than 5% after six months of life.[32]

While lipids can be synthesized *de novo* from carbohydrates or proteins,[33] endogenous net lipogenesis has a limited capacity and forms only non-essential fatty acids. Considerable amounts of energy are lost in

the process, because the synthesis of fatty acids from glucose consumes about 25% of the energy content of glucose invested. In contrast, fat deposition from dietary fat requires only about 1%–4% of the energy provided by the fat.[34]

In studies supplying isoenergetic diets with different fat contents, groups receiving higher fat intakes achieved a higher body weight and fat gain at a lower energy expenditure.[33,34]

The Committee on Nutrition of the European Society for Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) recommended a total fat intake of about 40% to 50%–55% of energy intake for young infants.[35] Similarly, global expert committees concluded that fat supply with infant and follow-up formulae should be in the range of 4.4–6.0 g/100 kcal which is equivalent to about 40%–54% of energy content and similar to values found typically in human milk.[29,30,36] Energy needs per kg bodyweight and the proportion of dietary energy deposited as fat decrease rapidly in the first months of life.[32] Therefore, in the second half of the first year lower dietary fat intakes can meet the needs for growth. In a randomized intervention study in Finland, infants receiving a mean dietary fat intake of about 28% of energy from the age of 6 months onwards showed normal growth and long term development.[37]

DIETARY FAT INTAKE IN CHILDREN

Similar to infants, also for children there is no firm evidence on which to set lower and upper limits of advisable lipid intakes. Both excessive and low supply of dietary lipids may be associated with adverse effects.

Concentrations of plasma total and LDL-cholesterol increase with high intakes of saturated and *trans* unsaturated fatty acids,[38] which is considered a risk factor for vascular lipid deposition and the occurrence of early vascular lesions.[39] Consequently, low intakes of saturated and *trans* unsaturated fatty acids and their replacement by unsaturated fats are recommended to reduce the long term risk of heart disease.[40] It has been recommended that saturated and *trans* fatty acid intake should not exceed 10% and 1% of energy intake, respectively.[41] While there is some concern that the advice to markedly lower fat intake, for example as part of dietary treatment of hypercholesterolemia, might cause adverse effects on child growth, observational studies did not report adverse effects of low fat diets on growth and biochemical measures of nutritional status.[42,43] A study in 663 children with hypercholesterolemia comparing reduced fat intake in

the intervention group (28.6%) to usual care (33%) did not demonstrate a correlation of percentage of dietary energy intake as fat to weight, height, body mass index or skinfold thickness.[44] However, other studies reported impaired growth in children with hypercholesterolemia treated with a low fat diet.[45] Modification of the quality of dietary fat intake rather than a reduction of the quantity of fat intake, which has beneficial effects on plasma lipoprotein concentrations, is possible without any adverse effects on child growth.[46]

It is unclear whether the adverse effects on growth and weight gain observed in some studies with a reduced fat intake are attributable to a low energy intake rather than lower fat intake per se. Diets with a low fat intake tend to have a lower energy density, which may reduce total energy intake and thereby reduce weight gain and eventually length gain.

It is controversial to which extent fat reduced diets may be associated with biologically significant reductions of the intakes of lipid soluble vitamins and micronutrients. Some studies showed low intakes of vitamins and micronutrients, accompanied by lower energy intake,[42,47] whereas others did not find a compromised intake of these nutrients associated with reduced fat intake.[43,48]

FATTY ACIDS

The fatty acids esterified into lipids comprise the major component of fats in the diet and in the body, and they play a key role in modulating the physicochemical, metabolic and functional properties of lipids. Fatty acids are conventionally classified according to their chain length as short chain fatty acids (chain length <8 carbon atoms), medium (8–11 carbons), intermediate (12–15 carbons), and long chain fatty acids (>16 carbons). Short formulas are used to describe fatty acids, which indicate the number of carbon atoms and double bonds as well as the position of the terminal double bond (Table 7-1). The chain length of a fatty acid is the major determinant of melting point, energy content (kcal/g), the degree of solubility in polar (e.g., water) and non-polar solvents (e.g., hexane) and hence the efficacy of absorption in relation to the availability of conjugated bile salts in the intestinal lumen, as well as metabolic properties such as effects on lipoprotein metabolism, oxidative degradation and tissue fat deposition. Fatty acid properties also depend on the number, type and position of the double bonds of which there may be none (saturated fatty acids), one (monounsaturated) or two or more (polyunsaturated) (Figure 7-3).

Unsaturated fatty acids are widely classified according to the number of carbon atoms between the methyl end of the acyl chain and the first double bond from this end of the chain, that is the omega end of the fatty acid (Figure 7-3). In human metabolism, enzyme-mediated biochemical reactions, such as chain elongation, chain shortening by β-oxidation or the introduction of new double bonds occur at the carboxyl end of the molecule. In contrast, the omega end of a fatty acid is usually unchanged in intermediary metabolism, unless the fatty acid undergoes complete β-oxidation. Since humans and other higher organisms cannot introduce double bonds in the n-6 or n-3 positions, n-6 PUFA (e.g., linoleic acid) and n-3 PUFA (e.g., α-linolenic acid) are essential nutrients that must be provided with the enteral or parenteral substrate supply.

TABLE 7-1 Biologically Important Fatty Acids

Name	Short formula
Saturated Fatty Acids	
Acetic acid	C2:0
Propionic acid	C3:0
Butyric acid	C4:0
Caproic acid	C6:0
Caprylic acid	C8:0
Capric acid	C10:0
Lauric acid	C12:0
Myristic acid	C14:0
Pentadecanoic acid	C15:0
Palmitic acid	C16:0
Margaric acid	C17:0
Stearic acid	C18:0
Arachidic acid	C20:0
Behenic acid	C22:0
Lignoceric acid	C24:0
***cis*-Monounsaturated Fatty Acids**	
Palmitoleic acid	C16:1n-7
Oleic acid	C18:1n-9
Vaccenic acid	C18:1n-11
Essential Fatty Acids	
Linoleic acid (LA)	C18:2n-6
α-Linolenic acid (ALA)	C18:3n-3

γ-Linolenic acid	C18:3n-6
Dihomo-γ-linolenic acid	C20:3n-6
Mead acid	C20:3n-9
Arachidonic acid (AA)	C20:4n-6
Eicosapentaenoic acid	C20:5n-3
Docosahexaenoic acid (DHA), Cervonic acid	C22:6n-3
***trans*-Fatty Acids**	
Elaidic acid	C18:1n-9t
trans-vaccenic acid	C18:1n-11t
Linolelaidic acid	C18:2n-6t
Conjugated linoleic acids (CLA)	C18:2-9c11t, C18:2-10t,12c, and others

Short formulas are used to describe the number of carbon (C) atoms, the number of double bonds, the omega-position of the last double bond (n-x) and the double bond configuration in *cis* (c) or *trans* (t) position.

There are two isomeric forms of double bonds, *cis* and *trans* (Figure 7-3). Plant and mammalian cells introduce almost entirely *cis*-configurated double bonds into fatty acids, in which both adjoining carbon chains point in the same direction of the double bond and, therefore, the molecule is bent and becomes more flexible and fluid. *Trans*-isomeric fatty acids are formed primarily by rumen bacteria (in the forestomach of cows, goats, and sheep) and during technical fat hydrogenation.[49] *Trans*-fatty acids have a straight carbon chain, with a geometry similar to saturated fatty acids (Figure 7-3), and they have higher melting points and less fluidity at body temperature than their respective *cis*-isomers. *Trans*-fatty acids may exert adverse effects on lipoprotein and essential fatty acid metabolism as well as fetal growth.[46,50,51] Therefore, the intake of dietary *trans* fatty acids from foods produced with partially hydrogenated fats, French fries and other deep fried foods, baked goods with hydrogenated fats and other foods high in *trans* fatty acids should be limited.

Positional and *trans* isomers of linoleic acid in which the double bonds are conjugated are called conjugated linoleic acids (CLAs). CLAs are formed primarily by biohydrogenation in ruminants. CLAs in the Western diet are predominantly supplied with cows' milk and cheese. CLA contents in bovine milk are in the order of about 1%–2% of total fatty acids, with higher amounts resulting from barn feeding in the winter than from pasture feeding in the summer. Human milk contents depend on maternal dietary intake primarily from dairy products and reach a mean of

about 0.4% CLA in human milk, with considerable intra- and interindividual variation.[52] CLA was found to be a high affinity ligand and activator of the peroxisome proliferator-activated receptor alpha and appears to have marked effects on lipid and energy metabolism. Studies in different species of experimental animals show that the consumption of CLA leads to a reduced energy intake, increased metabolic rate, and loss of fat and total body weight. In addition, these studies show that CLA also reduces the plasma concentrations of total and LDL cholesterol, has an anti-inflammatory effect, and can inhibit the development of mammary, skin and prostate cancers. While mixtures of CLA isomers as supplements or CLA-enriched products are widely marketed for human consumption, only a few studies in healthy humans with relatively small sample sizes have been published so far. A recent systematic review concluded that there is not enough evidence yet to show that CLA has an effect on weight and body composition in humans, while some of these studies have observed that the administration of various CLA isomers has adverse effects on lipid profile with a decrease of high-density lipoprotein (HDL) cholesterol concentration and increased Lp(a) levels, glucose metabolism, lipid oxidation, inflammation, or endothelial function.[53] Therefore, long-term randomized clinical trials with placebo controls with sufficient power to evaluate the efficacy and safety of CLA isomers in humans are needed before its use in human beings can be recommended.

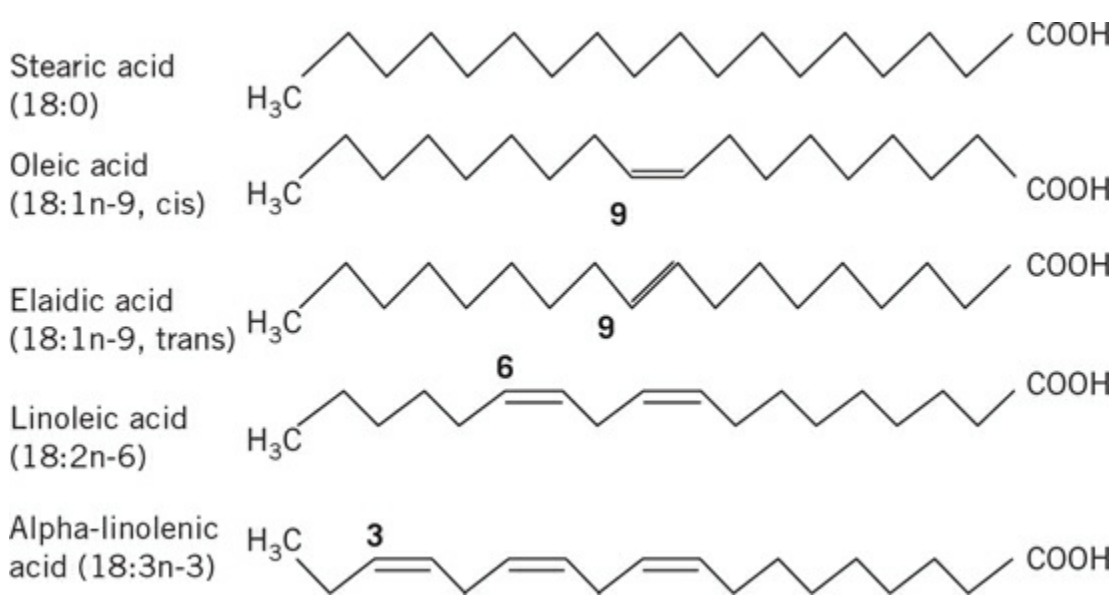


Figure 7-3 Structural formula of the saturated fatty acid stearic acid (18.0), the *cis*-monounsaturated fatty acid oleic acid (18:1n-9, one

double bond located 9 carbon atoms from the methyl end of the molecule), the *trans*-monounsaturated fatty acid elaidic acid (18:1n-9, *trans*, one *trans*-double bond located 9 carbon atoms from the methyl end of the molecule), the n-6 polyunsaturated fatty acid linoleic acid (C18:2n-6; n-6 fatty acid with the terminal double bond located 6 carbon atoms from the methyl end of the molecule), and the n-3 polyunsaturated fatty acid α-linolenic acid (18:3n-3; n-3 fatty acid with the terminal double bond 3 carbon atoms from the methyl end).

ESSENTIAL FATTY ACIDS

Saturated fatty acids, and unsaturated fatty acids of the n-9 and n-7 series can be synthesized *de novo* from acetyl CoA. In contrast, humans cannot introduce double bonds in the n-6 or n-3 positions, therefore, n-6 PUFAs (linoleic acid and metabolites) and n-3 PUFAs (α-linolenic acid and metabolites) are essential nutrients that must be provided with enteral or parenteral substrate supply. An inadequate dietary supply of linoleic acid induces a specific deficiency syndrome with a dry, scaly dermatitis and a variety of other signs and symptoms (Table 7-2). Linoleic acid deficiency has been observed mostly in infants, and a few adult patients with glucose-based, lipid-free parenteral nutrition. Infants are at a far higher risk of deficiency because of higher requirements for polyunsaturated fatty acids due to their rapid growth, while their body stores are very limited. Deficiency of n-3 fatty acids both in animals and in a limited number of human subjects on long-term tube feeding without adequate n-3 PUFA supply was reported to induce alterations of visual and neural functions.

The n-6 and n-3 fatty acids are not interconvertible, and they have different functions. Conversion of the dietary precursor essential fatty acids linoleic acid (18:2n-6) and α-linolenic acid (18:3n-3) occurs by alternating desaturation (insertion of double bonds) and elongation (addition of two carbon units) of the carbon chain (Figure 7-4) to highly unsaturated, long-chain polyunsaturated fatty acids (LC-PUFAs) with 20 and 22 carbon atoms and 3–6 double bonds. The LC-PUFA arachidonic acid (20:4n-6; AA) and docosahexaenoic acid (22:6n-3; DHA) are of particular importance for pregnancy outcomes, infant growth and development.[54]

TABLE 7-2 Signs and Symptoms of Essential Fatty Acid Deficiency

Scaly, hyperkeratotic dermatitis
Loss of hair
Growth faltering
Reduced utilization of dietary energy
Reduced nitrogen retention
Increased susceptibility to infection
Disturbed wound healing
Fatty liver, hepatomegaly, increased levels of ALAT & ASAT
Anemia
Thrombopenia, reduced platelet aggregation

PERINATAL SUPPLY OF LC-PUFA

There is accumulating evidence for an important functional role of the LC-PUFA supply and metabolism in nervous system development and function.[55] LC-PUFAs, especially AA (22:4 n-6) and DHA (22:6 n-3), are preferentially deposited in relatively high concentrations in developing neural cells during the perinatal brain growth spurt. In humans, most brain LC-PUFAs are accumulated during the phase of rapid brain growth in the last trimester of gestation and the first 2 years after birth. The importance of brain DHA accumulation mainly relates to its roles in maintaining neurological and visual development; decreases in brain and retina DHA result in altered visual acuity, disturbed electroretinographic measurements and learning impairment in various species. AA is a major component of structural phospholipids and serves as precursor for eicosanoids, which play important roles in cell division, signal transduction and many other physiological processes. The pathways to form DHA from the precursor essential fatty acid alpha-linolenic acid exist in humans (Figure 7-4), and the formation of LC-PUFA from the essential fatty acid precursors has been demonstrated even in newborn infants,[56] but the rate of essential fatty acid conversion is low.[57] The relative conversion of alpha-linolenic acid to n-3 LC-PUFA appears to be greater in women than in men, which may contribute to meeting the demands of the fetus and the breast fed neonate for DHA and hence represent a protective mechanism. In non-human primates, the supply of preformed dietary DHA is about an order of magnitude more efficient as a source for the neonatal brain accretion of DHA than is alpha-linolenic acid.[58] Therefore, adequate supplies of preformed n-3 LC-PUFA, and in particular DHA, during perinatal

development appear important for maintaining optimal tissue function.[59]

GENETIC VARIATION, ESSENTIAL FATTY ACID METABOLISM, AND HEALTH CONSEQUENCES

Recent data indicate a considerable inter-individual variation in the ability to convert the precursor alpha-linolenic acid to n-3 LC-PUFA, related to common polymorphisms in the human genes for the rate-limiting enzymes in this pathway delta-5 and delta-6 desaturases, that is the genes FADS1 and FADS2.[60] In 2006, the first genetic association study of the fatty acid composition in serum phospholipids revealed significant associations of SNPs in the *FADS* genes with several omega-6 and omega-3 PUFA.[61] Carriers of the minor alleles of the investigated SNPs showed higher concentrations of the desaturase substrates and lower concentrations of the desaturase products compared to major allele carriers. This led to the hypothesis that minor allele carriers have less ability to endogenously convert the precursor fatty acids to their longer-chain products. Several other (candidate-gene and genome-wide) studies involving populations of European, Asian, and African descent and including also other tissues such as erythrocyte membrane phospholipids, adipose tissue, and breast milk replicated the findings by Schaeffer et al.[61] Moreover, several genome-wide association studies on complex lipid traits reported associations of *FADS* polymorphisms with serum phosphatidylcholines, and the blood lipid parameters low-density lipoprotein (LDL), HDL, total cholesterol, and triacylglycerols.[60]

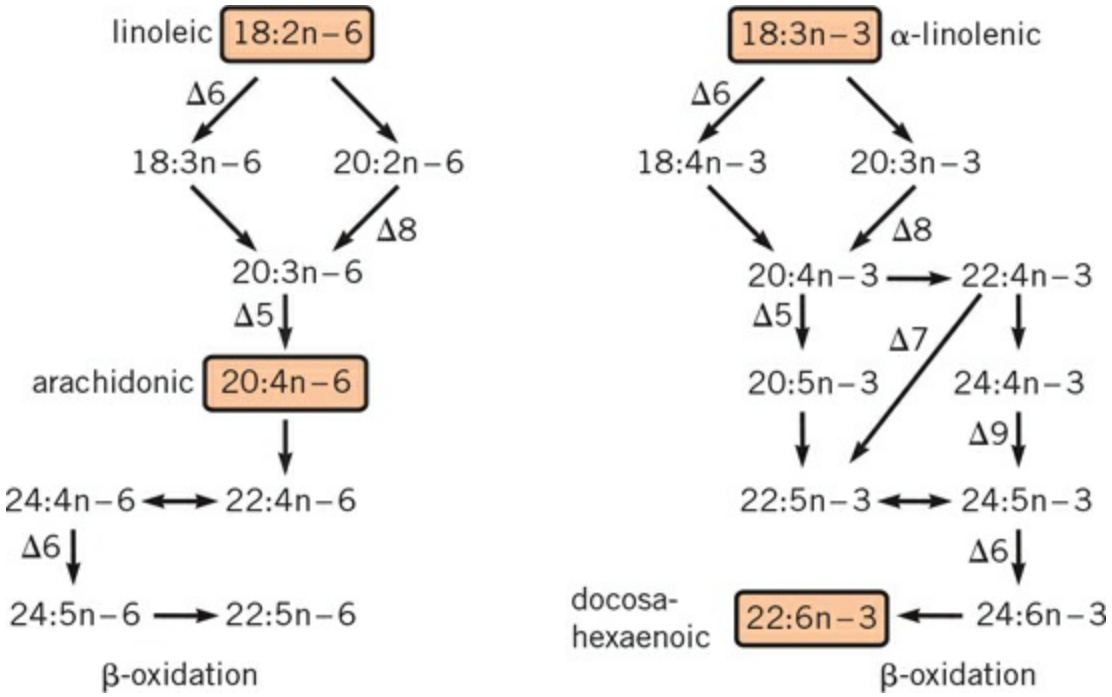


Figure 7-4 Pathways for the enzymatic conversion of the essential fatty acids linoleic acid (18:2n – 6) and α-linolenic acid (18:3n – 3) to long-chain polyunsaturated fatty acids (LC-PUFA).

FADS SNPs have also been associated with health endpoints including cardiovascular disease, type 2 diabetes mellitus, childhood eczema, and attention-deficit/hyperactivity and bipolar disorder.[60] Steer et al. found a strong modifying effect of genetic variation in the *FADS* SNPs on breastfeeding effects on later IQ in the Avon Longitudinal Study of Parents and Children.[62] There was an overall benefit from having been breastfed on full-scale IQ at 8 years, with higher mean values of eight points (unadjusted) or three points (adjusted) in previously breastfed compared to non-breastfed children. However, children homozygous for the minor allele had the lowest IQ scores when not having been breastfed (4.3 IQ points below not breastfed carriers of the major allele), but showed the highest benefit when having been breastfed (thereby reaching similar scores as breastfed major allele carriers) (*P*-value for interaction = .0091). Similar results were obtained in two Spanish birth cohorts.[63] These results suggest that breastfeeding is beneficial for cognitive development, but the effect size depends on the genetically determined variation in PUFA metabolism: children who have a limited ability to convert linoleic and alpha-linolenic acids to LC-PUFA have a greater benefit from

breastfeeding which provides preformed LC-PUFA. The data also provide evidence for a causal role of LC-PUFA availability of brain function.

We wish to emphasize that the polymorphisms referred to here are very frequent in the general population, with a minor allele frequency of around 30% and higher in Europeans. Thus, these nutrigenetic effects have a large public health importance.

PLACENTAL FATTY ACID TRANSFER

Numerous studies reported significant differences between the fatty acid composition of maternal plasma lipids at the time of delivery and cord plasma lipids. Percentages of saturated and monounsaturated fatty acids are very similar between maternal plasma phospholipids and cord blood phospholipids, but in cord blood the parent essential fatty acid levels are significantly lower whereas the LC-PUFA metabolites are clearly higher.[64] These observations point to a preferential transfer of LC-PUFA by the placenta.[65] Using stable isotope labeled fatty acids we investigated the placental transfer in vivo in humans.[66–68] Palmitic-, oleic-, linoleic- and docosahexaenoic acids labeled with the natural carbon variant ^{13}C were administrated orally 12 hours before elective caesarean section to pregnant women. Tracer concentration was analyzed in maternal blood samples until delivery, in cord blood and in placental tissue. The ratios between tracer concentrations in placental tissue or cord plasma, respectively, and in maternal plasma were similar to the saturated palmitic acid, the monounsaturated oleic acid, and the parent essential fatty acid linoleic acid, but were clearly higher for DHA pointing towards a preferential DHA transfer across the placenta (Figure 7-5). It can be deduced that incorporation of DHA into placental tissue and transfer of DHA across the placenta is more than three times as intense as of the other studied fatty acids.

The placental transfer of fatty acids is a complex process. Fatty acids bind to membrane proteins and cytoplasmatic transport proteins.[65,69] The placenta takes up circulating non-esterified fatty acids (NEFAs) as well as NEFA released from maternal circulating lipids by placental lipases and in addition human placental tissue express receptors to VLDL, LDL, and HDL lipoproteins,[70] which can provide fatty acids to the placenta for transfer once they are released by intracellular lipases. Nonesterified fatty acids may enter placental tissue both through passive diffusion, and by a carrier protein mediated transport (Figure 7-6). This facilitated transport

mechanism seems to involve plasma membrane fatty acid binding protein (FABPpm/GOT2). fatty acid translocase (FAT/CD36), fatty acid transport proteins (FATPs), and fatty acid binding proteins (FABPs).[71] It was proposed that a further fatty acid binding protein located exclusively on the maternal-facing membranes, placental plasma membrane fatty acid-binding protein (p-FABPpm), is also involved in the preferential uptake of LC-PUFA by trophoplast cells,[72] but definitive evidence on the possible structure and function of p-FABPpm must await analysis of its complete amino acid and/or cDNA sequence. Several FABPs are involved in the fatty acid transfer to the fetal circulation, for example heart-FABP (H-FABP), liver-FABP (L-FABP), and adipose tissue (A-FABP) have been identified in placenta.[73] The expression of the fatty acid transfer proteins FATP-1 and FATP-4 are correlated with placenta phospholipid DHA contents, which seems to indicate that they play a key role in facilitating preferential DHA transport from the mother to the fetus.[71] The presence of such an active and preferential materno-fetal DHA supports the hypothesis that fetal DHA supply is of biological importance.

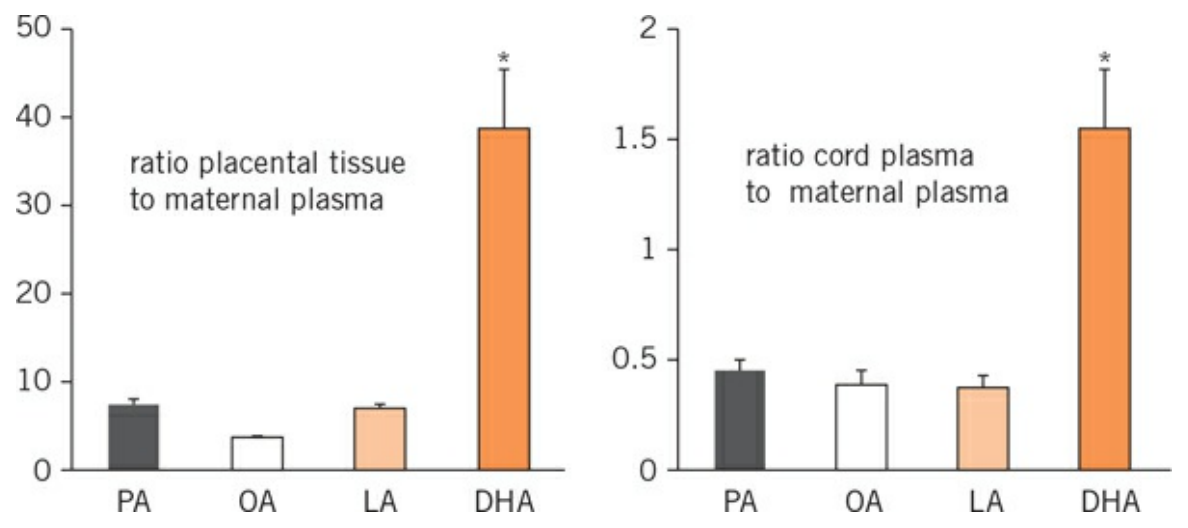


Figure 7-5 Ratios (%, *M*+SEM) between placental or cord plasma, respectively, and maternal plasma concentrations of studied ^{13}C-fatty acids. PA, palmitic acid; OA, oleic acid; LA, linoleic acid; DHA, doscosahexaenoic acid. $^{*}P<.05$ *post hoc* Bonferroni. Modified from Gil-Sanchez et al.[65]

DHA SUPPLY TO PREGNANT AND BREASTFEEDING WOMEN

Enhanced maternal dietary intakes of DHA increase fetal supply and lead to higher DHA concentrations in cord blood.[74] The effects of supplementing pregnant women with n-3 LC-PUFA from fish oil or DHA rich single cell oils on pregnancy outcomes have been evaluated in a number of randomized controlled clinical trials, which provided daily DHA intakes of 150–200 mg up to about 1200 mg/day, or up to 2.7 g total n-3 LC-PUFA/day. Systematic evaluation of these studies in recent meta-analyses revealed that n-3 LC-PUFA prolonged gestation by a mean of 1.6 or 2.6 days in two independent analyses, respectively, accompanied by a slight increase of birth weight by a mean 47 or 54 g, respectively.[75,76] Moreover, the supply of n-3 LC-PUFA reduced the risk of early preterm birth before 34 weeks of gestation by 31% in all pregnancies and by 61% in high-risk pregnancies.[54,76,77] Except for some reported discomfort associated with the intake of omega-3 oil capsules, such as belching and unpleasant taste, no adverse effects were detected up to the highest intake of 2.7 g total n-3 LC-PUFA/day tested in randomized controlled trials in pregnancy. A higher DHA supply to the fetus during pregnancy and to the infant after birth was associated with beneficial effects on the development of visual acuity, cognitive functions and attention, maturity of sleep patterns, spontaneous motor activity, immune phenotypes in a limited number of randomized clinical trials as well as in several prospective cohort studies.[59] Based on the information available, evidence-based consensus recommendations concluded that pregnant women should aim at achieving an average intake of at least 200–300 mg DHA/day.[59,59a] Supplementation of lactating women with 200 mg DHA/day increased human milk DHA content by about 0.2% of fatty acids to a level considered desirable for infant outcomes. Therefore, an average dietary intake of at least 200 mg DHA/day appears to be also adequate during lactation. An intake equivalent to 200 mg DHA/day is usually achieved by one to two meals of ocean fish per week, which includes fatty fish. This level of fish intake usually does not lead to an unacceptable daily intake of environmental pollutants.[59,78,79]

LC-PUFA SUPPLY TO INFANTS

Breastfed infants are provided with a dietary source of the LC-PUFA AA and DHA.[80] A systematic review of effects of LC-PUFA supplementation

of infant formulae on infant weight gain, based on original data analysis, indicates that the addition of DHA and AA to infant formula supports growth comparable to that seen in infants fed unsupplemented formula.[81] The available evidence indicated benefits of adding both DHA and AA to infant formula which has been supported by international expert recommendations.[30] DHA contents in infant formulas should reach at least 0.2%–0.3% of fatty acids, because lower contents have not been convincingly shown to provide benefits.[82] The amounts of added AA in infant formula should at least be equal to those of added DHA, and the amount of eicosapentaenoic acid added should not exceed the amount of added DHA. It appears advisable that dietary sources of DHA are also provided after the first six months of life because the effects of DHA status on indicators of neural function have been demonstrated in older infants and in children.[30] Recently, the European Food Safety Authority proposed that all infant formula should contain DHA levels of 20–50 mg/100 kcal (0.4–1% of fat), which are higher than the currently used DHA levels in term infant formula (near 0.3% of fat), but without the need to add AA.[83] This novel concept deviates from established European and global formula standards[84,85] that require AA contents in formula at least as high as DHA contents, which reflects the greater pre- and postnatal brain deposition of AA vs. DHA and the typically higher content of AA vs. DHA in human milk,[86] providing high amounts of n-3 LC-PUFA without AA-induced adverse effects on growth in preterm infants.[87,88] Concerns regarding adverse effects on development arise from the results of a double-blind, randomized trial in term infants fed formula either with no LC-PUFA or with DHA contents of 0.32, 0.64, and 0.96% at the same AA level of 0.64%, with developmental testing up to age 6 years.[89] Positive effects on word production, a card-sorting task, and an intelligence test were observed with the two lower DHA doses, but performance of children receiving 0.96% DHA with a reduced ratio of dietary AA to DHA was attenuated. Infant baboons fed the same formula showed reduced brain AA content with the formula providing 0.96% DHA but reduced dietary AA-to-DHA ratio.[90] Any major change in infant formula composition should be evaluated for nutritional adequacy and safety prior to market introduction.[91,92] Given the indications for adverse effects of providing formula with high levels of DHA without a concomitant increase in the provision of AA, it seems premature to accept the use of formula for infants from birth with the addition of 20–50 mg/100 kcal DHA without added AA in the absence of accountable data on the suitability and safety

from a thorough clinical evaluation of this novel approach.[86]

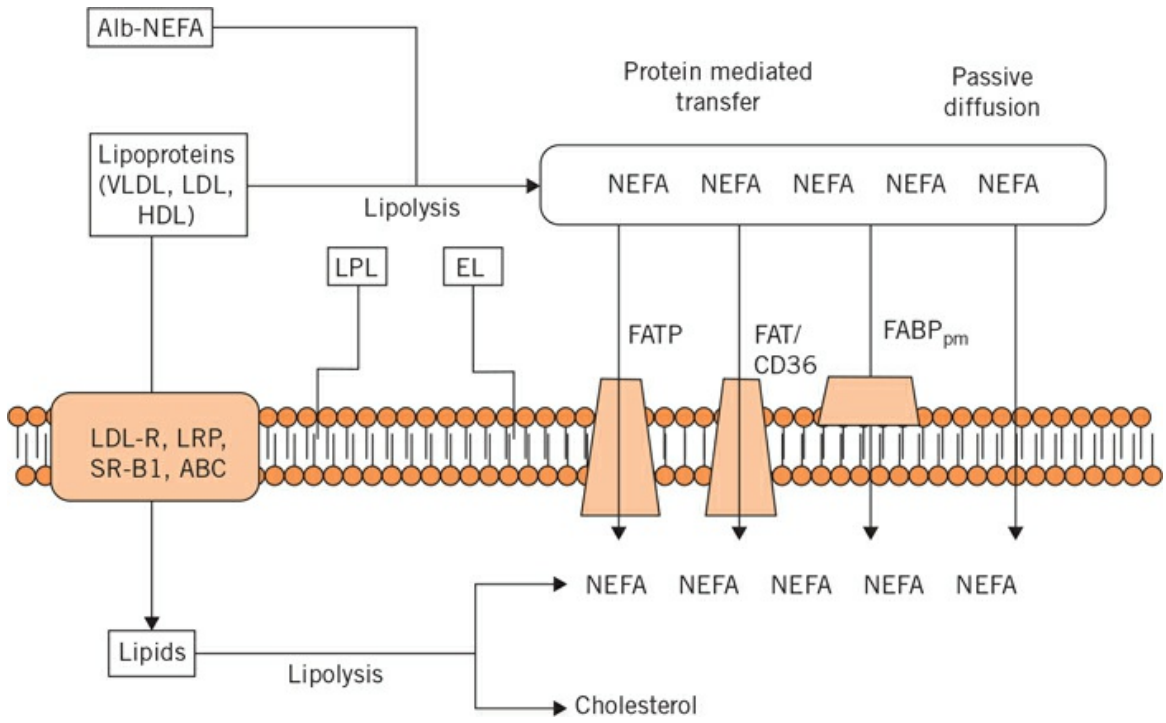


Figure 7-6 Mechanisms involved in the trophoblast fatty acid uptake. Intact lipoproteins can be taken up via lipoprotein receptors (LD-R), Scavenger receptor class B type 1 (SR-B1), lipoprotein related protein (LRP), ATP-binding cassette (ABC) transporters ABCA1 and ABCG1 or non-esterified fatty acids (NEFA) liberated by lipoprotein lipase (LPL) or endothelial lipase (EL). NEFA enter the cell by passive diffusion or carrier mediated. FATP, fatty acid transfer protein; FAT/CD36, fatty acid translocase: FABPpm, membrane bound fatty acid binding protein. Modified from Larque et al.[68]

CONCLUDING REMARKS

A large body of evidence has accumulated to show that the quantity and quality of dietary fat intake are of major importance for child growth and long-term health, including the modulation of risk of obesity, type 2 diabetes, and cardiovascular disease, visual, cognitive and neuromotor development, as well as immune response and the risk of allergy and autoimmune diseases. Enormous methodological progress enables future research to further explore lipid supply and metabolism as well as its effects at the molecular level, and to identify the effects of inter-individual

genetic variation in the relationship between fat intake and health outcomes. New scientific information should enable us to provide better advice on optimal lipid supplies to infants and children, both in health and disease.

ACKNOWLEDGMENTS

The work of the authors was financially supported by the Commission of the European Communities, specific RTD Programme “Quality of Life and Management of Living Resources”, within the 7th Framework Programme, contract no. FP7/2008-212652 (NUTRIMENTHE) and contract no. FP7/2007-13 (EarlyNutrition Project). This manuscript does not necessarily reflect the views of the Commission and in no way anticipates the future policy in this area. We also acknowledge financial support provided by the European Research Council, Advanced Grant META-GROWTH (grant agreement n° 322605). This manuscript does not necessarily reflect the views of the Commission and in no way anticipates the future policy in this area.

REFERENCES

1. Armand M, Hamosh M, Mehta NR, et al. Effect of human milk or formula on gastric function and fat digestion in the premature infant1. *Pediatr Res*. 1996;40(3):429–437.
2. Bernback S, Blackberg L, Hernell O. Fatty acids generated by gastric lipase promote human milk triacylglycerol digestion by pancreatic colipase-dependent lipase. *Biochim Biophys Acta*. 1989;1001(3):286–293.
3. Mu H, Hoy CE. The digestion of dietary triacylglycerols. *Prog Lipid Res*. 2004;43(2):105–133.
4. Armand M, Pasquier B, Andre M, et al. Digestion and absorption of 2 fat emulsions with different droplet sizes in the human digestive tract. *Am J Clin Nutr*. 1999;70(6):1096–1106.
5. Hernell O, Blackberg L. Molecular aspects of fat digestion in the newborn. *Acta Paediatr Suppl*. 1994;405:65–69.
6. Hernell O, Blackberg L, Bernback S. Digestion of human milk fat in early infancy. *Acta Paediatr Scand Suppl*. 1989;351:57–62.
7. Bernback S, Blackberg L, Hernell O. The complete digestion of human milk triacylglycerol in vitro requires gastric lipase, pancreatic colipase-dependent lipase, and bile salt-stimulated lipase. *J Clin Invest*. 1990;85(4):1221–1226.
8. Lindquist S, Hernell O. Lipid digestion and absorption in early life: an update. *Curr Opin Clin Nutr Metab Care*. 2010;13(3):314–320.
9. Montjaux-Regis N, Cristini C, Arnaud C, et al. Improved growth of preterm infants receiving mother’s own raw milk compared with pasteurized donor milk. *Acta Paediatr*. 2011;100(12):1548–1554.
10. Ling SC, Weaver LT. The fate of fat in the infant’s colon. *QJM*. 1997;90(9):553–

555.
11. Hamosh M, Bitman J, Liao TH, et al. Gastric lipolysis and fat absorption in preterm infants: effect of medium-chain triglyceride or long-chain triglyceride-containing formulas. *Pediatrics*. 1989;83(1):86–92.
12. Chappell JE, Clandinin MT, Kearney-Volpe C, et al. Fatty acid balance studies in premature infants fed human milk or formula: effect of calcium supplementation. *J Pediatr*. 1986;108(3):439–447.
13. Mattson FH, Volpenhein RA. Rearrangement of glyceride fatty acids during digestion and absorption. *J Biol Chem*. 1962;237:53–55.
14. Sauerwald TU, Demmelmair H, Koletzko B. Polyunsaturated fatty acid supply with human milk. *Lipids*. 2001;36(9):991–996.
15. Carnielli VP, Luijendijk IH, Van Goudoever JB, et al. Structural position and amount of palmitic acid in infant formulas: effects on fat, fatty acid, and mineral balance. *J Pediatr Gastroenterol Nutr*. 1996;23(5):553–560.
16. Motil KJ. Fat and calcium absorption in infancy revisited. *J Am Coll Nutr*. 1998;17(4):303–305.
17. Quinlan PT, Lockton S, Irwin J, Lucas AL. The relationship between stool hardness and stool composition in breast- and formula-fed infants. *J Pediatr Gastroenterol Nutr*. 1995;20(1):81–90.
18. Lien EL. The role of fatty acid composition and positional distribution in fat absorption in infants. *J Pediatr*. 1994;125(5 Pt 2):S62–S68.
19. Nelson SE, Frantz JA, Ziegler EE. Absorption of fat and calcium by infants fed a milk-based formula containing palm olein. *J Am Coll Nutr*. 1998;17(4):327–332.
20. Innis SM, Dyer R, Nelson CM. Evidence that palmitic acid is absorbed as sn-2 monoacylglycerol from human milk by breast-fed infants. *Lipids*. 1994;29(8):541–545.
21. Bach AC, Babayan VK. Medium-chain triglycerides: an update. *Am J Clin Nutr*. 1982;36(5):950–962.
22. Klenoff-Brumberg HL, Genen LH. High versus low medium chain triglyceride content of formula for promoting short term growth of preterm infants. *Cochrane Database Syst Rev*. 2003(1):CD002777.
23. Goulet O. Lipid requirements in infants with digestive diseases with references to short bowel syndrome. *Eur J Med Res*. 1997;2(2):79–83.
24. Jeppesen PB, Mortensen PB. The influence of a preserved colon on the absorption of medium chain fat in patients with small bowel resection. *Gut*. 1998;43(4):478–483.
25. Rodriguez M, Funke S, Fink M, et al. Plasma fatty acids and [13C]linoleic acid metabolism in preterm infants fed a formula with medium-chain triglycerides. *J lipid Res*. 2003;44(1):41–48.
26. Lehner F, Demmelmair H, Roschinger W, et al. Metabolic effects of intravenous LCT or MCT/LCT lipid emulsions in preterm infants. *J Lipid Res*. 2006;47(2):404–411.
27. McDonald GB, Weidman M. Partitioning of polar fatty acids into lymph and portal vein after intestinal absorption in the rat. *Q J Exp Physiol*. 1987;72(2):153–159.
28. Hayashi H, Fujimoto K, Cardelli JA, et al. Fat feeding increases size, but not number, of chylomicrons produced by small intestine. *Am J Physiol*. 1990;259(5 Pt 1):G709–G719.
29. Koletzko B, Baker S, Cleghorn G, et al. Global standard for the composition of infant formula: recommendations of an ESPGHAN coordinated international expert group. *J Pediatr Gastroenterol Nutr*. 2005;41(5):584–599.

30. Koletzko B, Bhutta ZA, Cai W, et al. Compositional requirements of follow-up formula for use in infancy: recommendations of an international expert group coordinated by the early nutrition academy. *Ann Nutr Metab*. 2012;62(1):44–54.
31. Ziegler EE, Fomon SJ. Methods in infant nutrition research: balance and growth studies. *Acta Paediatr Scand Suppl*. 1982;299:90–96.
32. Koletzko B. Response to and range of acceptable fat intakes in infants and children. *Eur J Clin Nutr*. 1999;53 Suppl 1:S78–S83.
33. Van Aerde JE, Sauer PJ, Pencharz PB, et al. Metabolic consequences of increasing energy intake by adding lipid to parenteral nutrition in full-term infants. *Am J Clin Nutr*. 1994;59(3):659–662.
34. Dauncey MJ, Bingham SA. Dependence of 24 h energy expenditure in man on the composition of the nutrient intake. *Br J Nutr*. 1983;50(1):1–13.
35. Aggett PJ, Haschke F, Heine W, et al. Comment on the content and composition of lipids in infant formulas. ESPGAN Committee on Nutrition. *Acta Paediatr Scand*. 1991;80(8–9):887–896.
36. Koletzko B, Rodriguez-Palmero M, Demmelmair H, et al. Physiological aspects of human milk lipids. *Early Hum Dev*. 2001;65 Suppl:S3–S18.
37. Talvia S, Lagstrom H, Rasanen M, et al. A randomized intervention since infancy to reduce intake of saturated fat: calorie (energy) and nutrient intakes up to the age of 10 years in the Special Turku Coronary Risk Factor Intervention Project. *Arch Pediatr Adolesc Med*. 2004;158(1):41–47.
38. Svahn JC, Feldl F, Raiha NC, et al. Fatty acid content of plasma lipid fractions, blood lipids, and apolipoproteins in children fed milk products containing different quantity and quality of fat. *J Pediatr Gastroenterol Nutr*. 2000;31(2):152–161.
39. Strong JP, Malcom GT, Newman WP 3rd, Oalmann MC. Early lesions of atherosclerosis in childhood and youth: natural history and risk factors. *J Am Coll Nutr*. 1992;11 Suppl:51S–514S.
40. Aggett PJ, Haschke F, Heine W, et al. Committee report: childhood diet and prevention of coronary heart disease. ESPGAN Committee on Nutrition. European Society of Pediatric Gastroenterology and Nutrition. *J Pediatr Gastroenterol Nutr*. 1994;19(3):261–269.
41. Woodside JV, Kromhout D. Fatty acids and CHD. *Proc Nutr Soc*. 2005;64(4):554–564.
42. Nicklas TA, Webber LS, Koschak M, Berenson GS. Nutrient adequacy of low fat intakes for children: the Bogalusa Heart Study. *Pediatrics*. 1992;89(2):221–228.
43. Boulton TJ, Magarey AM. Effects of differences in dietary fat on growth, energy and nutrient intake from infancy to eight years of age. *Acta Paediatr*. 1995;84(2):146–150.
44. Obarzanek E, Hunsberger SA, Van Horn L, et al. Safety of a fat-reduced diet: the Dietary Intervention Study in Children (DISC). *Pediatrics*. 1997;100(1):51–59.
45. Hansen D, Michaelsen KF, Skovby F. Growth during treatment of familial hypercholesterolemia. *Acta Paediatr*. 1992;81(12):1023–1025.
46. Koletzko B. Trans fatty acids may impair biosynthesis of long-chain polyunsaturates and growth in man. *Acta Paediatr*. 1992;81(4):302–306.
47. Vobecky JS, Vobecky J, Normand L. Risk and benefit of low fat intake in childhood. *Ann Nutr Metab*. 1995;39(2):124–133.
48. Tonstad S, Sivertsen M. Relation between dietary fat and energy and micronutrient intakes. *Arch Dis Child*. 1997;76(5):416–420.
49. Koletzko B. Potential adverse effects of trans fatty acids in infants and children. *Eur J Med Res*. 1995;1(2):123–125.

50. Carlson SE, Clandinin MT, Cook HW, et al. trans Fatty acids: infant and fetal development. *Am J Clin Nutr*. 1997;66(3):715S–736S.
51. Hunter JE. Dietary trans fatty acids: review of recent human studies and food industry responses. *Lipids*. 2006;41(11):967–992.
52. Precht D, Molkentin J. C18:1, C18:2 and C18:3 trans and cis fatty acid isomers including conjugated cis delta 9, trans delta 11 linoleic acid (CLA) as well as total fat composition of German human milk lipids. *Nahrung*. 1999;43(4):233–244.
53. Salas-Salvado J, Marquez-Sandoval F, Bullo M. Conjugated linoleic acid intake in humans: a systematic review focusing on its effect on body composition, glucose, and lipid metabolism. *Crit Rev Food Sci Nutr*. 2006;46(6):479–88.
54. Larque E, Gil-Sanchez A, Prieto-Sanchez MT, Koletzko B. Omega 3 fatty acids, gestation and pregnancy outcomes. *Br J Nutr*. 2012;107 Suppl 2:S77–S84.
55. Koletzko B, Uauy R, Palou A, et al. Dietary intake of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in children — a workshop report. *Br J Nutr*. 2010;103(6):923–928.
56. Demmelmair H, von Schenck U, Behrendt E, et al. Estimation of arachidonic acid synthesis in full term neonates using natural variation of 13C content. *J Pediatr Gastroenterol Nutr*. 1995;21(1):31–36.
57. Uauy R, Dangour AD. Nutrition in brain development and aging: role of essential fatty acids. *Nutr Rev*. 2006;64(5 Pt 2):S24–S33; discussion S72–S91.
58. Greiner RC, Winter J, Nathanielsz PW, Brenna JT. Brain docosahexaenoate accretion in fetal baboons: bioequivalence of dietary alpha-linolenic and docosahexaenoic acids. *Pediatr Res*. 1997;42(6):826–834.
59. Koletzko B, Cetin I, Brenna JT. Dietary fat intakes for pregnant and lactating women. *Br J Nutr*. 2007;98(5):873–877.
59a. Koletzko B, Boey CCM, Campoy C, Carlson SE, et al. Current information and Asian perspectives on long-chain polyunsaturated fatty acids in pregnancy, lactation, and infancy. Systematic review and practice recommendations from an Early Nutrition Academy workshop. *Ann Nutr Metab*. 2014;65(1):i49–80.
60. Glaser C, Lattka E, Rzehak P, et al. Genetic variation in polyunsaturated fatty acid metabolism and its potential relevance for human development and health. *Matern Child Nutr*. 2011;7 Suppl 2:27–40.
61. Schaeffer L, Gohlke H, Muller M, et al. Common genetic variants of the FADS1 FADS2 gene cluster and their reconstructed haplotypes are associated with the fatty acid composition in phospholipids. *Hum Mol Genet*. 2006;15(11):1745–56.
62. Steer CD, Davey Smith G, Emmett PM, et al. FADS2 polymorphisms modify the effect of breastfeeding on child IQ. *PloS ONE*. 2010;5(7):e11570.
63. Morales E, Bustamante M, Gonzalez JR, et al. Genetic variants of the FADS gene cluster and ELOVL gene family, colostrums LC-PUFA levels, breastfeeding, and child cognition. *PloS ONE*. 2011;6(2):e17181.
64. Berghaus TM, Demmelmair H, Koletzko B. Fatty acid composition of lipid classes in maternal and cord plasma at birth. *Eur J Pediatr*. 1998;157(9):763–768.
65. Gil-Sanchez A, Demmelmair H, Parrilla JJ, et al. Mechanisms involved in the selective transfer of long chain polyunsaturated Fatty acids to the fetus. *Front Genet*. 2011;2:57.
66. Larque E, Demmelmair H, Berger B, et al. In vivo investigation of the placental transfer of (13)C-labeled fatty acids in humans. *J Lipid Res*. 2003;44(1):49–55.
67. Gil-Sanchez A, Larque E, Demmelmair H, et al. Maternal-fetal in vivo transfer of [13C]docosahexaenoic and other fatty acids across the human placenta 12 h after maternal oral intake. *Am J Clin Nutr*. 2010;92(1):115–122.
68. Larque E, Demmelmair H, Gil-Sanchez A, et al. Placental transfer of fatty acids

and fetal implications. *Am J Clin Nutr*. 2011;94(6 Suppl):1908S–1913S.
69. Gil-Sanchez A, Koletzko B, Larque E. Current understanding of placental fatty acid transport. *Curr Opin Clin Nutr Metab Care*. 2012;15(3):265–272.
70. Wadsack C, Tabano S, Maier A, et al. Intrauterine growth restriction is associated with alterations in placental lipoprotein receptors and maternal lipoprotein composition. *Am J Physiol Endocrinol Metab*. 2007;292(2):E476–E484.
71. Larque E, Krauss-Etschmann S, Campoy C, et al. Docosahexaenoic acid supply in pregnancy affects placental expression of fatty acid transport proteins. *Am J Clin Nutr*. 2006;84(4):853–861.
72. Dutta-Roy AK. Transport mechanisms for long-chain polyunsaturated fatty acids in the human placenta. *Am J Clin Nutr*. 2000;71(1 Suppl):315S–322S.
73. Larque E, Demmelmair H, Klingler M, et al. Expression pattern of fatty acid transport protein-1 (FATP-1), FATP-4 and heart-fatty acid binding protein (H-FABP) genes in human term placenta. *Early Hum Dev*. 2006;82(10):697–701.
74. Krauss-Etschmann S, Shadid R, Campoy C, et al. Effects of fish-oil and folate supplementation of pregnant women on maternal and fetal plasma concentrations of docosahexaenoic acid and eicosapentaenoic acid: a European randomized multicenter trial. *Am J Clin Nutr*. 2007;85(5):1392–1400.
75. Szajewska H, Horvath A, Koletzko B. Effect of n-3 long-chain polyunsaturated fatty acid supplementation of women with low-risk pregnancies on pregnancy outcomes and growth measures at birth: a meta-analysis of randomized controlled trials. *Am J Clin Nutr*. 2006;83(6):1337–1344.
76. Makrides M, Duley L, Olsen SF. Marine oil, and other prostaglandin precursor, supplementation for pregnancy uncomplicated by pre-eclampsia or intrauterine growth restriction. *Cochrane Database Syst Rev*. 2006(3):CD003402.
77. Horvath A, Koletzko B, Szajewska H. Effect of supplementation of women in high-risk pregnancies with long-chain polyunsaturated fatty acids on pregnancy outcomes and growth measures at birth: a meta-analysis of randomized controlled trials. *Br J Nutr*. 2007;98(2):253–259.
78. European-Food-Safety-Authority. Opinion of the Scientific Panel on Contaminants in the food chain on a request from the Commission related to mercury and methylmercury in food (Request No EFSA-Q-2003–030, adopted on 24 February 2004). *Eur Food Safety Authority J*. 2004;34:1–14.
79. European-Food-Safety-Authority. Opinion of the Scientific Panel on Contaminants in the food chain on a request from the European Parliament related to the safety assessment of wild and farmed fish (Question No EFSA-Q-2004–22, adpted on 22 June 2005). *Eur Food Safety Authority J*. 2005;236:1–118.
80. Yuhas R, Pramuk K, Lien EL. Human milk fatty acid composition from nine countries varies most in DHA. *Lipids*. 2006;41(9):851–858.
81. Makrides M, Gibson RA, Udell T, Ried K. Supplementation of infant formula with long-chain polyunsaturated fatty acids does not influence the growth of term infants. *Am J Clin Nutr*. 2005;81(5):1094–1101.
82. European-Food-Safety-Authority. DHA and ARA and visual development. Scientific substantiation of a health claim related to docosahexaenoic acid (DHA) and arachidonic acid (ARA) and visual development pursuant to Article14 of Regulation (EC) No 1924/2006. *Eur Food Safety Authority J*. 2009;941:1–14.
83. EFSA-Panel-on-Dietetic-Products, -. N.-a.-A. *Draft Scientific Opinion on the Essential Composition of Infant and Follow-On Formulae*. Parma, Italy: EFS Authority; 2014.
84. European-Commission. Commission directive 2006/141/EC of 22 December 2006 on infant formulae and follow-on formulae and amending Directive 1999/21/EC.

Official Journal of the European Union (L 401/1);2006.
85. Codex-Alimentarius-Commission. *Standard for Infant Formula and Formulas for Special Medical Purposes Intended for Infants. CODEX STAN 72–1981. Rome, Italy: Codex-Alimentarius-Commission; 2007: 1–21.*
86. Koletzko B, Carlson SE, van Goudoever JB. Should infant formula provide both omega-3 DHA and omega-6 arachidonic acid? *Ann Nutr Metab*. 2015;65: in press.
87. Carlson SE, Cooke RJ, Werkman SH, Tolley EA. First year growth of preterm infants fed standard compared to marine oil n-3 supplemented formula. *Lipids*.1992;27(11):901–907.
88. Lapillonne A, and Carlson SE. Polyunsaturated fatty acids and infant growth. *Lipids*. 2001;36(9):901–911.
89. Colombo J, Carlson SE, Cheatham CL, Shaddy DJ, et al. Long-term effects of LCPUFA supplementation on childhood cognitive outcomes. *Am J Clin Nutr*. 2013;98(2):403–412.
90. Hsieh AT, Brenna JT. Dietary docosahexaenoic acid but not arachidonic acid influences central nervous system fatty acid status in baboon neonates. Prostaglandins *Leukot Essent Fatty Acids*. 2009;81(2-3):105–110.
91. Aggett PJ, Agostini C, Goulet O, Hernell O, Koletzko B, et al. The nutritional and safety assessment of breast milk substitutes and other dietary products for infants: a commentary by the ESPGHAN Committee on Nutrition. *J Pediatr Gastroenterol Nutr*. 2001;32(3):256–258.
92. Koletzko B, Ashwell M, Beck B, Bronner A, Mathioudakis B. Characterisation of infant food modifications in the European Union. *Ann Nutr Metab*. 2002;46(6):231–242.

Trace Elements

8

Nancy F. Krebs, MD, MS and K. Michael Hambidge, MD, ScD

More than 99.7% of the body weight is accounted for by 11 elements, termed the major elements (Table 8-1). The remainder of the body weight consists of approximately 25 trace elements, each of which contributes less than 0.01% to the body weight. Of these, a mammalian nutritional requirement has been established for iron, iodine, zinc, copper, chromium, selenium, molybdenum, manganese, cobalt, nickel, vanadium, silicon, arsenic, boron, and fluoride. Although it is reasonable to assume that the trace elements required by humans are similar to those required by other mammalian species, currently, a human nutritional requirement is accepted only for iron, iodine, zinc, copper, chromium, selenium, molybdenum, manganese, and cobalt (as vitamin B_{12}). Because of its beneficial effects in the prevention of dental caries, fluoride is also included in this list.

BIOLOGIC ROLES

Many enzyme systems depend on one or more specific trace elements, either as key components of metalloenzymes or as catalysts of metal ion–activated enzymes. The roles of metal ions in metalloenzymes may be structural, that is, maintaining the conformation of the protein, catalytic, or both. Iron, copper, zinc, molybdenum, selenium, and manganese have been identified as constituents of specific enzymes. Certain trace elements are of fundamental importance in the structure or metabolic activity of other compounds, including hemoglobin (iron), nucleic acids (multiple trace elements), transcription factor proteins (zinc), and vitamin B_{12} (cobalt). The roles of iodine in thyroxine (T_4) and triiodothyronine (T_3), zinc in steroid hormone receptors, and chromium in facilitating the action of insulin are examples of the importance of trace elements in the activity of certain hormones. The dependence of many vital metabolic processes on trace elements confers a physiologic importance of these micronutrients

that is analogous to that of vitamins and is out of proportion to their small contribution to body weight. Although considerable progress has been achieved in determining the functions of several trace elements, little is yet known about the biochemistry of others.

Table 8-1 Elements with a Role in Human Physiology

Major Elements	Recognized	Probable
Trace Elements		
Carbon (C)	Iron (Fe)	Nickel (Ni)
Hydrogen (H)	Iodine (I)	Vanadium (V)
Nitrogen (N)	Zinc (Zn)	Silicon (Si)
Oxygen (O)	Copper (Cu)	Arsenic (As)
Sodium (Na)	Chromium (Cr)	Boron (B)
Potassium (K)	Selenium (Se)	
Phosphorus (P)	Molybdenum (Mo)	
Sulfur (S)	Manganese (Mn)	
Chloride (Cl^-)	Cobalt (Co)	
Calcium (Ca)	Fluoride (F^-)	
Magnesium (Mg)		

DOSE RESPONSE

An elementary but important concept is that the biologic effect of any trace element depends on the dose or, more specifically, on the level of intake and on tissue concentrations (Figure 8-1). For each element, there is an optimal range of intake, the magnitude of which varies considerably between different elements and depends on a number of factors, including the efficiency of homeostatic mechanisms. Obviously, if intakes are sufficiently high, any element will prove toxic. In the case of non essential elements (e.g., mercury), harmful effects can result only from toxic levels of intake. However, in the case of the “essential” trace elements, problems may result from intakes that are too low as well as too high. In animal husbandry, the economic importance of trace deficiency states has been recognized for many years.[1] However, in human nutrition, only iron and iodine deficiencies were recognized prior to the 1960s, and it is only more recently that a broader concept of trace element nutrition has started to

develop with the recognition of zinc, copper, chromium, selenium, molybdenum, and possibly manganese deficiency states in humans. Likewise, greater appreciation has emerged for potential adverse effects of very high intakes, for example, from copper contamination of drinking water.

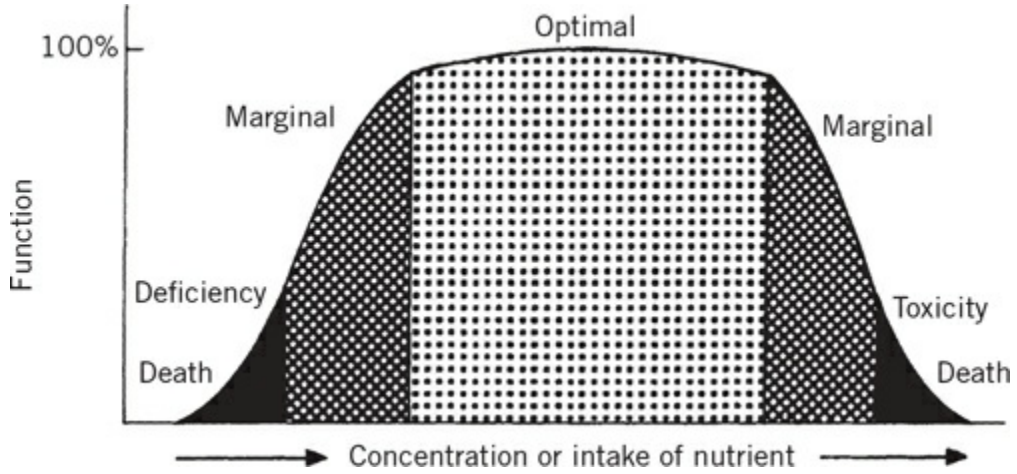


Figure 8-1 Dependence of biologic function on tissue concentration or intake of a nutrient. Reproduced from Mertz W. The essential trace elements. Science 1981; 213:1330.

ETIOLOGY OF HUMAN TRACE DEFICIENCY STATES

The last several decades have been notable not only for the number of new trace elements implicated in deficiency disease, but, perhaps more surprisingly, for the wide variety of different factors that have been discovered to play a role in the etiology of these deficiency states (Table 8-2). These include inherited or acquired alterations in metabolism and other special circumstances leading to increased nutritional requirements as well as factors that contribute to inadequate intakes of one or more of the trace elements.

TABLE 8-2 Etiology of Trace Mineral Deficiencies

Etiology	*Trace Elements* [a]
Inadequate Intake	

No special circumstances required but risk increased during infancy and pregnancy	Fe, Zn
Losses during food processing, including preparation of “humanized” infant milk formulas	Zn, Cr, Cu, Se
Poor bioavailability	Fe, Zn
Local deficiencies in geochemical environment	I, Se
In association with protein-energy malnutrition	Zn, Cu, Cr
Synthetic diets, including intravenous feeding	Zn, Cu, Cr, Se, Mo
Other Circumstances	
Prematurity	Fe, Cu, Zn
Diseases and other unusual circumstances	Zn, Cu, Cr causing impaired intestinal absorption or excessive losses of trace elements
Inborn errors of trace element metabolism	Zn, Cu, Mo

[a] Elements given are notable examples, but not necessarily a complete list.

Notable examples of the inborn errors of trace element metabolism are represented by Acrodermatitis enteropathica and “Menke steely hair syndrome” (MSHS), which are genetic defects in zinc absorption and copper trafficking, respectively.[2,3] Although these entities are rare and not strictly nutritional deficiency diseases, some of them are important in a nutritional context because of the insight they provide into the clinical and metabolic effects of severe nutritional deficiencies. Moreover, in some instances, manipulation of dietary intake has remarkable therapeutic benefits. Both of these points are well illustrated by Acrodermatitis enteropathica, which is discussed below. Although unresponsive to dietary supplements, Menkes’ is attributable in part to a de facto deficiency of copper in certain tissues and subcellular sites, resulting from deranged copper transport, including out of the enterocyte.

Diseases that cause intestinal malabsorption of fat and other nutrients may also lead to trace element deficiencies.[4] Excessive losses of trace elements occur in certain diseases through the kidneys, gastrointestinal tract, sweat, exudates, or hemorrhage. Low blood plasma concentrations of specific trace elements can occur in certain disease states in association with the redistribution of these elements within the body. For example, movement of zinc and iron from the plasma to the liver occurs in stress

situations, including infectious diseases. Correction of hypozincemia in these circumstances may be contraindicated, although the host response may depend on the specific cause of the stress.

The premature infant with very low birth weight provides a unique nutritional challenge that includes the provision of adequate trace elements.[5] Some elements, notably copper, iron, and zinc, are stored extensively by the fetus during the last trimester. These stores, which are available to the term infant in early postnatal life, are diminished in the premature infant. Immaturity of intestinal absorption mechanisms and relatively high losses of endogenous zinc have been reported. These problems are aggravated further by the high nutritional requirements that are imposed by the rapid growth of the premature infant after the early postnatal period. Nutritional requirements may also be relatively high during periods of rapid growth in the term infant and in later childhood, as well as during pregnancy and lactation.

Although nutritional requirements for trace elements are small and these nutrients are ubiquitous in the environment, the quantities present in food and water are usually correspondingly low. Hence, contrary to earlier concepts, it is not surprising that humans are at risk of nutritional deficiencies of the trace elements. Iron deficiency, although outside the scope of this chapter, remains one of the most common human nutritional disorders. Other trace deficiencies occur in more restricted geographic locations owing to localized deficiencies in soils, plants, or water. The classic example of a geographic deficiency is that of endemic goiter, and low body content of selenium is observed in some countries, for example New Zealand, while marked selenium depletion and deficiency were documented and subsequently treated through public health initiatives in large areas of China. Diets deficient in energy and other nutrients may also contain inadequate quantities of trace elements. Thus, deficiencies of zinc, copper, selenium and chromium have been documented in association with protein-energy malnutrition.

Variations in bioavailability are a major factor in determining the adequacy of dietary intake of some of the trace elements. This also applies especially to the infant because of its dependence on one major food staple. Thus, if the milk or milk substitute that provides most or all of the nutrients consumed by the infant is low in available trace elements, there is no chance that this deficit will be compensated for by a more favorable supply in other food items. Although our understanding of this complex subject is far from complete, it is known that human milk confers important advantages with respect to the bioavailability of iron and zinc.

Although not proven, it seems probable that this may also be true of other trace elements. As a corollary, the trace element concentrations in breast milk should be considered a minimum guideline for corresponding concentrations in infant milk formulas, and higher concentrations of at least some elements, including iron and zinc, are necessary in the latter. However, indiscriminate supplementation may have adverse consequences, especially through undesirable interactions with other trace elements. Additional research directed at mineral–mineral interactions is needed to determine optimal fortification levels in foods and formulas.

The use of synthetic diets in medical practice, including those used in intravenous feeding, has provided some of the most dramatic examples of acquired trace-element deficiency states. Deficiencies of zinc, copper, chromium, molybdenum, and probably selenium were all documented in association with long-term intravenous feeding without adequate provision of these micronutrients. Current pediatric trace mineral formulations routinely used in major pediatric centers have greatly reduced the incidence of deficiencies.

NUTRITIONAL REQUIREMENTS

The evolving nutritional importance of trace elements has been recognized by the publication of guidelines, including those of the Food and Nutrition Board of the National Academy of Sciences (see Appendix 2).[6] A formal Recommended Dietary Allowance (RDA) has been established for copper, iodine, iron, molybdenum, selenium, and zinc. An Adequate Intake (AI) has been set for chromium and manganese, which reflects the more limited data available on which to base an Estimated Average Requirement (EAR). Information is limited to the quantities of some of these elements in individual food items, which limits the application of these guidelines by nutritionists and other health professionals.

Guidelines have also been published for intravenous administration of trace elements to children who have to be maintained with total intravenous nutrition (Table 8-3).[7] Intravenous requirements for zinc and selenium vary according to the extent of ongoing losses, especially from the gastrointestinal tract and kidney, respectively. Hence, it is desirable to monitor status at regular intervals. In contrast, copper and manganese may accumulate in cholestatic patients who are receiving long-term parenteral nutrition support, in which case, monitoring and adjustment of the administered dose may also be warranted.

LABORATORY ANALYSES AND THE DETECTION OF TRACE ELEMENT DEFICIENCY STATES

Fundamental advances in analytic instrumentation, especially the advent and progressive refinement of atomic absorption spectrophotometry, have facilitated the quantitative measurement of trace elements that are present in the parts-per-billion as well as parts-per-million concentration ranges. Yet formidable analytic difficulties remain. For example, accurate measurement of chromium, which now appears to be present in blood plasma, urine, and milk in only parts-per-trillion concentrations, is extraordinarily difficult. Even for relatively simple measurements, such as those of zinc in plasma or serum, substantial inter-laboratory differences in analytic results continue to be evident, and it is ideal that each laboratory establish its own normal range. Even when analytic data are accurate, interpretation is frequently problematic. Plasma concentrations of zinc, for example, may be depressed for reasons other than a nutritional deficiency, and thus may fail to provide a sufficiently sensitive or reliable guide to body status. Established physiologic indices, such as the measurement of the activity of an enzyme that depends on a specific trace element, are generally unavailable. The depression of glutathione peroxidase activity in selenium deficiency states is one exception, but the clinical significance of this observation remains uncertain. Overall, laboratory indices for detection of “newer” trace element deficiencies are not as dependable as those available for the detection of iron deficiency. Hence, for research purposes, more cumbersome approaches are necessary to confirm the suspicion of a specific deficiency, including especially the effects of supplementation on specific physiologic parameters and functional outcomes under carefully controlled conditions. Meanwhile, the health professional needs to be aware of the circumstances in which there is a risk ofspecific trace deficiencies and of the clinical features that may be attributable to such a deficiency.

Table 8-3 Guidelines for Maintenance Intravenous Administration of Trace Elements during Parenteral Nutrition

Trace Element	Maintenance Requirement (µg/kg/day)
Zinc	100[a]

Copper	20
Chromium	0.2
Manganese	1
Selenium	2
Iodine	1

[a] Requirements for zinc may be much greater in patients who continue to excrete excessive quantities of zinc; also, in premature infants, 300 µg/kg/dayshould be administered

Hair analyses merit special mention because of their widespread misuse and the unsubstantiated claims for this diagnostic tool. Currently, the value of chemical analyses of hair in clinical practice is extremely limited.[8] Because of the inherent difficulties in interpretation of analytic data, it is unlikely that future research will lead to more useful clinical applications of hair analyses. Multi-element hair analyses continue to be available commercially, often accompanied by interpretations of questionable validity, and the results merit caution and additional confirmatory evidence prior to therapeutic interventions.

ZINC

A nutritional requirement for zinc was first recognized in microorganisms in 1869, in plants in 1914, and in mammals in 1934.[1] In 1954, zinc deficiency was shown to be the cause of porcine parakeratosis. The first of the zinc metalloenzymes to be identified was carbonic anhydrase in 1940. Subsequently, over 100 zinc metalloenzymes have been identified, including at least one in every major enzyme classification. Understanding of the role of zinc in cell growth and differentiation took a major step forward with recognition of “zinc fingers” as the most frequent deoxyribonucleic acid (DNA) binding motif for transcription factor proteins.[9] The first reports of human zinc deficiency attributable to dietary factors emerged in the Middle East in the early 1960s in relation to the syndrome of adolescent nutritional dwarfism.[10] In the 1990s, the importance of zinc deficiency on childhood morbidity and mortality in the developing world was documented by a number of carefully conducted, large-scale, randomized, controlled supplementation trials in vulnerable populations.[11]

CLINICAL FEATURES OF ZINC DEFICIENCY

The clinical features of human zinc deficiency range from a life-

threatening disease state, characterized by acro-orificial skin rashes, diarrhea, growth arrest, anorexia, depressed mood, and increased susceptibility to infection, to a mild impairment of growth velocity, often accompanied by other problems, including poor appetite (Table 8-4). In addition to the severity of the zinc depletion, the clinical sequelae appears to depend on other factors, including the speed of onset and associated clinical and dietary factors such as protein-energy malnutrition.

Decreased growth velocity or growth arrest is a consistent and early result of even mild zinc deficiency in young experimental animals and in infants, children, and adolescents. In severe acute zinc deficiency states (e.g., Acrodermatitis enteropathica), there is an abrupt cessation of weight gain when the skin rash becomes manifest, and "catch-up" growth occurs when adequate zinc supplements are administered.[12] A meta-analysis of trials in prepubertal children that met strict criteria indicated that supplementation was associated with significant responses in both height and weight increments. Responses were strongest in those initially underweight and in children >6 months of age with evidence of stunting.[13,14] These collective results suggest that zinc deficiency is a common cause of retarded growth velocity in the developing world. Zinc deficiency has also been linked to poor growth in association with certain diseases, including cystic fibrosis and sickle cell anemia.[15–17]

TABLE 8-4 Clinical Features of Zinc Deficiency[a]

Decrease in growth velocity
Impaired appetite and decreased food intake
Diarrhea
Increased susceptibility to infection with associated abnormalities of the immune system
Acro-orificial skin lesions; other epithelial lesions, including glossitis, alopecia, nail dystrophy, and hair loss
Behavioral abnormalities, including impairment of hedonic tone Delayed sexual maturation and impotence
Eye lesions, including photophobia and lack of dark adaptation. Delayed healing of wounds, burns, and decubitus ulcers
Hypogeusia or dysgeusia
Low birth weight
Prematurity

[a] Features are dependent on severity of deficiency and other factors.

Anorexia is an early manifestation of zinc deficiency in experimental animals. Although this has long been recognized as a feature of Acrodermatitis enteropathica and is also noted in association with milder zinc deficiency syndromes, it has been difficult to acquire objective evidence of a direct cause-and-effect relationship in humans. However, a significant effect of zinc supplementation on food intake has been observed in some double-blind, controlled studies.[18,19]

Diarrhea is one of the hallmark features of the early descriptions of acrodermatitis enteropathica and occurs in a high percentage of cases of severe zinc deficiency. There is growing evidence that zinc deficiency contributes to the incidence, severity, and duration of acute infectious and persistent diarrhea in infants and young children in the developing world. On the basis of pooled analysis of zinc supplementation trials, a substantial reduction in the rate of treatment failure or death in children given zinc supplements has been estimated. Diarrhea can itself also contribute to zinc deficiency because of the losses of endogenous zinc in diarrheal fluids. Thus, requirements for zinc are likely to be greater in the presence of diarrhea. In recognition of the beneficial effects of zinc supplementation and the high prevalence of marginal zinc status in young children in international settings, WHO and UNICEF have recommended routine administration of zinc supplements along with fluid replacement and continued feeding in all episodes of acute diarrhea.[20,21]

The skin lesions of severe acute zinc deficiency syndromes have a characteristic distribution, primarily in the extremities and adjacent to the body orifices (Figure 8-2). However, lesions do occur elsewhere and may become generalized. Most commonly, the rashes are erythematous, vesiculobullous, and pustular. Secondary infection is common, especially with staphylococci or *Candida*. In hereditary Acrodermatitis enteropathica, the skin lesions typically occur in early infancy, although they are often delayed until several months of age, especially after weaning if the infant is breast-fed. Other epidermal lesions include gingivitis, dermatitis, glossitis, conjunctivitis, blepharitis, alopecia, and, in chronic cases, nail dystrophies.

Behavioral abnormalities are a notable feature of severe zinc deficiency states. Irritability, lethargy, and depression occur typically in conjunction with the skin rash of severe zinc deficiency. Hedonic tone and motivation to engage in the environment, improve dramatically after the commencement of zinc therapy.[22] Cognitive development is impaired in young zinc-deficient animals, and there is some evidence that the same applies in the human, although results of trials have been inconsistent.[23,24]



Figure 8-2 The acute skin lesions of acrodermatitis enteropathica are demonstrated in this 10-month-old infant. Reproduced from Hambidge KM, Nichols PL Jr, editors. Zinc and copper in clinical medicine. Jamaica (NY): Spectrum Publications; 1978.

Frequent bacterial and monilial infections, associated with abnormalities of the immune system, contribute to the progressive and often fatal course that characterizes untreated acrodermatitis enteropathica (AE). Abnormalities of the immune system also occur in less severe zinc deficiency states.[25] Compared with studies of zinc and diarrhea, the data are less consistent linking zinc deficiency with incidence, prevalence and severity of respiratory infections, including pneumonia. A recent systematic review, based on six randomized, controlled trials in children from 2 to 59 months of age, found approximately a 13% reduction in the incidence of pneumonia, and a 41% reduction in prevalence; findings of subgroup analysis emphasized importance of clear definitions of

diagnostic criteria.[26] The distinction between an apparent response due to correction of pre-existing zinc deficiency versus a pharmacologic effect of zinc supplementation is rarely clear.

BIOCHEMICAL CORRELATES

Although there is now extensive knowledge about the biochemistry of zinc, especially its role in a wide range of enzyme systems and in gene transcription, the biochemical correlates of the clinical features of human zinc deficiency remain poorly defined. Many of the features of zinc deficiency, including growth retardation, poor wound healing, and abnormalities of fetal development and the immune system, are thought to be attributable, at least in part, to disturbances in nucleic acid metabolism and protein synthesis. Its role as a stabilizer of the molecular structure of subcellular constituents and membranes is exemplified by the observation of altered membrane signaling and postreceptor events that coordinate the response to insulin-like growth factor 1 when zinc availability is limited.[27,28] Such observations provide another plausible link to the growth retardation and impaired immune function during zinc deficiency. The so-called zinc fingers are the most common motif for the repetitive sites of transcription proteins that bind to DNA to initiate transcription.[9] Through this mechanism, which can be compromised by zinc restriction, zinc has an extraordinarily important role in gene expression. Additionally, several of the enzymes involved in nucleic acid synthesis are zinc dependent, including DNA and ribonucleic acid (RNA) polymerases, reverse transcriptase, and thymidine kinase, the activity of which is known to be reduced in human zinc deficiency.

Zinc is critical for normal immune function and interacts with several components of the immune system, ranging from the skin and its barrier function to aspects of both cellular and, to a lesser extent humeral, immunity.[25] In vitro studies emphasize the responsiveness of some components to zinc concentrations, with both deficiency and supraphysiologic levels leading to impaired immune function. Two recent studies examining zinc as an adjunct therapy for treatment of pneumonia[29] and probable bacterial sepsis[30] found reduction in case fatality rates and treatment failure, respectively, in young children and infants. Zinc in the form of lozenges has been investigated in developed countries for its ability to shorten the duration and reduce the severity of “the common cold,” or upper respiratory infections. This is an example of a

pharmacological therapeutic effect, since most individuals would be presumed to have sufficient zinc status at baseline. Reviews to date have concluded that, despite the likelihood of efficacy, data are insufficient to make recommendations regarding dose, formulation and duration.[31] Reviews address in vitro and in vivo studies of the role of zinc in normal immune function and the impact of zinc deficiency on common childhood illnesses.[25,32,33]

ETIOLOGIC FACTORS IN ZINC DEFICIENCY

The prototype of the severest forms of human zinc deficiency is provided by acrodermatitis enteropathica (AE), a rare autosomal recessively inherited disorder. The phenotypic expression of this disease is attributable entirely to a severe zinc-deficiency state. Although total body zinc is not greatly reduced in AE, plasma zinc concentrations are typically extremely low. Before the beneficial effects of zinc therapy were recognized, this was a severe, progressive, and often fatal disease. The zinc deficiency results from a partial block in the intestinal absorption of this trace metal due to mutations in the SLC39A4 gene, also known as the ZIP4 gene, that codes for a transmembrane, histidine-rich protein which functions in zinc uptake. The gene is expressed abundantly throughout the intestine, and accumulates at the apical surface of the enterocyte.[34] Mutations of SLC39A4 are now thought to be observed in only about half of the index AE cases tested, with ~40% of the patients found to be homozygotes or compound heterozygotes, and 8% found to be heterozygotes.[35] Clinical diagnosis by mutation analysis is now possible, although not yet routinely available.[36]

There have been numerous reports of severe zinc deficiency states in both term and pre-term breast-fed infants the presentations have been attributable to a condition now termed “acquired zinc deficiency of lactogenic origin,” which is characterized by an inability of the mother’s mammary gland to secrete normal quantities of zinc into the milk despite otherwise apparently normal maternal zinc status. Specific mutation(s) of the SLC30A2 gene for zinc transporters localized to the mammary gland is responsible for this condition.[37] The result is markedly low milk zinc concentrations (~25% of normal for stage of lactation), and severe zinc deficiency in the exclusively breastfed infant. Supplemental zinc to the infant easily treats the infant’s deficiency, as there is no absorptive defect in the infant. The prevalence of this defect is unknown, but clinical

impression is that it is not rare. In a large cohort of Chinese women, two polymorphisms of the SLC30A2 gene explained 3.23% of total variance in milk zinc concentrations.[38]

Zinc deficiency has also been well documented in premature and low-birth weight infants if supplemental zinc is not provided. Contributory factors may include immaturity of the gastrointestinal homeostatic processes, including both absorption of exogenous zinc and conservation of endogenous zinc; relatively high urine losses; and relatively high zinc requirements when rapid postnatal growth commences. In one large series from a hospital population, the strongest clinical risk factors include low gestational age, small for gestational age (SGA), and intestinal resection.[39] Fortification of human milk and use of fortified formulas designed to meet the needs of premature infants have been associated with positive net absorption and retention of zinc.[40] The results of zinc supplementation trials in low birth weight infants in developing countries have resulted in a striking reduction in morbidity and mortality, illustrating the apparent vulnerability of these infants with-out a generous dietary intake to zinc deficiency.[41,42]

A wide variety of etiologic factors may contribute to more moderate and mild zinc deficiency states. An “absolute” dietary deficiency is considered to be less common than relative deficiency due to reliance on dietary sources of poor bioavailability. Older breast-fed infants are susceptible to mild zinc deficiency because of the low zinc concentrations in human milk after 6 months. Deficiency has been documented in infants in developing countries, where a combination of complementary foods with low zinc content and/or bioavailability and a high infectious burden likely contribute to the risk of deficiency.[19] As phytate intakes are high in many developing countries owing to dependence on plant staples such as maize, wheat, or legumes, there is particular concern about the adequacy of zinc intake in infants and young children in these settings.[43] Even in western cultures, the traditional progression from iron fortified infant cereals to fruits and vegetables, with relatively late introduction of meats, may pose a risk for mild zinc deficiency in older breastfed infants.[44]

The risk of zinc deficiency is enhanced in conditions of gastrointestinal pathology such as short gut syndrome, intestinal failure, or other causes of malabsorption and/or increased endogenous intestinal losses.[45] Fat malabsorption or intestinal inflammation may impair zinc absorption and enhance losses of intestinal endogenous zinc, as in regional enteritis, celiac disease, and cystic fibrosis.[15,46] In children with celiac disease, institution

of a gluten free diet has been reported to result in normalization of plasma zinc levels, with or without zinc supplementation.[46,47] Excessive urinary losses of zinc occur in liver disease,[48] diabetes mellitus, sickle cell disease,[17] nephrosis, and catabolic conditions.[4] A number of chelating agents, diuretics, and other drugs can also cause excessive zinc excretion, but the extent to which this is of practical significance is unclear.

DIAGNOSIS OF ZINC DEFICIENCY

The characteristic distribution of the skin lesions facilitates the clinical diagnosis of severe zinc deficiency states. Unfortunately, however, the clinical features of moderate and mild zinc deficient states are nonspecific. Hypozincemia is not a consistent feature of mild chronic zinc deficiency states. Currently, laboratory parameters are of only limited value in the confirmation of zinc deficiency. Plasma zinc concentrations are markedly depressed (<40 μg/dL) in the severest deficiency states, and moderate hypozincemia (40–60 μg/dL) occurs in more moderate zinc deficiency states. However, plasma zinc levels are also depressed to this extent with the effects of various stresses, including infections, which result in hepatic sequestration of circulating zinc. Interleukin-6 and other cytokines are responsible for the decline in plasma zinc associated with infections. Underlying zinc deficiency can obviously co-exist with acute infection, and thus consideration of the benefit of zinc supplements should be given in settings of high risk of nutritional zinc deficiency.[49] Fasting is associated with higher plasma zinc levels, and the effects of meals results in progressively lower concentrations over the course of a day. An analysis of national data for children under 10 years suggested a lower cutoff of 65 μg/dL for morning non-fasting sampling, and 57 μg/dL for afternoon sampling.[50] Serum zinc levels have been reported to be higher than plasma zinc levels; this is attributable to a delay in separation from red cells. Erythrocyte zinc is little affected by zinc deficiency and is not a sensitive index, nor are leukocyte zinc concentrations. Urine excretion rates are diminished in severe zinc deficiency, but this is also not a sufficiently sensitive assay for clinical use. Serum alkaline phosphatase activity is depressed in moderate to severe zinc deficiency states and increases with zinc supplementation in cases of moderate zinc deficiency, but this assay is not a sufficiently specific diagnostic tool used alone. Because of the limitations of currently available laboratory indices in the detection of mild zinc deficiency states, the most reliable approach remains the demonstration of a functional response to a trial of zinc supplementation

under adequately controlled conditions.

TREATMENT OF ZINC DEFICIENCY

Acrodermatitis enteropathica can be treated effectively with 40 to 50 mg/day of elemental zinc administered as the sulfate, gluconate, or other salt. Requirements in infants and toddlers with AE may be considerably less (e.g., 20 to 30 mg/day). Most other milder cases of zinc deficiency can be treated effectively with 1 mg elemental zinc/kg body weight/day, typically up to a maximum of 20 mg zinc/day. Somewhat larger quantities may be required in the presence of excessive ongoing losses or malabsorption. In severe zinc deficiency in premature breast-fed infants, temporary zinc supplements are necessary to replete tissue pools until other sources of zinc are added to the diet. One milligram of elemental zinc is equivalent to 4.4 mg of $ZnSO_4 7H_2O$; 7 mg of zinc gluconate; and 2.8 mg of zinc acetate (anhydrous).

Maintenance intravenous requirements for adults are approximately 2 mg and for infants 100 µg/kg body weight/day. In premature infants, 300µg/kg body weight/day has been recommended.[7] Ongoing losses through the kidney or, more commonly, the gastrointestinal tract owing, for example, to intractable diarrhea may substantially increase the intravenous requirement. For intravenous administration, zinc chloride is frequently used; 1 mg zinc is equivalent to 2.1 mg $ZnCl_2$.

TOXICITY

Acute toxicity is rare but can occur, for example, from the ingestion of large quantities of pharmacological preparations of zinc or from the consumption of acidic drinks from galvanized cans. Symptoms are usually limited to diarrhea and vomiting, but there may be severe lethargy. Administration of zinc in quantities of 50 mg/day to adults can cause anemia owing to copper deficiency (see below) and may depress plasma high-density lipoprotein cholesterol levels. More moderate supplementation has generally not been found to be problematic. For example, 10 mg/day for 4 months in infants was not associated with hypocupremia.[51] Similarly, neither 5 or 10 mg of daily supplemental zinc in addition to dietary zinc for 4 months in adolescent boys was associated with adverse effects on an extensive array of indicators of copper status.[52]

ABSORPTION AND METABOLISM

The gastrointestinal tract, specifically the small intestine and the pancreas, have the major role in maintaining whole body Zn homeostasis.[4] Over 20 zinc transporters have been identified in these organs, with the ZnT family generally functioning as zinc exporters and the ZIP family functioning in zinc uptake into the cytoplasm. Gene expression of the transporters is a major component of homeostatic regulation, including regulation of zinc absorption via the small intestine and excreting zinc via both the pancreas and small intestine.

Of note, studies in humans applying stable isotope techniques indicate that the quantity of zinc ingested, rather than host zinc status, is the principal factor triggering regulation of expression of the transporters involved with zinc absorption into and through the enterocyte.[4] Zinc absorption is known to be a saturable transport process.[53] Fractional absorption, therefore, declines with increasing Zn intake and vice versa. Regulation of the quantity of endogenous Zn excreted has a major role in maintaining whole body Zn homeostasis in most circumstances, and appears to be influenced by host zinc status, with conservation in the face of marginal zinc intake.

Fractional absorption of zinc averages about 60% when ingested in solution in water, but is usually only in the range of 20%–40% from composite meals or infant formulas. The favorable bioavailability of zinc in human milk is undoubtedly in part due to the relatively modest concentrations; actual amount of absorbed zinc is lower than from fortified formulas.[54] On the other hand, fractional absorption is depressed by dietary phytate (inositol hexaphosphate), which is found in whole grains and legumes.[55] The intestine is the major route for excretion of endogenous zinc, and conservation of intestinal endogenous zinc appears to be critical in maintaining or restoring zinc balance.[4] The ability of the intestine to conserve endogenous zinc may be impaired by malabsorption, including cystic fibrosis[15], and possibly environmental enteropathy.[56]

About 10% of total body zinc, 100–200 mg in adults, intermixes with circulating plasma zinc within 2 days.[57] This zinc is thought to be of special physiologic importance to many of the zinc-dependent metabolic processes in the body, and it is also this zinc that is vulnerable to dietary zinc restriction. This is likely to explain why the symptoms of zinc deficiency occur with very little change in total body zinc. It has been found that the quantity of readily exchangeable zinc has a strong positive correlation with dietary zinc and an even stronger correlation with a

quantity of zinc absorbed.[4,44] A substantial part of this rapidly exchanging zinc is located in the liver.

In the systemic circulation, approximately 75% of the plasma zinc is loosely bound to albumin, and most of the remainder is tightly bound to α_2-macroglobulin. The normal zinc concentration in plasma averages about 0.85 µg/mL (13 µmol/L), with a range from 0.70 to 1.00 µg/mL. The zinc concentration in erythrocytes, most of which is incorporated in carbonic anhydrase, is approximately 12 µg/g wet weight. Bone, prostate, and hair all have concentrations near 100 µg/g. Muscle, liver, and kidney contain about 50 µg/g. The total body zinc content of an adult has been calculated to be about 2 g.[1]

DIETARY REQUIREMENTS

The results of recent studies of breast-fed and formula-fed infants using zinc stable isotope techniques allow reasonably accurate factorial estimates of zinc requirements. In fully breast-fed infants receiving mature human milk, conservation of endogenous zinc is critical to maintaining positive net absorption.[54] Because the amount of endogenous fecal zinc is positively correlated with dietary zinc intake and absorbed zinc, this figure is considerably higher in formula-fed infants receiving zinc-fortified formulas[58] Allowing for urine, sweat, and integumental losses as endogenous losses and average fractional absorption of approximately 0.5, a dietary zinc requirement of approximately 150 µg zinc/kg/day is estimated. Actual zinc intake of breast-fed infants varies considerably over the first 3 months of life as milk zinc concentrations decline. Mean intakes at 5 months of age have been found to be close to this projected dietary requirement. The physiologic requirement, the amount of zinc that must be absorbed to match endogenous losses and the zinc retained for new tissue synthesis, has been estimated to be 0.84 mg/day for infants 7–12 months of age, and 0.74 mg/day for 1–3 year olds.[6] Factorial estimations are higher for rapidly growing premature infants, for malnourished infants during the "catch-up" growth phase, and in situations with high endogenous losses, for example, with diarrhea and malabsorption.

The EARs and RDAs for older infants and children have been calculated by extrapolation from factorial data for infants, as described above.[6] The Tolerable Upper Intake Levels (ULs) failed to take into account the fact that many North American infants and young children who are not exclusively breast-fed consume substantially more zinc than

the ULs, with no detectable evidence of any harm.[59]

ZINC IN FOODS

The highest concentrations of zinc are found generally in animal products, especially meats; whole grains, nuts, and legumes also contain substantial quantities of zinc, but the absorption of zinc from these foods is less bioavailable due to phytic acid. The zinc content and bioavailability of typical complementary foods for the older breast-fed infant vary widely both in developed and developing countries.[44,60] As noted above, unfortified plant food sources of zinc are often inadequate to meet the needs of the older infant and toddler. Several ready-to-eat breakfast cereals and some infant cereals in the U.S. are now fortified with zinc as well as iron and other micronutrients. The zinc content of human milk declines by several-fold from colostrum to mature milk. Concentrations seem to be minimally influenced by maternal zinc intake.[61] Infant formulas and a wide range of special formulas are now routinely zinc fortified. An overview of the zinc and copper content of major food items is given in Table 8-5.

COPPER

Copper-dependent redox reactions developed very early in the evolutionary process. Because the earth's atmosphere changed from a reducing to an oxidizing environment, copper, together with iron, has been involved in the active sites of respiratory proteins. Recognition of copper as an essential trace mineral for humans followed demonstration of a role in the treatment of some cases of hypochromic anemia in infants. Copper deficiency has subsequently been identified in several special circumstances, but its frequency and the extent of its clinical importance in some conditions remain unclear, for example, in the very low birth weight premature infant.

Although beyond the scope of this chapter, an introduction to copper would not be complete without reference to two inborn errors of copper metabolism, which have attracted more clinical interest than has human dietary copper deficiencies. The central role of copper in the pathophysiology of Wilson's disease was recognized in 1945 and of Menkes' Steely Hair Syndrome (MSHS), or now termed simply Menkes disease, in 1962. The research stimulated by these inborn errors of copper metabolism has led to a clearer understanding of normal human copper

metabolism. It is now recognized that these two conditions represent a disruption of copper transport across cell membranes. The two homologous cation-transporting P-type adenosine triphosphatases (ATPases), the Menkes' P-type ATPase (MNK protein, ATP7A) and Wilson ATPase (Wilson protein, ATP7B), are the gene products primarily responsible for cellular copper homeostasis. The MNK functions in copper trafficking to the secretory pathway for efflux from cells, including enterocytes. In Menkes' syndrome, there is reduced copper absorption and placental copper transport, resulting in reduced availability of the mineral to peripheral organs and tissues. Although the pathophysiology of MSHS is complex, at least some of the principal clinical features can be attributed to a de facto severe copper deficiency state, including in the brain. The WND P-type ATPase also functions in copper trafficking to secrete excess copper into the biliary canalicular system. In Wilson's disease, mutations of the Wilson ATPase results in copper accumulation, especially in the liver, brain, and cornea.[62] Hence, recognition of the role of disordered copper metabolism in these diseases has helped to improve our understanding of the effects of severe human copper deficiency and of copper toxicity.[63,64]

TABLE 8-5 Trace Element Content of Common Foods

Item	Zinc (mg/100 g)	Copper (mg/100 g)
Meat group		
Regular ground beef	4.4	0.06
Round steak	5.8	0.05
Beef liver	5.1	4.6
Chicken—drumstick, fried	2.7	0.01
Pork chop	2.4	0.01
Bologna, all meat	1.8	0.02
Hot dog, all meat	1.6	0.08
Fish group		
Oyster, raw, Atlantic	74.7	3.62
Perch fillet	1.0	0.21
Tuna—in oil, drained, solids	1.1	0.01
Dairy and egg group		
Whole milk	0.4	0.005

Cheddar cheese	3.1	0.11
American cheese	3.0	0.11
Ice cream, vanilla	1.06	0.005
Eggs, hard-boiled	1.0	0.05
Infant formula	0.5	0.06
Nuts and legumes		
Common beans, cooked (red, kidney, pinto)	0.6	0.25
Peanut butter	2.9	0.61
Roasted peanuts	3.0	0.43
Bread and cereal group		
Whole wheat bread	1.0	0.17
Graham crackers	1.1	0.04
Corn flakes	0.3	0.02
Oatmeal, cooked	0.5	0.02
Macaroni, cooked	0.5	0.02
Rice	0.4	0.02
Vegetable and fruit group		
Green beans, canned	0.3	0.02–0.04
Potatoes, boiled	0.3	0.10
Potato chips	0.8	0.29
Lettuce	0.4	0.04
Apple	0.05	0.01
Banana	0.2	0.11
Orange juice	0.02	0.008

CLINICAL FEATURES OF COPPER DEFICIENCY

Copper deficiency usually occurs during development. The principal features of copper deficiency are hypocupremia, a hypochromic, normocytic anemia that is unresponsive to iron therapy, neutropenia, cardiovascular disease and osteoporosis later in life. The bone marrow shows megaloblastic changes and vacuolization of the erythroid series. Iron deposits may be seen by electron microscopy in mitochondria and in some of the cytoplasmic vacuoles in red blood cell precursors. Maturation arrest of the granulocytic series may be evident. Iron stores in the intestinal mucosa, reticuloendothelial system, and liver cells are increased. The anemia is initially hypoferremic, but later becomes hyperferremic because of the decreased uptake of transferrin-bound iron by the developing

erythrocytes. Early radiologic findings are osteoporosis of the metaphyses and epiphyses and retarded bone age. Typical findings in the established case are increased density of the provisional zone of calcification and cupping with sickle-shaped spurs in the metaphyseal region.[65] Other skeletal abnormalities include periosteal layering and sub metaphyseal and rib fractures.

Other clinical findings have been observed in association with copper deficiency, including pallor, decreased pigmentation of skin and hair, prominent superficial veins, skin lesions similar to those of seborrheic dermatitis, cardiac hypertrophy, and hepatosplenomegaly. Features suggestive of central nervous system involvement are hypotonia, lack of interest in outside surroundings, psychomotor retardation, ataxia, apparent lack of visual responses, seizures and apneic episodes.[66] Demyelination and myeloneuropathy have been reported in case reports of adults with hypocupremia, in one case with overuse of zinc supplements and in another with an apparent zinc overload syndrome without clear exposure to excessive zinc.[67,68] Thus, caution is warranted and zinc intake should be in proportion to copper in order to avoid potential adverse effects. In Menkes', there is severe and eventually fatal central nervous system involvement owing to disordered copper metabolism within the brain cells, which probably results in localized intracellular deficiencies at physiologically important subcellular sites. Other features attributable to copper deficiency in this complex entity include the hair changes, which are reminiscent of the wool of copper-deficient sheep and which led to the initial recognition of the role of disordered copper metabolism; hypothermia; and evidence of defective elastin and collagen formation.

BIOCHEMISTRY AND BIOCHEMICAL CORRELATES

Copper is an essential component of several oxidase enzymes, the reduced activity of which can explain many of the features of copper deficiency. Cytochrome oxidase is the terminal enzyme in the electron transport chain and catalyses the oxidation of reduced cytochrome *c* by molecular oxygen, which is itself reduced to water. This enzyme, which is necessary for normal cellular respiration, has been shown to be markedly reduced in the brains of copper-deficient lambs suffering from swayback.

More than 90% of the circulating copper in plasma is tightly and specifically bound to ceruloplasmin, a glycoprotein that contains six

copper atoms per molecule. The functions of this protein, which was first identified in 1948, have been and continue to be the focus of considerable interest and controversy. Roles in copper metabolism include copper transport, mobilization of iron to transfer in the plasma, and regulation of the biogenic amines.[69] Ceruloplasmin (ferroxidase I) exhibits enzymatic ferroxidase activity and is necessary for the oxidation of ferrous iron stored in the liver and bone marrow to ferric iron. This is an essential step in the release of iron from body stores and its attachment to transferrin for transport to developing erythrocytes in the bone marrow. Aceruloplasminemia, an autosomal recessive condition in humans, does not produce obvious disordered copper metabolism, but results in iron accumulation in tissues.[70] Depressed activity of the ferroxidase role of ceruloplasmin is likely to be related to the hypoferremic, hypochromic anemia of copper deficiency. However, there are, in addition, abnormalities of iron metabolism within the erythroid cell line, and it appears that intracellular transport of iron is defective, so that, despite an abnormal accumulation of iron, there is a lack of iron available for normal heme synthesis in the mitochondria.[69] The pathologic mechanisms for the neutropenia remain unexplained.

The bone lesions of copper deficiency are attributable, at least in part, to a lack of copper dependent lysyl oxidase, which acts on lysine and hydroxylysine side chains of collagen and elastin, and is required in the cross-linking of these connective tissue components. These defects are responsible for the gross structural defects seen in arteries and veins in copper-deficient animals and in MSHS.

Among other copper-containing oxidases is tyrosinase, which catalyzes the first two steps in the oxidation of tyro-sine to melanin. Lack of pigmentation is a prominent feature of copper deficiency in some species, but is only mild in human copper deficiency. Copper proteins previously given other names such as cerebrocuprein, hepatocuprein, and erythrocuprein are now known to be superoxide dismutase, which catalyzes the dismutation of superoxide-free radical ions.

Superoxide dismutase protects against oxidative damage. Detoxification of dangerous free radicals within the cell is of the greatest importance, but little is known about the activity of this enzyme in human copper deficiency states and what role decreased activity may play in the pathophysiology of copper deficiency.

ETIOLOGIC FACTORS IN COPPER DEFICIENCY

The first cases of human copper deficiency were reported in infants and children in Peru who were recovering from protein-energy malnutrition. Prior to their initial hospitalization, the infants had long histories of inadequate nutrition, which consisted mainly of diluted cow's milk, which is a relatively poor dietary source of copper. Copper supplementation was associated with improved growth rates in copper-deficient Chilean infants recovering from malnutrition.[71]

The term infant accumulates substantial hepatic stores of copper, which is attached to metallothionein, during the last three months of gestation. These stores are released postnatally and are thought to ensure protection of the young infant against copper deficiency. Very low birth weight infants born prematurely do not have the benefit of these stores and have very little copper at birth. Although premature infants are at risk for copper deficiency, positive copper balance is apparently achieved in early postnatal life in premature infants with current fortification of human milk and formulas, although the adequacy of retention is not yet clear.[5] Other causes of human copper deficiency include intestinal malabsorption syndromes, prolonged parenteral nutrition without copper, chronic diarrhea, short gut syndrome, and chelation therapy.[72,73] Extremely high intakes of zinc can precipitate a deficiency of copper,[67] but, as described above, this is unlikely under typical levels of zinc supplementation.

DIAGNOSIS OF COPPER DEFICIENCY

Both serum copper and ceruloplasmin levels are depressed in copper-deficiency states and are often used as biomarkers to determine moderate and severe copper deficiency. Unfortunately, these parameters are also influenced by other factors, some of which are likely to be operational when the possibility of copper deficiency, especially marginal, is considered. Ceruloplasmin synthesis is limited in the liver of the term neonate, and neither serum copper nor ceruloplasmin concentrations reach adult levels until at least 1 month of age. In the very low birth weight infant, copper concentrations are even lower, averaging about 0.30–0.40 μg/mL, and do not start to increase until 41 weeks of gestational age.[74] It has been suggested that concentrations lower than 0.25 μg/mL may be abnormal in the young preterm infant and may be associated with neutropenia. However, the interpretation and value of the data on serum copper levels prior to 40 to 44 weeks of gestational age remain uncertain. By 6 months of postnatal age, serum copper and ceruloplasmin

concentrations are similar to, or higher than, adult levels.[75] In addition, ceruloplasmin synthesis in the liver may be reduced in protein-energy malnutrition, and protein-losing enteropathy is associated with losses of ceruloplasmin. Based on a recent systematic review of 16 studies and 16 possible copper biomarkers, serum copper concentration remains the most commonly used population-based biomarker.[76] However, it is not reliable under specific health status/different population groups. For example, hypocupremia is a feature of both Wilson's disease (copper toxicity) and Menkes' disease (copper deficiency). Copper deficiency can also be masked and difficult to confirm by laboratory assays when there is an increase in synthesis of ceruloplasmin, which is regarded as an acute-phase reactant and is elevated in response to inflammatory stimuli. Hence, hypercupremia occurs, for example, in association with infections, in the postoperative period, and in patients with neoplasms. Several hormones cause hypercupremia, most notably oestrogens, and levels are increased substantially throughout pregnancy and for several weeks postpartum. Hypercupremia also occurs in obstructive liver disease owing to failure of the normal biliary excretory mechanisms.

Erythrocyte superoxide dismutase (ESOD), a copper dependent enzyme, has been investigated as an index of copper status and has been reported to be an early indicator of disturbed copper metabolism.[77] Recently, another copper transporter, copper chaperone for SOD1 (CCS) has been proposed to be more sensitive to changes in copper status.[52] Other potential indicators, such as leukocyte superoxide dismutase, plasma glutathione peroxidase and leukocyte cytochrome *c* oxidase, etc., require more research to draw confirm conclusions.[76]

Radiologically, separation of the epiphyses, an absence of metaphyseal fraying, and the increased density of the provisional zone of calcification may all help to distinguish copper deficiency from rickets. In contrast to scurvy, copper deficiency does not produce either a lucent line beneath the increased density of the provisional zone of calcification or a "corner sign," nor does it cause hemorrhages.

The diagnosis of copper deficiency should be considered in the presence of suggestive aetiologic circumstances, particularly when there is a hypochromic anemia unresponsive to iron therapy. One case series also documented improvement in erythropoietin-resistant anemia in adult hemodialysis patients after correction of copper deficiency.[78] If the anemia is accompanied by neutropenia and osteoporosis, together with low ceruloplasmin or plasma copper levels, the diagnosis becomes likely.

Confirmation will be provided by a prompt response to copper therapy with a reticulocytosis and increases in circulating neutrophils and in ceruloplasmin or plasma copper levels.

TREATMENT OF COPPER DEFICIENCY

Nutritional copper deficiency in infants has been treated successfully with 2–3 mg/day of copper sulfate as a 1% solution, which provides 400–600 μg of copper. Maintenance requirements for copper during intravenous nutrition are 0.5–1.0 mg/day for adults; as little as 20 μg/kg/day appears to be adequate for infants.[7] Although higher quantities may be required to achieve a positive balance, there is concern that the immature liver of the pre-mature infant may be prone to cholestasis and thus to hepatotoxicity from intravenous copper. Evidence suggests that parenteral copper supplementation (20 μg/kg/day) in cholestatic infants does not cause copper toxicity or impaired liver function.[79]

TOXICITY

Copper toxicity has resulted from the accidental or deliberate ingestion of excess copper, hemodialysis with copper-contaminated solutions, and absorption of topically administered copper salts. Acute poisoning by the oral route causes a metallic taste, nausea, epigastric pain, green-blue vomitus, and diarrhea. Jaundice and hepatomegaly may occur 2–3 days later. Laboratory parameters are indicative of hepatic injury, and there is histologic evidence of centrilobular necrosis and biliary stasis. Jaundice may also result from intravascular hemolysis caused by free copper ions, which also cause hematuria and a direct renal tubular toxic effect, leading to oliguria and anuria. Copper salts may have a cardiotoxic effect, which is responsible for the hypotonia and coma seen in severe cases. Chronic copper poisoning is a rare entity that has been associated with drinking water with a high copper content. Indian child-hood cirrhosis, a hereditary disease associated with greatly increased hepatic copper concentration, is manifest in conjunction with high copper intakes, such as with copper contaminated animal milk introduced at an early age.[80]

Acrodynia and cirrhosis have both been attributed to chronic copper poisoning. A concentration of 2 mg/L in potable water has been recommended as safe for infants without causing gastrointestinal symptoms, and this level has been provisionally endorsed by the WHO. However, higher levels (8 mg/L) in drinking water have not been

associated with apparent toxicity. The DRI's set an upper limit (UL) for copper at 10 mg/d.[6]

ABSORPTION AND METABOLISM

Approximately 35% of ingested copper is absorbed in the stomach and upper small intestine. The percentage of absorption is inversely related to the quantity ingested and has increased in copper deficiency states. The percentage of absorption also depends on the dietary form and on other dietary constituents. Studies using stable isotopes demonstrated a 40% absorption of copper in formula-fed premature infants, whereas absorption from human milk was approximately 70%. Studies in animal models have reported a range of copper absorption from infant formula, from 25% to 70%.[5] Zinc, iron, cadmium, calcium, sulfate, and molybdenum will interfere with copper absorption if given in sufficient quantities. Copper–iron interaction has been studied primarily in short terms. Iron supplementation may not affect copper absorption in infants in the longer term[81] and more research is still needed. In contrast to other trace elements, cellulose and phytate have not been found to have marked effects on copper absorption.

Absorbed copper passes through the portal circulation attached to albumin and is taken up by the hepatocytes. Copper may then be used by the hepatocytes for the synthesis of cuproproteins, especially ceruloplasmin, which is released 3 days later into the systemic circulation. Excess copper is excreted with the bile. Ceruloplasmin copper accounts for 90%–95% of the circulating copper in plasma. The highest copper concentrations are found in the liver, brain, heart, and kidneys. Because of their large mass, bone and muscle contain 50% of the body copper, whereas the liver contains only 10%. The liver of full-term infants contains 200–400 μg of copper/g of dry weight, which is 10–20 times the adult concentration (12–48 μg/g dry weight). Forty percent of neonatal hepatic copper occurs in the lysosomes attached to metallothionein. In contrast to liver copper, brain copper increases twofold from birth to adulthood. The highest concentrations are in the locus ceruleus of the brainstem and in the cortical gray matter. In erythrocytes, the copper concentration is similar to that of normal plasma (75–120 μg/dL). This copper concentration remains stable; 60% is associated with superoxide dismutase. The total copper content of an adult man is approximately 75 mg.

DIETARY REQUIREMENTS

Earlier, minimal estimates of copper requirements for older children and adults were 2 mg/day, and most diets were thought to contain at least this quantity. The most recent EAR, based on studies in normal adults under experimental depletion conditions, was set at 700 µg/day for adult men and women. The RDA was set at 900 µg/day.[6] Most recent national survey data indicate that median copper intakes for men and women are 1.0–1.6 mg/day. Six RDAs were established for children by extrapolation from the adult EAR; these are given in Appendix 2.

The young term infant is protected from copper deficiency by hepatic stores accumulated in utero. Moreover, the term infant usually achieves a positive copper balance by 1 week of age or a little later. The Food and Nutrition Board established an Adequate Intake of copper of 200 µg/day (30 µg/kg/day) for infants 0–6 months of age and 220 µg/day (24 µg/kg/day) for infants 7 to 12 months.[6] The estimated dietary requirement of the very low birth weight premature infant is approximately 100 µg/kg/day of copper.[5]

FOOD SOURCES

The richest sources of copper are oysters, followed by nuts, liver, kidney, corn oil, margarine, and dry legumes. The contribution of drinking water to the total copper intake varies with the type of piping and the hardness of the water. Information on the copper content of food items remains inadequate for optimal calculations of dietary intakes. Moreover, copper intake from diet is often overestimated. Several studies that compared calculated versus chemically analyzed dietary copper showed an average 77% higher estimation of dietary copper using food composition tables.[82] Thus, the actual copper intakes in adults from the national survey may be well below 1.0 mg/day, and more people may be consuming less than the RDA of copper.[83]

Infant formulas in North America now contain a minimum of 60 µg/100 kcal of copper, and those designed specifically for the premature infant contain substantially higher quantities, with a recommended range of 100–250 µg/100 kcal.[5] However, on a worldwide basis, there is still considerable variation in the copper content of infant formulas, and some contain higher quantities. Unprocessed cow's milk contains about 150 µg/L of copper, which is lower than that usually found in human milk (200–600 µg/L of copper) at any stage of lactation. Preterm infants have a

lower copper store and may require more copper from the diet. Preterm milk may not contain adequate copper to meet the needs of the infants.[84]

SELENIUM

Interest in the toxicity of selenium in mammals developed in the 1930s with the discovery of both acute and chronic forms of selenosis among grazing animals in Nebraska and Wyoming. The proved toxicity of selenium in animal husbandry in "natural" circumstances (in seleniferous areas) has resulted in extreme and only partially justified caution in adding selenium supplements to diets that are low in selenium. A beneficial role for selenium was not demonstrated until 1957, when selenium supplements were found to protect against liver necrosis in rats fed certain diets. The following year, selenium deficiency was shown to cause naturally occurring muscular dystrophy in lambs and calves, a problem that occurs in many areas of the world. Earlier nutritional studies indicated that there was a close association between selenium and vitamin E. In 1973, selenium was shown to be an integral component of glutathione peroxidase (GSHPx), an enzyme that destroys hydroperoxides. Although numerous other selenoproteins have been identified, the functions of many of these proteins are not well characterized. The selenoproteins iodothronine deiodinases (Types I–III) catalyze deiodination of thyroxine, triiodothyronine and reverse triiodothyronine, and as such play an active role in regulation of active hormone triiodothyronine. Experimentally, selenium restriction has been shown to cause hypothyroidism. The thioredoxin reductases, selenoprotein P, and selenoprotein W all are involved in oxidant defense systems.

Although low selenium states have been recognized in certain populations, notably in New Zealand, for many years, it was not until the 1970s, when a geochemical deficiency of selenium in a large area of China was recognized as the major etiologic factor in Keshan disease (KD), that the clinical importance of human selenium deficiency was unequivocally demonstrated. Geochemical depletion of both selenium and iodine occurs when surface soils are depleted by erosion and glacial washout, resulting in both plant and animal foods from such areas contain relatively low amounts of both trace elements. Individual cases of selenium related cardiomyopathy, as well as skeletal myopathies, have since been identified in North America and elsewhere, and less well-characterized milder selenium deficiency syndromes have also been reported.[85] Recent discovery of disease-associated polymorphisms in selenoprotein genes has

garnered attention to the broad roles of selenoproteins to health, including immune function, cancer and cognitive function. Selenium is recognized to have a U-shaped link with status, such that increased selenium intake may benefit people with low status, whereas those with adequate-to-high status might be affected adversely.[86]

CLINICAL FEATURES AND ETIOLOGY OF SELENIUM DEFICIENCY

Keshan disease (KD) is an endemic cardiomyopathy affecting children and women of childbearing age. The disease, named for a county in northeastern China, has a clear-cut geographic distribution in a broad area of China from the northeast to the southwest, an area in which the selenium content of food staples has been shown to be exceptionally low.[87] A series of controlled selenium supplementation studies was associated with an 80% reduction in the incidence of KD, whereas the mortality of identified cases was only 1% of that of untreated children.[88] Some of the etiologic circumstances of KD have not been explained clearly. The seasonal and annual fluctuations in KD have raised the possibility of an interaction between nutritional state and an infectious etiology. The recognition that selenium deficiency increases the cardiovirulence of coxsackievirus in animal models by changing the viral genome sequence is particularly intriguing.[89] Further supportive evidence is the isolation of coxsackievirus from patients with KD in the selenium-deficient area of China, although not all serotypes are equally pathogenic.[90] More studies will be required to determine whether the observations in animal models are relevant to humans with KD. However, the permissive role of selenium deficiency and the public health benefits of selenium supplementation in prevention of KD have been established convincingly.[91]

The main pathological feature of KD consists of multiple focal areas of myocardial necrosis. Presentation may be acute, with heart failure, arrhythmias, and cardiogenic shock, or chronic, with cardiomegaly with or without congestive failure. Electrocardiographic features include low-voltage ST-T wave changes, right bundle branch block, or complete atrioventricular block. Serum selenium levels in KD are extremely low, typically less than 10 μg/L. Other laboratory indices are correspondingly low. Long-term total parenteral nutrition devoid of selenium supplements can also cause severe selenium deficiency with cardiomyopathy or skeletal

myopathy.[92,93]

Kashin-Bek disease (KBD), a degenerative osteoarthropathy named for two investigators who studied patients in Siberia in the nineteenth century, has a geographic distribution similar to that of KD within China. KBD is an endemic osteoarthropathy involving children aged 5–13 years, and typically is endemic in selenium and iodine deficient geographic areas, including in China and central Africa. The disease is characterized pathologically by degeneration and necrosis of multiple articular cartilages and growth plates, causing enlargement, especially of fingers, toes, and knees and shortened extremities. Disabilities are typically permanent. Millions of people are likely to be affected with KBD. The relationship between selenium deficiency and KBD is unclear. Evidence from epidemiologic studies suggests that selenium and iodine deficiencies may interact to cause the condition. In Tibet, a comparison of those with and without KBD showed a selenium deficiency in both groups, but iodine deficiency was more severe in those with KBD. A randomized controlled trial of selenium supplementation, initiated after correction of iodine deficiency, in Tibetan children with KBD resulted in no effect on KBD, growth or thyroid function. The authors concluded that correction of iodine deficiency was more efficacious than treatment of selenium deficiency for KBD.[94]

An adequate supply of selenium is essential for normal thyroid hormone biosynthesis. For example, in goitrous children, the severity of selenium deficiency predicted the response to iodine supplementation, both with respect to reduction in thyroid size and to improved biochemical measures of thyroid function.[85] Impaired T_4 conversion to T_3 and low regulation of T_3 production have been reported in children with phenylketonuria (PKU) who were fed a phenylalanine-restricted diet that was also low in selenium,[95] but this has not been found consistently in children with PKU, even those with evidence of selenium deficiency and compromised antioxidant status.[96]

Moderately depressed levels of erythrocyte glutathione peroxidase activity, hair selenium, serum selenium, or blood selenium have been found in association with protein-energy malnutrition in developing countries and in low birth weight infants.[5] One report found significantly lower selenium levels in Malawian children with kwashiorkor who had congestive heart failure compared with those who did not develop this complication.[97] The clinical significance of these low selenium states remains unresolved. Milder degrees of selenium depletion, notably those

associated with low dietary levels in New Zealand and certain other geographic areas, have not been associated with adverse clinical consequences. There have been case reports of children on long-term parenteral nutrition or synthetic oral diets without selenium supplements who had macrocytosis and loss of hair pigment, both of which normalized following selenium supplementation.[98]

Interest in selenium in adult and pediatric patients with human immunodeficiency virus (HIV) has been prompted by observations suggesting an ameliorating role in the pathophysiologic processes of this infection. The effects may be mediated by its antioxidant effects, especially via glutathione peroxidase (GPx), and possibly via inhibition of viral replication.[99] In a large cohort study in Tanzania in children born to HIV-positive mothers, plasma selenium levels were inversely associated with risk of all-cause mortality.[100] Interpretation of these findings is complicated by the likelihood of co-existence of multiple nutritional deficiencies and by the fact that plasma selenium is impacted by acute inflammation.

BIOCHEMISTRY AND METABOLISM

Several selenium proteins have been identified; two of these have clearly established physiologic roles: glutathione peroxidase and iodothyronine deiodinase (ID). Selenium is an essential component of cellular glutathione peroxidase (GPx), its plasma homologue, and the membrane-bound phospholipid-hydroperoxide GPx. Glutathione peroxidase uses two molecules of reduced glutathione to reduce hydrogen peroxide and convert it to two molecules of water. This enzyme also catalyzes the reduction of fatty acid hydroperoxides to hydroxy acids in the tissues and thus helps to protect the lipids in cell membranes from peroxidation. GPx is present in a wide variety of tissues and accounts for 90% of the selenium in erythrocytes. Messenger RNA levels of liver GPx decrease with dietary selenium restriction.

ID has been identified as a selenoenzyme. This enzyme catalyses the conversion of T_4 to T_3 in the liver and other tissues. Selenium, as selenocysteine, is an integral component of ID, and its expression is influenced by dietary selenium. This enzyme provides the link between iodine and selenium because both are involved in thyroid hormone production.

Absorption of selenium depends to some extent on its chemical form.

Selenomethionine is better absorbed than inorganic forms of selenium. Intestinal absorption of food selenium is as high as 80%. Selenium homeostasis in the rat is controlled by the kidney, and the principal route of excretion in humans is the urine. The highest concentrations in the body are found in liver, tooth enamel, and nails. In the body, selenium is intimately associated with specific proteins, especially as selenocysteine. For example, more than 80% of the selenium in the rat is present as selenocysteine.

DIAGNOSIS AND TREATMENT OF SELENIUM DEFICIENCY

Selenium depletion is associated with low levels of selenium in whole blood, plasma, and hair and low erythrocyte GPx activity. Typical clinical assessment is achieved with plasma selenium concentrations; the 5th and 95th percentile range from the Third National Health and Nutrition Examination Survey (NHANES) was 100–154 μg/L (1.27–1.95 μmol/L). Plasma selenium concentrations less than 50 μg/L have been reported in healthy adults from low selenium areas. Selenium levels are low in young infants and gradually increase to adult values over a period of months. A strong positive correlation between erythrocyte GPx activity and whole blood selenium levels has been found in some populations. However, it is not yet clear how well these indicators may correlate with clinical features of selenium deficiency.[101]

The optimal treatment for selenium deficiency has not been determined. Currently, there are no clear-cut indications for oral selenium therapy apart from the prevention of KD, for which a weekly supplement of 500 μg in younger children and 1,000 μg in older children appears to suffice. On a daily basis, these quantities are somewhat above the RDA recommended by the Food and Nutrition Board.[101]

Selenium supplements should be added routinely to intravenous infusates, at least for those patients who receive a substantial portion of their nutrient intakes by the intravenous route for more than 2 weeks. The dose recommended is 2 μg/kg body weight/day.[7]

TOXICITY

Although excess selenium can cause undesirable consequences in agriculture, chronic selenium toxicity does not appear to be a problem in

humans. Blood selenium levels are elevated in children living in seleniferous areas of Venezuela, but the only clinical findings were the some-what poorly defined ones of nausea and pathologic nails. Loss of hair, brittleness of fingernails, garlic odor, a higher incidence of dental caries, and increased fatigue and irritability were reported to be signs of selenosis in an earlier survey of residents of South Dakota, where selenium toxicity occurred in farm animals.

Acute selenium poisoning is rare in humans but has been reported as a result of self-administration of selenium supplements. The outcome of acute toxicity appears to depend on both the amount ingested and the type of selenium compound. Acute ingestion of sodium selenite caused garlic odor on the breath and diarrhea in several cases. Evidence of mild, temporary liver damage and cardiac abnormality was seen in one case, but there were no permanent sequelae.

The highest selenium concentration observed in human milk in the United States, 60 µg/L, was used as the basis for the "no observed adverse effect level" in the 2000 DRIs,[101] and the DRI upper limit was set at 45 µg/day (7 µg/kg) for term infants 0–6 months. This amount was used parenterally in a supplementation trial of very low birth weight infants until the infants advanced to enteral feeds, at which time they received ~5µg/kg until 36 weeks postmenstrual age, with no apparent adverse effects.[102]

DIETARY REQUIREMENTS

Dietary surveys of selenium intake in endemic and adjacent nonendemic KD areas have led to the conclusion that selenium requirements for adult Chinese men and women are 19 and 13 µg/day, respectively.[132] This is the quantity calculated to be necessary to prevent KD. RDAs in this country have been set at 55 µg selenium/day for adult men and women.[101] Evidence from multiple studies suggests a link between selenium intake and an increased risk of type 2 diabetes in the US population, especially for those consuming higher than 55 µg selenium/day.

An AI was established for infants in 2000, recommending 15 µg/day (2.1 µg/kg/day) for infants up to 6 months of age, based on a mean selenium concentration of 18 µg/L in human milk and an average intake of 780 mL/day. The AI for older infants and young children (7–36 months) is 20 µg/day, for older children (4–8 years) 30 µg/day, and for adolescents 40–55 µg/day.[101] These recommendations have been extrapolated from adults with a generous addition for growth; actual requirements are

substantially less. Under conditions of illness in pediatric patients (i.e., burn injuries and sepsis), selenium requirement may be higher than the recommended level to maintain an optimal plasma glutathione peroxidase level.

FOOD SOURCES

The selenium content of human milk depends on maternal dietary selenium intake. Extremely low values, that is, less than 3 µg selenium/L, have been reported from areas where KD is endemic.[5] Concentrations between 5 and 10 µg selenium/L have been reported from other countries where maternal selenium intake is relatively low (e.g., New Zealand, Finland, and Poland).[103] In the United States, the mean value of the selenium content of human milk is approximately 18 µg selenium/L.[5]

The selenium content of cow's milk is generally lower than that of human milk in this country. The selenium content is reduced in the preparation of cow's milk–based infant formulas. The minimum for standard infant formulas was recommended to be 1.5 µg/100 kcal.[104] Whether the low plasma selenium and GPx levels in low birth weight infants are increased by providing generous selenium supplementation of formulas has been controversial, and feeding regimens that have provided amounts adequate to increase biochemical markers of selenium status in low birth weight infants have not demonstrated clear clinical benefit. Nevertheless, premature infants are born with strikingly low serum selenium levels and GSPx, and, without supplementation, these levels decline further.[102] An expert panel recently recommended a minimum concentration in formulas for preterm infants of 1.8 µg/100 kcal and a maximum of 5.0 µg/100 kcal, while acknowledging that it was unknown whether the lower amount would be adequate or optimal for preterm low birth weight infants.[5]

As noted above, concentrations of selenium in foods vary by geographic region and soil content. Generally, seafood (approximately 0.5 µg/g), kidney, liver, and meats (approximately 0.2 µg/g) are the best sources of selenium. Whole grains are also generally good sources of selenium provided that the soils in which they are grown are not low in selenium. Vegetables and fruits, with the exception of garlic, provide little. Daily intake in North America has been reported to range from 60 to 220 µg/day, but in New Zealand, intake is only 6 to 35 µg/day.

CHROMIUM

A role for chromium in mammalian physiology was indicated first in 1959, when it was reported that trace amounts of this element are necessary for normal glucose tolerance in the rat. Subsequent research suggested that chromium acted as a cofactor for insulin. These observations stimulated considerable interest in a possible role for chromium deficiency in the high incidence of impaired glucose tolerance in industrialized societies. Progress has been hampered, however, by a lack of laboratory indices of chromium status that would be of the greatest value in identifying appropriate subjects to include in clinical trials of chromium supplementation.

CLINICAL FEATURES AND ETIOLOGY OF CHROMIUM DEFICIENCY

Experimental chromium deficiency in several animal species has been reported to result in impaired glucose tolerance in the presence of normal concentrations of circulating insulin. Features of chromium deficiency have been reported to include impaired glucose tolerance with relative insulin resistance and peripheral and/or central neuropathy. Weight loss and poor childhood growth have also been reported.

In the pediatric age range, chromium supplementation studies have been undertaken in infants and young children suffering from protein-energy malnutrition. The results of these studies, which involved the use of a single oral dose of 250 μg trivalent chromium, suggested that chromium deficiency may be one factor responsible for the impaired glucose tolerance that occurs in association with severe generalized malnutrition. Chromium deficiency has been reported to complicate protein-energy malnutrition in some, but not all geographic areas. Lack of controls in some studies and limited subject numbers hamper the definitive interpretation of these data.

Several trials of chromium supplementation have been reported in adults, including elderly subjects.[105] Most of these trials have been small. Subjects have included noninsulin-dependent diabetic patients and otherwise normal subjects with evidence of impaired glucose tolerance. Supplements have always been trivalent chromium, on some occasions as an inorganic salt and on others as brewer’s yeast or in other forms that are thought to have favorable absorption. In several studies, chromium supplementation has been associated with improved glucose tolerance in

more than 50% of the chromium-supplemented subjects. Other apparent effects noted in some studies include a decrease in insulin response to an oral glucose load, a decrease in total blood lipids, and an increase in high-density lipoprotein cholesterol. A meta-analysis of chromium supplementation trials reported for healthy individuals and in those with glucose intolerance or type 2 diabetes found no effect of chromium on glucose or insulin concentrations in nondiabetic subjects, whereas the results from trials in diabetic subjects were inconclusive.[106]

Chromium deficiency has also been reported in patients receiving long-term parenteral nutrition without supplementation. Clinical features attributed to the chromium deficiency were weight loss, peripheral neuropathy, and encephalopathy. An abnormallylow removal rate of intravenous glucose and an abnormally low respiratory quotient indicating difficulty in metabolizing glucose for energy, dependence on exogenous insulin, and abnormalities of nitrogen and lipid metabolism all responded rapidly to intravenous chromium.[6]

BIOCHEMICAL CORRELATES

Although chromium may have several physiologic roles, for example, in nucleic acid metabolism, the role that has received most attention is its action as a cofactor for insulin. Experimental evidence is consistent with the hypothesis that chromium acts as a cofactor for insulin at the site of peripheral insulin receptors. Specifically, it has been hypothesized that a chromium-binding oligopeptide, termed chromodulin, facilitates the attachment of insulin to its receptors both at the cell membrane and intracellularly, and activates the insulin receptor kinase activity. Chromium may have to be in the trivalent ion form (Cr^{3+}) to be biologically active.[107] The biologic role of chromium as a cofactor for insulin requires further clarification and confirmation.

DIAGNOSIS OF CHROMIUM DEFICIENCY

The lack of established laboratory assays is especially evident for chromium, and deficiency states can be confirmed currently only by means of careful monitoring of the effects of chromium supplementation under controlled conditions. Normal values in such samples as blood plasma and urine have had a disconcerting tendency to fall as analytic techniques improve so that they remain near the detection limits of the equipment available at any particular time. Currently, normal levels for plasma

chromium, for example, are considered to be only about 0.1 parts per billion (100 pg/mL). This means that it continues to be extraordinarily difficult to measure low values accurately. An additional difficulty in interpretation results from uncertainties about the physiological meaning of low chromium levels. For example, it is thought that solitary plasma chromium levels do not reflect the body chromium status but only recent intake. However, low plasma chromium levels have been reported in chromium-deficient subjects, in pregnant women, and in association with experimental infections that cause impaired glucose tolerance.[6] There have been conflicting reports about a measurable increase in plasma chromium in response to a glucose load. At best, this is a tedious and difficult test for assessing chromium status. Absence of a detectable plasma chromium "response" has been reported in some diabetic patients, including gestational diabetic patients, and in some elderly people. An inverse correlation has been reported between the postprandial increase in plasma chromium and fasting blood glucose. Hair analyses provide only a crude index of chromium status. Theoretically, urine chromium levels, perhaps including measurement of the increase in urine excretion following a glucose load, should provide a useful index of chromium status. To date, however, this approach has not proved to have any conclusive value. Excessive urinary chromium losses have been noted in diabetic patients, and it has been suggested that the low chromium content of adult American tissues could result from excessive urine losses triggered by a diet high in sucrose. Confirmation of excessive chromium losses during the consumption of such diets is lacking. In summary, chromium measurements in tissue and body fluids are of no proven value in the diagnosis of chromium deficiency even if adequate laboratory facilities are available.

TREATMENT OF CHROMIUM DEFICIENCY

Suspected chromium deficiency states can be treated with 200 μg of trivalent chromium daily for several weeks or months in adults. In malnourished children, a single dose of Cr^{3+} 250 μg has been effective. Current recommendations for chromium during parenteral nutrition are 10–15 μg/day for adults or 0.1–0.2 μg/kg body weight/day for infants and young children. Levels considerably lower than this are probably adequate and may be preferable.

TOXICITY

Trivalent chromium has a low toxicity, and there are no reports of toxicity from oral administration. The minimum lethal dose of intravenous chromium as chromic chloride in mice is 800 mg/kg. Hexavalent chromium is more toxic, but 100 parts per million in drinking water can be tolerated by animals for many years. Chromate is a local irritant and is a cause of acute and chronic disease of the respiratory system, including bronchogenic carcinoma in workers in the chromate industry.

Plasma chromium concentrations have been reported to be elevated in patients receiving total parenteral nutrition, including standard quantities of intravenous chromium, who develop evidence of renal disease.[108] Chronic interstitial nephritis has been associated with ingestion of chromium picolinate as a dietary supplement.[109,110] It has been suggested that the renal disease is a result of chromium toxicity, although animal studies have not confirmed such toxicity.[111] It is also possible that the elevated plasma chromium levels are secondary to impaired chromium excretion via the kidneys. It appears prudent to reduce or withhold intravenous chromium supplements in the presence of renal disease.

ABSORPTION/METABOLISM

Inorganic trivalent chromium is absorbed poorly. Absorption of[51] Cr-labeled chromium chloride is limited to approximately 0.5%. A larger percentage of chromium is thought to be absorbed from organic chromium complexes found, for example, in brewer's yeast. Chromium picolinate has been marketed aggressively with claims that it is well absorbed and has some special beneficial metabolic effects, but documentation of these claims is poor. It is thought that the liver may be the site of a specific pool of biologically active chromium and that GTF chromium is released from this pool in response to a glucose load. This response may be mediated by the response of endogenous insulin. GTF has been reported to be the only form in which chromium can cross the placenta to the fetus. The major excretory route for chromium is through the kidney, and the urine contains at least 80% of excreted chromium. The total body chromium content of adults has been estimated to be about 6 mg.

DIETARY REQUIREMENTS AND FOOD SOURCES

Dietary chromium intakes for adults in the United States range from 25 µg/day for women to 50 µg chromium/day for men. The estimated AI for dietary chromium intake for adults set by the Food and Nutrition Board is 35 µg/day for men and 25 µg/day for women.[6] The AI recommended for infants 0 to 6 months is 0.2 µg/day and 5.5 µg/day for infants 7–12 months. The large increase in the second half of the first year of life reflects the contribution of complementary foods. The AIs for children and adolescents range from 11 to 35 µg/day.[6]

The mean chromium concentration of breast milk is approximately 0.25 µg/L.[112] Hence, the intake of the breast-fed infant would be about 0.2 µg/day. The bioavailability of this chromium may be high by comparison with that of other trace elements, but this has not been determined. Concentrations of chromium in cow's milk and infant formulas have been reported to be higher than those in human milk.[104]

Spices, brewer's yeast, liver, and kidney are especially good sources of chromium, but only small quantities are present in fish, vegetables, and fruit. Major losses of chromium have been reported during milling of wheat and refining of sugar, but stainless steel cooking containers may add to the chromium intake. Calculations of chromium in food have undoubtedly varied, depending on methods of sample preparation and analyses. Moreover, absolute quantities may not be as important as the form in which the chromium is present in foods.

MANGANESE

A nutritional requirement for manganese has been established in a wide variety of organisms and was first shown to be "essential" for animals in 1931. Interest was stimulated by the discovery in the 1930s that two economically important diseases of poultry, perosis and chondrodystrophy were caused by manganese deficiency. There are now extensive data on animal requirements, but knowledge of human manganese requirements and status is much more limited.[6]

One probable case of human manganese deficiency has been reported in a subject who was receiving an experimental synthetic diet that was retrospectively found to be low in manganese, providing only 350 µg/day of manganese for 3 months. Features attributed to manganese deficiency were hypocholesterolemia, depressed levels of clotting proteins in the plasma, weight loss, and slow growth of hair and nails. Low serum manganese levels have been reported in some diabetic and in epileptic children, and a negative manganese balance has been observed in children

with pancreatic insufficiency. The clinical significance of any of these observations remains obscure, and no cases of non-experimentally induced manganese deficiency have been reported. Experimental manganese depletion in animal models affects cartilage and bone structure and neurologic function and is associated with stunted growth and ataxia in newborns. The deficiency was also associated with lipid accumulation in the liver and kidneys and with hypocholesterolemia, a finding also observed in humans.

The low manganese concentrations found in blood plasma (approximately 1 ng/mL) and, until recently, the difficulty of obtaining accurate analyses has hindered progress in our understanding of manganese status in humans. Erythrocyte, plasma, and serum manganese concentrations demonstrate an age-dependent pattern, with the highest levels observed in young infants. Blood concentrations increase during pregnancy, which apparently result in high manganese concentrations in the fetus. Infants born prematurely have higher blood levels than term infants, and these relatively high levels persist for several weeks postnatally.[5]

Biochemically, manganese is required for the synthesis of mucopolysaccharides through the manganese-dependent enzymes polymerase and galactotransferase. Reduced activity of these enzymes, resulting in impaired mucopolysaccharide synthesis, accounts for the extensive skeletal abnormalities and the ataxia owing to impaired otolith synthesis that are seen in animals with manganese deficiency. Pyruvate carboxylase is a manganese metalloenzyme, and manganese is a potent, although not necessarily specific, promoter of oxidative phosphorylation. Peptidases, manganese superoxide dismutase, succinic dehydrogenase, and arginase appear to be manganese-dependent enzymes.

Adults absorb less than 5% of an ingested dose of ^{54}Mn.[113] Only half of this amount is retained 10 days later, but retention of the ingested dose at 10 days is 8% in the newborn and 16% in the premature infant. Breast-fed infants maintain positive balance on much lower manganese intakes than formula-fed infants. The higher retention of infants, especially those born prematurely, may reflect higher absorption efficiency, but may also reflect less efficient excretory mechanisms. Manganese absorption is enhanced by iron deficiency, but the long-term effects on retention are less clear. Neither the effects in infants of feeding iron-fortified formula for achieving optimal manganese status nor of iron deficiency on predisposing to manganese toxicity are well understood. The emerging understanding of shared transport of manganese and iron in the brain and the potential for

neurotoxicity highlight the potential importance of the interaction between these elements.[114] Manganese is excreted almost entirely through the bile, and urine losses are small.[113]

Chronic manganese toxicity, owing principally to inhalation from occupational exposure and from excessive parenterally administered manganese, causes extrapyramidal neurologic disease similar to Parkinson's disease, Alzheimer's disease, and Huntington's disease.[115] Such neurologic symptoms are associated with neuronal death in the globus pallidus and basal ganglia, regions which are rich in dopamine. Pilot data in children from the vicinity of a hazardous waste site indicated an inverse relationship between hair concentrations of manganese and arsenic and intelligence scores and memory tests.[116] Cholestatic liver disease, including that associated with total parenteral nutrition, has been associated with manganese accumulation in the liver and brain.[113] Manganese, as well as copper, should be reduced or withheld from parenteral nutrition patients with cholestatic liver disease, and iron deficiency should also be avoided if possible.

Adults are in positive manganese balance with an intake of 2.5 mg/day, but intakes of 0.7 mg/day result in a negative balance. Human milk contains only 3–4μg/L of manganese.[5] Hence, it appears that the requirements of young infants are extremely small. The Daily Recommended Intake (DRI) AI level for healthy 0–6-month-old infants was set at 3 μg/day (0.4 μg/kg), based on the mean manganese intake of exclusively breast-fed infants.[6] Formula-fed infants ingest much more manganese than breast-fed infants, with cow milk based formulas containing approximately a 10-fold greater content compared to human milk, and soy formulas being approximately 10-fold greater than cow milk formulas.[104,113] Although the toxicity of manganese administered by the oral route appears to be low and a wide range of intakes has appeared to tolerated without any clinical problems, recent reports in animal models have reported altered neurobehavioral measures in animals fed soy formula or given manganese supplementation.[117,118] Nuts and unrefined grains are rich sources of manganese, whereas animal products and seafood are relatively low in this trace element.

FLUORIDE

Fluoride has both beneficial (prevention of dental caries) and toxic (fluorosis) effects on teeth, with a small margin between intakes that are

beneficial and those that are toxic.

Fluoride has proved to have beneficial cariostatic properties during tooth development and, for this reason, is generally accepted as a necessary micronutrient. Other beneficial properties, for example, in ameliorating osteoporosis and protecting against arterial calcification, have not been substantiated convincingly.

PREVENTION OF DENTAL CARIES

Fluoride is effective in preventing dental caries. The principal means by which this protection is conferred is via a local effect on the tooth surface. Fluoride decreases demineralization of the enamel when the pH of the dental plaque is low owing to the action of oral bacteria on fermentable carbohydrates. Fluoride also accelerates remineralization when the pH rises, and it appears to decrease production of organic acids from fermentable carbohydrates on the teeth. The fluoride concentration in saliva and in dental plaque fluid necessary to achieve effects is only 0.1 part per million.[119] The systemic effect of fluoride on teeth prior to eruption appears to be small, although some protection may be conferred by incorporation of fluoride into enamel crystals during tooth development.

Prior to the widespread use of fluoridated dentifrices, lifelong use of a water supply containing 1 part per million fluoride resulted in a 50%–60% decrease in the prevalence of dental caries in permanent teeth. The beneficial effect on primary teeth was only slightly less. With the current wide-spread use of fluoridated dentifrices, the benefits of a fluoridated water supply have a somewhat decreased impact on the prevention of dental caries. Systematic reviews have confirmed the beneficial effects of topical fluoride (toothpaste, mouth rinses, varnishes, gels) on prevention of dental caries in children and adolescents.[120,121]

Near-maximal protective effect is achieved with a fluoride concentration of 1 part per million in the drinking water. The beneficial effects are likely to be greatest among children from low-income families, whose use of fluoridated dentifrices is more restricted.

FLUOROSIS

Dental fluorosis is characterized by undermineralization of the surface and subsurface enamel, resulting in increased porosity. This is a developmental abnormality affecting both primary and permanent teeth.[122] Although the

mechanism is not fully understood, fluoride appears to damage the ameloblasts that produce the enamel. The enamel appears opaque. Visible changes range from barely discernible fine white lines across the teeth to entirely chalky-white teeth. The outer enamel may break apart, and the exposed subsurface then becomes discolored.

The incidence of fluorosis has a linear relationship to calculated fluoride intake per unit body weight. This relationship is evident even at levels of fluoride intake that approach zero. The infant is thought to be especially susceptible to the effects of fluoride, with fluorosis resulting once the fluoride intake has reached a level of 40–100 µg/kg/day. The incidence of fluorosis, especially that of a mild degree, appears to have increased recently in communities with and without fluoridated water.[122] The teeth of greatest cosmetic concern are the permanent incisors and cuspids. Enamel formation in these teeth begins at 3 months and is completed by 7 years.

INTAKE OF FLUORIDE BY INFANTS

The fluoride content of breast milk is uniformly low whatever the maternal intake of this trace element. Remarkably, the incidence of dental caries is relatively low in children who have been breast-fed as infants. Fluorosis is uncommon in children who have been breast-fed as infants. The intake of fluoride from unmodified cow's milk is also low, although not as low as from human milk. Fluoride concentrations approximate 40 and 60 µg/L, respectively. The fluoride intake from formula tends to be higher and potentially in a range that can cause fluorosis, even though the fluoride content of water used in the manufacture of infant formulas has been restricted since 1978 and even when dried formulas are made up with water of relatively low fluoride content. For example, the calculated intake of fluoride of a young infant receiving 150 mL formula/kg body weight/day, prepared by mixing powdered milk–based formula (fluoride content = 690 µg/L) with water containing only 200 µg fluoride/L, is 41 µg/kg/day. Ready-to-feed formulas contain < 20 µg fluoride/L.[5] The fluoride content of bottled distilled water is low, but this does not apply to all bottled waters. Weaning foods may also provide significant quantities of fluoride, especially fruit juices, cereals, and poultry products.

Overall, the fluoride intake of young infants is less than it was two decades ago because of a decrease in fluoride content of formulas and because of the increased incidence of breast-feeding. On the other hand, the intake of older infants tends to be higher because of the progressive

substitution of infant formulas for whole cow's milk at this age. Older infants may also start to use fluoridated dentifrices. Small children ingest most of this dentifrice, and the fluoride is subsequently absorbed. The AI for 0–6 months is 10 μg/day (1.4 μg/kg/day).[123] The American Academy of Pediatric Dentistry and the American Academy of Pediatrics recommend no fluoride supplementation for infants under 6 months of age. For older infants and children from 6 months to 3 years for whom the community water supply is less than 0.3 parts per million, a supplement of 250 μg/day is recommended. These recommendations and a broad discussion of the use of fluoride to prevent and control dental caries have recently been published.[121]

IODINE

The clinical importance of adequate iodine intake has, like that of iron, been recognized for many years. Iodine was used in the treatment of goiter in the early years of the nineteenth century, and the major role of iodine deficiency in the etiology of endemic goiter was confirmed early in the twentieth century when the prophylactic value of iodine was demonstrated in Ohio schools. The early recognition of the nutritional role of this trace element can be attributed to the specific nature of the clinical features of iodine deficiency, the relative simplicity of measuring iodine in foods and other biologic samples, and the high concentration of iodine in one specific organ, the thyroid gland. Sixty percent of T_4 by weight is iodine, and the biologic importance of iodine is related entirely to its role in the thyroid hormones.[124]

ABSORPTION AND METABOLISM

Iodine is widely found in its negative ion (iodide, or I). Iodide is rapidly and almost completely absorbed by the human stomach and duodenum. Once enters the circulation, iodine is cleared by the thyroid gland to produce thyroid hormone. The effectiveness of iodine clearance varies by the host's iodine status and iodine intake. Once picked up by the thyroid gland, iodine is oxidized by thyroperoxidase, attaches to thyroglobulin and form T_4 and T_3.

Iodine is picked up by thyroid via active transportation by sodium/iodine synmporter. Some dietary substance found in cruciferous vegetables may compete with iodine for thyroid uptake. Flavonoids from

soy products can impair thyroperoxidase. This impairment does not reach a harmful extent in healthy adults, but may cause hypothyroidism in infants.

DEFICIENCY AND TREATMENT

Iodine deficiency disorders remain a global problem, despite aggressive prophylactic measures, and are the most prevalent cause of brain damage. WHO estimates that there may be nearly a billion people affected by iodine deficiency disorders, and as much as one-half of the world's population is at risk. Iodine deficiency is originally independent of socioeconomic status, but rather dependent on location. Places with low iodine content soil and water have iodine deficient plants and thus humans living in these regions will be iodine deficient as well. WHO recommended salt iodization as early as 1952 and over 70% of the world population now have access to iodized salt. Endemic goiter now occurs mainly in developing countries, typically mountainous areas such as the Andes and Himalayas and the mountainous chain extending through Southeast Asia and the South Pacific. Iodine deficiency also remains a problem in certain parts of Africa. Endemic goiter also persists in the Alps and in several Western and mid-European countries.[124] Iodine deficiency remains one of the four risk factors for impaired child development.[125]

Iodine deficiency causes inadequate thyroid hormone production. Thyroid hormone plays a functional role in different physiological processes, including modulating basal metabolic rate and promoting growth. The extreme result of hypothyroidism and iodine deficiency during development is cretinism. Two types of cretinism are described, neurologic and myxedematous, both of which exhibit severe mental retardation. The neurologic type prevails in most regions and is characterized by severe mental retardation, deaf mutism, spastic diplegia, and strabismus. The cretin appears normal at birth, but is obviously retarded by 6 months of age. There is a characteristic vacant face with a large protruding tongue. The head appears large, and a lordosis is usually prominent. Clinical evidence of hypothyroidism is usually absent. The neurologic damage does not appear to be attributable to maternal or fetal hypothyroidism; there has been speculation that it may be owing to the direct effects of fetal iodine deficiency or to an imbalance between serum T_4 and T_3. In iodine deficiency, T_4 (precursor of T_3) decreases, but T_3 remains normal or even elevated. The myxedematous type of cretin predominantly has the signs of congenital hypothyroidism but typically

has only modest enlargement of the thyroid gland. This type, which mayresult from a combination of a dietary goitrogen, and iodine deficiency, possibly complicated by co-existing selenium deficiency. Glutathione peroxidase is selenium dependent and selenium deficiency causes accumulation of peroxidase which damages the thyroid and impairs thyroid hormone production. Another selenium dependent enzyme, deiodinases converts T_4–T_3 (the active form) by removing one iodine atom. This type predominates in some Central African countries. Endemic cretinism is not a distinct entity in a region of endemic goiter but rather the severe end of a spectrum of neurologic damage.[124]

In mildly iodine-deficient areas, goiters (enlargement of the thyroid gland due to iodine deficiency) may not occur until adolescence or during pregnancy. These goiters disappear in adult males, but in women they become large. Degenerative changes then occur, and myxedema or, paradoxically, sometimes thyrotoxicosis, may follow. Milder cognitive and neuromuscular impairment due to chronic iodine deficiency is much more common.

Early in the 20th century, prophylaxis with iodized salt proved to be highly effective in North America. Although sporadic goiters still occur, these are apparently attributable primarily to unidentified goitrogens. Bread, water, milk, and chocolate are other vehicles that have been used to provide supplemental iodine. In areas of endemic goiter, intramuscular depot injections of 5 mL of iodized oil, providing 400 μg/mL of iodine, have also been used extensively. With the relatively high iodide concentrations now found in foods, especially milk, and the nationwide food distribution system in the United States, iodized salt is no longer considered to be necessary. In contrast, WHO and other international agencies have spearheaded national salt iodization programs in developing countries to eliminate the full spectrum of iodine deficiency disorders. In the 1990s, the number of countries with iodization programs doubled from 46 to 93, and there has been an accompanying improvement in the iodine status of these populations. The sustainability of these programs has been a challenge, however, resulting in persistence of widespread iodine deficiency.

DIAGNOSIS OF IODINE DEFICIENCY

Measurement of iodine excretion in casual or spot urine samples provides an excellent index of recent iodine intake and is a useful assay for assessing community iodide status, although it does not directly reflect

thyroid functions. Urinary iodine concentrations < 50 μg/L indicate moderate to severe deficiency; 100–199 μg/L indicates normal iodine status; > 200 μg/L indicates more than adequate; and > 300 μg/L indicates possible excess.[124,126] Once iodine deficiency is established, T_4 levels will be decreased (normal range equals 0.8–2.4 ng/dL). Thyroid size and thyroid-stimulating hormone and thyroglobulin concentrations reflect chronic iodine deficiency disorders. A urinary iodine concentration survey of school-aged children revealed almost 30% of school-aged children worldwide are iodine deficient and the majority of these children are from southeast Asia and Africa.[127]

IODINE TOXICITY

Excessive iodine consumption can also impair thyroid function. With the increasing levels of iodide in the US food supply, the risk of iodide toxicity is currently of greater concern than that of iodide deficiency. More cases of goiter in this country are currently attributable to excess rather than to a lack of iodide. The effects of excessive iodide intake include thyroiditis, goiter, hypothyroidism, and hyperthyroidism.[124] Large intakes of in adults may cause toxicity, especially in those who have previously had chronic iodine deficiency, which somewhat paradoxically predisposes to toxicity. A large prospective study of adults in China, which examined three distinct geographic areas with different levels of iodine intake, reported significantly higher risk of hypothyroidism and autoimmune thyroiditis in subjects with more than adequate or excessive iodine intake. Adverse effects were greatest in those who had previously been mildly deficient and subsequently increased iodine intake to greater than adequate intake.[128]

FOOD SOURCES

Plants grow in iodine-rich soils and animals feed on these plants have higher iodine content. Seaweed and fish are usually high in iodine. Because iodine distribution worldwide is not even, iodine fortification becomes the most effective way to prevent iodine deficiency. Cow’s milk is a major dietary source of iodide in the United States, one that has increased in recent years. This has resulted not only from use of iodized salt in the dairy industry but also from the use of iodates as sanitizers, especially in teat dips. Iodide concentrations in human milk are also

relatively high in this country (i.e., 140 to 180 µg iodide/L), much higher than the concentration in other countries.[6] The recommended range for term infant formulas is 53–230 µg iodide/L[104]; the comparable recommendation for preterm formulas is 48–280 µg iodide/L.[5] Fish is notable among other foods as naturally rich in iodide. Iodide in water does not usually contribute more than 10% of intake.

DIETARY REQUIREMENTS

The adult EAR of iodide is 95 µg/day or approximately 1 µg iodide/kg body weight/day; the RDA is 150 µg/day.[6] AIs for infants are 110 µg/day before the age of 6 months and 130 µg/day in older infants.[6] The upper limit (UL) for adults is 1,100 µg iodide/day; UL's for children are provided in the Appendix II.[6] These intake recommendations are generous. Some centers do not add iodide to intravenous nutrition because of that already present as contamination, including that derived from iodates used to clean tube connections. As centers have moved away from use of iodates for this purpose, the risk of deficiency may be increased, and routine parenteral administration may be warranted, but data are limited.[129] We recommend providing 1 µg iodide/kg body weight/day, a dose with minimal risk of toxicity.

CONCLUSION

Recognition of the practical importance of the trace elements in human nutrition continues to exponentially increase. Concurrently, there have been exciting advances in understanding of the biology, especially the molecular/subcellular biology of these micronutrients. Recent advances have been most notable for zinc, both in terms of basic biology and global public health. Identification of "new" biologic roles of selenium and increasing respect for its importance for human health serve as a reminder that knowledge of less prominent trace elements is likely far from complete. Although, globally, issues of trace element deficiencies pose major public health challenges, for example, iodine, there are also issues of overabundance owing especially to food fortification programs. The latter tend to underscore the importance of both vigilance toward and further research on interactions and other possible adverse effects.

REFERENCES

1. Hambidge KM, Casey CE, Krebs NF. Zinc. In: Mertz W, ed. *Trace Elements in Human and Animal Nutrition.* Vol. 2. Orlando, Florida: Academic Press; 1986:1–137.
2. Dufner-Beattie J, Wang F, Kuo YM, et al. The acrodermatitis enteropathica gene ZIP4 encodes a tissue-specific, zinc-regulated zinc transporter in mice. *J Biol Chem.* 2003;278(35):33474–33481.
3. Tumer Z. An overview and update of ATP7A mutations leading to Menkes' disease and occipital horn syndrome. *Hum Mutat.* 2013;34(3):417–429.
4. Krebs NF, Hambidge KM. Zinc metabolism and homeostasis: the application of tracer techniques to human zinc physiology. *Biometals.* 2001;2001(14):397–412.
5. Klein CJ. Nutrient requirements for preterm infant formulas. *J Nutr.* 2002;132(6 Suppl 1):1395S-1577S.
6. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Vitamin A, Vitamin K, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium and Zinc*. Washington, DC: National Academy Press; 2001.
7. Greene HL, Hambidge KM, Schanler R, Tsang RC. Guidelines for the use of vitamins, trace elements, calcium, magnesium, and phosphorus in infants and children receiving total parenteral nutrition: report of the Subcommittee on Pediatric Parenteral Nutrient Requirements from the Committee on Clinical Practice Issues of the American Society for Clinical Nutrition. [published errata appear in Am J Clin Nutr 1989 Jun;49(6):1332 and 1989 Sep;50(3):560]. *Am J Clin Nutr.* 1988;48(5):1324–1342.
8. Hambidge KM. Hair analyses: worthless for vitamins, limited for minerals. *Am J Clin Nutr.* 1982;36(5):943–949.
9. Andrews GK. Cellular zinc sensors: MTF-1 regulation of gene expression. *Biometals.* 2001;14(3-4):223–237.
10. Prasad AS, Miale A, Farid Z, et al. Zinc metabolism in patients with the syndrome of iron deficiency anemia, hepatosplenomegaly, dwarfism and hypogonadism. *J Lab Clin.* 1963;61:534.
11. Black RE. Zinc deficiency, infectious disease and mortality in the developing world. *J Nutr.* 2003;133(5 Suppl 1):1485S-1489S.
12. Sehgal VN, Jain S. Acrodermatitis enteropathica. *Clin Dermatol.* 2000;18(6):745–748.
13. Brown KH, Peerson JM, Rivera J, Allen LH. Effect of supplemental zinc on the growth and serum zinc concentrations of prepubertal children: a meta-analysis of randomized controlled trials. *Am J Clin Nutr.* 2002;75(6):1062–1071.
14. Ramakrishnan U, Nguyen P, Martorell R. Effects of micronutrients on growth of children under 5 y of age: meta-analyses of single and multiple nutrient interventions. *Am J Clin Nutr.* 2009;89(1):191–203.
15. Krebs NF, Westcott JE, Arnold TD, et al. Abnormalities in zinc homeostasis in young infants with cystic fibrosis. *Pediatr Res.* 2000;48(2):256–261.
16. Patrizi A, Bianchi F, Neri I, Specchia F. Acrodermatitis enteropathica-like eruption: a sign of malabsorption in cystic fibrosis. *Pediatr Dermatol.* 2003;20(2):187–188.
17. Zemel BS, Kawchak DA, Fung EB, et al. Effect of zinc supplementation on growth and body composition in children with sickle cell disease. *Am J Clin Nutr.* 2002;75(2):300–307.
18. Krebs NF, Hambidge KM, Walravens PA. Increased food intake of young children receiving a zinc supplement. *Am J Dis Child.* 1984;138(3):270–273.

19. Umeta M, West CE, Haidar J, et al. Zinc supplementation and stunted infants in Ethiopia: a randomised controlled trial. *Lancet.* 2000; 355(9220):2021–2026.
20. WHO U. *WHO-UNICEF Joint Statement on the Clinical Management of Acute Diarrhea*. Geneva, Switzerland: World Health Assembly; 2004:2004.
21. Lazzerini M, Ronfani L. Oral zinc for treating diarrhoea in children. *Cochrane Database Syst Rev.* 2008(3):CD005436.
22. Walravens PA, Van Doorninck WJ, Hambidge KM. Metals and mental function [letter]. *J Pediatr.* 1978;93(3):535.
23. Surkan PJ, Siegel EH, Patel SA, et al. Effects of zinc and iron supplementation fail to improve motor and language milestone scores of infants and toddlers. *Nutrition.* 2013;29(3):542–548
24. Siegel EH, Kordas K, Stoltzfus RJ, et al. Inconsistent effects of iron-folic acid and/or zinc supplementation on the cognitive development of infants. *J Health Popul Nutr.* 2011;29(6):593–604.
25. Shankar AH, Prasad AS. Zinc and immune function: the biological basis of altered resistance to infection. *Am J Clin Nutr.* 1998;68(2 Suppl):447S-463S.
26. Lassi ZS, Haider BA, Bhutta ZA. Zinc supplementation for the prevention of pneumonia in children aged 2 months to 59 months. *Cochrane Database Syst Rev.* 2010(12):CD005978.
27. Hall AG, Kelleher SL, Lonnerdal B, Philipps AF. A graded model of dietary zinc deficiency: effects on growth, insulin-like growth factor-I, and the glucose/insulin axis in weanling rats. *J Pediatr Gastroenterol Nutr.* 2005;41(1):72–80.
28. Hojyo S, Fukada T, Shimoda S, et al. The zinc transporter SLC39A14/ZIP14 controls G-protein coupled receptor-mediated signaling required for systemic growth. *PLoS ONE.* 2011;6(3):e18059.
29. Srinivasan MG, Ndeezi G, Mboijana CK, et al. Zinc adjunct therapy reduces case fatality in severe childhood pneumonia: a randomized double blind placebo-controlled trial. *BMC Med.* 2012;10:14.
30. Bhatnagar S, Wadhwa N, Aneja S, et al. Zinc as adjunct treatment in infants aged between 7 and 120 days with probable serious bacterial infection: a randomised, double-blind, placebo-controlled trial. *Lancet.* 2012;379(9831):2072–2078.
31. Singh M, Das RR. Zinc for the common cold. *Cochrane Database Syst Rev.* 2011(2):CD001364.
32. Fraker PJ, King LE. Reprogramming of the immune system during zinc deficiency. *Annu Rev Nutr.* 2004;24:277–298.
33. Cuevas LE, Koyanagi A. Zinc and infection: a review. *Ann Trop Paediatr.* 2005;25(3):149–160.
34. Wang F, Kim BE, Dufner-Beattie J, et al. Acrodermatitis enteropathica mutations affect transport activity, localization and zinc-responsive trafficking of the mouse ZIP4 zinc transporter. *Hum Mol Genet.* 2004;13(5):563–571.
35. Kury S, Kharfi M, Schmitt S, Bezieau S. Clinical utility gene card for: acrodermatitis enteropathica. *Eur J Hum Genet.* 2012;20(3) doi:10.1038/ejhg.2011.227.
36. Lehnert T, Kury S, Burk G, et al. [Acrodermatitis enteropathica (AE) is caused by mutations in the zinc transporter gene SLC39A4]. *Klin Padiatr.* 2006;218(4):221–223.
37. Michalczyk A, Varigos G, Catto-Smith A, et al. Analysis of zinc transporter, hZnT4 (Slc30A4), gene expression in a mammary gland disorder leading to reduced zinc secretion into milk. *Hum Genet.* 2003;113(3):202–210.
38. Qian L, Wang B, Tang N, et al. Polymorphisms of SLC30A2 and selected perinatal factors associated with low milk zinc in Chinese breastfeeding women.

Early Hum Dev. 2012;88(8):663–668.
39. Wulf K, Wilhelm A, Spielmann M, et al. Frequency of symptomatic zinc deficiency in very low birth weight infants. *Klin Padiatr.* 2013;225(1):13–17.
40. Jalla S, Krebs NF, Rodden D, Hambidge KM. Zinc homeostasis in premature infants does not differ between those fed preterm formula or fortified human milk. *Pediatr Res.* 2004;56(4):615–620.
41. Sazawal S, Black RE, Menon VP, et al. Zinc supplementation in infants born small for gestational age reduces mortality: a prospective, randomized, controlled trial. *Pediatrics.* 2001;108(6):1280–1286.
42. Sur D, Gupta DN, Mondal SK, et al. Impact of zinc supplementation on diarrheal morbidity and growth pattern of low birth weight infants in Kolkata, India: a randomized, double-blind, placebo-controlled, community-based study. *Pediatrics.* 2003;112(6 Pt 1):1327–1332.
43. Gibson RS, Ferguson EL, Lehrfeld J. Complementary foods for infant feeding in developing countries: their nutrient adequacy and improvement. *Eur J Clin Nutr.* 1998;52(10):764–770.
44. Krebs NF, Westcott JE, Culbertson DL, et al. Comparison of complementary feeding strategies to meet zinc requirements of older breastfed infants. *Am J Clin Nutr.* 2012;96(1):30–35.
45. Yang CF, Duro D, Zurakowski D, et al. High prevalence of multiple micronutrient deficiencies in children with intestinal failure: a longitudinal study. *J Pediatr.* 2011;159(1):39–44.e31.
46. Hogberg L, Danielsson L, Jarleman S, et al. Serum zinc in small children with coeliac disease. *Acta Paediatr.* 2009;98(2):343–345.
47. Rawal P, Thapa BR, Prasad R, et al. Zinc supplementation to patients with celiac disease--is it required? *J Trop Pediatr.* 2010;56(6):391–397.
48. Mohammad MK, Zhou Z, Cave M, et al. Zinc and liver disease. *Nutr Clin Pract.* 2012;27(1):8–20.
49. Duggan C, MacLeod WB, Krebs NF, et al. Plasma zinc concentrations are depressed during the acute phase response in children with falciparum malaria. *J Nutr.* 2005;135(4):802–807.
50. Hotz C, Peerson JM, Brown KH. Suggested lower cutoffs of serum zinc concentrations for assessing zinc status: reanalysis of the second National Health and Nutrition Examination Survey data (1976–1980). *Am J Clin Nutr.* 2003;78(4):756–764.
51. Sazawal S, Malik P, Jalla S, et al. Zinc supplementation for four months does not affect plasma copper concentration in infants. *Acta Paediatr.* 2004;93(5):599–602.
52. Bertinato J, Simpson JR, Sherrard L, et al. Zinc supplementation does not alter sensitive biomarkers of copper status in healthy boys. *J Nutr.* 2013;143(3):284–289.
53. Hambidge KM, Miller LV, Westcott JE, et al. Zinc bioavailability and homeostasis. *Am J Clin Nutr.* 2010;91(5):1478S-1483S.
54. Krebs NF, Reidinger CJ, Miller LV, Hambidge KM. Zinc homeostasis in breast-fed infants. *Pediatr Res.* 1996;39(4 Pt 1):661–665.
55. Miller LV, Krebs NF, Hambidge KM. A mathematical model of zinc absorption in humans as a function of dietary zinc and phytate. *J Nutr.* 2007;137(1):135–141.
56. Manary MJ, Abrams SA, Griffin IJ, et al. Perturbed zinc homeostasis in rural 3-5-y-old Malawian children is associated with abnormalities in intestinal permeability attributed to tropical enteropathy. *Pediatr Res.* 2010;67(6):671–675.
57. Miller LV, Hambidge KM, Naake VL, et al. Size of the zinc pools that exchange rapidly with plasma zinc in humans: alternative techniques for measuring and

relation to dietary zinc intake. *J Nutr.* 1994;124(2):268–276.
58. Krebs N, Reidinger CJ, Miller LV, Borschel M. Zinc homeostasis in normal infants fed a casein hydrolysate formula. *J Pediatr Gastro Nutr.* 2000;30:29–33.
59. Arsenault JE, Brown KH. Zinc intake of US preschool children exceeds new dietary reference intakes. *Am J Clin Nutr.* 2003;78(5):1011–1017.
60. Gibson RS, Bailey KB, Gibbs M, Ferguson EL. A review of phytate, iron, zinc, and calcium concentrations in plant-based complementary foods used in low-income countries and implications for bioavailability. *Food Nutr Bull.* 2010;31(2 Suppl):S134-S146.
61. Krebs NF, Hambidge KM. Complementary feeding: clinically relevant factors affecting timing and composition. *Am J Clin Nutr.* 2007;85(2):639S-645S.
62. Ferenci P. Pathophysiology and clinical features of Wilson disease. *Metab Brain Dis.* 2004;19(3-4):229–239.
63. Daniel KG, Harbach RH, Guida WC, Dou QP. Copper storage diseases: Menkes, Wilsons, and cancer. *Front Biosci.* 2004;9:2652–2662.
64. Kaler SG. ATP7A-related copper transport diseases-emerging concepts and future trends. *Nat Rev Neurol.* 2011;7(1):15–29.
65. Grunebaum M, Horodniceanu C, Steinherz R. The radiographic manifestations of bone changes in copper deficiency. *Pediatr Radiol.* 1980;9(2):101–104.
66. Kodama H, Fujisawa C, Bhadhprasit W. Pathology, clinical features and treatments of congenital copper metabolic disorders--focus on neurologic aspects. *Brain Dev.* 2011;33(3):243–251.
67. Hedera P, Peltier A, Fink JK, et al. Myelopolyneuropathy and pancytopenia due to copper deficiency and high zinc levels of unknown origin II. The denture cream is a primary source of excessive zinc. *Neurotoxicology.* 2009;30(6):996–999.
68. Rowin J, Lewis SL. Copper deficiency myeloneuropathy and pancytopenia secondary to overuse of zinc supplementation. *J Neurol Neurosurg Psychiatry.* 2005;76(5):750–751.
69. Hellman NE, Gitlin JD. Ceruloplasmin metabolism and function. *Annu Rev Nutr.* 2002;22:439–458.
70. Dusek P, Schneider SA. Neurodegeneration with brain iron accumulation. *Curr Opin Neurol.* 2012;25(4):499–506.
71. Castillo-Duran C, Uauy R. Copper deficiency impairs growth of infants recovering from malnutrition. *Am J Clin Nutr.* 1988;47(4):710–714.
72. Marquardt ML, Done SL, Sandrock M, et al. Copper deficiency presenting as metabolic bone disease in extremely low birth weight, short-gut infants. *Pediatrics.* 2012;130(3):e695–698.
73. Dembinski K, Gargasz AE, Dabrow S, Rodriguez L. Three distinct cases of copper deficiency in hospitalized pediatric patients. *Clin Pediatr (Phila).* 2012;51(8):759–762.
74. Hillman LS. Serial serum copper concentrations in premature and SGA infants during the first 3 months of life. *J Pediatr.* 1981;98(2):305–308.
75. Salmenpera L, Siimes MA, Nanto V, Perheentupa J. Copper supplementation: failure to increase plasma copper and ceruloplasmin concentrations in healthy infants. *Am J Clin Nutr.* 1989;50(4):843–847.
76. Harvey LJ, Ashton K, Hooper L, et al. Methods of assessment of copper status in humans: a systematic review. *Am J Clin Nutr.* 2009;89(6):2009S-2024S.
77. Milne DB. Copper intake and assessment of copper status. *Am J Clin Nutr.* 1998;67(5 Suppl):1041S-1045S.
78. Higuchi T, Matsukawa Y, Okada K, et al. Correction of copper deficiency improves erythropoietin unresponsiveness in hemodialysis patients with anemia.

Intern Med. 2006;45(5):271–273.
79. Frem J, Sarson Y, Sternberg T, Cole CR. Copper supplementation in parenteral nutrition of cholestatic infants. *J Pediatr Gastroenterol Nutr.* 2010;50(6):650–654.
80. Tanner MS. Role of copper in Indian childhood cirrhosis. *Am J Clin Nutr.* 1998;67(5 Suppl):1074S–1081S.
81. Domellof M, Hernell O, Abrams SA, et al. Iron supplementation does not affect copper and zinc absorption in breastfed infants. *Am J Clin Nutr.* 2009;89(1):185–190.
82. Klevay LM. Is the Western diet adequate in copper? *J Trace Elem Med Biol.* 2011;25(4):204–212.
83. Klevay LM. Intakes of copper in nutrition surveys are falsely high. *Am J Clin Nutr.* 2012;95(5):1293–1294; author reply 1294.
84. Kim SY, Park JH, Kim EA, Lee-Kim YC. Longitudinal study on trace mineral compositions (selenium, zinc, copper, manganese) in Korean human preterm milk. *J Korean Med Sci.* 2012;27(5):532–536.
85. Zimmermann MB, Kohrle J. The impact of iron and selenium deficiencies on iodine and thyroid metabolism: biochemistry and relevance to public health. *Thyroid.* 2002;12(10):867–878.
86. Rayman MP. Selenium and human health. *Lancet.* 2012;379(9822): 1256–1268.
87. Moreno-Reyes R, Suetens C, Mathieu F, et al. Kashin-Beck osteoarthropathy in rural Tibet in relation to selenium and iodine status. *N Engl J Med.* 1998;339(16):1112–1120.
88. Keshan Disease Research Group. Observations of effects of sodium selenite in prevention of Keshan disease. *Chin Med J (English).* 1979;92:471–476.
89. Beck MA, Shi Q, Morris VC, Levander OA. Rapid genomic evolution of a non-virulent coxsackievirus B3 in selenium-deficient mice results in selection of identical virulent isolates. *Nat Med.* 1995;1(5):433–436.
90. Peng T, Li Y, Yang Y, et al. Characterization of enterovirus isolates from patients with heart muscle disease in a selenium-deficient area of China. *J Clin Microbiol.* 2000;38(10):3538–3543.
91. Tanguy S, Grauzam S, de Leiris J, Boucher F. Impact of dietary selenium intake on cardiac health: experimental approaches and human studies. *Mol Nutr Food Res.* 2012;56(7):1106–1121.
92. Yusuf SW, Rehman Q, Casscells W. Cardiomyopathy in association with selenium deficiency: a case report. *JPEN J Parenter Enteral Nutr.* 2002;26(1):63–66.
93. Brown MR, Cohen HJ, Lyons JM, et al. Proximal muscle weakness and selenium deficiency associated with long term parenteral nutrition. *Am J Clin Nutr.* 1986;43(4):549–554.
94. Moreno-Reyes R, Mathieu F, Boelaert M, et al. Selenium and iodine supplementation of rural Tibetan children affected by Kashin-Beck osteoarthropathy. *Am J Clin Nutr.* 2003;78(1):137–144.
95. Jochum F, Terwolbeck K, Meinhold H, et al. Effects of a low selenium state in patients with phenylketonuria. *Acta Paediatr.* 1997;86(7):775–777.
96. van Bakel MM, Printzen G, Wermuth B, Wiesmann UN. Antioxidant and thyroid hormone status in selenium-deficient phenylketonuric and hyperphenylalaninemic patients. *Am J Clin Nutr.* 2000;72(4):976–981.
97. Manary MJ, MacPherson GD, McArdle F, et al. Selenium status, kwashiorkor and congestive heart failure. *Acta Paediatr.* 2001;90(8):950–952.
98. Yannicelli S, Hambidge KM, Picciano MF. Decreased selenium intake and low plasma selenium concentrations leading to clinical symptoms in a child with

propionic acidaemia. *J Inherit Metab Dis.* 1992;15(2):261–268.
99. Ogunro PS, Ogungbamigbe TO, Elemie PO, et al. Plasma selenium concentration and glutathione peroxidase activity in hiv-1/aids infected patients: a correlation with the disease progression. *Niger Postgrad Med J.* 2006;13(1):1–5.
100. Kupka R, Msamanga GI, Spiegelman D, et al. Selenium levels in relation to morbidity and mortality among children born to HIV-infected mothers. *Eur J Clin Nutr.* 2005;59(11):1250–1258.
101. Food and Nutrition Board, Institue of Medicine. *Dietary reference Intakes for vitamin C, vitamin E, selenium, and carotenoids.* Washington, DC: National Academy Press; 2000.
102. Darlow BA, Winterbourn CC, Inder TE, et al. The effect of selenium supplementation on outcome in very low birth weight infants: a randomized controlled trial. The New Zealand Neonatal Study Group. *J Pediatr.* 2000;136(4):473–480.
103. Zachara BA, Pilecki A. Selenium concentration in the milk of breast-feeding mothers and its geographic distribution. *Environ Health Perspect.* 2000;108(11):1043–1046.
104. Raiten DJ, Talbot JM, Waters JH. Assessment of nutrient requirements for infant formulas. *J Nutr.* 1998;128:2059S–2294S.
105. Anderson RA. Chromium as an essential nutrient for humans. *Regul Toxicol Pharmacol.* 1997;26(1 Pt 2):S35–S41.
106. Althuis MD, Jordan NE, Ludington EA, Wittes JT. Glucose and insulin responses to dietary chromium supplements: a meta-analysis. *Am J Clin Nutr.* 2002;76(1):148–155.
107. Vincent JB. Recent advances in the nutritional biochemistry of trivalent chromium. *Proc Nutr Soc.* 2004;63(1):41–47.
108. Moukarzel AA, Song MK, Buchman AL, et al. Excessive chromium intake in children receiving total parenteral nutrition. *Lancet.* 1992;339(8790):385–388.
109. Cerulli J, Grabe DW, Gauthier I, et al. Chromium picolinate toxicity. *Ann Pharmacother.* 1998;32(4):428–431.
110. Wasser WG, Feldman NS, D'Agati VD. Chronic renal failure after ingestion of over-the-counter chromium picolinate. *Ann Intern Med.* 1997;126(5):410.
111. Anderson RA, Bryden NA, Polansky MM. Lack of toxicity of chromium chloride and chromium picolinate in rats. *J Am Coll Nutr.* 1997;16(3):273–279.
112. Anderson RA, Bryden NA, Patterson KY, et al. Breast milk chromium and its association with chromium intake, chromium excretion, and serum chromium. *Am J Clin Nutr.* 1993;57(4):519–523.
113. Aschner JL, Aschner M. Nutritional aspects of manganese homeostasis. *Mol Aspects Med.* 2005;26(4-5):353–362.
114. Smith EA, Newland P, Bestwick KG, Ahmed N. Increased whole blood manganese concentrations observed in children with iron deficiency anaemia. *J Trace Elem Med Biol*2013;27(1):65–69.
115. Abdalian R, Saqui O, Fernandes G, Allard JP. Effects of manganese from a commercial multi-trace element supplement in a population sample of canadian patients on long-term parenteral nutrition. *JPEN J Parenter Enteral Nutr.* 2013;37(4):538–543.
116. Wright RO, Amarasiriwardena C, Woolf AD, et al. Neuropsychological correlates of hair arsenic, manganese, and cadmium levels in school-age children residing near a hazardous waste site. *Neurotoxicology.* 2006;27(2):210–216.
117. Golub MS, Hogrefe CE, Germann SL, et al. Neurobehavioral evaluation of rhesus monkey infants fed cow's milk formula, soy formula, or soy formula with added

manganese. *Neurotoxicol Teratol.* 2005;27(4):615–627.
118. Tran TT, Chowanadisai W, Crinella FM, et al. Effect of high dietary manganese intake of neonatal rats on tissue mineral accumulation, striatal dopamine levels, and neurodevelopmental status. *Neurotoxicology.* 2002;23(4–5):635–643.
119. Dawes C, Weatherell JA. Kinetics of fluoride in the oral fluids. *J Dent Res.* 1990;69 Spec No:638–644; discussion 682–633.
120. Marinho VC, Higgins JP, Logan S, Sheiham A. Topical fluoride (toothpastes, mouthrinses, gels or varnishes) for preventing dental caries in children and adolescents. *Cochrane Database Syst Rev.* 2003(4):CD002782.
121. Palmer CA, Gilbert JA. Position of the Academy of Nutrition and Dietetics: the impact of fluoride on health. *J Acad Nutr Diet.* 2012;112(9):1443–1453.
122. Centers for Disease Control and Prevention. Recommendations for using fluoride to prevent and control dental caries in the United States. Centers for Disease Control and Prevention. *MMWR Recomm Rep.* 2001;50(RR-14):1–42.
123. Food and Nutrition Board, National Reseach Council. *Dietary Reference Intakes for Calcium, Phosphorus, Magnesium, Vitamin D, and Flouride.* Washington, DC: National Academy Press; 1997.
124. Dunn J. Iodine. In: Shils ME SM, Ross AC, Caballero B, Cousins RJ, eds. *Modern Nutrition in Health and Disease*. 10th ed. Baltimore, MD: Lippincott Williams and Wilkins; 2006.
125. Walker SP, Wachs TD, Gardner JM, et al. Child development: risk factors for adverse outcomes in developing countries. *Lancet.* 2007;369(9556):145–157.
126. Zimmermann MB, Andersson M. Assessment of iodine nutrition in populations: past, present, and future. *Nutr Rev.* 2012;70(10):553–570.
127. Andersson M, Karumbunathan V, Zimmermann MB. Global iodine status in 2011 and trends over the past decade. *J Nutr.*2012;142(4):744–750.
128. Teng W, Shan Z, Teng X, et al. Effect of iodine intake on thyroid diseases in China. *N Engl J Med.* 2006;354(26):2783–2793.
129. Vanek VW, Borum P, Buchman A, et al. A.S.P.E.N. position paper: recommendations for changes in commercially available parenteral multivitamin and multi-trace element products. *Nutr Clin Pract.* 2012;27(4):440–491.

Iron

9

Amina Khambalia, PhD, Ashley Aimone, MSc, RD, and S. H. Zlotkin, MD, PhD

INTRODUCTION

Iron is a constituent of many enzymes in the human body and is especially important to ensure the oxygen carrying capacity of hemoglobin. The most prevalent iron disorder is iron deficiency. Iron deficiency (ID) is the most common nutritional deficiency in children. While iron deficiency is far more prevalent than iron overload, both deficiency and excess are associated with significant morbidity and mortality. This chapter reviews issues related to iron metabolism, clinical manifestations, assessment, and prevention in infants, young children and pregnant women. The specific sections covered in this chapter are as follows: (1) What is iron? (2) Why is iron important? (3) How much iron does the body need? (4) Clinical case study; (4) What are dietary sources of iron? (4.1: Special topic—Iron deficiency and obesity; (5) How does the body maintain iron homeostasis? (5.1 Clinical case study); (6) How does the body regulate iron absorption? (7) How does the body transport iron? (8) What about too *little* iron in the body? (9) What about too *much* iron in the body? (10) How do we assess iron?

SECTION 1: WHAT IS IRON?

Iron is a versatile and highly reactive group-VIII transition metal with electrons distributed throughout five 3d orbitals.[1] Depending on the chemical environment iron can exist in several oxidation states (between –2 and +6). At physiological oxygen concentrations, iron exists in its stable ferric (Fe^{3+}) state for most of its biological complexes.

The only stable iron states in the aqueous environment of the human body and in food are ferric and ferrous iron. Ferrous (Fe^{2+}) iron is used in

transmembrane transport of iron, deposition of iron in the storage protein and synthesis of heme.[2] The human body contains 2–4 g of elemental iron, most of which is in the hemoglobin of erythrocytes.[3] Over 65% of body iron is present in the hemoglobin, up to 10% is found in myoglobin and approximately 1%–5% is present as part of enzymes.[4]

SECTION 2: WHY IS IRON IMPORTANT?

With the exception of certain members of the bacterial genera, iron is an essential nutrient for all living organisms. Iron is vital for metabolic processes such as oxygen transport, DNA synthesis and electron transport.

Iron's key function is tissue oxygenation. Oxygen is bound to an iron-containing porphyrin ring, either as part of the prosthetic group of hemoglobin within erythrocytes, or as part of myoglobin located within the cytoplasm of muscle cells.[5] Some critical proteins and enzymes that have iron as a component or cofactor include hemoglobin, myoglobin, catalase, xanthine oxidase, aconitase, reduced nicotinamide adenine dinucleotide, ribonucleotide reductase, peroxidases and cytochrome oxidase. Iron's role in electron transport and reversible ligand binding is accredited to its ability to interconvert between oxidation states and achieve various redox potentials during electrochemical reactions. Heme is contained in the active sites of cytochromes that serve as electron carriers within the cell, in iron–sulfur proteins (flavoproteins, heme-flavoproteins) required for the first reaction in the electron transport chain, and in hydrogen perioxidasess (e.g., catalase and perioxidase).[1] The cytochrome P450 family of enzymes located in microsomal membranes of liver and intestinal mucosal cells also contain heme. Cytochrome P450 is involved in detoxification of foreign substances in the liver and the synthesis of steroid hormones and bile acids.

SECTION 3: HOW MUCH IRON DOES THE BODY NEED?

Iron requirements are high during periods of rapid growth such as: (a) *infancy*, (b) *adolescence* and (c) *pregnancy* (Table 9-1). During infancy the amount of iron needed for growth is so great that the abundant iron stores present at birth are usually exhausted by 4–6 months of age. After the first year of life, adolescence is the second most critical period of physical growth in the life cycle. During pregnancy, iron requirements rise

significantly due to increased red cell mass, the expansion of plasma volume and growth of the fetal-placental unit.[6]

Infancy

Fetal hemoglobin (Hb) has a high affinity for oxygen; thereby, giving the fetus precedence for oxygen over the mother at the placental interface.[7] Delivery of oxygen from fetal Hb to tissues is lowered. As a consequence, the fetus needs more Hb to deliver a certain amount of oxygen to tissues (regulated by erythropoietin). Daily fetal iron accretion occurs at a rate of approximately 1.6–2.0 mg/kg.[8] Fetal iron is present in three forms: hemoglobin iron, tissue iron and storage iron, comprising 77%, 9% and 14% of total body iron, respectively. In late fetal and early postnatal life the greatest need for iron is for Hb synthesis.[9] At delivery, the production of fetal Hb is exchanged for the production of normal adult Hb, and oxygen is more readily available from the lungs.[7]

TABLE 9-1 Main Factors Negatively Influencing Iron Balance According to Life Stage

Birth	Infancy	Childhood and Adolescence
Before birth	0–6 months	Antacid therapy
Maternal iron deficiency	Low/depleted body iron stores	Low dietary iron supply
Intrauterine growth retardation	Rapid weight gain	Low dietary iron availability
At birth	Blood loss due to frequent sampling	Intense physical activity
Low body iron stores at birth	Blood loss due to cow milk feeding	Adolescent pregnancy
Early onset of postnatal erythropoiesis	6–12 months	Loss or dysfunction of absorptive enterocytes
Rapid postnatal catch-up growth	Low/depleted iron stores	Bowel resection

Low birth weight	Rapid weight gain	Celiac disease
Premature birth	Blood loss due to cow milk feeding	Intrinsic enterocyte defects
Fetal-maternal hemorrhage	Low dietary iron supply	Gastrointestinal blood loss: epistaxis, varices, gastritis, ulcer, tumor, meckel's diverticulum, parasitosis, vascular malformations, inflammatory bowel disease, diverticulosis, hemorrhoids, menorrhagia, cancer, chronic infection, pulmonary hemosiderosis, trauma, excessive phlebotomy, large vascular malformations
Perinatal hemorrhage	Low dietary iron availability	
Twin-to-twin transfusion	Prolonged exclusive breastfeeding	
Umbilical cord preclamping	Frequent infections	
Phlebotomy losses		
Restrictive RBC transfusion practice		
Recombinant erythropoietin administration (preterm infants)		

After birth, shifts in iron compartments occur; erythropoiesis falls as Hb concentration tends to return to normal levels around 120 g/L instead of neonatal levels around 170 g/L.[9] Iron derived from this physiologic process is accumulated in iron stores and successively used for Hb production. An infant is born with about 250 mg of total body iron (75 mg/kg), over two-thirds of which is accreted during the third trimester at a rate proportional to an increase in body weight.[12] The amount of iron present at birth depends on the length of gestational period and the weight of the baby.[10] At birth a normal full-term infant has iron stores to cover iron needs during the first 4–6 months of life.[11] After 6 months the assumed amount of iron stores endowed at birth have been utilized.

Exogenous sources of iron are required as erythropoiesis is more active.

During years of growth, iron absorption must exceed iron losses by approximately 0.5 mg daily.[5] From 4 to 12 months of age an infant's blood volume doubles, with each additional gram of Hb that is synthesized requiring an additional 3.5 mg of iron.[8] During the second half of infancy exogenous iron requirements rapidly increase to concentrations that are higher than any other time in life on a body weight basis.[16] It is only after approximately two years of life that iron requirements begin to be reduced.[7]

Adolescence

During adolescence iron requirements nearly double in both boys and girls as a result of the expansion of the total blood volume and the increase in lean body mass.[13] Estimates of physiological iron requirements during the adolescent period have been computed using a factorial method of iron components (basal losses, menstrual losses, and accretion).[14] Preadolescent levels of overall iron requirements (0.7–0.9 mg Fe/day) increase to as much as 2.2 mg Fe/day.[15,16]

Female adolescents require an additional amount of iron to account for iron losses during menses. Menstrual losses may increase daily requirements to as much as 2.1 mg/day in girls at the 75th percentile of blood loss.[13] Among 14 year old girls, median iron requirements can be 30% higher than for their mothers.[16] Iron requirements may increase further in heavily menstruating young women.

Pregnancy

Pregnancy is associated with an increased red cell mass, expansion of plasma volume and growth in the fetal-placental unit, all of which contribute to a significant increase in iron requirements. The additional iron requirement for pregnancy (3–4 mg/day) is substantial and it is estimated that fewer than 50% of women have adequate iron stores to meet this additional need.[6] Furthermore, iron requirements are not distributed equally over the duration of pregnancy. Requirements are low in the first trimester (due to the absence of menstrual iron losses and the negligible needs of the fetus) but rise to between 4 and 6 mg in the second and third trimesters, respectively.[17] During the last 6–8 weeks of pregnancy requirements may reach as much as 10 mg/day.[7]

By the end of pregnancy, approximately 1200 mg of iron is required,

either from body iron stores or from the diet.[18] The average requirement for a menstruating woman for the same period of time is roughly 400 mg. In order to accommodate this increased requirement of 800 mg and avoid a negative iron balance the average woman must absorb 500 mg (2 mg/day) more iron than she required while menstruating.[18] In most cases, dietary intakes alone are insufficient to meet the requirements of pregnancy, especially in the latter part of pregnancy.[18] It is, therefore, estimated that a female needs 500 mg of stored iron to cover iron requirements during pregnancy.[7] A woman who routinely eats a diet high in bioavailable iron and enters pregnancy with sufficient iron stores will likely meet her requirements.[17] However, a higher amount of stored iron is needed when the diet is less than optimal.[7]

SECTION 3.1: A STUDY OF CLINICAL CASE

There are wide variations in iron supplementation practices among neonatal units worldwide. Since at least the 1950s, the need for iron therapy in preterm infants has been debated.[*] There is indication for iron supplementation because premature infants have a propensity to develop iron deficiency. Conversely, the pro-oxidant role of non-protein bound iron and the poorly developed antioxidant measures in preterm infants cautions against aggressive iron supplementation in this population.[†] The goal of nutrient delivery to preterm infants is to mimic the intrauterine accretion rate and maintain normal serum levels. However, unlike intrauterine life, growth and erythropoiesis cease soon after birth making iron requirements lower. Well-executed, randomized controlled trials have been conducted to determine the iron requirements of preterm infants. Trials have established the beneficial effect of iron supplementation via fortification of human milk, iron-fortified formula and medicinal iron in decreasing the risk of iron deficiency.[†] Most studies have only evaluated short-term benefits on hematological parameters and a paucity of information on the long-term effects of iron supplementation.

Further discussion points:

What factors predispose premature infants to iron deficiency?

What iatrogenic factors are responsible for excessive iron accumulation in preterm infants?

What are the consequences of iron deficiency in premature infants?

What are the consequences of iron overload in premature infants?

What is the best route for supplementation (i.e., parenteral, intramuscular,

intravenous)?

* Halvorsen S, Seip M. Erythrocyte production and iron stores in premature infants during the first months of life; the anemia of prematurity-etiology, pathogenesis, iron requirement. Acta Paediatr Scand. 1956;45:600–617.

† Raghavendra R, Georgieff MK. Iron therapy for preterm infants. Clin Perinatol. 2009;36:27–42.

SECTION 4: WHAT ARE DIETARY SOURCES OF IRON?

A normal diet contains approximately 13–18 mg of iron per day, of which only 1 mg of iron per day is needed in healthy persons to replace basal iron losses.[18] There are two forms of iron in the diet—inorganic iron and heme iron. While many foods are good sources of iron, some are limited by the bioavailability of iron. Iron-rich foods include cocoa, liver, kidney, dried fish, shell-fish, sesame seed, soy flour, and pulses.[20] Foods with low-iron content include milk, cheese, sugar, sweets, jams, and unfortified milled cereals (Table 9-2).

Heme iron is present in the hemoglobin and myoglobin of animal tissues and is thus mainly associated with meat, poultry and fish intake.[22] Heme iron may contribute up to 50% of the iron that actually enters the body.[9] In a single meal 5–35% of heme iron is absorbed.[23] Studies using radiolabeled heme or hemoglobin have established that the intestinal absorption of heme is far more efficient than inorganic iron.[11] Heme iron is well absorbed regardless of meal composition with the exception of meat and calcium. Meat has been shown to enhance heme iron absorption.[24] Calcium is the only dietary factor known to negatively influence the absorption of heme iron and does so to the same extent that it influences non-heme iron (Figure 9-1).[25]

TABLE 9-2 Iron Content of Various Foods[21]

Food	One Serving	Iron Content (mg)
Clam, mixed species, canned, drained solids	3 oz	23.8
Cereals, ready-to-eat, GENERAL MILLS, CHERRIOS	1 cup	10.3
Soybeans, mature cooked, boiled, without salt	1 cup	8.84

Cereals, ready-to-eat, KELLOGS, CORN FLAKES	1 cup	8.12
Infant cereals, including rice, oats, mixed cereals	15 g	7.10
Lentils, mature seeds, cooked, boiled, without salt	1 cup	6.59
Chickpeas, matured seeds, cooked, boiled, without salt	1 cup	4.74
Rice, white, long-grained, parboiled, enriched, cooked	1 cup	3.17
Peas, edible-podded, boiled, drained, without salt	1 cup	3.15
Infant formula—with iron, ready-to-feed	240 mL	2.90
Sardine, Atlantic, canned in oil, drained solids with bone	3 oz	2.48
Chicken liver, cooked, simmered	1 liver	2.28
Potatoes, baked, flesh and skin, without salt	1 potato	2.18
Beef, 75% lean/25% fat, cooked, broiled	3 oz	2.01
Almonds	1 oz (24 nuts)	1.22
Tuna, light, canned in oil, drained solids	3 oz	1.18
Bread, whole, commercially prepared	1 slice	0.93
Egg, whole, raw, fresh	1 large	0.92
Blueberries, raw	1 cup	0.89
Rice, brown, long-grained, cooked	1 cup	0.82
Spinach, raw	1 cup	0.81
Broccoli, raw	1 cup	0.64
Bread, white, commercially prepared	1 slice	0.73
Infant formula- low iron, ready-to-feed	240 mL	0.30
Cheese, cheddar	1 oz	0.19
Apples, raw, with skin	1 apple	0.17
Milk, cow, whole, 3.25% milk fat	1 cup	0.07

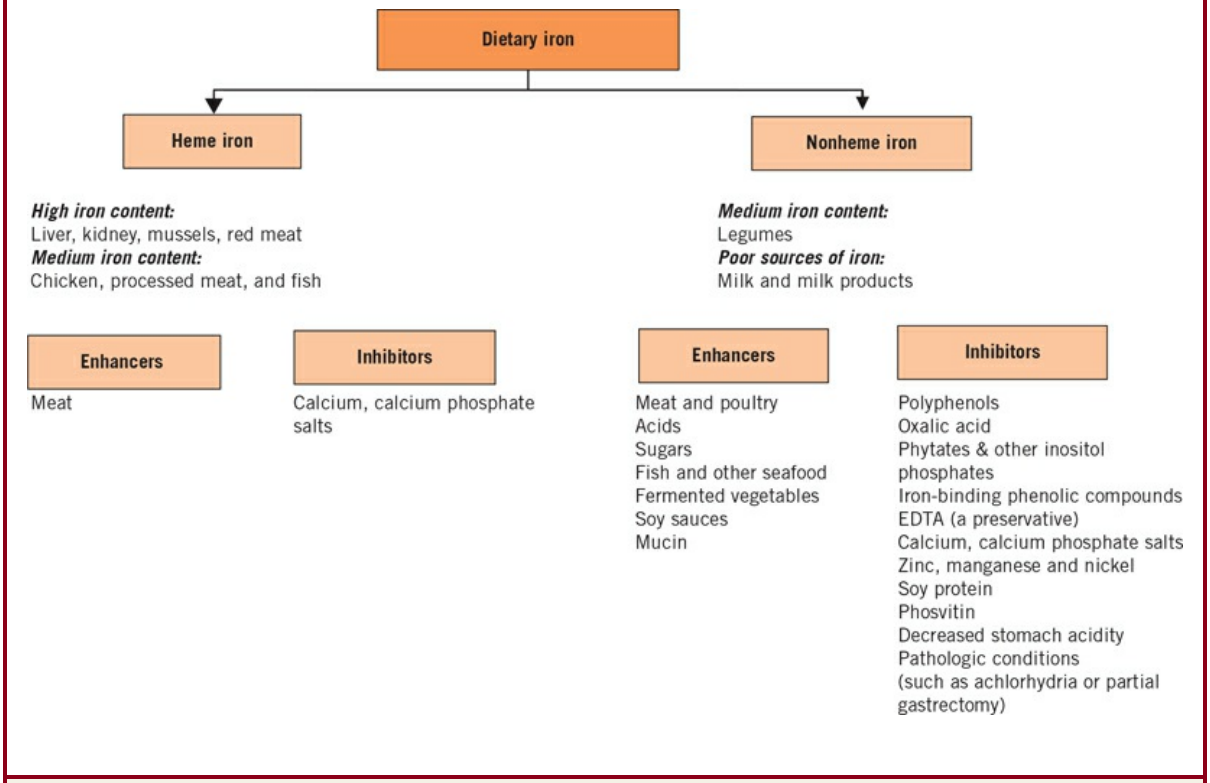


Figure 9-1 Factors affecting dietary iron absorption

Inorganic iron, predominantly found in components of plant origin, is the most prevalent form of dietary iron.[26] Inorganic iron accounts for 80–90% of the iron in a standard diet.[1] Non-heme iron is generally poorly absorbed and greatly influenced by enhancing or inhibiting substances (Figure 9-1).[20] Meat, fish amino acids and organic acids (i.e., ascorbic acid, lactic acid, citric acid, and malic acid) promote non-heme iron absorption.[26,27] These non-heme iron enhancers form soluble complexes with iron at low gastric pH levels and prevent the formation of insoluble hydroxides at the higher pH of small intestine.[9] Phenols, calcium and phytates which are present in cereals and legumes, impair non-heme iron absorption (Figure 9-1).[9] Phenolic compounds, which bear catechol or galloyl groups, have iron-binding properties.[20] Tea, coffee, cocoa, red wine, many vegetables (such as spinach), grains, herbs and spices (such as oregano and cinnamon) contain phenolic compounds. Vitamin A may be capable of preventing the inhibiting effect of polyphenols on iron absorption.[20] Phytates are a storage form of phosphates and minerals in cereals, seeds, nuts, vegetables, and fruit. Bran, oats, and fiber-rich foods are rich in phytates. The effect of phytates is counteracted by ascorbic

acid, meat, fish and poultry.[20]

SPECIAL TOPIC—IRON DEFICIENCY AND OBESITY

Results from cross-sectional studies performed in industrialized countries have consistently shown that obese individuals are at increased risk of iron deficiency.[28]

Obesity is a worldwide health problem. While large segments of the population in lower- to middle-income countries in Asia, Africa, the Middle East, and Latin America face under nutrition and poverty-related diseases, increasing proportions of the population are developing obesity and non-communicable diseases such as diabetes and coronary problems.[29] The co-occurrence of over nutrition and undernutrition is attributed to the "nutrition transition"—a term used to describe shifts in diet, physical activity, health, and nutrition.[30] The nutrition transition is characterized by a shift toward more energy-dense diets and a reduction in physical activity due to higher incomes and the influence of mass media and food marketing.[29]

Data from the American National Health and Nutrition Examination Survey III as well as data obtained in children from transition countries have suggested that among children, the prevalence of iron deficiency increases as body mass index increases from normal weight to at risk for obesity to obesity.[31,32]

The underlying mechanisms of the association between obesity and iron deficiency are not clear. The data from small studies suggest that iron deficiency in obese individuals is due to lower dietary iron intake or increased iron requirements from a larger blood volume.[28] Despite their excessive dietary and caloric intake, obese children and adolescents may be at risk of ID because they tend to consume unbalanced meals, particularly rich in carbohydrates and fat. Obesity and iron deficiency are more prevalent in lower socioeconomic groups, who consume low-cost fast foods that are low in essential nutrients and rich in fats, sugars and preservatives. It is possible that obese children have increased iron needs because of their increased growth and body surface area.[33] Higher risk of iron deficiency in obese individuals may also be related to chronic adiposity-related inflammation which increases hepcidin concentrations, which in turn, decreases intestinal iron absorption and/or increased reticuloendothelial iron sequestration.[34]

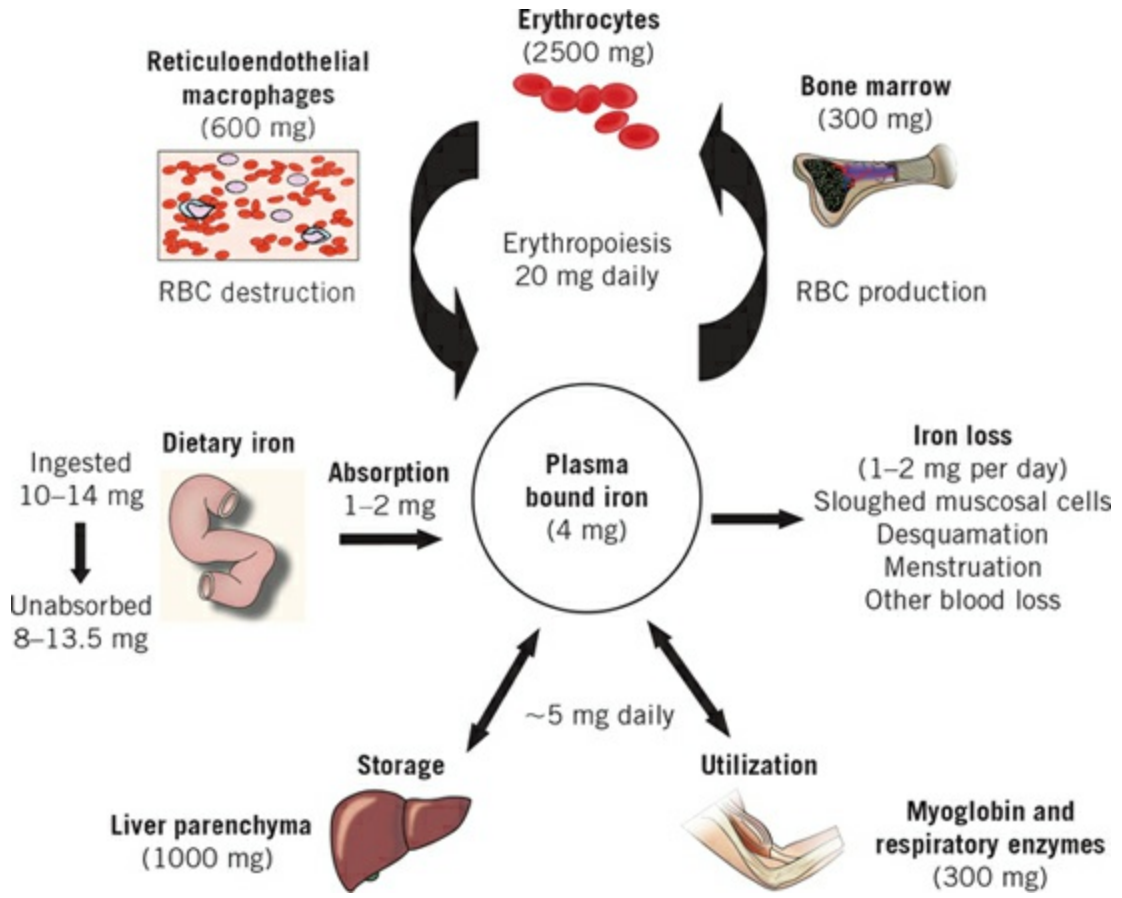


Figure 9-2 Iron distribution and exchange pools.[36–38]

SECTION 5: HOW DOES THE BODY MAINTAIN IRON HOMEOSTASIS?

Iron balance in the human body is maintained by: (a) *absorption*, (b) *turnover*, (c) *storage*, *and* (d) *excretion* (Figure 9-2).

Absorption

Humans lack a physiological pathway for iron excretion therefore, iron homeostasis in the body is determined by the regulation of iron absorption in the proximal small intestine.[7] Several processes have been found to influence the rate of iron absorption, including diet, body iron stores, marrow erythropoietic activity, blood hemoglobin concentration, blood oxygen content, and the presence of systemic inflammation.[35] Iron absorption decreases in the presence of iron overload and inflammation

and increases in response to iron deficiency, accelerated erythropoiesis, and hypoxia.[35] Some of these processes are inter-related and may occur simultaneously.

Turnover

A steady supply of iron is needed for cellular turnover. Ferroportin, a transmembrane protein exports iron from cellular storage sites into the circulation where it is rapidly bound to transferrin.[39] The export rate is closely matched to tissue requirements. Almost all functional requirements of cells located throughout the body are met from this small dynamic pool.[40] Iron is constantly being delivered from stores to maturing red blood cells to make hemoglobin and, after 4 months, returned to the stores when senescent red blood cells are catabolized by specialized macrophages in the spleen, liver, and bone marrow and the iron salvaged from the hemoglobin.[40,41]

Storage

When intracellular iron exceeds cellular requirements, excess iron is kept in storage. All cells are capable of storing iron; however, principal storage sites in humans are the liver, spleen, and bone marrow.[2] The concentration of storage iron in the body varies by gender and iron status.

Excretion

Iron in the human body is highly conserved. The majority of iron is lost in feces (derived from shed enterocytes, extravasated red blood cells, and biliary heme breakdown products that are poorly absorbed), with minimal losses from the urine, gastrointestinal tract and skin amounting to 0.08, 0.6, 0.2–0.3 mg/day, respectively.[1]

Absorption

Iron balance is highly regulated by the control of absorption in humans. Iron absorption occurs in the proximal duodenum and involves a sequence of events: (i) *reduction*; (ii) *apical uptake*; (iii) *intracellular storage or trafficking* and; (iv) *basolateral release*.

Reduction

Most inorganic iron in the diet is in the oxidized or ferric form. Before iron can be taken up by the intestinal epithelial cells or enterocytes, iron must

first be reduced from its ferric to ferrous form in the luminal brush border of the enterocyte (Figure 9-3).[35] A strong candidate for the brush border ferrireductase is duodenal cytochrome *b* (Dcytb).[42] Ferrous iron is more readily absorbed than ferric iron due to higher solubility in the proximal small intestine. Iron reduction is facilitated by the acidic milieu of the gastric content and the meal composition.[12]

Apical Uptake

In order to reach the plasma, iron must pass from the gut through the apical (lumenal) and basolateral (serosal) membranes of the enterocyte.[43] Absorptive cells located in the crypts of the duodenum are organized as finger-like villous structures to increase surface area. Beginning as stem cells, these absorptive cells proliferate and migrate out of the crypt and up to the villus over a period of several days.[44] During migration the cells differentiate and perform their functions.[45] During the intestinal phase of digestion iron is present in the lumen as either heme or non-heme iron. Non-heme iron that is liberated by gastric and pancreatic proteases becomes bound to intraluminal molecules. These iron-binding proteins prevent non-heme iron from being readily oxidized from ferrous to ferric iron in the alkaline environment and rendered insoluble and biologically unavailable.

The pathway by which heme iron is absorbed is less well defined than that of inorganic iron.[43] Heme iron is released from proteins such as hemoglobin and myoglobin in the lumen of the stomach and small intestine.[43] Luminal absorption of heme does not require binding proteins because heme iron is soluble in an alkaline environment. After binding to its receptor, heme iron is absorbed intact by the intestinal enterocytes.[11] Iron from heme is not released prior to its cellular uptake, making heme iron less susceptible than inorganic heme to variations in dietary composition.[23] The percent iron absorption from heme iron is five- to ten-fold higher than that from non-heme iron. Most studies have shown that iron deficiency is able to stimulate the absorption of both heme iron and inorganic iron.[23] The enterocyte's apical membrane transports heme and ferrous iron into the cell.[46] Iron transport is meticulously regulated by chelating compounds that are produced to detoxify extra- and intra-cellular iron.[43]

Intracellular Storage or Trafficking

Once iron has been absorbed by enterocytes it is bound by, as of yet

unidentified, chaperones. Inside the absorptive enterocyte, iron can either be stored as ferritin or transferred across the basolateral membrane into the circulation where it will be bound to apoferritin.[46] Proteins related to iron metabolism on the basolateral membrane of the precursor and mature enterocyte either sense body iron stores or facilitate regulated iron transport into the plasma. Iron that remains in the form of ferritin as the enterocyte completes its life cycle will be sloughed off with the senescent cell and exit the body through the gastrointestinal tract (Figure 9-3). This maturation process takes approximately 3–5 days.[19]

The discovery of the iron regulatory hormone hepcidin supports the liver's central role in determining the amount of iron that is absorbed by the gut and released from storage sites.[48] It has long been known that the liver is the major depot for iron; hepatocytes are the first cells in the body to have access to absorbed iron entering the portal circulation from the gut.[49] Hepcidin (also called HAMP) is a 25-amino acid peptide that is primarily produced by the liver. It is secreted into plasma, and is cleared by the kidneys.[50] Hepcidin acts as an inhibitor of iron absorption and iron release. Important cellular targets for hepcidin are the villus enterocyte, the reticuloendothelial macrophage and the hepatocyte (Figure 9-3). Hepcidin modulates the ferroportin-mediated release of cellular iron by binding to ferroportin and causing it to be degraded.[48] Decreased functional activity of ferroportin causes a decrease in basolateral iron transfer in the enterocyte and a decrease in iron export from reticoloendothelial macrophages and hepatocytes (increase in stored iron).[48]

The production of hepcidin is inversely related to iron stores, erythropoietic activity, hemoglobin, oxygen content, and inflammation.[48] Hepcidin expression is decreased when body iron requirements are high, such as when iron stores decrease, during iron deficiency, accelerated erythropoiesis, and hypoxia.[49] Increased levels of hepcidin are observed in non-genetic iron overload and inflammation.[50] While it is clear that hepcidin is a strong negative regulator of iron absorption, there is much to be learned regarding the mechanisms by which the liver senses body iron status and erythropoietic demand.[51]

Basolateral Release

The pathways by which inorganic and heme iron are taken up from the intestinal lumen by the enterocytes vary; however, it has been proposed that their basolateral export from enterocytes, that is, the step involving hepaestin and IREG1, is common to iron from both sources (Figure 9-3).

The basolateral membrane mediates the transfer of iron from the intestinal epithelial cells to the rest of the body.[45] Once inside the epithelial cell heme is either split into ferrous iron and bilirubin by heme oxygenase, or transported as the intact porphyrin ring.[23] The released ferrous iron likely enters the pool of iron in the enterocyte and exits the enterocyte via a pathway common to inorganic iron.[51] If required by the body, iron rapidly moves across the basolateral membrane and into the circulation. Once iron transverses the basolateral membrane it is bound to transferrin.[50]

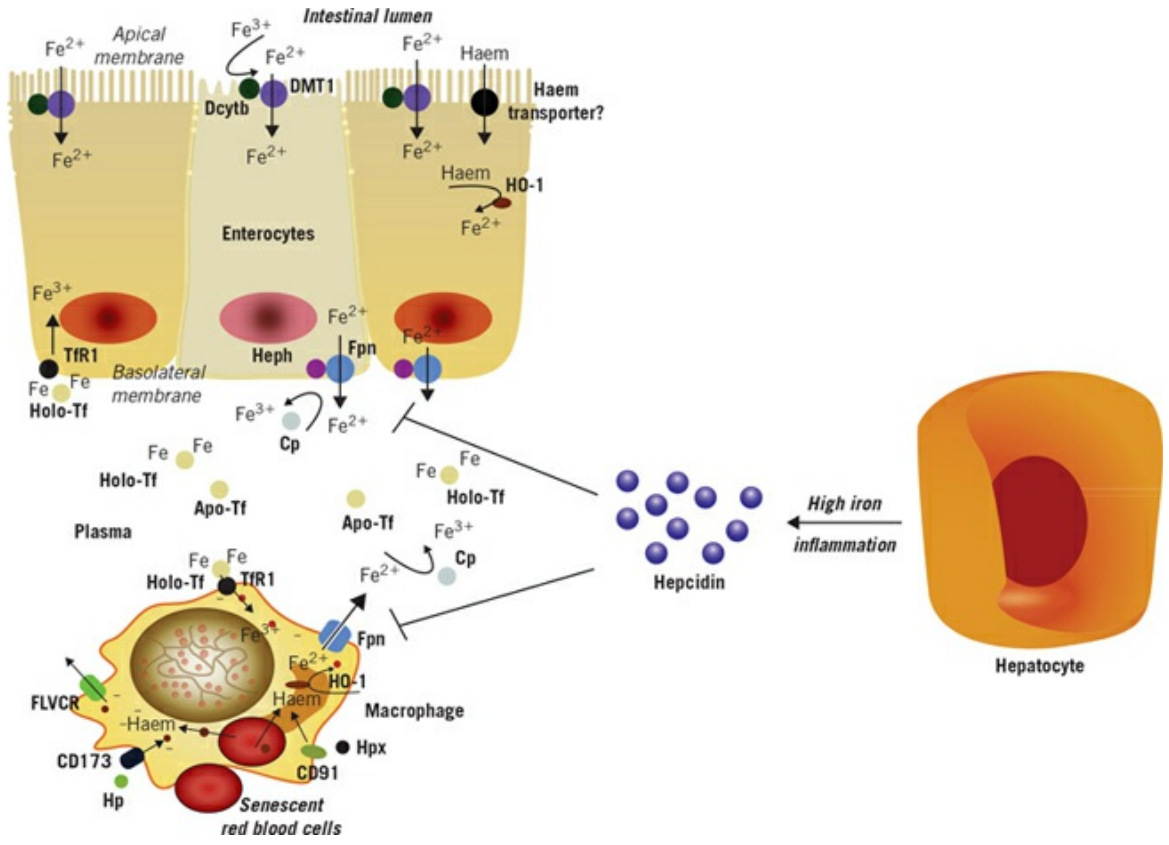


Figure 9-3 Hormonal regulation of iron efflux from duodenal enterocytes and reticuloendothelial macrophages by hepcidin.[47] Enterocytes absorb inorganic or heme iron from the diet and macrophages phagocytose iron-loaded senescent red blood cells, or acquire iron by other mechanisms. Both cell types release Fe2+ into the plasma via ferroportin (Fpn), which is incorporated into apo-Tf following oxidation to Fe3+ via hephaestin (Heph) or ceruloplasmin (Cp). Hepatocytes generate the iron-regulatory hormone hepcidin in response to high iron or inflammatory signals, which inhibits the efflux of iron via ferroportin and promotes its retention within enterocytes and

macrophages. (Copyright © 2011 The Authors have paid for this article to be freely available under the terms of the Creative Commons Attribution Non-Commercial License [http://creativecommons.org/licenses/by-nc/2.5/] which permits unrestricted non-commercial use, distribution and reproduction in any medium provided the original work is properly cited.)

Turnover

At the end of their functional lifetime, erythrocytes undergo changes in the structure of their membrane, causing them to be recognized as senescent cells.[1] Senescent red blood cells are taken up by macrophages and phagocytized in the spleen, bone marrow (reticuloendothelial cell macrophages) and liver (Kupffer cells) (Figure 9-2). This internal erythroid circuit removes and replaces 0.66% of the body's total iron content each day.[1] The daily requirement for the erythron alone is 20 mg of iron and yet only 1–2 mg of iron normally enters the body each day through the intestine.[36] The heme portion of hemoglobin is catabolized by hemoxygenase to biliverdin and subsequently, bilirubin.[50] Bilirubin is secreted into the bile for excretion from the body. The amount of iron that is recycled from effete red cells and returned to plasma transferrin approximates the amount of iron needed for the synthesis of new erythrocytes.[50]

Storage

Most of the iron store is located in the spleen, liver, and bone marrow in the form of ferritin.[37] Iron is stored in the form of ferritin or hemosiderin. Ferritin is comprised of 24 similar polypeptide subunits that form a cage isolating iron from the cellular milieu. Ferritin exists in at least two different isoforms (H and L).[2] The relative proportions of the two subunits vary by tissue type and development. The H form is predominant in the heart; whereas the L form is predominant in the liver. The liver contains about 60% of the ferritin in the body with the remaining amount present in muscle tissues and cells of the reticuloenthelial system. Macrophages act as a storage depot for iron recovered from effete erythrocytes or from medicinal forms of iron administered by injection. Their storage capacity is evident when tissues from patients with transfusional iron overload are stained for iron; large amounts of iron accumulate in macrophages before deposition in parenchymal cells becomes apparent.[50]

Excretion

Only a small amount of iron is lost each day (Figure 9-2). Daily basal losses of iron are limited to 0.90 to 1.02 mg/day in non-menstruating women.[37] The amount of iron lost during menstruation is variable. The mean loss is estimated to be between 0.6 and 0.7 mg/day, which translates into a loss of 17.5 mg iron per period.[52] However, basal losses have been reported to decline to about 0.5 mg/day in an iron-deficient state and increase to about 2 mg/day during iron overload.[53] Non-physiologic causes of iron losses include bleeding from gastrointestinal ulcers, intestinal parasites, cow's milk feeding to infants, aspirin, non-steroidal anti-inflammatory drugs or corticosteroid administration, or hemorrhage induced by surgery or injury.[3]

SECTION 5.1: A STUDY OF CLINICAL CASES

Since the 1970s, many studies have shown that feeding cow's milk to infants is associated with diminished iron stores and an increased probability of iron deficiency.* Several mechanisms have been postulated including: low iron content of cow's milk, which makes it difficult for infants to obtain iron requirements for growth; occult intestinal blood loss associated with cow's milk and inhibition of non-heme iron absorption by calcium and casein, which are present in high amounts in cow's milk.* There is concern that the feeding of unmodified pasteurized cow's milk also places infants at high risk of severe dehydration and may increase the risk of obesity in childhood.

Further discussion points:

What are the possible adverse effects of cow's milk owing to it high protein and electrolyte content?

Why would the feeding of unmodified pasteurized cow's milk be associated with increased risk of severe dehydration?

Why would the feeding of cow's milk be associated with increased the risk of obesity in childhood?

* Ziegler E. Consumption of cow's milk as a cause of iron deficiency in infants and toddlers. Nutr Rev. 2011;69:S37–S42.

SECTION 6: HOW DOES THE BODY REGULATE IRON ABSORPTION?

Humoral regulatory mechanisms for iron absorption include: (a) *dietary regulator*, (b) *stores regulator* and (c) *erythropoietic regulator*.

Dietary Regulator

Absorption of intestinal iron can be modulated by the amount of iron recently consumed. Absorptive enterocytes are resistant to acquiring additional iron several days after a dietary bolus.[36] This phenomenon is commonly referred to as the "mucosal block" theory. The hypothesis is that the amount of iron that is absorbed is dependent on previous exposure of the gastrointestinal tract to iron.[44] It is postulated that multipotent precursor cells in the epithelial layer of the duodenum act as sensors of body iron needs. Iron levels are programmed by enterocytes in the crypt thus modulating iron absorption at the villus.[19] Upon differentiation new proteins required for iron absorption, storage and export are expressed in the enterocyte. The lag period between injection of iron and change in iron absorption is thought to represent the time taken for cell differentiation and migration.

Stores Regulator

The store regulator senses iron levels, but instead of responding to dietary iron, appears to respond to total body iron.[36] After ingestion of foods or iron supplements, higher non-heme iron absorption occurs when stores are depleted and lower absorption occurs when stores are replete or enlarged.[44,54] An inverse linear relationship has been described between iron absorption and serum ferritin (a measure of iron stores). It has been suggested that once iron stores are replete there is a "set-point" or steady-state above which they do not further increase iron absorption. This predetermined "set-point" for iron stores is believed to vary between individuals.

Erythropoietic Regulator

The erythropoietic regulator has been postulated to modulate iron absorption in response to acute marrow iron needs.[7] It is thought that this mechanism senses ineffective erythropoiesis such as occurs in congenital disorders of red cell maturation (i.e., thalassemia, sideroblastic anemia). Anemic states that stimulate iron absorption, such as thalassemia syndromes, congenital dyserythropoietic anemias and sideroblastic anemias all involve destruction of erythroid cells near the site of their development, within the bone marrow. Anemias that are characterized by

peripheral destruction of red cells, such as sickle cell anemia, hereditary spherocytosis and autoimmune hemolytic anemia, do not cause increases in iron absorption leading to the speculation that another component of erythropoiesis other than its rate must explain the absorptive response.[40]

SECTION 7: HOW DOES THE BODY TRANSPORT IRON?

The processes that transport iron from stores in the circulating transferrin pool and out of it to supply cellular demands are both highly regulated. Iron is transported by transferrin (Tf), a plasma protein predominantly synthesized in the liver. Tranferrin can acquire iron from dietary absorption or from heme that has been degraded by the phagocytosis of senescent red blood cells. High-affinity binding of iron to Tf at physiological pH is required to prevent the toxic effect of iron ions, making it difficult to develop iron chelators that compete with transferrin for iron binding.[11]

The 80-kDa serum iron-binding protein reversibly binds to two atoms of iron.[4] The polypeptide chain is arranged in two lobes, representing the N-terminal and C-terminal halves of the molecule (Figure 9-4). When devoid of iron, a transferrin lobe takes an open conformation with the domains separated. Once iron is bound, the domains undergo a rigid rotation to enclose their iron-binding site, thereby guarding bound iron from hydrolysis and too-facile release.

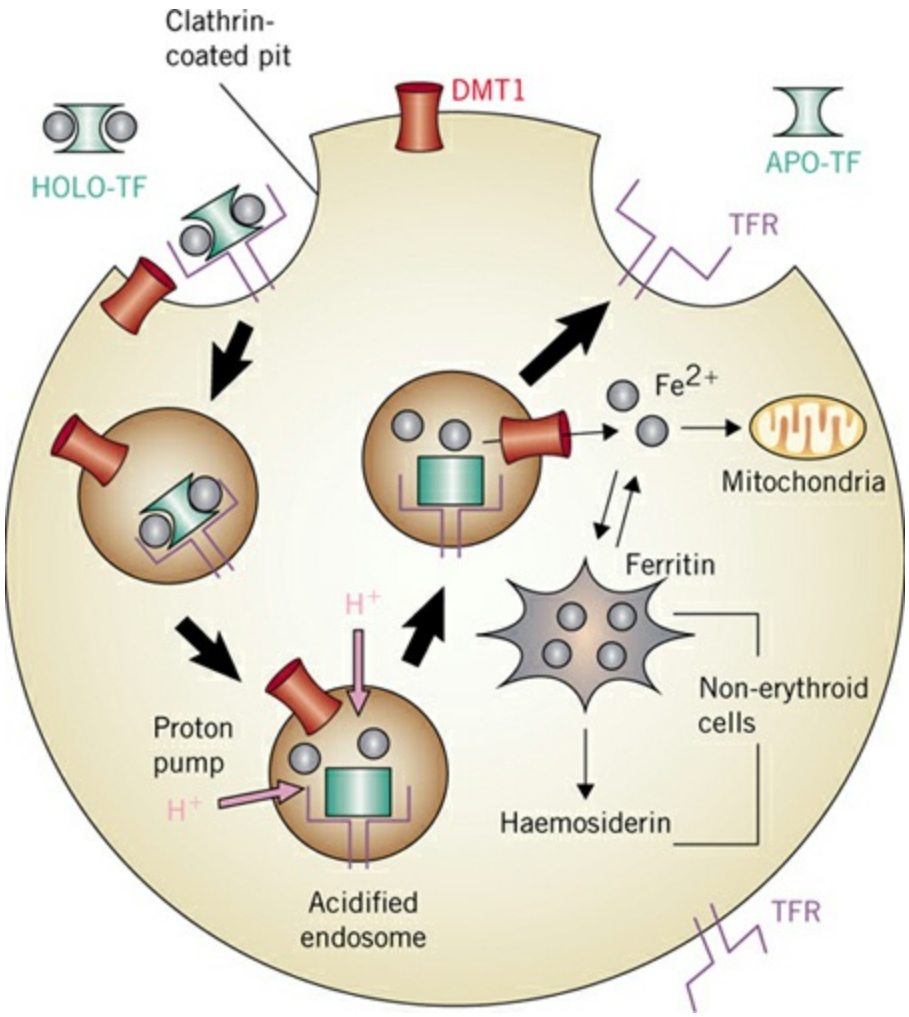


Figure 9-4 Cellular iron transport. The transferrin cycle. HOLOTRANSFERRIN (HOLOTF) binds to transferrin receptors (TFR) on the cell surface. The complexes localize to clathrin-coated pits, which invaginate to initiate endocytosis. Specialized endosomes form, and become acidified through the action of a proton pump. Acidification leads to protein conformational changes that release iron from transferrin. Acidification also enables proton-coupled iron transport out of the endosomes through the activity of the divalent metal transporter 1 protein (DMT1). Subsequently, APOTRANSFERRIN (APO-TF) and the transferrin receptor both return to the cell surface, where they dissociate at neutral pH. Both proteins participate in further rounds of iron delivery. In non-erythroid cells, iron is stored as ferritin and hemosiderin.

Normally, all non-heme iron in the circulation is bound to transferrin. However, only ~30% of transferrin binding sites are occupied, so most of the protein is free of iron. Normal individuals never have a full saturation

of all Tf iron-binding sites, thus providing a safety margin to accommodate fluctuation of iron levels from dietary and environmental changes.[2] Complete saturation is only seen in pathological states such as hemochromatosis and transfusional iron overload. Transferrin receptors are able to distinguish between iron-free and iron-loaded transferrin, ensuring cells do not waste efforts taking up the former.

Transferrin receptors are firmly anchored in cell membranes; with the major portion of the protein being external to the cell.[1] The density of surface transferrin receptors on a given cell is proportional to the cell's iron requirements. In most tissues, however, the receptor number falls sharply as the cell matures. Two exceptions are placental cells, and erythroid precursors—both rapidly dividing cells where large amounts of iron are required throughout the life span.

Transferrin-iron complexes attach to transferrin receptors (TfR) on the cell surface. The entire complex of Tf-iron and TfR are invaginated by part of the cell wall to form a vacuole. The complex is internalized by receptor-mediated endocytosis into clathrin-coated endosomes.[19] The pH in endosomal compartments is rapidly lowered through the action of an ATP-dependent proton pump.[4] The acidic milieu facilitates conformational changes in the transferrin and transferrin receptor proteins.[50] Iron is released from the complex and reduced to its ferrous form. Iron is then transported across the endosomal membrane into the cytoplasm by divalent metal transporter 1 (DMT1, previously known as Nramp2, DCT1). The TfR/Tf complex returns to the cell surface where the physiological pH causes transferrin to be released and recycled into the circulation.[43]

SECTION 8: WHAT ABOUT TOO *LITTLE* IRON IN THE BODY?

Iron deficiency disproportionately affects infants, young children, adolescents, and women of childbearing age, particularly individuals who live in developing countries because their diets provide insufficient bioavailable iron to meet their needs for growth, the replacement of menstrual losses, and pregnancy. The most important clinical clue of moderate to severe chronic iron deficiency in adults is chronic fatigue. Other features include: pallor of the skin and of the conjunctiva, stomatitis, ice eating (pagophagia), leg cramping, koilonychia, glossitis, gastric atrophy and dysphagia.[55] It is also possible that some individuals with iron

deficiency may not experience any symptoms.[1] Since clinical signs of anemia are not specific, it is recommended that biochemical tests be used to determine iron deficiency and its magnitude.[56]

Etiology

The etiology of iron deficiency can be categorized into either physiological or pathological causes.[55] Physiological causes relate to a disparity between iron requirements and the maximum absorption of iron from the diet.[57] Nutritional iron deficiency, a term that has been widely used to describe conditions where the diet cannot cover physiologic iron requirements is the most common cause of iron deficiency worldwide.[55] Segments of the population at risk for not meeting high iron requirements during periods of rapid growth include infants, adolescents and child-bearing females.[55] Impaired absorption of iron can occur in patients with achlorhydria and who have undergone gastric surgery. Gluten enteropathy is also associated with IDA, especially in young patients.[22]

Pathological iron deficiency results from excessive blood loss, which usually occurs in the GI tract.[55] In clinical practice a diagnosis of iron deficiency includes a search for pathologic causes of blood loss (e.g., tumors in the gastrointestinal tract or uterus, especially if uterine bleedings have increased or changed in regularity). Blood loss can occur from chronic use of aspirin or other non-steroidal anti-inflammatory drugs or parasitic infestations (i.e., hookworm).[22]

Epidemiology

In the World Health Report 2002 (“Reducing Risks, Promoting Healthy Life”) nutritional iron deficiency was identified as one of the 10 leading risk factors for disease, disability and death in the world today, with an impact greater than that of either zinc or vitamin A deficiency.[58] In 2008, the Copenhagen Consensus (a not-for-profit “think-tank” lead by a panel of economic experts) ranked “malnutrition and hunger” as the top ranked global challenge and iron fortification as the third highest priority for addressing this challenge.[59] Up to five billion people (80% of the world’s population) are affected by iron deficiency, most of whom live in developing countries.[60]

While it is well recognized that iron deficiency is the single most common nutritional disorder worldwide and the leading cause of anemia during infancy, childhood and pregnancy[61]; obtaining accurate estimates

of its prevalence has proven to be a challenging task. Lack of national data and a single, inexpensive and easy to use indicator of iron status, are just two of the many hurtles in assessing the prevalence of iron deficiency and its anemia.[62]

In 1991, a global Vitamin and Mineral Nutrition Information System (VMNIS) was established by the Nutrition Unit of the World Health Organization to strengthen worldwide surveillance of micronutrient deficiencies. The VMNIS supports Member States in monitoring the vitamin and mineral nutrition status of populations at the national, sub-national, regional and first-administrative level.[63] These data are compiled in a large comprehensive Micronutrients Database, which is used to monitor micronutrient status around the world, and provide global estimates of the prevalence and trends in micronutrient deficiencies over time. Currently the Micronutrients Database contains information on anemia, vitamin A and iodine deficiency.[63]

In the United States, data from the National Health and Nutrition Examination Survey (1999–2006) found the prevalence of iron deficiency to be 14.4 ± 1.9% in toddlers aged 1–2 years, 3.7 ± 0.8% in children aged 3–5 years, 9.3 ± 1.0% in adolescent females, 9.2 ± 1.6% in women,[64] and 18.0 ± 1.4% in pregnant women.[65]

Definition

Broadly defined, iron deficiency is a condition in which there is a lack of cellular iron to the tissues.[6] Iron deficiency represents a spectrum of mild to severe forms characterized by the presence or absence of physiological, biochemical, hematological and functional changes. Therefore, various definitions of iron deficiency have been formulated. The development of iron deficiency anemia is commonly thought of as a progression of *three* successive stages.[66]

The first stage—iron depletion (ID), is characterized by a mobilization of storage iron. A fall in serum iron reflects the progressive reduction of storage iron in the liver; however, the supply of iron to the functional compartment remains uncompromised. Levels of transport iron and hemoglobin remain normal.

The second stage—early functional iron deficiency (previously called iron-deficient erythropoiesis), involves a decrease in transport iron resulting in low levels of serum ferritin and low serum transferrin saturation and elevated levels of serum-transferrin receptor (TfR) and free erythrocyte protoporphyrin (FEP).[41] This stage is commonly referred to as

iron deficiency without anemia. The iron supply required for full maturation of eyrthropoietic cells is markedly reduced; however, mature red blood cells stay unaffected. Hemoglobin concentrations usually remain within in the normal range, although they may decrease slightly. Unlike iron deficiency anemia, in which iron stores are totally absent, the IDE can occur with normal or increased amounts of stored iron due to the impaired release of iron in the plasma from the storage iron compartment.[55]

The third and final stage is established functional iron deficiency, often referred to as: iron-deficiencyanemia (IDA). IDA develops when the supply of transport iron decreases to the point where hemoglobin synthesis is restricted. During this stage iron stores are exhausted, circulating levels of iron are declining and microcytic, hypochromic anemia is evident. A restricted supply of iron to the bone marrow causes a reduction in the concentration of hemoglobin in the red blood cells and an observable decline in hematocrit and red cell indices.

Risk Factors

Iron deficiency can arise from several risk factors, alone or in combination. Examples from the range of possible risk factors include: (a) inadequate iron supply, (b) increased iron requirements imposed by growth, and (c) increased iron losses.

Inadequate iron supply can be the consequence of low iron stores or low iron in the diet. At birth, low iron stores can result from low birthweight, perinatal bleeding, early clamping of umbilical cord and maternal iron deficiency. Low birthweight influences iron stores in a linear proportion; a newborn weighing 1500 has roughly half the iron content of a 3000 g well-grown term newborn.[9] Preterm infants (born before 32 weeks gestation) encounter low body iron stores because they miss out on the accretion of iron during last trimester of pregnancy.[8] In addition, preterm infants are at increased risk of iron deficiency from postnatal practices such as uncompensated phlebotomy losses, administration of recombinant human erythropoietin (rhEpo) and inadequate iron supplementation.[8] The timing of the clamping of the umbilical cord at delivery has been shown to impact the amount of blood transferred to the infant.[67] The timing of umbilical cord clamping depends on whether the cord is clamped early (less than a minute), intermediately (1–3 min), or late (after pulsations cease). For example, if the cord is not cut until pulsations cease, the infants will receive a 20–30 mL/kg body weight transfusion, which is the equivalent to approximately 30–35 mg of

additional iron.[67] To put this amount of iron into perspective, this is the equivalent to the amount of iron in 100 l of human milk.[67]

Maternal iron deficiency has been shown to affect infant iron stores and iron status. It was previously held that infants acted as parasites with respect to sequester iron from their mother in utero. However, current evidence suggests that maternal iron stores over a wide range determine iron stores in the neonate[68–73] and that even moderate degrees of maternal ID (Hb 85–100 g/L) may result in lower body iron stores during infancy in the offspring.[68] In developing countries, maternal ID (Hb < 85 g/L) affects 30%–50% of pregnancies and is arguably the most common cause of perinatal iron deficiency.[8]

Nutritional iron deficiency results from reduced dietary iron and/or low bioavailability of dietary iron. Despite the high bioavailability of the iron in human milk, human milk contains very little iron, thus iron stores in the exclusively breastfed infant will ultimately be depleted after 6 months of age.[74] If human milk remains the only food source, iron deficiency anemia will ultimately develop. The choice of complementary foods will markedly influence the risk of iron deficiency and anemia. Unfortunately, in developing countries typical home-prepared grain or rice-based complementary foods are poor sources of iron and may contain phytic acid, which is a potent inhibitor of iron absorption. For example, per 100 g, unfortified rice-based complementary foods contain 1 mg of iron and maize and wheat-based complementary foods contain 0.8 mg of iron.[75]

Adolescents at risk for nutritional iron deficiency include those on a restrictive diet, suffering from an unhealthy body image or practicing vegetarianism (restrictive intake of heme iron sources).[74] Many adolescent girls inadvertently limit iron intake by trying to control their weight or by passing through temporary period of vegetarianism because of concerns for animal warfare.[76] Vegetarians can achieve an adequate intake of iron, but have to purposely choose foods with a high concentration of non-heme iron or choose foods fortified with iron (see Table 9-2). Women who conceive during or shortly after adolescence are at increased risk for entering pregnancy with low or absent iron stores and subsequently developing iron deficiency during pregnancy.[18] The available data on iron intake in adolescents suggest that adolescent girls are unlikely to acquire substantial iron stores during this time period because intakes may average as little as 10–11 mg iron/day.[18] The bioavailability from diets in developing and industrialized countries indicates a negative iron balance is likely in many female populations.

An important and common risk factor for ID is increased iron requirements imposed by growth. During the first 2 years of life, infants require relatively large amounts of iron due to their high growth rate. On average, during the first year of life, the birth weight and blood volume of the full-term, normal birth weight infant will triple.[77] In the postnatal period preterm infants are risk of ID because of rapid growth rates, uncompensated iron losses from phlebotomy and low nascent iron stores.[30] Other periods of high iron requirements include the growth spurt during adolescence and pregnancy. Iron requirements during adolescent pregnancies are particularly high because girls in their teens may not have reached their full growth.[11]

Iron deficiency can also result from increased iron losses. In young women of child-bearing age menstrual blood loss (both "heaviness" and duration) can be a major determinant of iron status.[78] In hospitalized infants the most common cause of blood loss is frequent blood sampling.[9] Since the majority of fetal iron is present as Hb, any blood loss that lowers neonatal Hb concentration (including twin-to-twin transfusion and fetal-maternal hemorrhage, perinatal hemorrhage, considerable blood draws after birth) will have an impact on iron balance.[9] Gastrointestinal bleeding is uncommon in otherwise healthy infants, but when it does occur, it is usually due to food allergies, repeated episodes of acute diarrhea or *Helicobacter pylori* infections.[61] In tropical areas, gastrointestinal bleeding may be due to infestation with parasites such as hookworms, whipworms and round worms or hemoglobinuria may cause blood loss due to malaria.[79] Occult gastrointestinal bleeding among formula-fed infants because of cow's milk protein sensitization or among healthy infants owing to feeding with fresh cow's milk may result in negative iron balance in the first year of life.[9] The precise cause of blood loss not well understood but is likely to be an allergic-type reaction between a protein in cow's milk and enterocytes of GI tract.[14] Increased fecal blood loss in response to cow's milk is more common among infants who were breastfed early in life than in infants who were never breastfed.[9] Earlier exposure to cow's milk proteins may sensitize infants who are predisposed to allergy.[80]

Consequences of Iron Deficiency

Clinical sequelae of iron deficiency include impaired cognitive performance, anemia, poor immune function and decreased work performance (Table 9-3).[81] In children, numerous studies have consistently

demonstrated that even moderate anemia (hemoglobin < 100 g/L) is associated with depressed mental and motor development in children.[82–84] Although the mechanism whereby iron influences development is not fully understood, older research identified the role of iron in central nervous system neurotransmitter function, and more recent work has shown that brain iron is essential for normal myelination.[85–87]

TABLE 9-3 Functional Consequences of Iron Deficiency

Pregnancy	Infants and Young Children
Increased risk of anemia and maternal morbidity	Motor and cognitive developmental delays in infancy
Increased risk of prematurity and lower birth weight	Effects on emotional maturation
Higher infant mortality	Poorer academic achievement in school-age children who were iron deficient in early childhood
	Increased risk of severe morbidity from malaria in children
	Increased prevalence and duration of upper respiratory tract infections in children

It used to be presumed that iron deficiency anemia had few deleterious effects unless severe enough to compromise cardiovascular function.[88] However, over the last two decades, the negative impact of iron-deficiency anemia (IDA) in infancy on psychomotor development has been well documented in more than a dozen studies.[89] Longitudinal studies have indicated that children who were anemic in early childhood continue to have poor cognitive and motor development and depressed school achievement into middle childhood.[83] Studies in human infants with iron deficiency anemia confer findings from rodent models which document poorer performance on global assessments of cognitive, motor, and social-emotional behavior[90] and alterations in regulatory processes such as the sleep-wake cycle.[91] Iron deficiency in infancy has been associated with short- and long-term alterations related to major dopamine pathways (mesocortical, mesolimbic, nigrostriatal, tuberohypophyseal).[92] For anemic children less than 2 years of age, there is no good evidence from randomized controlled trials that iron treatment helps their cognitive and motor development.[82] For children above 2 years of age, short-term

treatment is associated with improvement in cognition but not in school achievement.

The study of cognitive function and iron status has been extended to older children and adolescents. In a randomized controlled trial, adolescent girls with depleted iron stores, who received iron supplements improved their scores on a test of memory and verbal learning compared with the placebo-treated group.[93] Another large study in preadolescents and adolescents described an association between iron status and standardized mathematics scores.[94] Those with iron deficiency anemia or iron deficiency without anemia were 2.3 and 2.4 times more likely to have low mathematics scores than those with no iron deficiency. The authors of this report suggested that screening for iron deficiency may be warranted for all children and adolescents.

During pregnancy, the negative implications of anemia caused by iron deficiency alone or in combination with other factors, e.g., folate deficiency, vitamin A deficiency, and malaria, include increased risk of preterm delivery and infant low birth weight.[95–98] There is some evidence to suggest an association between a higher risk of maternal mortality and severe anemia.[68,99] However, data were predominantly retrospective observations of maternal hemoglobin concentrations at, or close to, delivery and subsequent mortality.[68] Furthermore, the iron deficiency component of this relationship is unknown.[79] Mechanisms by which iron deficiency anemia could increase maternal mortality need to be elucidated.

Increased risk of maternal mortality has also been shown in women with severe anemia.[99] Children born to adolescent mothers are more likely to be of poor health and have a higher likelihood of infant mortality.[99] Concern for the health of young mothers and their infants has led to suggestions that adolescence may be an ideal time to optimize iron status and prevent iron deficiency during pregnancy.[100]

Rapid expansion of maternal blood volume and red cell mass during pregnancy and placental and fetal growth leave many women, even in developed countries, with low iron stores and tissue iron deficiency at parturition.[101] The consequences of poor maternal iron status postpartum are increasingly being recognized as a non-benign condition, shown to be related to maternal fatigue, depression and ability to care and nurture offspring.[102–104]

Included in an exciting surge of research in the last decade on iron's role during brain development has been recent work in the rodent model that shows that brain iron is essential for normal myelination. Regional

differences in brain iron concentration and cytochrome *c* oxidase (CytOx) activity were measured in young rats born to dams on iron-deficient or iron-sufficient diets throughout gestation and early lactation.[105] Markedly reduced neuronal metabolism in all regions of the hippocampus was found, demonstrating the vulnerability of the developing hippocampus.[105] Studies not only show that iron deficiency during gestation and lactation in the rat results in reduced brain iron, but also that the degree varies greatly by brain region.[106] New research also shows altered gene and protein profiles.[106]

Prevention

In light of accumulating evidence regarding the adverse effects of iron deficiency anemia on maternal and infant outcomes, prevention rather than treatment is highly recommended. If implemented successfully, there are three interventions that are likely to prevent anemia. These include (a) ***dietary diversification*** to foods with more bioavailable iron; (b) ***fortification*** of foods targeted to full-term infants and children; and (c) ***iron supplementation***.

Diet

For infants 0–6 months, it is assumed that iron provided by human milk is adequate to meet the iron needs of exclusively breast fed infants born at normal birth weight from mothers with good prenatal iron status.[107,108] Milk iron composition is unaffected by maternal iron intake or status.[109,110] While human milk iron content is low (0.2–0.4 mg/L) it is highly bioavailable (45–100 per cent) compared to cow's milk-based infant formula (10 per cent).[9,14] Since the average iron concentration in human milk is 0.35 mg/L, the adequate intake (AI) level is set at 0.27 mg/day (0.78 L/day × 0.35 mg/L).[14] However, even after taking a high estimate of iron absorption from human milk (50%) into consideration and assuming an iron intake of 0.075 mg/kg/day from human milk, it is estimated that term infants will absorb less than 0.04 mg/kg/day.[110] Since this iron intake is less than the amount required for growth, storage iron will utilized until dietary iron intake increases. After 6 months of age, the introduction of heme-iron containing foods or iron-fortified foods may support normal iron status; therefore, it is recommended that iron-fortified formulas and iron-containing foods be introduced once exclusive breast-feeding has stopped. The iron content of preterm human milk is approximately 0.5 mg/L.[8] Because preterm infants are at high risk for iron

deficiency because of a lack of iron accretion during the last trimester of gestation, preterm infants fed exclusively human milk are recommended to receive an iron supplement at a dose of 2–3 mg/kg per day from 1 to 12 months.[107,112]

In contrast to human milk, cow's milk has a low concentration of iron (around 0.4 mg/L) and low bioavailability because of its high calcium and phosphorus and low ascorbic acid content.[9] Cow's milk and unfortified cow's milk formulas cannot supply the infant with sufficient amounts of iron to maintain an optimal iron balance during the first 6 months of life.[9] Introduction of cow's milk is not recommended until 9–12 months of age according to the Canadian Pediatrics Society and the American Academy of Pediatrics.[107,113]

The WHO (2004) recommends introducing iron-rich complementary foods at 6 months of age to meet iron needs.[108] For infants over the age of 6 months, dietary diversification is recommended as an intervention strategy for preventing iron deficiency. Dietary diversification involves promotion of a diet with a wider variety of natural iron-containing foods, especially red meat, poultry, fish and food rich in vitamin C. The selection of weaning foods determines in large part whether an infant will be at risk of developing iron deficiency and associated anemia.[114] Typical home-made complementary foods used in developing countries are poor sources of bioavailable iron and thus, are inadequate to meet infants' high iron requirements for rapid growth and blood volume expansion.[108]

The US/Canadian recommended dietary allowance for iron (mg/day) is 11 (6–12 months), 7 (1–3 years) and 10 (4–8 years) (Table 9-4). Two major factors should be taken into consideration when determining the iron requirements of children. The first is the iron need of the individual and the second is the bioavailability of iron in the diet. Estimates of dietary iron requirements for infants over the age of 6 months have been modeled using the factorial components of absorbed iron requirements. The four major components of iron need have been assessed for infants aged 7–12 months and children aged 1–8 years: obligatory fecal, urinary, and dermal losses; the increase in hemoglobin mass; the increase in tissue iron, and the increase in storage iron. Included in the factorial model are the type of iron, heme or non-heme, and other dietary factors that may influence the iron bioavailability in the diet. The diets of infants tend to be high in cereals and vegetables with little meat, and are therefore low in heme iron. The iron in high cereal diets is considered to have a medium bioavailability of 10%.[74]

TABLE 9-4 Recommended Dietary Allowance (RDA) of Iron for Both Sexes from Birth to 18 Years[14]

Age	RDA for total iron intake, mg/day
0–6 months	0.27
6–12 months	11
1–3 years	7
4–8 years	10
9–13 years	8
14–18 years	11 for boys; 15 for girls
Pregnancy	27
Breastfeeding mothers under the age of 19 years	10

The "estimated adequate requirement" (EAR) for children and adolescents ages 9 through 18 years has been determined by factorial modeling of the median components of iron requirements.[14] The physiological processes associated with puberty that have a major impact on iron requirements are the growth spurt in both sexes, menarche in girls, and the major increase in hemoglobin concentrations in boys. Total iron requirements for adolescent boys are computed from increased iron requirements required for the expansion of the total blood volume and the increase in the lean body mass. The increase in mean total iron requirements may exceed 1.8 mg/day, or more than double those requirements of preadolescent males.[13] Iron requirements for adolescent girls include the amount of iron lost during menses in addition to the growth requirements. In female adolescents, the recommended dietary allowance (RDA) increases from 8 mg Fe/day for those 9–13 years of age to 15 mg Fe/day for those 14–18 years of age (Table 9-4).

Fortification

There are three types of fortification strategies: mass fortification; open market fortification; and targeted fortification. Mass fortification refers to the addition of micronutrients to foods consumed by all or most of the population. Mass fortification is regulated by governments. Wheat flour has been fortified in Canada, the United Kingdom and the United States since the 1940s. National fortification programs were also in place in

Denmark until 1987 and in Sweden until 1994.[115] Wheat is the cereal flour that is fortified most often, but several countries—particularly in Central and South America—also have programs for iron fortification of maize flour.[115] In developing countries mass fortification programs of fish sauce (in Thailand and Vietnam), sugar (in Guatemala), curry powder (in South Africa), and soy sauce (in China) have resulted in improved iron status of their populations.[115]

Open market fortification refers to the initiative of a food manufacturer to fortify one or more products with the aim of improving both public health and product profitability. Open market fortification of several processed foods, particularly breakfast cereals, is widely practiced, primarily in developed countries. Fortification of staple foods is likely to increase iron intake for those who eat the most (i.e., adult males). The level of fortification is purposely kept low to avoid iron overload in populations most likely to eat the most fortified foods (adult males). However, infants and children are not likely to benefit significantly from this strategy because they have a limited capacity to eat large quantities of food and the concentration of iron in fortified staples is purposely low.

Targeted fortification (e.g. infant formula and infant cereals) describes the specifically directed fortification of foods consumed by high-risk groups. Targeted fortification provides an excellent source of iron to those who need it the most. Infant cereals, for example, typically contain 7.1 mg of iron per 15-g serving, and breakfast cereals for children contain over 4 mg of iron per 30-g serving.[21] A single 30-g serving of these “ready-to-eat” cereals provides 60% of the RDA for children aged between 1 and 3 years, 45% of the Recommended Daily Allowance (RDA) for children aged 4–8 years and 75% for boys and girls aged 9–13 years. For adolescents, especially those who are restricting their total food intake or who are vegetarians, the use of iron-fortified breakfast cereals that typically contain 4–8 mg/serving is an excellent way to meet their iron requirements and to prevent anemia. In fact, ready-to-eat cereals are among the top contributors of iron, folate, vitamin A and C, and zinc intakes.[116]

The American Academy of Pediatrics (AAP) recommends that infants who are partially breastfed (receive more than half of their daily feedings as human milk) should also receive an iron-fortified formula or 1 mg/kg/day of supplemental iron beginning at 4 months of age.[107] For term formula fed infants, the AAP recommends a standard infant formula with an iron content of 10–12 mg/L for the first 12 months of life,[107] which will

provide approximately 2 mg/kg/day if the infant's energy intake from formula is adequate. Most infants, including VLBW infants should be offered formulas with iron at the time formula is introduced.

Term infants who are exclusively breast-fed do not need supplemental iron until they are 6 months of age. If solid foods are introduced earlier, they should contain an adequate amount of iron. After 6 months of age, breast-fed infants should receive extra iron in the form of iron-fortified infant cereals and other iron-rich foods. These infants should be offered an iron-fortified infant formula after they have been weaned from breast milk. Term infants who are not breast-fed should be given an iron-fortified infant formula from birth. Studies are under way to determine the optimal iron content of these formulas: and further studies are encouraged.

For premature infants, an iron supplement should be started by at least 1 month of age and continued until the first birthday.[107] Iron-fortified formula for bottle-fed infants or commercial iron drops for breast-fed infants are the recommended source of supplemental iron. Current preterm formulae available in USA fortified with 14.6 mg/L of iron provide 2.2 mg/kg per day if consumed at 150 mL/kg per day.[107] Despite the use of iron-containing formula, some preterm infants may require an additional iron supplement, although there is currently insufficient evidence to make a general recommendation in terms of the dose, route and commencement of supplementation to prevent ID in preterm infants.[107]

Iron Supplements

Another approach for preventing or treating iron deficiency is through supplementation of individuals or communities at risk. This approach would be implemented for the treatment of individuals with anemia or in situations where at-risk communities of infants and young children do not have ready access to targeted iron-fortified foods. Global guidelines for iron supplementation have been published by the Nutritional anemia Consultative Group/World Health Organization/UNICEF.[117] For children 6 to 24 months of age 12.5 mg/day of iron is recommended, in tandem with 50 μg/day of folic acid until 12 months of age. Recommendations for children and adolescents are as follows: 20–30 mg/day for children 2 to 5 years old; 30–60 mg/day for children 6 to 11 years old; 60 mg/day for adolescents and adults (plus 400 μg/day of folic acid for child-bearing aged females); 60 mg/day in addition to 400 μg/day of folic acid for pregnant women, continuing for 3 months of lactation where anemia prevalence is high (>40%).

The most common form of iron in iron supplements is ferrous sulfate;

however, fumarate and gluconate are also used. Infants are most commonly supplemented with a liquid formulation. When a soluble form of iron (such as ferrous sulfate or fumarate) is ingested in the proper dose, this intervention is efficacious. However, adherence to long-term ingestion of oral iron drops is often poor because of the unpleasant metallic taste of drops; drops can stain a baby's teeth unless wiped off immediately after use; and if the dose is high, the infant may demonstrate abdominal discomfort.

In many women, it is uncertain whether diet alone can provide the additional iron needs of pregnancy.[118] The need for iron supplementation during pregnancy is actively debated.[119–121] Most studies in pregnant women supplemented with iron show a change in total blood volume of <45%, with an increase in plasma volume of <50% and an increase in red blood cell mass of <35%.[17] While controlled trials of iron supplementation during pregnancy have consistently demonstrated positive effects on maternal iron status at delivery; the impact of iron supplementation in reducing factors that are associated with maternal anemia, that is, increased risk of preterm delivery and infant low birth weight is less clear.[95] While micronutrient deficiencies during pregnancy are thought to contribute to neonatal mortality; few trials have sufficient data or sample sizes to report on neonatal outcomes. However, two large double-blinded, placebo-controlled randomized trials conducted in Nepal have found a significant impact of early iron supplementation during pregnancy on low birth weight (LBW), as defined as < 2500 g.[122–124]

Currently, iron supplements are almost universally recommended for pregnant women. In developed countries, pregnant women are recommended to take 27 mg of iron daily.[14] For iron supplementation programs where anemia prevalence is high, the International Nutritional anemia Consultative Group (INACG) has recently changed its recommendation from 120 to 60 mg/day. However, if duration of iron supplementation during pregnancy is short, or if anemia is present, 120 mg/day is still recommended. The administration of iron supplements weekly instead of daily in humans (in which gut mucosal turnover occurs every 5–6 days) is actively being investigated as a viable means of controlling iron deficiency in populations, including pregnant women.[18]

When considering iron supplements, attention should be paid to possible interactions between iron and nutrients. Iron deficiency impairs vitamin A and iodine metabolism and may hinder the effectiveness of interventions aimed at improving these deficiencies.[125] A direct

relationship has been found between hemoglobin and serum retinol concentrations.[126] Dietary deficiencies of vitamin A and iron frequently co-exist in developing countries and vitamin A deficiency appears to affect iron transport and red blood cell production.[126] Effects have not been demonstrated in vitamin A sufficient subjects.[126] Studies in anemic children and pregnant women in regions with endemic vitamin A deficiency have shown improved iron status post vitamin A supplementation.[61] Due to the anti-infective properties of vitamin A, it has been suggested that the benefits of vitamin A on iron status may be a result of reduced level of infection.[61] Some concern has been raised regarding the potential adverse effects of food iron supplements on zinc nutrition. While reduced absorption of one of the metals may occur when solutions of inorganic iron and zinc are administered, there appears to be little support for a significant effect when the metals are given with food in doses that are considered optimal for fortification.[126]

The interaction between iron and infection has been a continuing debate, largely due to associations between iron treatment and exacerbations of infection, in particular malaria.[127] A review of 13 randomized controlled trials determined that the known benefits of iron supplementation likely outweighs the risk of adverse effects in regions where malaria is endemic.[117] However, the results of two large-scale randomized controlled trials designed to evaluate the impact of iron and zinc supplementation on mortality and morbidity in young children found that iron supplementation may be associated with adverse effects in areas where malaria transmission is high.[128,129] Following these two trials, recommendations were made to target supplementation of iron and folic acid to children who are anemic or iron deficient and to provide children with concurrent protection from malaria and other infectious diseases.[130,131] To date few studies of oral iron supplementation have shown deleterious effects in non-malaria or low transmission malaria regions.[132,134,135] A recent study by Soofie et al. reported increased rates of bloody diarrhea and signs of respiratory infections in a malnourished population of children receiving multimicronutrient supplements with iron.[132] In a malaria endemic region, the use of a similar micronutrient formulation reduced overall incidence of malaria and did not impact diarrhea or respiratory infections.[133]

SPECIAL TOPIC—IRON DEFICIENCY AND

ANEMIA IN INFANTS AND. YOUNG CHILDREN IN. MALARIA-ENDEMIC AREAS

The World Health Organization (WHO) coordinated and supported two large trials in Zanzibar, Tanzania, and in Nepal to evaluate the impact of zinc and/or iron–folic acid supplementation on the mortality and severe morbidity of preschool children.[128,129] In Zanzibar, an area of stable, perennial, and intense transmission of *Plasmodium falciparum* malaria, routine iron–folic acid supplements given with or without zinc resulted in an increased rate of severe adverse events in children (morbidity and mortality). In Nepal, the trial showed no difference in mortality or the incidence of the common infections. Results from the study in Zanzibar, Tanzania raised the issue of the safety of administering additional iron to infants and young children in areas of malaria endemicity.[134]

After the results of the trials were released, the WHO and UNICEF released a joint statement recommending that in malaria endemic areas, rather than using universal supplementation strategies, iron supplements should be targeted only to children who are anemic and at risk of iron deficiency.[135] This led to much debate among the nutrition research community on how to move forward, and country policy and program decision makers were uncertain as to how to address young child iron deficiency and anemia in their countries.[136] Since the release of the WHO/UNICEF recommendation, a systematic review has been published supporting the use of iron supplements in malaria endemic regions, when regular malaria surveillance and treatment services are also provided.[137] Thus, the WHO updated its recommendation and currently advises that in malaria-endemic areas, the provision of iron should be implemented in conjunction with measures to prevent, diagnose, and treat malaria.[138]

SECTION 9: WHAT ABOUT TOO *MUCH* IRON IN THE BODY?

There is no evidence that large iron stores provide a benefit to the individual. Although there is concern that a high daily intake of iron may cause decreased absorption of competing minerals; pro-oxidant effects; and aggravating symptoms in diseases in which iron absorption is increased,[12] there have no clinical studies that have demonstrated negative pro-oxidant effects of iron in healthy term infants. The decision to select 60 mg/day as The Lowest Observed Adverse Effect Level (LOAEL) was

based on a study that found a significantly increased risk of constipation, higher rates of gastric pain and diarrhea and more total gastrointestinal symptoms (nausea, gastric pain, constipation, and diarrhea) compared to placebo.[139]

Iron toxicity is related to the generation of free radicals. Normally enzymes such as superoxide dismutase, glutathione peroxidase and catalase metabolize and neutralize free radicals. Free iron has the ability to generate oxidative radicals that damage lipids, proteins and DNA. Normal and pathological cellular processes produce superoxide (O_2^-) and hydrogen peroxide (H_2O_2) byproducts. The redox capabilities of iron underlie its toxicity. The one-electron reduction of dioxygen by Fe^{2+} results in superoxide formation, which in turn, leads to the well-known Haber-Fenton sequence generating hydroxyl radical OH°(2):

$$Fe^{2+} + O_2 \rightarrow Fe^{3+} + O_2^{-\cdot}$$
$$2O_2^{-\cdot} + 2H+ \rightarrow H_2O_2 + O_2$$
$$Fe^{2+} + H_2O \rightarrow OH\cdot + OH^- + Fe^{3+}$$

The hydroxyl radical (HO), perhaps the most powerful oxidant encountered in biological systems, can inactivate enzymes, cause DNA strand breaks, depolymerize polysaccharides, and initiate chain-propagating lipid perioxidation.[140]

The body's innate mechanisms for preventing tissue damage from free radical formation are to keep iron chelated during most absorption and distribution phases, keep iron in a trivalent redox state and to store iron as ferritin.[5] Tissue iron loading occurs in two characteristic patterns. One type of tissue iron loading occurs when the amount of available plasma iron exceeds the iron-binding capacity of transferrin, thus requiring parenchymal cells of the liver to avidly take up iron. As iron overload progresses, iron is accumulated in cardiac myocytes, pancreatic acinar cells and other cells of the body.[36] A different pattern of tissue iron loading occurs when iron overload results from increased catabolism of erythrocytes, such as is often the case in patients with repeat blood transfusions. Iron first accumulates in reticuloendothelial macrophages and only later deposits in parenchymal cells. Parenchymal iron loading leads to tissue damage and fibrosis. The reticuloendothelial system is generally a safe sink for iron because reticuloendothelial macrophage sequester iron and attenuate its activity.

Iron Poisoning

Well-described cases of iron poisoning first appeared in publication in the 1940s and 1950s.[141] Classic animal experiments were conducted in the 1950s and 1960s and the chelator deferoxamine, used as an antidote for iron poisoning, was introduced in the 1960s. Almost all deaths from iron poisonings are in children under 3 years of age, resulting from ingestion of adult iron preparations (30–40 pills). However, deaths after exposure to as few as 5 pills has been reported.[142] Despite a vast number of exposures to iron-containing products, deaths rarely occur.[141] Hospitalization due to iron poisoning is an infrequent occurrence; the hospitalization rate among children aged 0 to 4 years has been estimated at 8.7 per 100,000 children.[143–145]

It is currently unknown how the highly regulated process of iron absorption is affected during an iron overdose.[146] Rapid absorption occurs through intact intestinal mucosa. In volunteer studies, a peak serum iron is observed within several hours of ingestion of supratherapeutic oral doses.[141] The clinical manifestations of iron poisoning occur in progressive stages (Table 9-5). Clinical management of iron poisoning includes clinical evaluation, fluid resuscitation, laboratory evaluation, abdominal imaging, chelation therapy, and gastrointestinal decontamination.[141]

Genetic Disorders of Iron Overload

Hemochromatosis refers to the accumulation of iron in tissue. Since the late 19th century hemochromatosis has been recognized as a distinct clinical syndrome.[147] In the 1930s a genetic predisposition for the disease was discovered; a single gene defect was found that leads to an inborn error of iron metabolism.[36] In 1976, a French physician named Marcel Simon and colleagues found that the genetic predisposition to hemochromatosis was closely linked to the human major histocompatibility complex.[3] This finding led to the positional cloning of the hemochromatosis gene 20 years later.[3] Since the discovery of the HFE gene, hemochromatosis conditions have been classified as primary (genetic) hemochromatosis and secondary.[19]

TABLE 9-5 Stages of Acute Iron Toxicity[141]

Stage 1: Local gastrointestinal (GI)	Usually occurs within several hours

effects	
	Nausea, vomiting, diarrhea
	GI bleeding may lead to hematemesis or blood diarrhea
Stage II: Quiescent phase	Resolution of GI symptoms
	Rarely seen today because children presenting with Stage I would receive fluid resuscitation and chelation therapy
Stage III: Systemic toxicity	Hypovolemic shock and acidosis
	Hypoperfusion- pallor, cold extremities, tachycardia and tachypnea
	Hypotension may be a late finding
	Myocardial dysfunction may be seen in severe iron poisoning
Stage IV: Recovery	Begins after initiation of fluid and antidotal therapy
Stage V: Gastrointestinal obstruction	Gastric and pyloric strictures (frequently require surgical therapy)
	May occur 2–8 weeks after initial injury

Hereditary Hemochromatosis (HH) is an autosomal recessive trait most commonly seen in people of Northern European descent.[148] Several mutations of the HFE gene have been described. Two main point mutations are either homozygotes for a gene designated C282Y (tyrosine, Y, substitution for a cysteine, C, mutation at position 282) or heterozygotes with C282Y/H63D (aspartic acid for histidine).[36]

Compared to unaffected individuals, patients with HH absorb two to three times more dietary iron.[36] The consequences of HH are severe; iron deposits first in hepatocytes, then in other parenchymal cells including heart, pancreas, and adrenals.[149] Excess iron in affected tissues catalyzes oxidative damage, resulting in cirrhosis, hepatoma, cardiomyopathy, diabetes, hypogonadism, and arthritis.[149] Because iron accumulation occurs gradually, the effects of the disease largely remain undetected until middle age or later.[36] In middle age, patients often present with diabetes from iron damage to islet cells and skin pigmentation from increased corticotrophin secretion (ATCH) secondary to adrenal insufficiency. Liver and gonadal dysfunction often accompany these features. One of the dilemmas of HH is that only half of patients with genetic mutations for HH show evidence of iron overload.[19] Population screening for hereditary hemochromatosis has not been supported because the penetrance is very

low.[147] Previous diagnosis depended on analysis of iron by liver biopsy, an expensive procedure not without risk; however, genetic testing has recently become available.[149] The most efficient approach to diagnose patients suspected to have the disease is to measure the serum transferrin saturation and ferritin levels.[147]

Not all patients with familial hemochromatosis disorders have mutations in the HFE gene. A subgroup of patients with non-HFE hemochromatosis that are genetically predisposed to severe iron loading during the first 3 decades of life are those with juvenile hemochromatosis (Table 9-6). These patients develop iron overload is the same tissue patterns as patients with classical HFE-associated hemochromatosis but at an accelerated rate.[3] Another disorder not associated with mutations in *HFE* is neonatal hemochromatosis (Table 9-6). Characterized by liver failure in newborn infants, iron overload occurs in the myocardium and in pancreatic acinar cells.[3] It is not clear whether the disease occurs as a result of iron loading or some other insult to developing hepatocytes. While medical therapies have not proven to be effective, the lives of some patients have been prolonged after liver transplantation.[3]

There is a distinct iron overload disorder in sub-Saharan African. Previously termed “Bantu siderosis,” African overload affects up to 10 per cent of some rural populations.[36] African iron overload results from a predisposition to iron loading that is exacerbated by excessive intakes of dietary iron. Iron overload was observed in Africans who drank a traditional beer brewed in non-galvanized steel drums. However, since serious iron overload does not develop in all beer drinkers and not all patients with iron overload consume excessive amounts of beer, it is postulated that a currently unidentified gene may confer susceptibility to the disorder. African overload is not due to mutations in the *HFE* gene and is not linked to the HLA locus.[36] Moreover, the pattern of iron disposition differs from that seen in HH. In African iron overload, marked iron loading occurs in hepatocytes and in hepatic Kuffer cells.

TABLE 9-6 Diseases of Iron Metabolism

Disorder	Etiology	Characterization
Neonatal hemochromatosis	Uncertain if fibrosis is from iron overload or whether fibrosis leads to iron accumulation in	Usually diagnosed at autopsy. Hepatic iron staining with variable total iron stores, liver fibrosis and bile duct proliferation. Iron deposits also found in heart, kidney, pancreas and mucosal

	remaining hepatocytes and shunting of iron to other tissues	salivary glands. The reticuloendothelial system relatively spared from iron overload. At birth liver already heavily fibrotic
Juvenille hemochromatosis	Autosomal recessive. Gene has been mapped to chromosome I; however, exact location of gene and its functional gene product are still being investigated	Phenotypically similar to HH. Onset is earlier than HH. Patients have hypgonadism, joint pain and unexplained stomach pain and die of cardiac dysfunction
African iron overload (also termed Bantu Siderosis)	Hypothesized to be related to increased dietary intake from using iron pots to dominant inheritance of an uncharacterized defect or to increased beer from non-galvanized iron brew pots	Liver macrohages and hepatocytes show increase iron loading, suggesting a defect in iron recirculation
Sideroblastic anemias	Involves first step of heme synthesis (catabolized by erythroid aminolevulinic acid-e-ALAS) and the last step of loading iron into the protoporphyrin ring	Accumulation of mitochrondrial iron, along with anemia
X-linked sideroblastic anemia	Missense mutation of the gene on the X chromosome for e-ALAS causes iron to accumulate in the mitochrondria of developing red blood cells	Prevents red blood cells from making sufficient hemoglobin. Signs and symptoms usually begin during the first three decades of life. Characteristic features include pale skin, fatigue, dizziness, and an enlarged spleen and liver. Heart disease, liver damage, and kidney failure are severe medical problems that can result from these increased levels of iron
X-linked sideroblastic anemia with ataxia	Mutation in the ATP-binding cassette 7 gene (ABC7) on the X chromosome	Distinct form of X-linked iron overload. Associated with onset of nonprogressive ataxia by early childhood. Anemia is microcytic and hypochromic
Pearson’s	Maternal	Vacuolization of marrow precursors and

syndrome	inheritance pointed to mutation in mitochrondrial DNA, with chromoral deletions commonly described	exocrine pancreatic dysfunction. Anemia is diagnosed before one year of age with macrocytic red cells and low neutrophil and platelet counts. Patients are short and have malabsorption
Friedreich ataxia	Autosomal recessive disorder	Degeneration of the central and peripheral nervous system, cardiomyopathy and diabetes. Iron accumulation has been detected in heart myocytes. Deficiency in protein fraxatin also observed. Reduced levels of fraxatin may lead to mitochrondrial respiratory failure and cellular damage and death
Hallervorden–Spatz syndrome	Family studies reveal a linkage to chromosome 20	Iron accumulation is limited to central nervous system, especially the basal ganglia. Clinical signs include dystonia, rigidity, and dementia
Aceruloplasminemia	Several mutations in the ceruloplasmin gene on the short arm of chromosome 3 have been discovered in affected families	Iron accumulation is observed in the spleen, heart, kidney, thyroid, and retina

CLINICAL CASE STUDY

Transfusion is a frequently employed therapy in sickle-cell disease (SCD). The major and unavoidable complication of transfusions in SCD is iron overload. Therefore, knowledge of iron toxicity in SCD is of paramount importance. The mechanism of end organ damage in iron overload is saturation of transferrin by excess circulating iron, which enters tissues and results in formation of reaction oxygen species (ROS).* Excess iron tends to deposit in the hepatic parenchyma, in endocrine organs, and in cardiac myocytes, causing end organ damage by ROS-mediated lipid peroxidation. Patients with SCD suffer from recurrent infections and ongoing endothelial damage by reperfusion injury that results in a chronic inflammatory-like state. The gold standard for assessing liver iron stores in the absence of cirrhosis is the hepatic iron content, determined by liver biopsy and quantitation with atomic absorption spectrophotometry. Non-invasive methods for assessing iron stores include blood tests (ferritin and iron saturation) and imaging techniques. Chelation therapy is routinely employed to prevent and treat iron overload in chronically transfused SCD patients.

Further discussion points:

Why does the role of hepcidin in the pathophysiology of iron overload in SCD remains controversial?

How do chelators work as a pharmacological therapy for treating iron overload?

What chelating agents are available and commonly used?

* Raghupathy R, Manwani D, Little JA. Iron overload in sickle cell disease. *Adv Hematol*. 010;2010:272940. doi:10.1155/2010/272940.

The aim of treatment of iron storage diseases is to remove excess iron from the body. In patients with one of the forms of primary hemochromatosis (i.e., hereditary hemochromatosis) removal of excess iron is best achieved by phlebotomy.[147] Treatment of secondary hemochromatosis consists of administration of iron chelating agents.[147]

SECTION 10: HOW DO WE ASSESS IRON STATUS?

Biomarkers for iron status were determined by physiologists who studied iron metabolism in animal models and humans. Thus, biomarkers were selected because of their specificity for identifying functional aspects of iron storage, transport and use and were focused on red blood cell production and turnover because the circulating red blood cells constitute the largest and most accessible iron containing compartment in the body.[150]

Among the wide variety of laboratory methods that are available to assess iron status (i.e., hematocrit, hemoglobin, mean cell volume, erythrocyte mean index, free erythrocyte protoporphyrin, serum iron, total iron-binding capacity, serum transferrin, transferrin saturation, serum ferritin, and serum transferrin receptor) there is no one single test that is ideal. Diminished stainable iron in a bone marrow aspirate is generally accepted to be the reference standard for iron status.[151] However, the method is invasive, costly and subject to varying interpretation.[152]

Rather than using a single measurement, a valid assessment of iron status is best characterized by multiple indices.[153] Presently, there is no consensus on the best definition of iron deficiency using multiple measures. In the United States, two combinations of multiple indices were used to assess low iron status and anemia in the National Health and Nutrition Examination Surveys II and III.[47,154] One included serum

ferritin, transferrin saturation and FEP. The other method involved MCV, transferrin saturation and FEP. Selecting the most appropriate combination of iron status indicators depends on the study objectives and the health of the study population.[151,155] Features of the more commonly accepted diagnostic indices are summarized in Tables 9-7 and 9-8. For a detailed review see Gibson.[156]

Hemoglobin

Hemoglobin (Hb) measures the presence of anemia. While it is the most widely used screening tool for iron deficiency, hemoglobin determination has serious limitations when relied upon as the only laboratory measurement because of its low specificity and sensitivity.[55] When used alone hemoglobin concentration does not distinguish between anemia due to iron deficiency and that due to infection (e.g., malaria, HIV), other micronutrient deficiencies (e.g., folate, vitamin B12, vitamin A) or other disease states (e.g., sickle cell disease, thalassemia). Furthermore, the wide range of the values in normal subjects means that Hb values overlap considerably between normal and iron deficient anemic individuals.[7] Affected by diurnal variations; Hb values in the evening tend to be lower than the values in the morning by up to 10 g/L.[156]

As with any other parameter, it is essential to interpret hemoglobin concentration according to age-specific and sex-specific reference standards. In infants, reference values for iron status variables, including hemoglobin have not been well developed and may not be accurate since they are based on extrapolation on data from older age groups.[157,158] The cut-off for anemia at sea level recommended by the WHO for infants 6 months to 5 years of age is 110 g/L. This cutoff concentration is based on studies in young children and has been extrapolated to cover infancy because of lack of more appropriate data.[156] However, several studies have indicated that this concentration may be too high.[12] Furthermore, there is growing opinion that the normative hemoglobin distribution curve is not the same throughout the world.[159]

Ferritin

Serum and erythrocyte ferritin concentrations reflect the level of storage iron.[11] Serum measurements are simpler and are the more common of the two tests.[152] Having been found to be directly proportional to the abundance of storage iron in normal subjects, serum ferritin concentration

is used as a sensitive and specific index for iron deficiency. When used in combination with the measurement of hemoglobin, serum ferritin distinguishes between the anemias of chronic disease and iron deficiency.

A low serum ferritin concentration is usually interpreted as depleted iron stores, but does not necessarily indicate systemic iron deficiency. Adequate iron for erythropoiesis can be maintained even in the presence of low stores, as occurs in frequent blood donors.[152] Once iron stores have been completely exhausted the concentration of serum ferritin no longer reflects the severity of iron deficiency. The absence of iron stores may not be associated with any adverse functional implications and should therefore only be regarded as a risk factor for the development of iron deficiency anemia.[7]

The most important limitation of ferritin is its acute phase response, which can mask an otherwise iron-depletion state in clinical conditions such as infection, inflammation, and malignancy.[152] To overcome this limitation, simultaneous measurement of a biomarker of the acute-phase response such as C-reactive protein and, where appropriate, screening for the presence of malaria parasites should be conducted.[7]

TABLE 9-7 Cutoff Points by Age Used to Identify Abnormal Values of Iron Status Measures

Iron Measure	Age (Years)	Abnormal Values
Hemoglobin (g/L)[a,b]	0.5–5	<110
	5–11	<115
	12–15 (females)	<119
	12–15 (males)	<126
	16–19 (females)	<120
	16–19 (males)	<136
	Pregnant women	<110
Hematocrit[a]	0.5–5	<0.33
	5–11	<0.34
	12–13	<0.36
	Pregnant women	<0.33
Mean cell volume (fL)[c]	1–3	<77
	3–5	<79
	6–11	<80

	11–14	<82
	15–74	<85
Serum ferritin (µg/L)[b]	1–25	<10
	6–16	<12
Transferrin saturation (%)[b]	1–2	<10
	3–5	<12
	6–11	<14
	12–15	<14
	≤16	<15
Erythrocyte protoporphrin (µmol/L RBCs)[b]	1–2	>1.42 (80 µg/dL RBCs)
	3–5	>1.24 (70 µg/dL RBCs)
	6–11	>1.24
	12–15	>1.24
	≥16	>1.24

[a] Ref. 163
[b] Ref. 154
[c] Ref. 164

The most commonly used cut-off value for serum ferritin indicating depleted iron stores is 12 µg/L, however the appropriate cut-off value that is indicative of depleted iron stores in neonates and infants, with their rapidly changing erythropoietic activity, is still under discussion.[152] Current WHO cut-offs, including those for serum ferritin, are based on populations with an unknown prevalence of infection. Research in Honduras and Sweden established cut-offs at 9 µg/L at 6 months of age and at 5 µg/L at 9 months of age to define depleted iron stores. Some authors use the cutoff of 10 µg/L for infants and young children, which is based on the distribution of serum ferritin concentrations in a large survey in the United States (NHANES III), in which the fifth percentile indicated abnormal values.[12]

TABLE 9-8 Biochemical Indices for Measuring Iron Status

Indicator	Measurement	Advantages and Disadvantages	Factors That Affect Measurement

Bone marrow examination	Performed to judge the amount of hemosiderin in the reticulum cells; absence of stainable reticular iron is characteristic of latent iron deficiency; quantity of visible hemosiderin after staining with potassium ferrocyanide correlates well with the concentration of non-heme iron	Advantages High specificity for storage iron assessment Disadvantages Invasive technique; subjective nature (result depends on the skill of the investigator); requires a sufficient amount of available marrow stroma and a meticulous staining technique. Invasive and often painful. Not appropriate for children	
Hemoglobin (Hb)	Hemoglobin is the protein molecule within red blood cells that carries oxygen; decreased concentration of Hb measures presence of anemia; most widely used test for IDA	Advantages Inexpensive; universally available Disadvantages Low sensitivity; low specificity	Diurnal variation, age, sex, race, pregnancy, high altitudes, other micronutrient deficiencies, parasitic infections, disease states
Hematocrit	Ratio of the volume of packed red blood cells to the total blood volume; indicator of the concentration of RBCs in the blood and impaired Hb formation; in presence of anemia decrease in red blood cells is observed	Advantages Inexpensive; requires a small sample volume; relatively fast analysis Disadvantages Sensitive to changes in plasma volume, as well as red cell volume; difficult to standardize because it is a calculated parameter	Measurement errors (poorly packed iron-deficient cells, improper mixing of blood because of intermittent blood flow, excessive anticoagulant in collection tube); elevated white blood cell counts; vitamin B12 deficiency, bleeding, kidney disease, cirrhosis of the liver, and malignancies; pregnancy
Mean cell volume (MCV)	Average volume of a red cell; calculated by	Advantages Inexpensive; fairly	Chronic inflammation, certain

	dividing hematocrit (volume fraction) by red blood cell count (102/L)	specific indicator of iron deficiency once thalassemia and the anemia of chronic disease have been excluded	hemoglobinopathies, lead poisoning, age, race
Mean cell hemoglobin concentration (MCHC)	The average amount of hemoglobin in the average red cell; determined by dividing hemoglobin (g/L) by hematocrit (volume fraction)	Advantages Widely available; well established Disadvantages Late indicator; low specificity	Age
Red cell distribution width (RDW)	A measurement of the variability of red cell size; calculated by multiplying the standard deviation of mean cell volume by 100 and then dividing the product by the mean cell volume	Advantages More sensitive than serum iron, transferrin saturation, or serum ferritin level Disadvantages Low specificity; worthwhile when used in combination with other parameters	Age, micronutrient deficiencies, hemoglobinopathies
Serum iron	In fasting state serum iron levels reflect transit iron going from reticulo-endothethial system to the bone marrow	Advantages Sensitive to the stage of mild iron deficiency Disadvantages Wide diurnal variations; low specificity	Diurnal variation, age, sex, inflammation, chronic neoplastic diseases, blood loss or donation, pregnancy, with chronic infections, shock, pyrexia, rheumatoid arthritis and malignancy
Total iron-binding concentration (TIBC)	Measures the extent to which iron-binding sites in the serum can be saturated; an indirect measurement of the amount of transferrin in the blood	Disadvantages Low specificity; to properly understand a value for TIBC, one also must know the serum iron, the percent transferrin saturation	Sex, age, oral contraceptives, infection, inflammation, chronic neoplastic diseases, liver disease
Transferrin saturation	Ratio of serum iron to iron-binding capacity; calculated by dividing serum iron (μmol/L) by	Advantages Most accurate indicator of iron supply to the bone	Diurnal variation; oral contraceptives, infection, inflammation,

	TIBC (μmol/L) and multiplying the product by 100 to obtain a percentage	marrow Disadvantages Wide diurnal variation and low specificity	chronic neoplastic diseases
Ferritin	Serum or plasma iron concentration reflects total amount of storage iron	Advantages Reliable and sensitive parameter for iron stores in healthy subjects; high specificity for iron deficiency Disadvantages Does not indicate the severity of the iron deficiency due to higher assay variability; acute phase reactant	Diurnal effect is minimal, age, sex, race, inflammatory diseases, infections, certain neoplastic diseases and liver disorders
Zinc protoporphyrin (ZnPP); Free erythrocyte protoporphyrin (FEP)	Zinc protoporphyrin is a product of disordered heme biosynthesis; about 90% of protoporphyrin in red blood cells is ZPP, while the remainder is present as FEP (not bound to zinc); testing method for ZPP measures both substances, and the results may be reported as either ZPP or FEP; elevated in the more advanced stage of iron deficient erythropoiesis	Advantages Stability within the individual; easily measured using a hematofluorometer (which only a couple of drops of blood and minimal technical experience) Disadvantages Skin contamination may lead to false elevations; both test and blood lead values are needed for full evaluation; fluorescent substances in plasma may interfere with hematofluorometer results	Raised levels of EP found in subjects with lead poisoning and acquired defects in hemoglobin synthesis
Serum transferrin receptor (sTfR)	Transferrin receptors are found on cell membranes and allow iron-bound transferrin to enter the cell; an increase in serum receptor levels is seen	Advantages Remains normal in patients with acute or chronic inflammation or liver disease; effective in	Hematologic disorders associated with expansion of erythroid marrow

	in patients with iron deficient erythropoiesis or iron deficiency anemia	distinguishing iron deficiency anemia from anemia of chronic disease Disadvantages Lack of standardization for assay and reference values. Expensive assay cost	

Transferrin Receptor

Soluble transferrin receptor (sTfR), a critical iron binding protein in iron metabolism, has only recently been added to the available screening tests for iron deficiency. Using sensitive immunological assays, Kohgo et al. were the first to detect small amounts of a transferrin receptor-like protein in human serum.[160] The tranferrin receptor (TfR) reflects the degree of deficiency in functional iron after iron stores have been depleted.[152] Inclusion of serum TfR in the determination of iron status is recommended because it can distinguish anemia of chronic disease from IDA in the presence of chronic infection or inflammation.[7] This noteworthy advantage contrasts with other biochemical tests for iron status, which are influenced by anemia of chronic disease. It should be emphasized that the sTfR cannot be used in isolation of other laboratory measurements to identify iron deficiency anemia (IDA) in anemic patients, because it is also elevated in hematologic disorders associated with expansion of the erythroid marrow.[161]

Certain conditions affecting erythropoiesis such as hematological disorders (e.g., sickle cell anemia, autoimmune hemolytic anemia, thalassemia) and nutritional deficiencies other than iron (e.g., folate, vitamin B12) may confound the diagnosis of iron deficiency since they have been demonstrated to raise sTfR concentration.[161] Hemolysis associated with malaria may elevate sTfR concentration and confuse the detection of iron deficiency, although the magnitude of this effect remains unknown.

A major laboratory issue that remains unsolved is the lack of proper standardization of the sTfR assay: there are several commercial assays available for sTfR, but no common reference material.[162] Over the past decade, a number of laboratories have developed different assays for measuring sTfR concentration and have explored its normal range. The

normal range for sTfR reported in the literature varies considerably.[152] To date, there are no standards for measuring sTfR. The ratio of serum transferrin receptor (sTfR) to serum ferritin (*R/F* ratio) has been shown to perform well in estimating body iron stores.[162] Further research is needed regarding age appropriate cut-offs and the standardization of laboratory techniques such as the use of anticoagulant, blood collection methods, time delay between blood collection and centrifugation, incubation time, and the extent to which samples have to be diluted.[162]

SUMMARY

The primary role of iron, an essential nutrient for living organisms, is tissue oxygenation. The amount of iron in the body is determined by the regulation of iron absorption in the proximal small intestine. The diet, body iron stores, marrow erythropoietic activity, blood hemoglobin concentration, blood oxygen content, and the presence of systemic inflammation influence the rate of iron absorption. The most prevalent iron disorder is iron deficiency. Negative iron balance can occur from consuming a diet low in bioavailable iron, during periods of rapid growth (i.e., infancy, adolescence, and pregnancy), from malabsorption secondary to pathological conditions associated with the proximal small bowel (e.g., gluten induced enteropathy) or excessive blood loss.

Iron deficiency has been identified as one of the ten leading risk factors for disease, disability and death in the world today and the leading cause of anemia during infancy, childhood and pregnancy. Consequences of iron deficiency include poor immune function and decreased work performance. In children, moderate anemia has been associated with depressed mental and motor development. Anemia during pregnancy has been associated with increased risk of preterm delivery and infant low birth weight. Preventive strategies for iron deficiency include dietary diversification of foods with more bioavailable iron; fortification (commercial or in the home) of foods targeted to full-term infants and children; and iron supplementation.

Iron deficiency remains the most prevalent nutritional disorder in the world, especially among children and pregnant women in developing countries. Further research is needed to elucidate the pathophysiological mechanisms of iron absorption and body iron homeostasis and to develop specific and sensitive measures for detecting iron deficiency in large-scale studies. In order to increase the effectiveness of public health prevention and treatment strategies further efforts need to be made towards

developing products which improve compliance, and towards improving health service delivery and partnerships between the public and private sectors.

REFERENCES

1. Beard J, Dawson H, Pinero DJ. Iron metabolism: a comprehensive review. *Nutr Rev.* 1996;54:295–317.
2. Aisen P, Enns C, Wesslin-Resnick M. Chemistry and biology of eukaryotic iron metabolism. *Int J Biochem Cell Biol.* 2001;**33**:940–959.
3. Andrews N. Iron metabolism: Iron deficiency and iron overload. *Annu Rev Genomics Hum Genet.* 2000;1:75–98.
4. Wessling-Resnick M. Iron transport. *Annu Rev Nutr.* 2000; 20:129–151.
5. Conrad M, Umbreit JM. Iron absorption and transport—an update. *Am J Hematol.* 2000;64:287–298.
6. Thornburg K, Jacobson, SL, Giraud, GD, Morton, MJ. Hemodynamic changes in pregnancy. *Semin Perinatol.* 2000;24:11–14.
7. Hallberg L. Perspectives on nutritional iron deficiency. *Annu Rev Nutr.* 2001;21:1–21.
8. Rao R, Georgieff MK. Perinatal aspects of iron metabolism. *Acta Paediatr Suppl.* 2002;438:124–129.
9. Faldella G, Corvaglia L, Lanari M, Salvioli GP. Iron balance and iron nutrition in infancy. *Acta Paediatr Suppl.* 2003;441:82–85.
10. Zlotkin S. The role of nutrition in the prevention of iron deficiency anemia in infants, children and adolescents. Clinical nutrition: 8. *CMAJ.* 2003;168:59–63.
11. Hallberg L, Hulthen L. Perspectives on iron absorption. *Blood Cells Mol Dis.* 2002;29:562–573.
12. Aggett P, Agosttoni C, Axelsson I, et al. Iron metabolism and requirements in early childhood: Do we know enough? A commentary by the ESPGHAN Committee on Nutrition. *J Pediatr Gastroenterol Nutr.* 2002;34:337–345.
13. Beard J. Iron requirements in adolescent females. *J Nutr.* 2000;130:437S–439S.
14. NRCFNB: *National Research Council Food and Nutrition Board. Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc.* Washington, DC: National Academy Press; 2001.
15. Fairweather-Tait S. Iron requirements and prevalence of iron deficiency in adolescents. An overview. In: Hallberg L, editor. *Iron Nutrition in Health and Disease*. London, UK: John Libbey & Co; 1996. pp. 137–148.
16. Rossander-Hulthen L, Hallberg L. Prevalence of iron deficiency in adolescents. In: Hallberg L, editor. *Iron Nutrition in Health and Disease*. London, UK: Libbey; 1996. pp. 149–156.
17. Bothwell T. Iron requirements in pregnancy and strategies to meet them. *Am J Clin Nutr.* 2000;72:257S–264S.
18. Tapiero T, Gate L, Tew KD. Iron: deficiencies and requirements. *Biomed Pharmacother.* 2001;55:324–332.
19. Hagar W, Theil WC, Vichinsky EP. Diseases of iron metabolism. *Pediatr Clin N Am.* 2002, 49:893–909.
20. Zijp I, Korver O, Tijburg LB. Effect of tea and other dietary factors on iron absorption. *Crit Rev Food Sci Nutr.* 2000;40:371–398.
21. USDA: United States Department of Agriculture. Nutrient database for standard

reference: release 14. Beltsville (MD), editor. Nutrient Data Laboratory, Human Nutrition Research Center of the Agricultural Research Service (ARS).10-2-2006. 2001.
22. WHO: World Health Organization. Iron. In: Report of a joint FAO/WHO expert consultation, editor. *Human Vitamin and Mineral Requirements*. Bangkok, Thailand: FAO and WHO; 2002.
23. Anderson G, Frazer, DM, McKie AT, et al. Mechanisms of haem and non-haem iron absorption: lessons from inherited disorders of iron metabolism. *BioMetals.* 2005;18:339–348.
24. Hallberg L, Bjorn-Rasmussen E, Howard L, Rossander L. Dietary heme iron absorption. A discussion of possible mechanisms for the absorption-promoting effect of meat and for the regulation of iron absorption. *Scand J Gastroenterol.* 1979;7:769–779.
25. Harris S. The effect of calcium consumption on iron absorption and iron status. *Nutr Clin Care.* 2002;5:231–235.
26. Teucher B, Olivares M, Cori H. Enhancers of iron absorption: ascorbic acid and other organic acids. *Nutr Res.* 2004;74:403–419.
27. Hallberg L, Brune M, Rossander L. Effect of ascorbic acid on iron absorption from different types of meals. Studies with ascorbic-acid-rich foods and synthetic ascorbic acid given in different amounts with different meals. *Hum Nutr Appl Nutr.* 1986, 40:97–113.
28. Cepeda-Lopez A, Osendarp SJ, Melse-Boonstra A, et al. Sharply higher rates of iron deficiency in obese Mexican women and children are predicted by obesity-related inflammation rather than by differences in dietary iron intake. *Am J Clin Nutr.* 2011;93:975–983.
29. Popkin B. *Urbanization and the Nutrition Transition*. Washington, DC: International Food Policy Research Institute; 2000. Available at: http://www.ifpriorg/publication/urbanization-and-nutrition-transition Accessed November 16, 2011.
30. Popkin B. Nutritional patterns and transitions. *Popul Dev Rev.* 1993;19:138–157.
31. Nead K, Halterman JS, Kaczorowski JM, et al. Overweight children and adolescents: a risk group for iron deficiency. *Pediatrics.* 2004;114:104–108.
32. Gillum R. Association of serum ferritin and indices of body fat distribution and obesity in Mexican American men: the Third National Health and Nutrition Examination Survey. *Int J Obes.* 2001;25:639–645.
33. Pinhas-Hamiel O, Newfield RS, Koren I, et al. Greater prevalence of iron deficiency in overweight and obese children and adolescents. *Int J Obes Relat Metab Disord.* 2003;27:416–418.
34. McClung J, Karl JP. Iron deficiency and obesity: the contribution of inflammation and diminished iron absorption. *Nutr Rev.* 2009;67:100–104.
35. Fleming R. Advances in understanding the molecular basis for the regulation of dietary iron absorption. *Curr Opin Gastroenterol.* 2005;21:201–206.
36. Andrews N. Disorders of iron metabolism. *N Eng J Med.* 1999;341:1986–1996.
37. Bothwell T, Cook CR, Cook J, Finch CE. *Iron Metabolism in Man.* Oxford, England: Blackwell Scientific; 1979.
38. Conrad M, Umbreit JM. A concise review: iron absorption—the mucin-mobilferrin-integrin pathway. A competitive pathway for metal absorption. *Am J Hematol.* 1993;42:67–73.
39. Donovan A, Lima CA, Pinkus JL, et al. The iron exporter ferroportin/Slc40a1 is essential for iron homeostasis. *Cell Metab.* 2005;1:191–200.
40. Finch C. Regulators of iron balance in humans. *Blood Cells Mol Dis.*

1994;84:1697–1702.
41. Cook J, Finch CA. Assessing iron status of a population. *Am J Clin Nutr.* 1979;32:2115–2119.
42. McKie A, Barrow D, Latunde-Dada GO, et al. An iron-regulated ferric reductase associated with the absorption of dietary iron. *Science* 2001;291:1755–1759.
43. Andrews N, Fleming MD, Gunshin H. Iron transport across biologic membranes. *Nutr Rev.* 1999;57:114–123.
44. Frazer D, Anderson GJ. The orchestration of body iron intake: how and where do enterocytes receive their cues? *Blood Cells Mol Dis.* 2003;30:288–297.
45. Roy C, Enns CA. Iron homeostatis: new tales from the crypt. *Blood Cells Mol Dis.* 2000;96:4040–4027.
46. Schneider B, Leipold EA. Regulation of mammalian iron homeostasis. *Curr Opin Clin Nutr Metab Care.* 2000;3:267–273.
47. *Assessment of the Iron Nutritional Status of the U.S. Population Based Data Collected in the Second National Health and Nutrition Examination Survey, 1976–1980.* Bethesda, MD: Life Sciences Research Office, Federation of the American Societies for Experimental Biology; 1984.
48. Fleming R, Bacon BR. Orchestration of iron homeostasis. *N Eng J Med.* 2005;352:1741–1744.
49. Frazer D, Anderson GJ. Iron imports. I. Intestinal iron absorption and its regulation. *Am J Physiol Gastrointest Liver Physiol.* 2005;289:631–635.
50. Andrews N. Molecular control of iron metabolism. *Best Pract Res Clin Haematol.* 2005;18:159–169.
51. Miret S, Simpson RJ, McKie AT. Physiology and molecular biology of dietary iron absorption. *Annu Rev Nutr.* 2003;23:283–301.
52. Hallberg L, Hulthen L, Bentgsson C, et al. Iron balance in menstruating women. *Eur J Clin Nutr.* 1995;49:200–207.
53. Green R, Charlton RW, Seftel H, et al. Body iron excretion in man: A collaborative study. *Am J Med.* 1968;45:336–353.
54. Tondeur M, Schauer CS, Christofides AL, et al. Determination of iron absorption from intrinsically labeled microencapsulated ferrous fumarate (sprinkles) in infants with different iron and hematologic status by using a dual-stable-isotope method. *Am J Clin Nutr.* 2004;80:1436–1444.
55. Cook J. Diagnosis and management of iron deficiency anemia. *Best Pract Res Clin Haematol.* 2006;18:319–332.
56. DeMaeyer E, Dallman PR, Gurney JM, et al. *Preventing and Controlling Iron Deficiency anemia Through Primary Health Care: A Guide for Health Administrators and Programme Managers*. Geneva, Switzerland: World Health Organization; 1989.
57. Yip R. The challenge of improving iron nutrition: limitations and potentials of major intervention approaches. *Eur J Clin Nutr.* 1997;51:516–524.
58. WHO: *The World Health Organization. Reducing Risks, Promoting Healthy Life*. Geneva, Switzerland: World Health Report; 2002.
59. CC08: *Copenhagen Consensus 2008 Results*. Copenhagen, Denmark: Copenhagen Consensus Center; 2008.
60. Battling iron deficiency anemia: The challenge Micronutrient deficiencies. 2011; Available from: https://apps.who.int/nut/ida.htm. Accessed October 2012.
61. Olivares M, Walter T, Hertrampf E, Pizzaro F. Anemia and iron deficiency in children. *Br Med Bull.* 1999;55:534–543.
62. Florentino R, Guirriec RM. Prevalence of nutritional anemia in infancy and childhood with emphasis on developing countries. In: Stekel A, editor. *Iron*

nutrition in infancy and childhood. New York: Nestle, Vevey/Raven Press; 1984. pp. 61–72.
63. WHO. The Vitamin and Mineral Nutrition Information System (VMNIS). 2012; Available from: http://www.who.int/vmnis/database/en. Accessed October, 2012.
64. Cogswell ME, Looker AC, Pfeiffer CM, et al. Assessment of iron deficiency in US preschool children and nonpregnant females of childbearing age: National Health and Nutrition Examination Survey 2003–2006. *Am J Clin Nutr.* 2009;89:1334–1342.
65. Mei Z, Cogswell ME, Looker AC, et al. Assessment of iron status in US pregnant women from the National Health and Nutrition Examination Survey (NHANES), 1999–2006. *Am J Clin Nutr.* 2011;93:1312–1320.
66. Panagiotou J, Douros K. Clinicolaboratory findings and treatment of iron-deficiency anemia in childhood. *Pediatr Hematol Oncol.* 2004;21:519–532.
67. Mercer J, Nelson C, Skovgaard R. Umbilical cord clamping: Beliefs and practices of American nurse-midwives. *Clin Pract Exch.* 2000;45:58–66.
68. Allen L. Anemia and iron deficiency: effects on pregnancy outcome. *Am J Clin Nutr.* 2000;71:1280S–1284S.
69. Choi J, Kim CS, Pai SH. Erythropoietic activity and soluble transferrin receptor level in neonates and maternal blood. *Acta Paediatr Suppl.* 2000;89:675–679.
70. Gaspar M, Ortega RM, Moreiras O. Relationship between iron status in pregnant women and their new-born infants. *Acta Obstet Gynecol Scand.* 1993;72:534–537.
71. Halvorsen S. Iron balance between mother and infant during pregnancy and breastfeeding. *Acta Paediatr Suppl.* 2000;89:625–627.
72. Singla P, Gupta VJ, Agarwal KN. Storage iron in human foetal organs. *Acta Paediatr Scand.* 1985;74:701–706.
73. Singla P, Tyagi M, Hankar R, et al. Fetal iron status in maternal anemia. *Acta Paediatr Scand.* 1996;85:1327–1330.
74. Zlotkin S. Strategies for the prevention of iron deficiency anemia in infants and children. *Heinz Infant Nutrition Institute* 2003;20:1–4.
75. Gibson R, Ferguson EL, Lehrfeld J. Complementary foods for infants feeding in developing countries: their nutrient adequacy and improvement. *Eur J Clin Nutr.* 1998;52:764–770.
76. Wharton B. Iron deficiency in children: detection and prevention. *Br J Haematol.* 1999;106:270–280.
77. Zlotkin S. Iron deficiency in Canadian children? Implications and prevention. *Can J Pediatr.* 1993;4:18–23.
78. Gibson R, Health ALM, Ferguson EL. Risk of suboptimal iron and zinc nutriture among adolescent girls in Australia and New Zealand: causes, consequences and solutions. *Asia Pacific J Clin Nutr.* 2002;11:S543–S552.
79. Brabin B. The role of malaria in nutritional anemias. In: Fomon SJ and Zlotkin S, editors. *Nutritional Anemias*. New York: Nestle, Vevey/Raven Press; 2006. pp. 65–77.
80. Jarvinen K, Suomalainen H. Development of cow's milk allergy in breast-fed infants. *Clin Exp Allergy.* 2001;31:978–987.
81. Scholl T. Iron status during pregnancy: setting the stage for mother and infant. *Am J Clin Nutr.* 2005;81:1218S–1222S.
82. Grantham-McGregor S, Ani C. A review of studies on the effect of iron deificiency on cognitive development in children. *J Nutr.* 2001;131:649S–668S.
83. Lozoff B, Jimenez MD, Hagen J, et al. Poorer behavioural and developmental outcome more than 10 years of treatment for iron deficiency in infancy. *Pediatrics.* 2000;105:51–62.

84. Pollitt E. Iron deficiency and cognitive function. *Ann Rev Nutr.* 1993;13:521–537.
85. Angulo-Kinzler R, Peirano P, Lin E, et al. Spontaneous motor activity in human infants with iron deficiency anemia. *Early Hum Dev.* 2002;66:67–79.
86. Roncagliolo M, Garrido M, Walter T, et al. Evidence of altered central nervous system development in infants with iron deficiency anemia at 6 mo: delayed maturation of auditory brainstem responses. *Am J Clin Nutr.* 1998;68:683–690.
87. Youdin M. Neuropharmacological and neurobiochemical aspects of iron deficiency. In: Dobbing J, editor. *Brain, Behaviour and Iron in the Infant Diet.* London, UK: Springer-Verlag; 1990. pp. 83–106.
88. Lozoff B. Iron and learning potential in childhood. *Bull NY Acad Med.* 1989;65:1050–1066.
89. Walter T. Effect of iron-deficiency anemia on cognitive skills and neuromaturation in infancy and childhood. *Food Nutr Bull.* 2003;24:104–110.
90. Lozoff B. Iron deficiency and child development. *Food Nutr Bull.* 2007;28:S560–S571.
91. Peirano P, Algarín CR, Chamorro RA, et al. Sleep alterations and iron deficiency anemia in infancy. *Sleep Med.* 2010;11:637–642.
92. Lozoff B. Early iron deficiency has brain and behavior effects consistent with dopaminergic dysfunction. *J Nutr.* 2011;141:740S–746S.
93. Bruner A, Joffe A, Duggan AK, et al. Randomized study of cognitive effects of iron supplementation in non-anaemic iron-deficient adolescent girls. *Lancet.* 1996;348:992–996.
94. Halterman J, Kaczorowski JM, Aligne A, et al. Iron deficiency and cognitive achievement among school-aged children and adolescents in the United States. *Pediatrics.* 2001;107:1381–1386.
95. Scholl T, Reilly T. Anemia, iron and pregnancy outcome. *J Nutr.* 2000;130:443S–447S.
96. Steer P. Maternal hemoglobin concentration and birth weight. *Am J Clin Nutr.* 2000;71:1288S–1297S.
97. Xiong X, Buekens P, Alexander S, et al. Anemia during pregnancy and birth outcome: a meta-analysis. *Am J Perinatol.* 2000;17:137–146.
98. Yip R. Significance of an abnormally low or high hemoglobin concentration during pregnancy: special consideration of iron nutrition. *Am J Clin Nutr.* 2000;72:272S–279S.
99. Brabin B, Hakimi M, Pelletier D. An analysis of anemia and pregnancy-related maternal mortality. *J Nutr.* 2001;131:604S–614S.
100. Kurz K, Galloway R. Improving adolescent iron status before childbearing. *J Nutr.* 2000;130:437S–439S.
101. Picciano M. Pregnancy and lactation: physiological adjustments, nutritional requirements and the role of dietary supplements. *J Nutr.* 2003;133:1997S–2002S.
102. Beard J, Hendricks MK, Perez EM, et al. Maternal iron deficiency anemia affects postpartum emotions and cognition. *J Nutr.* 2005;135:267–272.
103. Perez E, Hendricks MK, Beard JL, et al. Mother-infant interactions and infant development are altered by maternal iron deficiency anemia. *J Nutr.* 2005;135:850–855.
104. Ross S, Fahrbach K, Frame D, et al. The effect of anemia treatment on selected health related quality life domains: a systematic review. *Clin Ther.* 2003;25:1786–1805.
105. Lozoff B. Perinatal iron deficiency and the developing brain. *Pediatr Res.* 2000;48:137–139.
106. Lozoff B, Beard J, Connor J, et al. Long-lasting neural and behavioral effects of

iron deficiency in infancy. *Nutr Rev.* 2006;64:S34–S43.
107. Baker RD, Greer FR, Pediatrics CoNAAo. Diagnosis and prevention of iron deficiency and iron-deficiency anemia in infants and young children (0–3 years of age). *Pediatrics.* 2010;126:1040–1050.
108. WHO: *Guiding Principles for Complementary Feeding of the Breastfed Child.* Geneva, Switzerland: World Health Organization; 2004.
109. Viteri FE. Iron endowment at birth: maternal iron status and other influences. *Nutr Rev.* 2011;69 (Suppl 1):S3–S16.
110. Tawia S. Iron and exclusive breastfeeding. *Breastfeed Rev.* 2012;20:35–47.
111. Hicks PD, Zavaleta N, Chen Z, et al. Iron deficiency, but not anemia, upregulates iron absorption in breast-fed peruvian infants. *J Nutr.* 2006;136:2435–2438.
112. Civardi E, Tzialla C, Garofoli F, et al. Nutritional needs of premature infants. *J Matern Fetal Neonatal Med.* 2011;24 (Suppl 1):27–29.
113. CPS: Nutrition for healthy term infants: a joint statement of the Canadian Paediatric Society, Dietitians of Canada and Health Canada. *Paediatr Child Health* 1998;3:109–112.
114. Meyer R. Infant feeding in the first year. 2: feeding practices from 6–12 months of life. *J Fam Health Care.* 2009;19:47–50.
115. Lynch S. The impact of iron fortification on nutritional anemia. *Best Pract Res Clin Haematol.* 2005;18:333–346.
116. Subar A, Krebs-Smith SM, Cook A, Kahle LL. Dietary sources of nutrients among US children, 1989–91. *Pediatrics* 1998;102:913–923.
117. INACG: *International Nutritional Anemia Consultative Group. Safety of Iron Supplementation Programs in Malaria-Endemic Regions*. Washington, DC: ILSI Press; 2001.
118. Allen L. Pregnancy and iron deficiency: unresolved issues. *Nutr Rev.* 1997;55:91–101.
119. Beard J. Weekly iron intervention: the case for intermittent iron supplementation. *Am J Clin Nutr.* 1998;68:209–212.
120. Hallberg L. Daily iron supplementation: why it is necessary. *Am J Clin Nutr.* 1998;68:213–217.
121. Viteri F. Iron supplementation for the control of iron deficiency in populations at risk. *Nutr Rev.* 1997;55:195–209.
122. Christian P, Shrestha, SR, LeClerq SC, et al. Supplementation with micronutrients in addition to iron and folic acid does not further improve the hematologic status of pregnant women in rural Nepal. *J Nutr.* 2003;133:3492–3498.
123. Christian P, Khatry SK, Katz J, et al. Effects of alternative maternal micronutrient supplements on low birth weight in rural Nepal: double blind randomised community trial. *Br Med J.* 2003;326:571.
124. Osrin D, Vaidya A, Shrestha Y, et al. Effects of antenatal multiple micronutrient supplementation on birthweight and gestational duration in Nepal: double-blind, randomized controlled trial. *Lancet.* 2005;365:955–962.
125. Allen L. Iron supplements: Scientific issues concerning efficacy and implications for research and programs. *J Nutr.* 2002;132:813S–819S.
126. Lynch S. Interaction of iron with other nutrients. *Nutr Rev.* 1997;55:102–110.
127. Oppenheimer S. Iron and its relation to immunity and infectious disease. *J Nutr.* 2001;131:616S–635S.
128. Sazawal Z, Black RE, Ramsan M, et al. Effects of routine prophylactic supplementation with iron and folic acid on admission to hospital and mortality in preschool children in a high malaria transmission setting: community-based, randomised, placebo-controlled trial. *Lancet* 2006;367:133–143.

129. Tielsch J, Katry S, Stoltzfus RJ et al. Effect of routine prophylactic supplementation with iron and folic acid on preschool child mortality in southern Nepal: community-based, cluster-randomised, placebo-controlled trial. *Lancet.* 2006;367:144–152.
130. English M, Snow RW. Iron and folic acid supplementation and malaria risk. *Lancet.* 2006;367:90–91.
131. WHO: *World Health Organization. Iron Supplementation of Young Children in Regions Where Malaria Transmission is Intense and Infectious Disease Highly Prevalent*. Geneva, Switzerland: World Health Organization; 2004.
132. Soofi S, Cousens S, Iqbal SP, Akhund T, et al. Effect of provision of daily zinc and iron with several micronutrients on growth and morbidity among young children in Pakistan: a cluster-randomised trial. *Lancet*. 2013;382:29–40.
133. Zlotkin S, Newton S, Aimone AM, Azindow I, et al. Effect of iron fortification on malaria incidence in infants and young children in Ghana: a randomized trial. *JAMA*. 2013;310:938–47.
134. WHO: Conclusions and recommendations of the WHO Consultation on prevention and control of iron deficiency in infants and young children in malaria endemic areas. Lyon, France: World Health Organization Secretariat on behalf of the participants to the Consultation 2006, 12–14 June:Accessed October 6, 2012.
135. WHO/UNICEF. Iron supplementation of young children in regions where malaria transmission is intense and infectious disease highly prevalent: Joint statement. Geneva, Switzerland: World Health Organization and UNICEF; 2006.
136. Harding K, Neufeld LM. Iron deficiency and anemia control for infants and young children in malaria-endemic areas: a call to action and consensus among the research community. *Adv Nutr.* 2012;3:551–554.
137. Ojukwu JU, Okebe JU, Yahav D, Paul M. Oral iron supplementation for preventing or treating anaemia among children in malaria-endemic areas. *Cochrane Database Syst Rev*. 2009:CD006589.
138. WHO. Iron supplementation for children in malaria-endemic regions. Updated February 20, 2013. Available from: http://www.who.int/elena/titles/iron_infants_malaria/en/. Accessed March 19, 2014.
139. Fryman E, Bystrom M, Jansson U, et al. Side effects of iron supplements in blood donors: superior tolerance of heme iron. *J Lab Clin Med.* 1994;123:561–564.
140. McCord J. Iron, free radicals, and oxidative injury. *Semin Hematol.* 1998;35:5–12.
141. Fine J. Iron poisoning. *Curr Probl Pediatr*. 2000;30:71–90.
142. Anonymous. Toddler deaths resulting from ingestion of iron supplements- Los Angeles, 1992–1993. *MMWR Morb Mort Wkly Rep.* 1993;42:111–113.
143. Fazen L, Lovejoy FH, Crone RK. Acute poisoning in a children's hospital: A 2-year experience. *Pediatrics.* 1986;77:144–151.
144. Ferguson J, Sellar C, Goldace MJ. Some epidemiological observations on medicinal and non-medicinal poisoning in preschool children. *J Epidemiol Community Health.* 1992;44:207–210.
145. Trinkoff A, Baker SP. Poisoning hospitalizations and deaths from solids and liquids among children and teenagers. *Am J Public Health.* 1986;76:657–660.
146. Tenenbein M. Toxicokinetics and toxicodynamics of iron poisoning. *Toxicol Lett.* 1998;102:653–656.
147. Beutler E, Hoffbrand V, Cook JD. Iron deficiency and iron overload. *Hematology Am Soc Hematol Educ Program.* 2003:40–61.
148. Yen A, Fancher TL, Bowlus CL. Revisiting hereditary hemochromatosis: current concepts and progress. *Am J Med.* 2006;119:391–399.

149. Whitlock E, Garlitz BA, Harris EL, et al. Screening for hereditary hemochromatosis: a systematic review for the U.S. Preventive Services Task Force. *Ann Intern Med.* 2006;145:209–223.
150. Lynch S. Case studies: iron. *Am J Clin Nutr.* 2011;94:673S–678S.
151. Umbreit J. Iron deficiency: a concise review. *Am J Hematol.* 2005;78:225–231.
152. Labbe R, Dewanji A. Iron assessment tests: transferrin receptor vis-a`-vis zinc protoporphyrin. *Clin Biochem.* 2004;37:165–174.
153. Cook J, Skikne BS, Baynes RD. Screening strategies for nutritional iron deficiency. In: Fomon SJ and Zlotkin S, editors. *Nutritional Anemias*. New York: Nestle, Vevey/Raven Press. *Nestle nutrition workshop series*; 1992:159–168.
154. Looker A, Dallman PR, Carroll MD, et al. Prevalence of iron deficiency in the United States. *JAMA.* 1997;277:973–976.
155. Sharma N, Butterworth J, Cooper BT, et al. The emerging role of the liver in iron metabolism. *Am J Gastroenterol.* 2005;100:201–206.
156. Gibson R. Assessment of iron status. In: Gibson RS, editor. Principles of Nutritional Assessment. New York: Oxford University Press; 2005. pp. 443–476.
157. Emond AM, Hawkins N, Pennock C, Golding J. Haemoglobin and ferritin concentrations in infants at 8 months of age. *Arch Dis Child.* 1996;74:36–39.
158. Domellöf M, Dewey KG, Lönnerdal B, et al. The diagnostic criteria for iron deficiency in infants should be reevaluated. *J Nutr.* 2002;132:3680–3686.
159. Stoltzfus R. Defining iron deficiency anemia in public health terms: a time for reflection. *J Nutr.* 2001;131:565S–567S.
160. Kohgo Y, Nishisato T, Kondo H. Circulating transferrin receptor in human serum. *Br J Haematol.* 1986;64:277–281.
161. Cook J. The measurement of serum transferrin receptor. Symposium: new perspectives on iron. *Am J Med Sci.* 1999;318:269.
162. Brugnara C. Iron deficiency and erythropoiesis: new diagnostic approaches. *Clin Chem* 2003;49:1573–1578.
163. Guidelines for the Use of Iron Supplements to Prevent and Treat Iron Deficiency Anemia. *International Nutritional Anemia Consultative Group (INACG), World Health Organization (WHO), United Nations Children's Fund (UNICEF).* Washington, DC: International Life Sciences Institute Press; 1998.
164. Dallman P. Influence of age on laboratory criteria for the diagnosis of iron deficiency anemia and iron deficiency anemia in infants and children. In: Hallberg L and Asp N-G, editors. London, UK: John Libbey and Co.;1996. pp. 65–74.

Vitamins

10

Roland Kupka, DSc and Eduardo Villamor, MD, DrPH

INTRODUCTION

Since ancient times, dietary approaches have been used to prevent and treat ailments such as night blindness, pellagra, scurvy, and rickets. However, it was not until the end of the 19th century that specific dietary compounds other than carbohydrates, fats, and proteins began to be identified as responsible for vital functions. In 1912, the term "vitamin" was coined to denote "vital amines" deemed necessary to prevent dietary deficiency diseases. It was later shown that not all vitamins were amines, and the term, was subsequently shortened to "vitamin."

Vitamins are currently defined as organic compounds which are essential for health and are required in minute quantities in the diet because they cannot be synthesized in sufficient quantities by an organism. A total of 13 vitamins are required in humans: The fat-soluble vitamins A, D, E, and K, and the water-soluble vitamins thiamin, riboflavin, niacin, pyridoxine, folate, cobalamin, biotin, and pantothenic acid. Water-soluble vitamins are carried in the body's tissues, but are not easily stored and must therefore be consumed frequently. Fat-soluble vitamins are absorbed along with fats in the diet and in contrast to water-soluble vitamins can be stored in the body. Vitamin deficiencies are classified as "primary" when the diet provides insufficient amounts or "secondary" when absorption or utilization of the vitamin is impaired.

From its birth a century ago, the field of vitamin research remains active today. While the initial research focused on the diseases of deficiency, current research now examines a much broader role of vitamins in optimizing health. For instance, vitamin D is now considered to function not only in its traditional role in calcium absorption and bone health, but also in muscle and immune function and the prevention of

chronic diseases.

The vitamin and mineral status has improved greatly in the Western world in the past 50 years, especially among women and children, two of the most at-risk populations for deficiencies. In North America, a variety of factors can be credited for these improvements, including the introduction of fortified foods, agricultural policies leading to more equitable access to foods, and targeted programs such as the Women, Infants, and Children program.[1] Nevertheless, intakes of selected nutrients may remain inadequate among children and adults in North America and other Western countries.[2,3] Furthermore, vitamin inadequacies remain highly prevalent among elderly parts of the populations in these countries.[4]

In resource-poor settings, micronutrient deficiencies remain a major public health problem. The underlying factors are insufficient dietary intake, oftentimes due to monotonous plant-based diets, and a high prevalence of infectious diseases leading to anorexia, malabsorption, and impaired storage and metabolism of vitamins and minerals. As a result, an estimated 190 million children of preschool-aged and 19 million pregnant women are deficient in vitamin A.[5] Deficiencies in folic acid, vitamin B_{12}, and vitamin D are also of concern, but their burden is insufficiently characterized.[6] See Part V: Nutrition in Low- and Middle-Income Countries for further details.

In this chapter, we present a brief discussion on each vitamin, emphasizing the basic chemistry, physiological, and current epidemiological knowledge with a special focus on issues pertaining to child health. A summary of information on the clinical examination of nutritional deficiencies and excesses, as well as on important food sources of vitamins, is provided in Table I-8 in Appendix I. The reader is referred to Chapter 11 for a discussion on current United States Dietary Reference Intakes (DRIs) set forth by the Food and Nutrition Board of the Institute of Medicine (IOM) and to Appendix II for a listing of recommended vitamin intakes.

VITAMIN A

History, Structure, and Metabolism

The importance of a substance present in certain foods for the treatment of night blindness was already known in ancient Egypt. The Greek physician Hippocrates (460–327 B.C.) recommended the intake of “raw beef liver, soaked in honey, once or twice by mouth” for the treatment of

“nyktalopia,” or the total inability to see in darkness. However, it was only in the late 19th and early 20th centuries when the biochemistry of this compound was clarified. In 1881, Lunin reported that while mice could not survive on a purified diet of fats, carbohydrates, proteins and salts alone, their survival was drastically improved after the addition of whole milk. Similar findings led Hopkins to postulate in 1906 the existence of “unsuspected dietetic factors” required for growth and life. Three years later, a fat-soluble factor was shown to promote growth in rats and was dubbed “fat-soluble factor A.” The name was changed to “vitamin A” in 1920. In 1930, Karrer completed long-standing work on carotenoid precursors of vitamin A by identifying the structure of β-carotene, the major pro-vitamin A carotenoid.

The term vitamin A groups a number of retinoid compounds with the biological activity of all-*trans*-retinol, the alcohol form of vitamin A. Retinoids usually consist of four isoprenoid units with five conjugated carbon–carbon double bonds. 11-*cis*-Retinal is the oxidation product of retinol primarily involved in visual functions. Retinal can be further oxidized to retinoic acid, which functions in growth but not a vision or reproduction. Retinyl palmitate is the principal storage form of vitamin A. Of the more than 600 carotenoids that have been identified in nature, about 50 have can serve as vitamin A precursors. Carotenoids without vitamin A activity may still have antioxidant properties, however.

Vitamin A is usually ingested as retinyl palmitate from animal sources, and as carotenoids, mainly β-carotene, from plants. In the stomach, preformed vitamin A and carotenoids are released from protein by proteolysis, then aggregate with lipids and pass into the upper part of the small intestine. Bile salts stimulate pancreatic lipase and esterases that hydrolyze retinyl esters to retinol in enterocytes. Retinol subsequently binds to the cellular retinol-binding protein CRBP(II). Carotenoids, on the other hand, pass into the mucosal cells unaltered, and are cleaved by a 15-15´-oxygenase into retinal which also binds to the CRPB(II). Retinal reductase and lecithin-retinol acyltransferase (LRAT) recognize the CRPB(II) complexes and synthesize retinol, which is then packed into chylomicrons before entering the circulation via the lymphatic system. Triacylglycerol is hydrolyzed from circulating chylomicrons by plasma lipoprotein lipase to form chylomicron remnants. These remnants deliver retinol to the liver, where it is taken up by hepatocytes through high-affinity receptors of apolipoproteins E and B48. In hepatic lysosomes, an acidic hydrolase releases retinol to bind with apo-CRBP. At least three pathways could follow after this CRBP-retinol complex is formed: First,

synthesis of CRBP-retinal through retinol dehydrogenase (RoDH) and later retinal through retinal dehydrogenase (RALDH); second, retinol esterification mediated by LRAT; and third, rapid mobilization of retinol catalyzed by the bile salt-independent reductase (bsiREH).[7]

From the hepatocyte, vitamin A can be locally transported to fat-storing stellate cells. Plasma concentrations of unbound retinol binding protein RBP (also known as apo-RBP) regulate the mobilization of retinol from the liver. Increased concentrations of apo-RBP signal stellate cells to release retinyl esters for de-esterification to retinol. The retinol bound to RBP (also known as holo-RBP) is secreted in plasma together with transthyretin, a tetramer that minimizes the loss of RBP in urine. The complex is then distributed to peripheral tissues or recycled back to the liver in a tightly regulated system that allows retinol molecules to recirculate at least seven times before final excretion. In the retina, holo-RBP binds to membrane receptors for internalization by endocytosis. These receptors have been found also in the skin, placenta, and the blood-testes and blood-brain barriers. In the lungs, kidneys, muscle, and possibly other tissues, retinol may be taken up directly by lipid membranes in a receptor-independent fashion. Vitamin A metabolites are primarily excreted in bile. However, pathological conditions may lead to substantial losses of retinol in urine.

Bioconversion of carotenoids is inversely regulated by vitamin A status. The bioavailability of vitamin A and carotenoids is influenced by the integrity of the intestinal mucosa and the intake of other nutrients. The presence of fat in a meal as well as food processing and heat treatment improve bioavailability of β-carotene and its conversion to vitamin A in humans, while dietary fiber reduces absorption. Among genetic determinants, variant allelic polymorphisms in the BCMO1 enzyme reduce the conversion of β-carotene to retinal.[8]

Traditionally, the vitamin A activity of β-carotene has been expressed as retinol equivalents (REs). However, recent data indicate that the vitamin A activities of purified β-carotene in oil as well as dietary β-carotene are lower than what had previously been assumed. For this reason, the contribution of β-carotene, and in fact also of other provitamin A carotenoids, to dietary vitamin A intakes had been overestimated. To take into account the new data, requirements for vitamin A are now expressed as retinol activity equivalents (RAEs), rather than REs. While the RE for dietary β-carotene was 6 μg, the RAE for dietary β-carotene is 12 μg, indicating that 12 μg (instead of 6 μg) of β-carotene needs to be consumed to yield 1 μg of available retinol. The corresponding RAE is 24 μg for both

α-carotene and β-cryptoxanthin, which are other important pro-vitamin A carotenoids.[9]

Functions

Vitamin A plays a key role in gene expression of numerous physiological processes. In the form of its metabolite all-*trans* retinoic acid, vitamin A is a ligand for a family of the nuclear retinoic acid receptors (RARs), whereas 9-*cis* retinoic acid activates RARs and retinoid X receptors (RXRs). These groups of receptors form heterodimers and bind to genes in regulatory regions known as retinoic acid response elements (RAREs).

Specific physiological functions exhibited by vitamin A include vision, immunity, cell differentiation, and growth. The visual role of vitamin A occurs in the rod cells of the eye and enables sight in dim-light conditions. In this biochemical process, all-*trans*-retinol undergoes isomerization and oxidation to yield the light-sensitive 11-*cis*-retinal, which binds to opsin to form rhodopsin (Figure 10-1). As rhodopsin absorbs light, 11-*cis*-retinal changes to all-*trans*-retinal, causing it to dissociate from opsin. As all-*trans*-retinal is released, opsin undergoes a conformational change and thus alters the permeability to cations, increases membrane polarization, and generates a nerve impulse. Rhodopsin can be regenerated, which completes the vitamin A-dependent visual cycle.

The role of vitamin A in immunity is mediated mainly by the metabolites all-*trans* retinoic acid and 9-*cis* retinoic acid. At the level of mucosal immunity, vitamin A deficiency (VAD) is related to alterations in the integrity of the ocular, respiratory, gastrointestinal (GI), and genitourinary epithelia. These alterations include replacement of normal ciliated and mucus producing cells by keratinized epithelium. Keratinization and mucin production may be additionally regulated by vitamin A at the transcriptional level. Local secretion of humoral immune factors such as secretory IgA and lactoferrin may be also impaired by VAD.

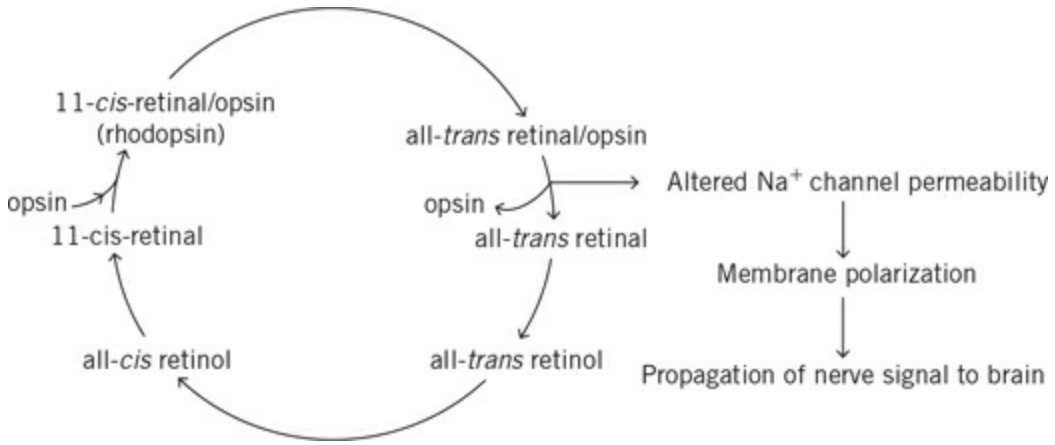


Figure 10-1 The vitamin A visual cycle.

Vitamin A influences the differentiation of white cell lines in the bone marrow, as well as the function of circulating immune cells. Phagocytosis, chemotaxis, and production of free radicals carried out by neutrophils are altered in vitamin A-deficient animals, as well as the number and cytotoxic activity of natural killer cells. Retinoic acid enhances various functions of mononuclear cells in vitro, such as phagocytic activity of macrophages, and release of tumor necrosis factor-α, interleukin 1 (IL-1), transforming growth factor-β (TGF-β), and expression of immunoglobulin receptors by monocytes that modulate proliferation of B lymphocytes. It also increases the capacity of Langerhans cells to present antigens to T lymphocytes in the skin.

Vitamin A appears to influence the antigen-specific proliferation of T lymphocytes. In animals, VAD impairs cytotoxic T lymphocyte activity. The growth and differentiation of B lymphocytes into immunoglobulin-producing cells also requires retinol, possibly through the induction of cytokine secretion by T helper lymphocytes. The antibody response against T cell-dependent antigens is usually impaired by VAD in animals, particularly during infections that would normally induce a T helper type-2 like response. In this case, instead of high production of IgG, IL-4, IL-5, and IL-10, a Th1 response consists of low antibody production and increased levels of IL-2 and interferon-γ. During the Th1 response to infections, it is thought that VAD further up-regulates such a response.

During the first 6 months of life, the most important sources of vitamin A are breast milk and hepatic stores. Breast milk levels decrease in vitamin A-deficient mothers, which may thus also compromise infant vitamin A

status.[10] After 6 months of age, complementary foods increase in importance to meet the infant's vitamin A requirements. Among US children age 2–18 years, ready-to-eat cereals and milk supply more than 40% of dietary vitamin A.[11] In poor settings, food-based approaches should ideally help meet requirements, but the poorer conversion ratios of carotenes from plant based foods may require other deficiency control mechanisms.

High-dosages of vitamin A are adequately absorbed and retained, replenish liver stores, and are released to meet tissue requirements. Even though high-dose vitamin A supplementation only transiently improves vitamin A status (as measured through serum retinol),[12] its benefits for child survival have been well documented. Summary estimates from the 17 mortality trials published to date indicate that vitamin A supplementation reduces all-cause mortality by 24% and diarrhea-related mortality by 28% among children aged 6–59 months. High-dose supplements among pregnant[13] and postpartum women[14] have no effect on maternal and infant mortality, and supplementation among neonates[15–17] and infants aged 1–5 months[15] has no consistent mortality effect. Based on these findings, WHO recommends one high-dose supplement in the form of 100,000 IU retinol to infants 6–11 months, and doses of 200,000 IU retinol every 4–6 months to children aged 12–59 months in settings where VAD is a public health problem (defined as the prevalence of night blindness is ≥1% in children 24–59 months of age or ≥20% prevalence of serum retinol ≤0.70 μmol/L among children 6–59 months of age). Vitamin A does not have any negative effect on seroconversion of childhood vaccines and supplements are recommended to be delivered during routine health system contacts such as vaccinations and integrated into the other child health and survival programs such as national immunization days and child health days.[18] Vitamin A supplementation programs among preschoolers should continue until their population serum retinol values have normalized.[12]

Among children with measles, vitamin A supplementation improves survival, reduces pneumonia incidence, and speeds recovery from diarrhea.[19–21] Based on the beneficial effects of vitamin A for recovery from measles infection, the American Academy of Pediatrics (AAP) recommends vitamin A supplementation for children aged 6 months to 2 years who are hospitalized with measles and children older than 6 months with measles and additional risk factors for VAD.[22] Children 1 year of age and older should receive 200,000 IU (60,000 μg) retinol by mouth, while

children aged 6 months to 1 year should receive 100,000 IU (30,000 μg) retinol.

Vitamin A was discovered as a growth-promoting factor in animals, but its role in growth and body composition in humans may be limited. Positive relations between vitamin A status and growth have been reported from observational studies, particularly in malnourished populations. However, these observational studies are limited by potential confounding by other nutrient deficiencies and infection. In randomized trials conducted in Asia and Africa, vitamin A supplementation generally did not improve child growth. Among children at highest risk of VAD and those with debilitating infectious diseases, including HIV, however, malaria, and persistent diarrhea, supplementation may improve growth.[23]

The industrial fortification of staple foods with vitamin A plays an increasingly important role in VAD control, but staple food consumption among young children may be too low to impact status.[24] Biofortification, that is, fortifying food by breeding crops to increase their nutritional value, either through conventional selective breeding or genetic engineering, is being evaluated as a VAD control measure. The provision of specially designed fortified complementary foods may have higher impacts but access of economically disadvantaged groups to these products may be low.[25] Increasing the amount of bioavailable nutrients through home fortification of complementary foods with low-dose lipid-based nutritional supplements or multiple micronutrient powders is promising but the impact of current formulations on vitamin A status remains unclear.[26] Until such dietary strategies reach scale, the most widely used program to alleviate the mortality effects of VAD is the twice-yearly supplementation of children aged 6–59 months.

Hepatic reserves accurately reflect vitamin A status, but cannot be measured in human studies. Serum retinol is a useful biomarker but is under homeostatic control until hepatic stores are depleted. Dose–response tests and isotope dilution assays are useful to determine marginal vitamin A status, but only dose–response tests are field friendly.[27]

Retinol dosages ≥ 150,000 μg among adults, and lower dosages among children proportional to body weight, induce transient toxic effects that include nausea, vomiting, headache, increased cerebrospinal fluid pressure, vertigo, blurred vision, and muscular incoordination. Chronic exposure to retinol at dosages of approximately 6000 μg/day or higher have also been associated with bulging fontanel among infants. Using these data and applying a large safety margin, the IOM recommends that children from 0 to 36 months should have preformed retinol intakes from

dietary and supplemental sources below 600 μg/day.[9] It has been pointed out, however, that this estimate may be overly conservative, given that 25%–30% of young children, as assessed by the Continuing Survey of Food Intakes by Individuals, have intakes above that level without evidence of adverse effects.[28] Women of childbearing age should be advised against consuming more than 2800–3000 μg/day of retinol to lower the risk of teratogenicity.[9]

VITAMIN D

History, Structure, and Metabolism

Rickets, a disorder due to vitamin D deficiency (VDD), has afflicted humanity since Neanderthal times some 200,000 years ago. Nevertheless, it was not described scientifically until the 17th century. In 1919, Mellanby linkend rickets to a nutritional deficit by inducing rickets in dogs through a synthetic diet and preventing the condition by administering cod liver or butter to diets.

This led him to propose that the dietary factor responsible for rickets was vitamin A. In 1922, McCollum distinguished antixerophthalmic factors in the diet from antirachitic factors, which he called vitamin D. Huldschinsky showed that rickets could be cured with exposure to ultraviolet (UV) rays and Hess and Weinstock demonstrated that UV irradiation induced the formation of an antirachitic factor in the skin.

Vitamin D designates a group of compounds derived from the cyclopentanoperhydrophenanthrene molecule, which have antirachitic activity. All these substances have a conjugated triene system of double bonds. The two most abundant forms of the vitamin are D_3 and D_2. Vitamin D_3 is also called cholecalciferol and is produced in the skin from 7-dehydrocholesterol, by solar radiation. Vitamin D_2, or ergocalciferol, is derived industrially from ergosterol, a plant steroid. Given that vitamin D can be produced by the human body, there is no dietary requirement for the vitamin as long as access to sunlight is available. For these reasons, vitamin D does not strictly fulfill the definition of a vitamin, and based on its various metabolic functions, the active metabolite 1,25-dihydroxycholecalciferol (1α, $25(OH)_2D_3$) is classified as a steroid hormone.

Few foods naturally contain vitamin D. The best natural sources are fatty fish such as salmon, tuna, and mackerel, and beef liver, cheese, and

egg yolks also contain small amounts. In the US, breakfast cereals and milk are commonly fortified with vitamin D. Absorption of vitamin D from the diet is relatively low, as only about 50% of dietary vitamin is absorbed. The major source of the vitamin in humans is from photochemical conversion of 7-dehydrocholesterol to previtamin D in the skin, and posterior isomerization to vitamin D (Figure 10-2). Vitamin D then passes to the systemic circulation bound to its transporter, the vitamin D-binding protein, which carries most biological forms and metabolites of the vitamin. It is taken up rapidly by the liver, but it is not preferentially stored there. The highest concentrations of vitamin D are found in blood, although adipose and muscle tissue in humans are also major storage sites for vitamin D. During deprivation, turnover from these sites is slow, and the supply of vitamin D to peripheral organs can be sustained for a period of time.

Vitamin D_3 does not have known biological functions, as it must first be hydroxylated to $25(OH)D_3$ in the liver. $25(OH)D_3$ is the most abundant circulating form of the vitamin, and is a good proxy of vitamin D_3 status. It is transported to the proximal tubules of the kidney, where a 1-hydroxylase converts it into $1\alpha,25(OH)_2D_3$. This hydroxylase is considered the most important regulatory mechanism of the vitamin D endocrine system. Its activity is regulated by the concentration of $1\alpha,25(OH)_2D_3$, parathyroid hormone (PTH), calcium, and phosphate. The second hydroxylated metabolite of D_3 produced in the kidney is $24R,25(OH)_2D_3$. Although this metabolite may play unique roles in vitamin D functions such as regulating PTH and bone mineralization, it is likely that both $25(OH)_2D_3$ and $24R,25(OH)_2D_3$ may be required. Vitamin D is mostly excreted in bile through catabolic pathways that are not well understood yet. Only small amounts of the vitamin are recovered during enterohepatic circulation. Some water-soluble metabolites of vitamin D, especially calcitroic acid (1α-hydroxy-23 carboxy-24,25,26,27-tetranorvitamin D_3), are also excreted in urine.

Functions

Vitamin D has a well-described role in bone health and calcium and phosphorous homeostasis. It also regulates genes that function in immune responses as well as cellular growth, differentiation, and apoptosis. As part of its steroid form $1\alpha,25(OH)_2D_3$, vitamin D is biologically active by rapidly activating several signal transduction pathways through the

membrane receptor VDR_{mem} as well as by regulating gene transcription through its nuclear receptor VDR_{nuc}.[29]

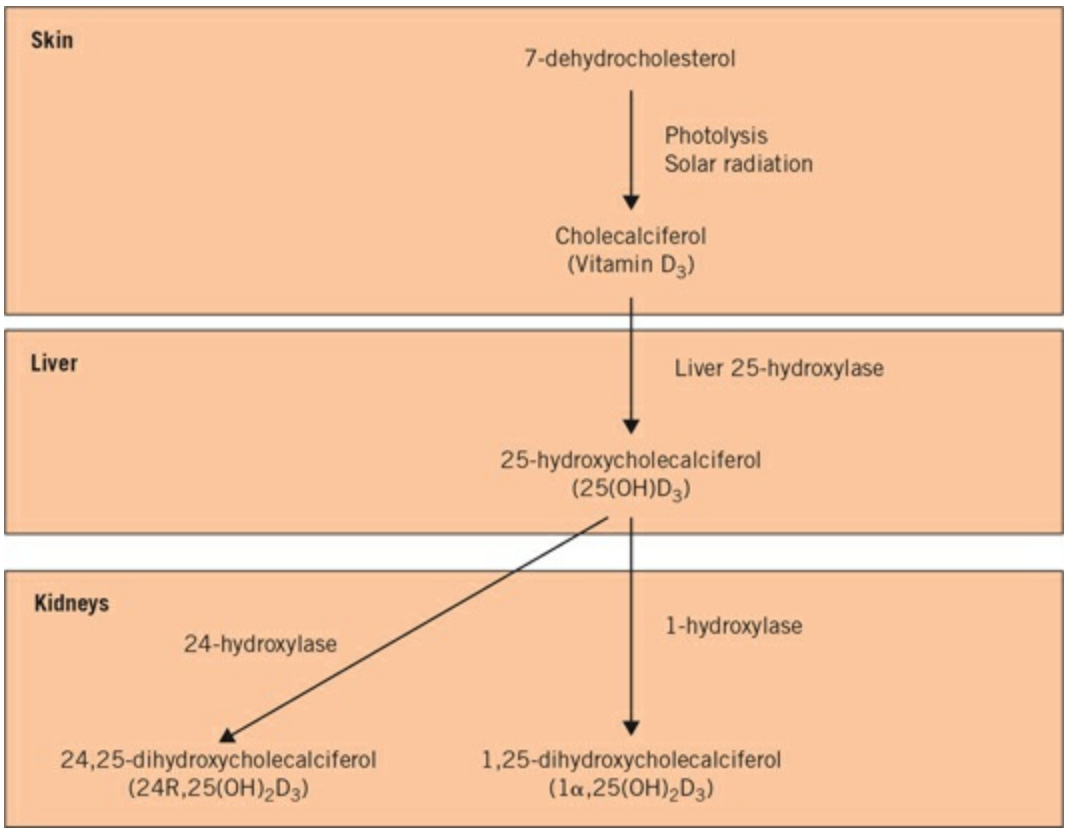


Figure 10-2 Overview of vitamin D metabolism (R).

The rapid, non-genomic physiological responses triggered by the interaction of $1\alpha,25(OH)_2D_3$ with the membrane receptor VDR_{mem} include the stimulation of Ca^{2+} absorption in the intestine (transcaltachia), opening of Ca^{2+} and Cl^- channels, activation of protein kinase C and mitogen-activated kinase, Ca^{2+} entry in skeletal muscle cells, and uptake of Ca^{2+} by osteoblasts.[29]

The mechanisms involving gene transcription are initiated when $1\alpha,25(OH)_2D_3$ binds in target organs to its nuclear receptor, VDR_{nuc}, which has domains also found in other steroid hormone receptors. After binding, VDR forms a heterodimer with RXR and later, the complex interacts with the vitamin D response element in the transcription control region of target genes. These activated promoter genes may also bind to coactivator proteins such as TATA, TBP, and TFIIB, and the resulting

transcriptional complex is then able to modulate DNA transcription.[30] Of the more than 60 genes regulated by 1α,25$(OH)_2D_3$, the gene linkend to the induction of the calcium binding protein calbindin D is one of the most important ones because it is directly related to many of the known physiological functions of vitamin D.

Regulation of mineral homeostasis is the classical function attributed to vitamin D. This process occurs at various target organs. In the intestine, 1α,25$(OH)_2D_3$ induces the transport of calcium and phosphate from lumen to plasma, possibly by activating calbindin, and also through changes in the membrane composition of the brush border cells. In the kidney, 1α,25$(OH)_2D_3$ downregulates 25$(OH)D_3$-1α-hydroxylase, thus controlling its own production. 1αx,25$(OH)_2D_3$ also stimulates the activity of 25$(OH)D_3$-24-hydroxylase. In bone, 1α,25$(OH)_2D_3$ and PTH mobilize monocytic stem cells to become mature osteoclasts. 1α,25$(OH)_2D_3$ may promote bone growth and mineralization through its effect on increasing intestinal calcium absorption, and possibly also by promoting differentiation of growth plate cells via chondrocyte membrane receptors and augmenting osteoblast proliferation and calcium uptake.

In skeletal muscle, vitamin D may be an important stimulant of calcium release during hypocalcemia. Myopathy frequently accompanies bone diseases related to VDD. In the skin, 1α,25$(OH)_2D_3$ appears to foster differentiation of keratinocytes, and protects them from UV stress. Some types of psoriasis can improve with the topical application of vitamin D analogs. Pancreatic β-cells respond to vitamin D with insulin secretion and VDD is accompanied with impaired insulin production. High doses of vitamin D may decrease the risk of type 1-diabetes in children.[31]

Various actions of vitamin D on the immune system have been reported. Immune cells, such as T and B lymphocytes, monocytes, and dendritic cells may be dependent on vitamin D as they contain VDR_{nuc} and some exert hydroxylase activity. Children with low serum 25(OH)D concentrations may be at increased risk of respiratory and GI infections[32] and supplemental vitamin D may reduce the incidence of influenza A[33] and acute respiratory infections[34] in this group. VDD in adults may be related to reactivation of latent tuberculosis, but more research is needed. The immune effects of vitamin D are explained by upregulation of Th2-immune responses that protect against extracellular pathogens[35] and induction of the antimicrobial peptide cathelicidin in the innate response against intracellular bacteria.[36] Vitamin D may also down regulate Th1-

immune responses, which may explain why vitamin D suppresses various autoimmune diseases and prolongs allograft survival.

In observational epidemiologic studies, VDD is related to early sexual maturation[37] and increased risks of obesity[38] and asthma.[39] Randomized trials are needed to demonstrate whether these relations are causal.

Recognition is growing that VDD may be a major public health problem worldwide, even in equatorial regions where it had not been suspected before. In the US, up to 20% of children ages 6–11 years are vitamin D-deficient (serum levels of 25-hydroxyvitamin D [25(OH)D] <50 nmol/L), while 52% have vitamin D insufficiency (25(OH)D ≥50 to <75 nmol/L).[40] Hispanic and black children are at the highest risk for VDD. Poor maternal vitamin D status impairs fetal vitamin D accretion during pregnancy and further compromises child vitamin D status due to reduced breastmilk vitamin D concentrations.

Even though regular brief exposure to sunlight can theoretically offset low dietary intakes, skin production of vitamin D is variable and depends on many factors, including skin color, time of the day and season of the year, geographical latitude, and age. The optimal amount of vitamin D intake is a matter of debate. In 2008, the AAP recommended a minimal intake of 400 IU/day for infants and children.[41] By contrast, in 2010, the Institute of Medicine set a recommended dietary allowance of 600 IU/day for children 1 to 18 years of age with maximum intakes of 2500 IU, 3000 IU, and 4000 IU for age groups 1–3 years, 4–8 years, and 9–18 years, respectively.[42] For infants <1 year of age, adequate intake was set at 400 IU/day. Some prominent vitamin D researchers argue that a higher recommendation is needed to decrease the prevalence of deficiency and improve vitamin D-related health outcomes considering that tolerable upper intake level is likely much higher than currently established. Nevertheless, additional evidence from randomized trials will be needed before a new revision of recommended intake takes place.

VITAMIN E

History, Structure, and Metabolism

Vitamin E was discovered by Bishop and Evans in 1922 as a fat-soluble compound in wheat and lettuce that was necessary to avoid fetal death in rats. The vitamin was shown to be essential among humans, not until the late 1960s, when its deficiency was linked to hemolytic anemia among premature infants. The potential protective role of vitamin E as an

antioxidant against chronic and degenerative illness is currently an active field of research.

Natural vitamin E occurs in the form of eight different isomers, four tocopherols and four tocotrienols. A chromanol ring is common to all isomers, but while tocopherols have a saturated phytyl tail, tocotrienols have an unsaturated tail. Based on the number of methyl groups on the chromanol ring, the isomers are grouped into α-, β-, γ-, and δ forms of tocopherol and tocotrienol. The various isomers of vitamin E are not interconvertible in humans and that they have different metabolic functions. Only certain forms of α-tocopherol are maintained in the plasma and are thus considered to be the active form that is relevant to establish nutritional requirements. These active forms are the *2R*-stereoisomers of α-tocopherol (classified as *RRR*-, *RSR*-, *RRS*-, and *RSS*-α-tocopherol), while the *2S*-stereoisomeric form (SRR-, SSR-, SRS-, and SSS-α-tocopherol) are not maintained in plasma and are thus not considered to be active. Synthetic forms of vitamin E are a mixture of all eight α-tocopherol steroisomers (prefixed "*all-rac-*," for *all-racemic*), while only the *RRR*-stereoisomer occurs in food.

After ingestion, all forms of vitamin E combine with bile salts and pancreatic secretions in the intestinal lumen, form micelles, and are taken up into enterocytes. Absorption of all forms of vitamin E is low, but appears to be inversely related to intake. It is also reduced by long-chain polyunsaturated fatty acids (PUFAs), but increased by medium-chain triglycerides. At the enterocyte, all forms of vitamin E are incorporated into chylomicrons and enter the circulation via the lymphatic system. As a result of chylomicron catabolism, vitamin E is distributed to tissues by means of circulating lipoproteins. Chylomicron remnants that contain the remainders of newly absorbed vitamin E are taken up by the liver. A selection process occurs in the liver, as RRR-α-tocopherol, the active form of-tocopherol, is preferentially resecreted into nascent VLDLs by-tocopherol transfer protein and recirculated through the body. During the conversion of VLDL to form LDL, some α-tocopherol is transferred to HDLs, which transport it to all of the circulating lipoproteins. The α-tocopherol remaining in LDL enters the cells of peripheral tissues through the LDL receptor pathway. The metabolic products of α-tocopherol after oxidation are dimers and trimers, as well as tocopheryl chromanoxy radicals that can be reverted to tocopherol by reducing agents such as vitamin C and glutathione. Subsequent oxidation of the tocopheroxyl radical produces tocopheryl quinone that, after reduction, can be excreted in the feces, or converted into tocopheronic acid by side chain oxidation

and excreted in urine as glucuronate conjugates. Other non-oxidation metabolites of the tocopherol phytyl tail are excreted in urine. These include 2,5,7,8-tetramethyl-2-(2′-carboxyethyl)-6-hydroxychroman, or α-CEHC, and γ-CEHC. High amounts of supplemental vitamin E result in increases in urinary α-CEHC concentrations. Due to the low bioavailability of ingested vitamin E, large amounts of the vitamin are excreted in feces. Absorbed forms of the vitamin that are in excess of requirements are excreted unchanged in the bile.

Functions

Vitamin E deficiency in animal models results most commonly in necrotizing myopathy, but may also lead to encephalomalacia, exudative diathesis, and embryonic degeneration. Deficiency symptoms also appear to depend on the status of other nutrients such as selenium and certain amino acids. In humans, vitamin E deficiency due to a poor diet is rare. However, it may occur among premature infants and secondary to pathologies causing fat malabsorption, such as abetalipoproteinemia, cystic fibrosis, and cholestatic disease. Neurological alterations are the most common manifestation of vitamin E deficiency in humans.

The specific mechanisms of vitamin E deficiency syndromes are not clearly understood. However, it is accepted that the most important function of vitamin E is protection against tissue damage caused by lipid peroxidation. Primordial free radicals (e.g., superoxide) can be originated in normal metabolism and through the effect of toxins. These free radicals may contribute to the formation of lipid free radicals when conjugated with PUFAs. The lipid radicals undergo a propagation sequence whose final products are lipid hydroperoxides (LOOH), which enter an autooxidative chain sequence until two radicals combine in a 2-electron bond. Vitamin E prevents auto-oxidation in at least two steps: (a) it traps peroxyl radicals; thus, a stable LOOH molecule is formed with a vitamin E radical subproduct; and (b) this vitamin E radical can still bind to other lipid radical. It is estimated that one tocopherol molecule is enough to protect 100 PUFA molecules from auto-oxidation. Some functions of vitamin E are strongly dependent on interactions with other nutrients. The antioxidant efficacy of tocopherol is enhanced by the presence of other antioxidant systems. The glutathione peroxidase system is selenium-dependent, and may explain the fact that selenium reduces the severity of vitamin E deficiency and that both may act synergistically against oxidative stress. Vitamin C (ascorbic acid) appears to reduce the vitamin E requirement by regenerating tocopherol after oxidation. β-Carotene also

may be a co-adjuvant of vitamin E in its membrane antioxidant functions; on the other hand, vitamin E increases the lymphatic uptake of β-carotene and its conversion to retinol. High intakes of PUFAs increase vitamin E requirements, but the extent remains unclear.

Vitamin E may improve immune function at various levels. In addition to its ability to scavenge lipid soluble free radicals generated during the immune response, vitamin E improves T-cell function by decreasing macrophage PGE-2 production and supports lymphocyte maturation by stabilizing membranes and facilitating the binding of antigen-presenting cells to immature T cells.[43] Other vitamin E-related functions include the regulation of xanthine oxidase, an important enzyme in the conversion of purine bases to uric acid, and the modulation of the activity of microsomal oxidases, protein kinases, and proliferation of smooth-muscle and endothelial cells.

Whether vitamin E supplementation has positive effects on health outcomes is controversial. Supplementation with vitamin E decreased the incidence of upper, but not lower, respiratory infection in elderly nursing home residents.[44] Evidence from other trials does not clearly support a preventive role against chronic disease, including cardiovascular [45] or Alzheimer's disease,[46] and some studies, in fact, suggest a potential deleterious role on certain cancers.[47] Information is scant on the effects of vitamin E supplementation alone on pediatric health. Gestational vitamin E supplementation has no effect on the offspring's risk of asthma[48] and supplementation in preterm infants increased the risk of sepsis.[49]

There is no evidence that toxic amounts of vitamin E can be ingested from food alone. However, supplemental intakes of vitamin E should not exceed 1000 mg among adults to prevent hemorrhagic toxicity and diminished blood coagulation among individuals who are on anticoagulant therapy or who are vitamin K deficient. Lower ULs have been defined for children older than one year to safeguard against potential toxic effects. For infants, the IOM has not determined a UL, given that there is a paucity of data and considering that food should be the only source of the vitamin for infants. While supplemental vitamin E may lower the risk of retinopathy of prematurity, there are no convincing data to suggest that supplemental vitamin E reduces the occurrence of bronchopulmonary dysplasia or intraventricular hemorrhage. According to the AAP, large parenteral doses of vitamin E in preterm infants, which elevate serum levels >40 μg/mL, are contraindicated as they may increase the risk of bacterial and fungal sepsis.[50] The IOM recommends that premature infants

should not ingest more than 21 mg of α-tocopherol to prevent hemorrhagic toxicity.[51]

VITAMIN K

History, Structure, and Metabolism

In 1929, Dam described a bleeding condition in chicks fed diets formulated to investigate cholesterol metabolism. He suggested the existence of a new vitamin in 1935 after this fat-soluble compound effectively prevented the bleeding condition in chicks. In 1939, a research group led by Doisy attributed its chemical and physical properties to a substituted 1,4-naphthoquinone. Dam and Doisy jointly received the Nobel Prize for medicine in 1943 for the discovery of the vitamin and for the elucidation of the chemical structure.

Vitamin K is a fat-soluble vitamin that consists of a family of compounds with a common 2-methyl-1,4-naphthoquinone ring structure. Vitamin K_1, or phylloquinone, is synthesized by plants and has a phytyl side chain. Vitamin K_2, is synthesized by bacteria as multiple homologs called menaquinones, which are characterized by unsaturated isoprenyl side chains of varying lengths. Menaquinone-4 is the most common homolog because animal enzymes can generate it from plant phylloquinone.

Excellent sources of phylloquinone are green vegetables such as kale, spinach, collards, parsley and broccoli, and plant oils such as soybean and canola account for large quantities in the diet. Menaquinone is found in meat, eggs, and dairy products. Vitamin K is absorbed from the first half of the small intestine alongside dietary fat. Absorption of pharmacological preparations of phylloquinone in adult humans is about 80%, but may only be about 20% from food sources.[52] After incorporation into chylomicrons in the intestinal mucosa, the vitamin is secreted into lymph and is taken up by the liver from chylomicron remnants through apolipoprotein E receptors. There is no known carrier protein and phylloquinone is principally transported with triacylglycerol-rich lipoproteins. Vitamin K is principally stored in the liver, albeit at lower levels than other fat-soluble vitamins. Menaquinones account for a higher proportion than phylloquinone, possible because of their slower hepatic turnover. Menaquinones are produced in the large intestine, but they may have very limited bioavailability and therefore may only contribute little to satisfying human vitamin K requirements. Phylloquinone, generally in the form of a

metabolite, is excreted in the feces via bile and to a lesser degree in urine. Little is known on the degradative metabolism of menaquinones.

Functions

Vitamin K serves as a cofactor in the carboxylation of the amino acid glutamic acid to form γ-carboxyglutamic acid (Gla), which is critical to the calcium-binding function of a number of proteins. After carboxylation, vitamin K is oxidized to vitamin K 2,3-epoxide, which can be successively recycled to quinine and hydroquinone in reactions catalyzed by dithiol-dependent reductases. These reductases can be inhibited by coumarinic, anticoagulant drugs such as warfarin. Undercarboxylated proteins have historically been referred to as proteins induced by vitamin K absence or antagonism, or PIVKAs. Undercarboxylated prothrombin, also known as PIVKA-II, is a sensitive marker of subclinical vitamin K deficiency after infancy.[53]

In the blood coagulation cascade, seven vitamin K-dependent factors and proteins play a major regulatory role (Figure 10-3). Factors IX and VII activate factor X through the intrinsic and extrinsic pathways, respectively, while factor X activates the synthesis of prothrombin from thrombin. The vitamin K-dependent proteins C, S, and Z function as anticoagulants in hemostasis.

Vitamin K also functions in bone health and calcification of tissues as a cofactor for the bone Gla proteins osteocalcin, matrix Gla protein (MGP), and protein S. Osteocalcin, also known as bone Gla protein, is produced in osteoblasts and odontoblasts and binds to hydroxyapatite crystals through its three Gla residues. Its synthesis is regulated by the active form of vitamin D (calcitriol). The percentage of undercarboxylated osteocalcin (%ucOC) is a sensitive marker of vitamin K deficiency and is related to lower bone resorption and formation in growing children.[54] In turn, supplementation with vitamin K_2 may maintain bone mineral density and prevent fractures among osteoporotic patients.[55] Vitamin K activates the calcification inhibitor MGP, which is expressed in various soft tissues and cell types, and may therefore prevent vascular calcification.[56] Protein S deficiency has been associated with osteopenia and osteonecrosis, as well as impaired vascular repair relevant for cardiovascular health, but its exact role in bone metabolism remains unclear.[57]

Recent research on vitamin K has identified the vitamin K dependent protein Gas6, which is a growth factor with mitogenic and antiapoptotic properties that helps maintain cells in the rest phase of the cell cycle.[58]

This research also helped identify that vitamin K_2 serves as a mitochondrial electron carrier, helping to maintain normal ATP production, which may make it a target for the treatment of mitochondrial pathologies.[59]

High intake of natural sources menaquinones, generally through cheese, is associated with a reduced risk of cancer[60] and coronary heart disease.[61] Oral phylloquinone and menaquinone supplementation is related to reduced bone loss.[62] More research is needed on the role of vitamin K in these conditions, however.

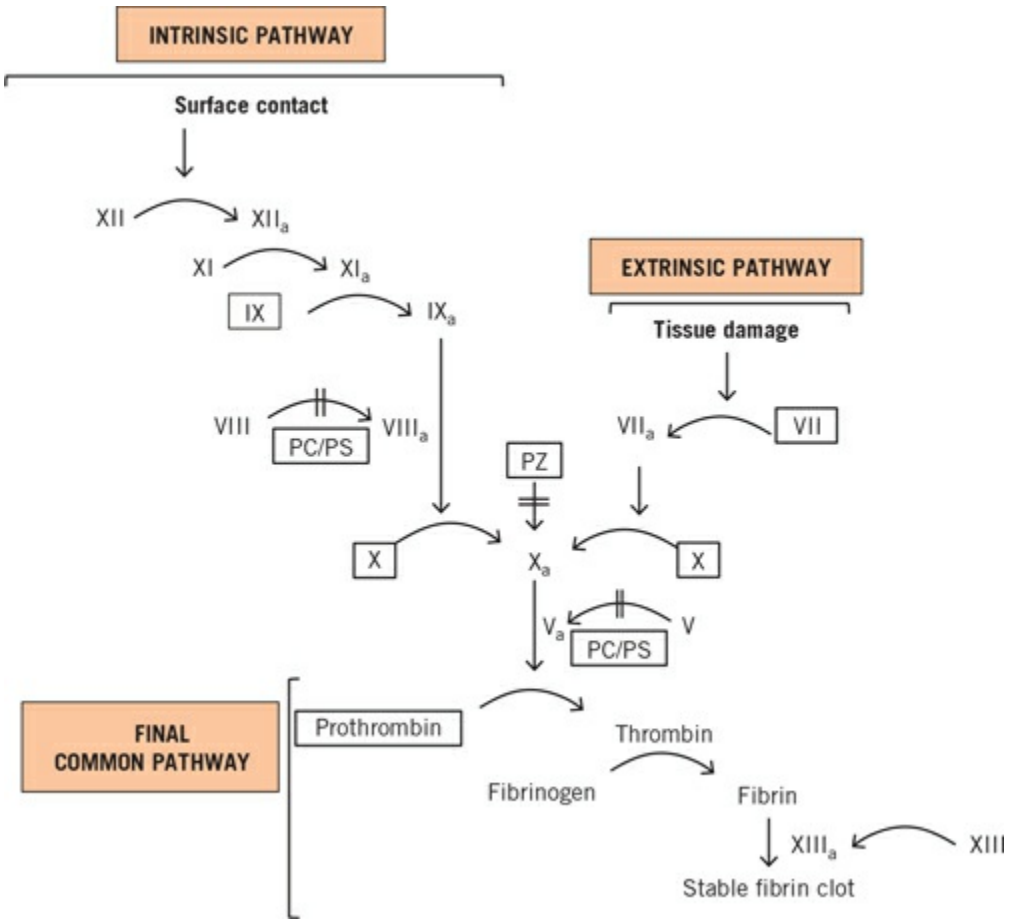


Figure 10-3 Role of Vitamin K in the blood coagulation cascade. Proteins C, S, and Z are shown as PC, PS, and PZ, respectively. Active proteins are shown with the subscript "a". Boxed factors and proteins are vitamin K-dependent.

Among adults without fat malabsorption, abnormal hemostatic control and other signs of vitamin K deficiency are extremely uncommon. In

contrast, liver menaquinone levels are either absent or extremely low at birth because of poor placental transfer and therefore place newborns at high risk of vitamin K deficiency bleeding (VKDB). Breast milk contains 2 μg/L of phylloquinone in colostrum and 1 μg/L in mature milk, and MK-4 levels at about half those levels. These low levels make exclusive breastfeeding a risk factor for VKDB, but exclusive breastfeeding should still be promoted for the early months of life given its other benefits and while other vitamin K deficiency control strategies are employed. Infant formulas are typically fortified to contain 55 μg/L of phylloquinone and are therefore a better source than breast milk.[53]

VKDB among infants is due to an insufficient production of clotting factors in the liver and presents in several forms. The earliest form of the disorder (occurring within the first 24 hours of life) is rare, but may be fatal due to bleeding from intracranial, intrathoracic, intra-abdominal and GI sites. It is generally caused by maternal use of drugs that inhibit vitamin K metabolism. Classical VKDB (occurring between days 2 and 7 of life) is typically restricted to the GI tract, umbilicus, skin, nose, or site of circumcision and is generally not lethal. It is considered as idiopathic but may be related to poor feeding practices and low intakes of breast milk or formula. Late VKDB peaks between weeks 3 and 8, but may occur as late as 6 months. It generally occurs among exclusively breast-fed infants and among infants malabsorption of vitamin K caused by undiagnosed cholestasis. It is the form with the most severe public health consequences and is often marked by acute intracranial hemorrhage and therefore higher rates of neurological handicaps or death.[53]

Without vitamin K prophylaxis, the incidence of late VKDB is an estimated 7 per 100,000 in the Europe but may be higher in less developed countries.[53] VKDB is effectively prevented by administering a single intramuscular dose of 0.5–1 mg to all newborns, as currently recommended by the AAP.[63] Weekly oral vitamin K supplementation during the first 3 months of life is effective among most children but not those with latent cholestasis. Research conducted in the1990s implicated intramuscular vitamin K with an increased risk of childhood cancer, but these findings have not been replicated in other studies.

High dietary intakes of vitamin K decrease the therapeutic effectiveness of anticoagulant medications such as warfarin. Therefore, dietary and supplementation habits should not be altered once the anticoagulant dose has been established. There is no evidence that high intakes of phylloquinone or menaquinones from food or supplements lead to adverse effects among persons who are not on anticoagulant therapy.

VITAMIN C

History, Structure, and Metabolism

Medical writings from ancient Egyptians, Greeks and Romans were the first to describe a syndrome akin to scurvy, the disease associated with vitamin C deficiency. The first modern accounts of scurvy appear in the sailing logs of the Portuguese and Spanish marine expeditions of the early 16th century. In 1747, Lind conducted his famous clinical experiments on board of the British vessel Salisbury in which he cured scurvy with oranges and lemons. In 1932, research groups led by Szent-Györgyi and King isolated and identified vitamin C. Haworth subsequently determined the structure of ascorbic acid and synthesized the vitamin. For their groundbreaking work on vitamin C, Szent-Györgyi and Haworth received the Nobel Prize in 1937.

The term vitamin C, a six-carbon lactone, encompasses a redox system composed of L-ascorbic acid, monodehydro-L-ascorbic acid (a free radical), and oxidized ascorbate (dehydro-L-ascorbic acid, or DHA). Vitamin C is synthesized from glucose by many animals. Humans lack L-gluconolactone oxidase, an enzyme necessary for the synthesis of ascorbic acid, and therefore require dietary supply of the vitamin. Vitamin C is widely distributed in fruits and vegetables. Excellent sources include citrus fruits and their juices, kiwis, and red and green peppers. Other good sources include broccoli, baked potatoes, and tomatoes. After ingestion, the vitamin is absorbed in the small intestine, preferentially in the distal segment, through the sodium-ascorbate co-transporters (SVCTs) and hexose transporters (GLUTs). At higher GI concentrations, it is also absorbed by passive diffusion. Absorption is 70%–90% of usual intake levels, but decreases to below 50% at intakes exceeding 1 g/day. The bioavailability from foods is similar to that from synthetic sources. The vitamin enters cells as DHA, where it is reduced to ascorbate by glutathione-dependent mechanisms. Relatively low amounts of the vitamin are lost through catabolism. The vitamin is excreted as unmetabolized ascorbic acid and oxalate. Renal excretion of vitamin C increases proportionally with higher intake levels and reaches up to 80% after intravenous dosages of approximately 100 g.[64]

Functions

Vitamin C plays important antioxidant roles and serves as a cofactor for enzyme-mediated hydroxylation reactions. The vitamin C-dependent enzymes prolyl 4-hydroxylase, prolyl 3-hydroxylase are required for the

hydroxylation of proline and lysyl-hydroxylase for the hydroxylation of lysine residues in the synthesis of collagen. Without this hydroxylation, precursor molecules of collagen are unable to fold into their final triple helical structure. Non-folding collagen explains many of the symptoms of scurvy, such as gum bleeding, bruising, and inadequate repair of damaged tissue. In carnitine synthesis, Vitamin C is a cofactor for 6-*N*-trimethyl-L-lysine hydroxylase and γ-butyrobetaine. Carnitine deficiency and decreased oxidation of fatty acids in muscle may be responsible for the muscular weakness characteristic of scurvy. Vitamin C is a cofactor of dopamine-β hydroxylase in the synthesis of norepinephrine and epinephrine, and peptidylglycine α-amidating monooxygenase in the activation of hormones and neurotransmitters. Ongoing work is examining the role of vitamin C in cell differentiation and intracellular signaling cascades.[65]

Ascorbic acid has strong antioxidant properties by scavenging reactive oxygen species. It thus neutralizes phagocyte-derived oxidants and protects plasma lipids, lipid membranes, and ocular tissue from oxidative damage. The antioxidant properties of ascorbic acid suggest that the vitamin may have protective properties against degenerative diseases such as atherosclerosis and cancer.

In observational studies, consumption of vitamin C-rich diets is related to a lower cancer risk, but there is concern that vitamin C is merely a marker of better diets or healthier general lifestyles in these studies. In line with this evidence, there are no clear benefits of oral vitamin C supplementation for cancer risk.[66] Vitamin C-rich diets and intakes of >700 mg supplemental vitamin C/day relate to a lower risk of coronary heart disease.[67] However, evidence on the benefits of supplemental vitamin C from randomized, controlled trials are inconclusive.[68]

For decades, vitamin C has been implicated in preventing and treating the common cold. However, a review of all studies administering oral dosages of 0.2 g/day of vitamin C did not reduce the incidence and severity of colds in the general population; however, supplemental vitamin C may reduce incidence among subgroups engaging in heavy physical exercise or living in cold environments.[69] There is evidence to suggest that vitamin C is related to several aspects of the immune function. Ascorbic acid supplementation improves neutrophil chemotaxis.

Vitamin C interacts with other nutrients in vivo. Ascorbic acid positively interacts with the endogenous antioxidant glutathione, and may contribute to antioxidant protection by maintaining glutathione in its reduced form. In iron absorption, vitamin C reduces ferric iron found in

dietary nonheme iron in the more absorbable ferrous iron form, and may impair the formation of unavailable iron complexes with ligands normally present in the GI lumen. Dietary vitamin C of about 25–50 mg/meal improves nonheme iron absorption and may improve iron status.[70]

Vitamin C deficiency among infants manifests itself as infantile scurvy, in the form of bone abnormalities, hemorrhage symptoms, and anemia. However, vitamin C deficiency is unlikely to occur when infants are exclusively breastfed during the first 6 months of life or when fortified infant formulas are used.

High vitamin C intakes from food are unlikely to lead to adverse effects; however, intakes should be limited to the RDA levels among those with hemochromatosis, glucose-6-phosphate dehydrogenase deficiency, and renal disorders. Among healthy adults, excess intake through supplements (>3000 mg/d) has been associated with mild adverse effects, such as osmotic diarrhea and intestinal discomfort. To prevent vitamin C-related adverse effects, healthy adults should not consume vitamin C at levels above the UL set at 2000 mg/d; taking into account their lower body weight, ULs are lower for children.

THIAMIN

History, Structure, and Metabolism

Descriptions of beriberi or *kakke*, the disease caused by dietary thiamin deficiency, appeared in Chinese medical manuscripts more than 4 millennia ago. It became a major public health problem in Eastern and Southern Asia and Europe when polished rice, devoid of the thiamin-rich aleurone layer between the germ and the endosperm, became a major staple food. The first reference to a potential nutritional cause of beriberi appeared in 1884 when a Japanese surgeon, Takaki, showed that the prevalence of the disease in the navy could be reduced by feeding sailors meat, wheat and milk instead of their usual diet, which consisted of polished rice and dried fish. In 1890, Eijkman documented that a diet of polished rice caused polyneuritis in chicken. In 1901, Grijns found that the antineuritic factor in rice bran was water-soluble, and Funk first referred to the factor as “vitamine” in 1911. Frazier and Stanton successfully used extracts of rice polishing to cure beriberi in humans in 1909–1915, and Williams published its structure in 1936.

Thiamin, also called vitamin B_1, vitamin F, and aneurin, consists of a pyrimidine nucleus and a thiazole ring linkend by a methylene bridge. The

natural form of the vitamin is a base, and the commercial form is usually a chloride hydrochloride salt. Bacteria, fungi, and plans can synthesize thiamin, but humans and animals depend on dietary intakes.

After ingestion, dietary thiamin phosphate esters are hydrolyzed to thiamin or thiamin monophosphate (ThMP) by an intestinal alkaline phosphatase and transported across the brush-border membrane to the portal system. The vitamin is absorbed in the proximal small intestine by an active, saturable, and energy-dependent transport. At high luminal concentrations, passive diffusion also becomes important. The liver, and to a lesser extent erythrocytes, capture and phosphorylate most of the thiamin entering the bloodstream.[71] Even though urinary thiamin excretion is downregulated when tissue levels decrease, tissue thiamin depletion occurs relatively rapidly under low dietary intake.

Functions

In the form of thiamin diphosphate (ThDP) (also known as thiamin pyrophosphate), the coenzymatic and most abundant form of the vitamin in tissues, thiamin is an indispensable cofactor in key metabolic pathways. In eukaryotes, ThDP accounts for 80%–90% of cellular thiamin, ThMP and free thiamin for less than 15%, and thiamin triphosphate (ThTP) for less than 1%.

One major ThDP-dependent biochemical reaction is the oxidative decarboxylation of α-keto acids. It involves the oxidative decarboxylation of pyruvic acid, the first step in the conversion of pyruvate to acetyl CoA in the mitochondria; the oxidative decarboxylation of α-ketoglutaric acid, a step within the tricarboxylic acid cycle in which α-ketoglutarate is converted into succinyl CoA; and the oxidative decarboxylation of the branched-chain α-keto acids α-ketoisocaproate, α-keto-β-methylvalerate, and α-ketoisovalerate, to produce isovaleryl CoA, α-methylbutyryl CoA, and isobutyryl CoA, respectively, in high energy-generating pathways.

ThDP is also required in the transketolase reaction occurs in the cytosol, an essential reaction for the synthesis of ribose and oxidation of glucose by the pentose phosphate pathway. In the peroxisome, ThDP functions in the α-oxidation of 3-methyl-branched fatty acids, primarily phytanic acid, a degradation product of chlorophyll ingested through the diet, in order to produce long-chain aldehydes. The failure of this α-oxidation reaction and concomitant accumulation of phytanic acid leads to Refsum disease, an autosomal recessive neurological disease.

Thiamin deficiency impairs numerous enzymes and cellular processes. Animal studies suggest that maternal thiamin deficiency impairs brain

lipogenesis[72] and energy metabolism.[73] In humans, maternal thiamin deficiency may increase the risk of intrauterine growth retardation[74] and in conjunction with niacin and pyridoxine deficiencies increase the risk of oral facial clefts.[75] Thiamin deficiency has also been linkend to increased risk of Alzheimer's disease. Oral and parenteral thiamin may improve cognitive function in patients with Alzheimer's disease, but more research is needed in this field.[76] High intakes of milled grains, a poor source of thiamin, increase thiamin needs owing to thiamin's role in carbohydrate metabolism. Consumption of thiaminases, which destroy thiamin, in fermented raw fish, betel nuts, and fermented tea leaves also contributes to thiamin deficiency. Thiamin deficiency used to be an important nutritional problem in emergency settings such as refugee camps, but can now be controlled through fortified blended foods used in these camps. Beriberi, the classical thiamin deficiency disease, generally manifests itself as the so-called dry form of the disease, a chronic, generally non-fatal condition characterized by peripheral neuropathy with sensory, motor, and autonomic compromises, predominantly of the distal segments of the limbs. Wet beriberi is less common and is characterized by edema, lactic acidosis, pulmonary hypertension and may lead to heart failure. Thiamin deficiency may also be common, but often unrecognized among critically ill patients and appropriate supplementation necessary.[77] Infantile beriberi may be found in children 2–3 months of age whose mothers have low thiamin concentrations in breast milk. In 2003, a defective soy-based infant formula caused an outbreak of thiamin deficiency among Israeli infants.[78] Cases were effectively treated with intramuscular doses of thiamin (50 mg/day) for two weeks.

Chronic, excessive alcohol consumption is linkend to low thiamin intake and impaired absorption and storage of the vitamin, and therefore predisposes to the thiamin deficiency disorder Wernicke–Korsakoff syndrome. The syndrome generally starts with Wernicke's encephalopathy, characterized by ataxia of gait and paralysis of eye movements, which at an early stage is responsive to intravenous thiamin. Prolonged deficiency may result in Korsakoff psychosis, which consists of mental alterations, including amnesia and irreversible brain lesions. Genetic factors, AIDS, or cancers may also predispose to Wernicke–Korsakoff syndrome.

There are no reports of toxicity from excess thiamin intake from food or supplements, presumably due to the low absorption efficiency of intakes >5 mg and rapid urinary excretion when tissue levels are elevated.[9]

RIBOFLAVIN

History, Structure, and Metabolism

Riboflavin, or vitamin B_2, was discovered as a fluorescent "yellow growth factor" in the heat-stable fraction of anti-pellagra extracts found by McCollum and collaborators. It was synthesized by Kuhn and Karrer in 1935, and its structure was elucidated by Warburg and Christian in 1938.

All riboflavin-related compounds have an isoalloxazine ring. Riboflavin itself does not have significant physiological functions, but is converted to the coenzymes flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD) in reactions catalyzed by riboflavin kinase and FAD synthetase, respectively (Figure 10-4). Hydrolysis reactions catalyzed by phosphatases can reverse this process. About two-thirds of the riboflavin in food is present as FAD, and smaller amounts as FMN and the free form. Riboflavin is very sensitive to UV radiation and can be destroyed during phototherapy of neonatal jaundice.[79]

The most common dietary sources of riboflavin are milk, eggs, meat, green leafy vegetables and enriched breakfast cereals. After ingestion, the riboflavin coenzyme derivatives are hydrolyzed to riboflavin in the brush border of enterocytes and absorbed mainly in the proximal small intestine by an active, saturable, carrier-mediated transport system. Two intestinal riboflavin transporters have recently been identified.[80,81] In enterocytes, free riboflavin is converted to FMN, most of which is further converted to FAD. After the action of nonspecific phosphatases, riboflavin enters the blood as FMN alongside the free form of the vitamin.

The liver captures large parts of the vitamin after absorption. In the blood, riboflavin is transported mainly bound to immunoglobulins and to a lesser degree to albumin. The vitamin is converted to its coenzymatic forms in the cytoplasm of many tissues, especially in the small intestine, liver, heart, and kidney. About 90% of the vitamin form in cells are FAD and the remainder FMN. Riboflavin is excreted in urine, predominantly in its free form.

Functions

Riboflavin functions via the cofactors FMN and FAD, which are used by more than 150 proteins involved in energetic metabolism, redox homeostasis and numerous regulatory events.

In the respiratory chain, FAD plays a key role in energy production. FAD helps protect tissues against lipid peroxidation by regenerating glutathione in the glutathione peroxidase antioxidant system. The role of

FAD in the activation of NADPH oxidase, needed for cell defense and production of signaling molecules related to innate immunity and other cellular responses, has recently been identified.[82]

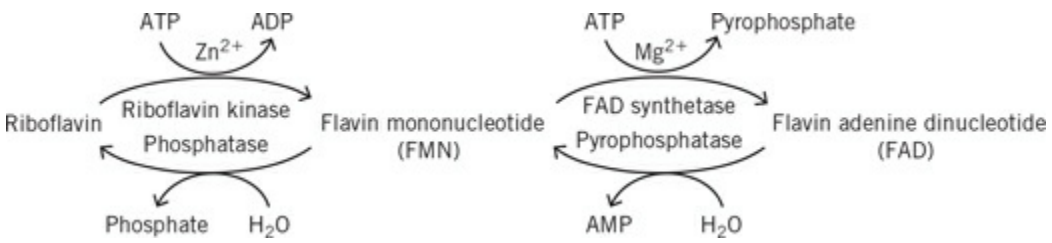


Figure 10-4 Interconversions of riboflavin and its cofactors.

Riboflavin enzymes are also involved in the metabolism of drugs, lipids, and several nutrients. FAD is important for folate and vitamin B_{12} metabolism. It is a cofactor for the enzyme 5,10-methylenetetrahydrofolate reductase, which catalyzes the conversion of methylene tetrahydrofolate to methyl tetrahydrofolate. The latter is required for the remethylation of homocysteine to methionine. In the metabolism of vitamin B_6, FMN is required for the conversion of pyridoxal-5-phosphate, while in niacin metabolism, the FAD-dependent kynurenine hydroxylase is needed for the conversion of tryptophan to niacin. Riboflavin deficiency may also impair iron absorption.

Inadequate food intake is a major cause of riboflavin deficiency, especially among those consuming dairy-free diets and the poor. High consumption of alcohol and certain psychotropic, chemotherapeutic, and antimalarial medications are related to poor riboflavin status. Poor riboflavin status may be common among critically ill patients[83] and translates into poor riboflavin levels in breast milk.[84] Among the several types of riboflavin-responsive genetic disorders that have been identified recently, the most common one is multiple acyl-CoA dehydrogenase deficiency, an autosomal recessive disease caused by defects in the mitochondrial electron transfer system and fatty acid metabolism.[85]

Riboflavin deficiency generally coexists with other vitamin deficiencies. Clinical deficiency symptoms in humans include stomatitis, glossitis, dermatitis, keratitis, anemia, and neurological dysfunction. Riboflavin deficiency may also impair cardiovascular health by increasing homocysteine concentrations and blood pressure[86] and possibly increase

cancer risk.[87] Recently, high-dose riboflavin supplements were identified as a potential prophylactic treatment in children and adolescents suffering from migraine.[88]

High riboflavin is not thought to lead to adverse effects, presumably due to limited GI absorption and rapid urinary excretion of the vitamin, but more information is needed on effects related to the use of pharmacological dosages over extended periods of time.

NIACIN

History, Structure, and Metabolism

Pellagra, the manifestation of niacin deficiency in humans, was first described in Spain in 1735. The disorder was soon recognized throughout large parts of Europe and Northern Africa when corn became the major food crop. The disease was also common in the United States in the early twentieth century, where it affected hundreds of thousands of people, with up to 7000 reported annual deaths.[89] In 1914, Goldberger began to demonstrate the nutritional origins of pellagra and in 1937, niacin was shown to cure the manifestation of pellagra in dogs and humans.

Niacin is found in a variety of foods, including beef, chicken, fish, liver, peanuts, and legumes. It is absorbed at physiological concentrations in the stomach and intestine by pH-dependent facilitated diffusion, while at higher concentrations passive diffusion predominates. NAD and NAD phosphate (NADP), the major forms of niacin in food, are converted by enzymatic action in the gut mucosa to nicotinamide. In the blood, nicotinamide constitutes the major form of vitamin and serves in the tissue synthesis of NAD when needed. However, both NAD and NADP can be synthesized in all tissues from nicotinic acid or nicotinamide. NAD is produced in different metabolic pathways that all involve the enzyme nicotinamide mononucleotide phosphoribosyltransferase.

Humans cannot synthesize nicotinic acid or nicotinamide endogenously and are therefore reliant on dietary intakes. However, niacin is unique among the B-vitamins in that it can be synthesized from a precursor molecule. The essential amino acid tryptophan can be converted to NAD via the formation of quinolinic acid and nicotinic acid ribonucleotide. An estimated 60 mg of dietary tryptophan yields 1 mg niacin,[90] but the conversion may be inhibited by deficiencies in other nutrients and by certain disease states, as explained in the next section. In typical Western adult diets, tryptophan contributes as much as two thirds

of the required niacin activity.

Excess niacin is excreted in the urine, mainly in the forms of N^1-methylnicotinamide (NMN) and 2-pyridone.

Functions

The metabolically active forms of niacin are NAD and NADP, whose nicotinamide moiety serves as an electron carrier or hydrogen donor in numerous redox reactions that are essential in carbohydrate, lipid, and amino acid metabolism. The NAD coenzyme generally functions as an electron acceptor in catabolic reactions that produce energy (e.g., the citric acid cycle). It also serves as the substrate for enzymes in non-redox pathways that cleave the β-*N*-glycosylic bond of NAD to free nicotinamide and catalyze the transfer of ADP-ribose. These non-redox pathways may modulate or regulate DNA and genome fidelity, and inflammatory and immune responses. NADP participates in reductive biosynthetic pathways involving fatty acid and cholesterol synthesis. It functions with NAD as a cohydrogenase in the pentose phosphate pathway to oxidize glucose 6-phosphate to ribose 5-phosphate. A novel area of research examines the role of NAD as a substrate for sirtuins (SIRTs), NAD-dependent protein deacetylases, of which seven members have been identified in humans. SIRTs have diverse cellular locations and functions, including epigenetic gene silencing, DNA repair and recombination, cell cycle regulation, apoptosis, and regulation of metabolism.[91]

The classical symptoms of the niacin-deficiency disease pellagra can be summarized as dermatitis, diarrhea, dementia, and potentially death (often called “the four Ds”). Pellagra develops when dietary niacin and tryptophan are insufficient to meet niacin requirements. Human experiments indicate that pellagra could develop after 50–60 days on a corn diet, which is low in both bioavailable niacin and tryptophan. The conversion of tryptophan to niacin is augmented among pregnant women and women taking oral contraceptives.[92] On the other hand, excessive intake of the amino acid leucine and inadequate intakes of iron, riboflavin, or vitamin B_6 may inhibit the conversion of tryptophan to niacin. In humans, prolonged isoniazid treatment, malignant carcinoid syndrome, chronic alcoholism, and Hartnup’s disease may result in pellagra-like symptoms.

Pharmacological levels of niacin are used to treat lipid disorders and prevent cardiovascular disease. They lower VLDL, LDL, and triglyceride levels while increasing beneficial HDL levels through mechanisms remain

incompletely understood.[93] Pharmacologic doses of nicotinic acid (but not nicotinamide) in the range of 1.5–4 g/day have proven effective at reducing the risk of atherosclerotic heart disease.[94] Several lines of evidence support niacin as a potential protective agent against skin cancer[95] and niacin's role in colon cancer is under investigation.[96] Low dietary niacin intakes may increase risk of Alzheimer's disease[97] and possibly multiple sclerosis and other neurodegenerative diseases.[98]

High niacin intakes from food are not known to cause adverse effects. Excess pharmacological dosages may lead first lead to flushing and subsequently to liver toxicity, impaired glucose tolerance, and nausea and vomiting. For children younger than one year, no UL has been defined due to a lack of data and suggestions that intakes should occur from food alone. For older children, ULs were defined based on adult data on flushing and subsequent adjustments based on body weight.[9] However, it has been argued that data on flushing is not relevant for children, and that there are therefore insufficient data to define a UL for children.[28]

VITAMIN B_6

History, Structure, and Metabolism

Vitamin B_6 was discovered by Gyoergy in 1934 during investigations on the etiology of the niacin deficiency disease pellagra. In 1939, Harris and Folkers elucidated the structure and achieved the synthesis of pyridoxine.

The term vitamin B_6 encompasses a group of compounds that are metabolically interchangeable and that, in their free form, are named pyridoxine (an alcohol), pyridoxal (an aldehyde), and pyridoxamine (an amine). These forms also exist in their respective 5´-phosphate ester forms pyridoxine phosphate (PNP), pyridoxal phosphate (PLP), and pyridoxamine phosphate (PMP).

The major forms of vitamin B_6 in animal foods are PLP and PMP, and pyridoxine and PNP in plant-derived foods. Rich sources of vitamin B_6 in U.S. diets are beef, poultry, and fortified cereals; other excellent sources are chickpeas, fish, beef liver, and other organ meats.

In the jejunum, phosphate ester forms of the vitamin are dephosphorylated, and free vitamin B_6 is absorbed by passive diffusion. The bioavailability of vitamin B_6 obtained from a mixed diet is approximately 75%. Vitamin B_6 is also synthesized by bacterial sources in

the colon and taken up by a regulatable carrier-mediated process, but its contribution to the human nutritional status remains to be defined.[99]

The free forms of vitamin B_6 are taken up by the liver and converted to PLP. Bound to albumin, PLP is also the major transport form carried to organs and tissues. About 75% of the body's vitamin B_6 pool is present in muscle. This pool is resistant to depletion even after prolonged intake of a vitamin B_6-deficient diet, which indicates that muscle vitamin B_6 is not a storage form of the vitamin that can be drawn upon in times of inadequate vitamin B_6 intakes. Catabolism of vitamin B_6 occurs mostly in the liver and excretion of the vitamin occurs in the urine, predominantly as 4-pyridoxic acid.

Functions

PLP facilitates the transfer of amino groups and is thus critical for the production of nonessential amino acids. For example, PLP is a cofactor for the enzyme glutamate-oxaloacetate aminotransferase, which catalyzes the formation of aspartic acid from glutamate. As a cofactor for kynureninase, PLP facilitates the conversion of the amino acid tryptophan to active niacin and the decarboxylation of precursor amino acids to generate several neurotransmitters, such as serotonin, taurine, dopamine, norepinephrine, histamine, and γ-aminobutyric acid.

In cellular one-carbon metabolism, the PLP-dependent enzyme serine hydroxymethyltransferase catalyzes the reversible transfer of the three-carbon of serine to tetrahydrofolate (THF) in the production of 5,10-methylene THF and glycine, which can be converted back to THF by the PLP-dependent glycine decarboxylase multienzyme system (Figure 10-5). With cystathione synthase, PLP converts homocysteine to cystathione, and with cystathione γ-lyase cystathione to cysteine. In carbohydrate metabolism, PLP functions in the production of glucose 1-phosphate from glycogen, in lipid metabolism in the production of carnitine and phospholipids, and in heme synthesis in the rate-limiting formation of γ-aminolevulinate.

Vitamin B_6 deficiency is first evidenced by decreases in plasma PLP levels and lowered amounts of the excretory product 4-pyridoxic acid in the urine. Continued vitamin B_6 deficiency leads to seborrheic dermatitis and decreased synthesis of neurotransmitters, resulting in depression, confusion, and abnormal brain wave patterns as indicated by abnormal electroencephalograms. Infants fed vitamin B_6-deficient formula are also

at risk of developing convulsions.[100] Vitamin B_6 deficiency may depress humoral and cell-mediated immunity by impairing the production of one-carbon units necessary nucleic acid synthesis. It may also impair heme synthesis and thus cause hypochromic microcytic anemia.

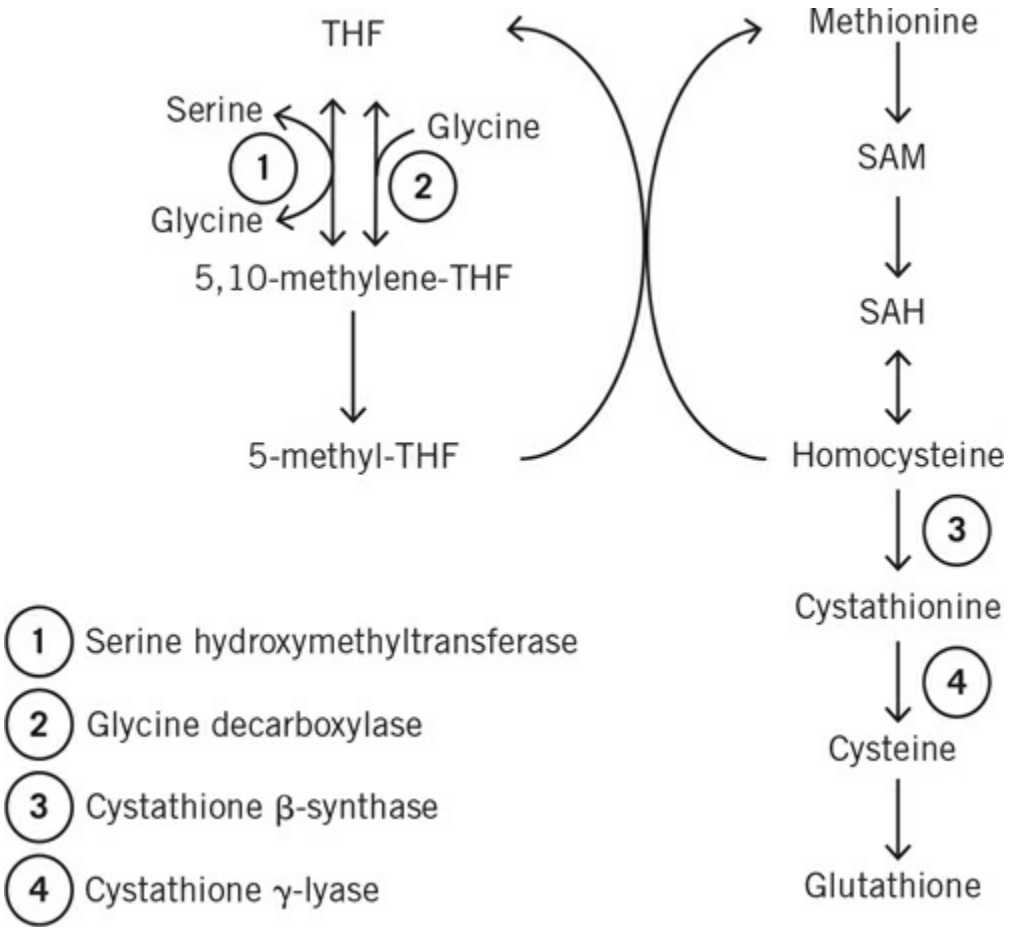


Figure 10-5 Vitamin B_6-dependent enzymes in one-carbon metabolism.

In observational epidemiologic studies, plasma PLP concentrations are inversely related to risk of colorectal cancer,[101] lung cancer,[102] coronary artery disease,[103] cerebrovascular disease,[104] and inflammatory conditions.[103] However, the biologic mechanisms are unclear. In fact, supplemental vitamin B_6 did not lower cardiovascular events or mortality in a high-risk populations either alone or when administered with other B vitamins.[105,106]

Levels of vitamin B_6 in breast milk are dependent on maternal intakes

of the vitamin and there is evidence that lactating mothers who consume inadequate levels of vitamin B_6 pass on deficient amounts of the vitamin to the infant.[84] Risk factors for human vitamin B_6 deficiency are old age, alcoholism, and use of penicillamine, isoniazid, and L-DOPA. Lower PLP levels have also been documented among oral contraceptive users.[107] High doses of vitamin B_6 may be beneficial in special conditions that elevate PLP requirements, such as intractable epilepsy among infants and inborn errors of vitamin B_6 metabolism. However, peripheral neuropathy and other adverse side effects must be considered.[108]

To safeguard against toxic effects of vitamin B_6, it is recommended that adults should not consume >100 mg/day. ULs have been set for children aged 1–18 years that are based on extrapolations from the adult UL.[9]

FOLATE

History, Structure, and Metabolism

In 1931, Wills described clinical folate deficiency as a tropical "macrocytic anemia" curable by a yeast extract. Ten years later, folate was isolated and extracted from spinach leaves in 1941 by Mitchell and others.

Folate is a generic term for a family of compounds that have nutritional properties of folic acid, the simplest form of the vitamin. Folate and folic acid derive their names from the Latin word for "leaf" (*folium*). Folic acid (pteroylglutamic acid) consists of a pteridine bicyclic ring system linkend to *p*-aminobenzoic acid, which is connected by a peptide bond to glutamic acid. Folic acid is the chemical form used in synthetic folate supplements, but does not occur in appreciable amounts in foods. Most folates exist in nature as polyglutamates, with five to eight glutamate residues joined to the first glutamate. The vitamin may be modified by reduction of the pteridine ring system to generate dihydrofolic acid and tetrahydrofolic acid (THF). The N-5 and N-10 positions of THF, either individually or in concert, can carry 1-C units, including methyl ($–CH_3$), formyl (–HCO), methylene ($–CH_2$), formyl (–CH=O), or formimino (–CH=NH) groups.

Good dietary sources of folate are liver, green leafy vegetables, citrus fruits, beans, and peas. Since 1998, folic acid has been added to enriched grain products includes breads, cereals, flours, and rice in the United States, and these products have become important contributors to dietary folate intakes. Currently, 75 countries mandate the addition of folic acid

(alongside iron) to grain products, most commonly wheat flour.[109]

Food folate occurs mostly as polyglutamates, which undergo hydrolysis in the jejunum by glutamate carboxypeptidase II in a pH-dependent mechanism. Transport into the intestinal cell is mediated by protein-coupled folate transporter (PCFT) and to a lesser degree by the reduced folate carrier. At high luminal levels, absorption can also occur by non-saturable passive diffusion. The monoglutamate from 5-methyl-THF is the predominant form in the portal circulation and in plasma. In tissue, most of folate is converted to polyglutamate forms by the enzymes methionine synthase and folylpolyglutamate synthetase to increase enzyme affinity and to retain and concentrate folate intracellularly.

Excess folate is excreted in the form of cleavage products in the urine. Fecal losses of folate are difficult to quantify because of the presence of folate-producing microbes in the colon.

Functions

Folate, in the form of its primary coenzyme THF, serves in the transfer of 1-C compounds needed in cycles for methionine, thymidylate, and purine synthesis. In these interconnected cycles, folate coenzymes are interconverted by C1-THF synthase in reactions producing 10-formyl THF, 5,10-methenyl THF and 5,10-methylene THF (Figure 10-6).

In the metabolism of amino acids, serine can transfer a methylene group to THF, to form glycine and 5,10-methylene-THF, which in turn can be reduced to 5-methyl-THF, followed by the transfer for the methyl group to homocysteine in a reaction catalyzed by the vitamin B_{12}-dependent enzyme methionine synthase to produce methionine and regenerate THF. This reaction is important in sustaining sufficient methionine pools for *S*-adenosylmethionine (SAM), which is involved in more than a hundred physiological reactions, including the methylation of DNA, RNA, and membrane phospholipids.

In a reaction catalyzed by thymidylate synthase, 5-10-methylene-THF donates a methyl group for the formation of the thymidylate, a necessary precursor of DNA synthesis and erythrocyte formation. THF is subsequently regenerated from DHF. The trifunctional enzyme C1-THF synthase converts 5,10-methylene-THF to 10-formyl THF, which is necessary for the *de novo* synthesis of the purines adenine and guanine. However, 5,10-methylene-THF may not be a major precursor in the production of 10-formyl-THF because the cellular environment favors the conversion of 10-formyl THF to 5,10-methylene-THF. A more important

reaction of the entry of 1-C units for folate-dependent biosynthetic reactions is the formulation of THF catalyzed by C1-THF synthase.

The first manifestation of folate deficiency is a decline in serum folate levels, occurring after approximately two weeks of consuming a folate-deficient diet. Deficiencies may also occur due to impaired absorption of the vitamin and after intake of a range of medications, including methotrexate, anticonvulsants, and anticancer drugs with anti-folate activity. An increasing number of folate-related polymorphisms resulting in impaired folate metabolism are being discovered.[110]

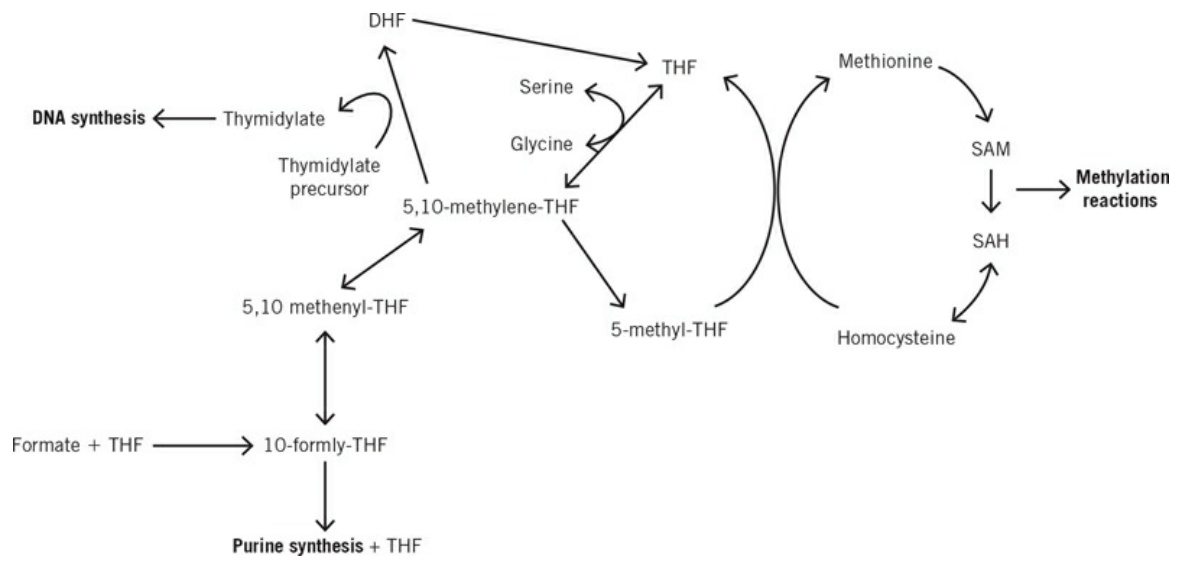


Figure 10-6 Summary of folate metabolism.

Folate deficiency leads to hyperpigmentation of neutrophils and a rise in homocysteine levels and abnormally nucleated erythrocytes in the bone marrow. Continued folate deficiency may lead to megaloblastic anemia. In the rapidly dividing cells of the GI tract, folate deficiency may impair cell division and lead to GI symptoms.

Increased folate consumption is related to lower plasma homocysteine levels and may therefore reduce vascular disease. However, folic acid therapy tested in randomized controlled trials had no effect on the incidence of major cardiovascular events.[111]

Owing to its role in DNA methylation, synthesis, and repair, dietary folate deficiency has been implicated in enhanced carcinogenesis, particularly in rapidly proliferative tissues such as the colorectal mucosa. In observational studies, higher dietary folate intakes are related to lower

risk of colorectal and other types of cancers.[112] However, high-dose folic acid does not reduce colorectal cancer risk.[111] There are concerns that folic acid supplementation may increase the growth of pre-existing tumors but more research is needed in this area.

Adequate folate status early in pregnancy greatly decreases the risk of NTDs, the most common major congenital abnormality in many countries,[113] and possibly congenital heart disease as well.[114] For this reason, folic acid has been added to enriched grains and mixed food items containing these grains in the United States since 1998 at levels of 0.14 mg/100 g flour.[115] This program has been credited with reducing the prevalence of low serum folate (<3 ng/mL) in the general population from 21% to <1% and with reducing NTD prevalence from 10.8 to 6.9 per 10,000 population.[116,117]

Deficiencies in either folate or vitamin B_{12} impair DNA synthesis in erythrocyte formation and thus lead to megaloblastic anemia. Deficiency in vitamin B_{12}, but not folate, also leads to neurological complications. There is concern that high intakes of folate may obscure or delay the diagnosis of vitamin B_{12} deficiency and associated neurological complications, especially among older adults with atrophic gastritis and those who follow a vegan diet or who suffer from pernicious anemia and bacterial overgrowth in the gut. The recommended ULs for folate proposed by the IOM attempt to safeguard against the masking of vitamin B_{12}.[118]

VITAMIN B_{12}

History, Structure, and Metabolism

The symptoms of the main vitamin B_{12} deficiency disorder, pernicious anemia, were first described in the mid-19th century by Addison. In 1926, Minot and Murphy showed that large quantities of liver normalized the hematological symptoms of pernicious anemia and in 1948, Folkers and Smith isolated the vitamin in the crystalline form from liver.

Vitamin B_{12} (cobalamin) is a large molecule with a complex corrin ring structure and a cobalt molecule at its center. Vitamin B_{12} is produced exclusively by microbes. Good dietary sources are meats (especially organ meats), fish, shellfish, eggs, fortified cereals and to a lesser extent dairy products. The vitamin is not generally present in plant products or yeast,

although high levels are present in dried green and purple lavers (seaweed). About 50% of dietary vitamin B_{12} is absorbed from mixed diets.[119]

Dietary vitamin B_{12} is mostly bound to proteins and released by pepsin in the acidic gastric environment. Free vitamin B_{12} binds to R protein (haptocorrin) and in the alkaline environment of the duodenum, R protein is degraded by pancreatic enzymes and the vitamin B_{12} binds to intrinsic factor (IF). In the ileum, the IF-B_{12} complex is taken up by the cubam receptor and is released as transcobalamin (TC). Absorption via passive diffusion may also occur, but only accounts for about 1% of absorbed vitamin B_{12}. After absorption, vitamin B_{12} is transported as part of TC to the liver and other tissues, where the TC-B_{12} complex is taken up by receptor-mediated endocytosis.[120]

Vitamin B_{12} is readily converted to its coenzyme forms after release from lysosomes. Among healthy omnivores, body vitamin B_{12} stores amount to about 2–3 mg, of which 50% is stored in the liver.

The main form of vitamin B_{12} excretion is bile, but daily losses are minimized to 0.1%–0.2% of the body pool due to effective reabsorption during enteropathic circulation. At very high plasma concentrations, such as after injections of the vitamin, urinary excretion also occurs.

Functions

In humans, vitamin B_{12} functions as part of the coenzyme methylcobalamin for methionine synthase and as part of 5-deoxyadenosylcobalamin for L-methylmalonyl-CoA mutase. Methylcobalamin is required by methionine synthase for the methyl transfer from 5-methyl-THF to homocysteine, to yield the end products methionine and THF. Methionine is required for the synthesis of the methyl donor SAM used in numerous methylation reactions.[121] In the absence of vitamin B_{12}, folate becomes trapped in its inactive form 5-methyl-THF, DNA synthesis is impaired, and megaloblastic anemia develops.

As part of 5-deoxyadenosylcobalamin, vitamin B_{12} is required for the enzyme L-methylmalonyl-CoA mutase to convert L-methylmalonyl-CoA to succinyl-CoA in an isomerization reaction. Lack of production of succinyl-CoA due to vitamin B_{12} deficiency is unlikely to have serious

metabolic consequences as succinyl-CoA is abundantly derived from fats and carbohydrates. However, accumulation of L-methylmalonyl-CoA in the cell can lead to increased production of odd-chain and branched-chain fatty acids. These fatty acids may be abnormally incorporated into nerve cell membranes and may account for neurological symptoms due to degenerated myelin nerve sheaths.[122] Vitamin B_{12} deficiency generally increases levels of methylmalonic acid, a marker for vitamin B_{12} deficiency, and of homocysteine, a less specific marker.

Foods of animal origin are the best naturally occurring sources of vitamin B_{12} and those who follow a vegan diet are therefore at risk for vitamin B_{12} deficiency. Vegan mothers may pass on inadequate levels of vitamin B_{12} through their breast milk are thus advised to supplement their infants from birth with vitamin B_{12} at the level of the adequate intake. Vitamin B_{12} deficiency may be a common public health problem in South Asia, selected parts of Africa and the Americas, but more data are needed to characterize the magnitude of the problem.[123,124]

In the United States, dietary intakes of vitamin B_{12} in the general, non-vegan population generally meet dietary needs. Vitamin B_{12} deficiency is therefore more commonly due to poor absorption rather than inadequate intakes. The most common cause of severe vitamin B_{12} absorption is pernicious anemia, a chronic atrophic gastritis in which autoantibodies are formed to IF producing parietal cells or to IF directly. In children, pernicious anemia is generally the result of a genetically determined defect in secretion of IF or the secretion of a defective IF. The prevalence of pernicious anemia increases with age and undiagnosed pernicious anemia may affect 2% of individuals over 60 years of age in the United States.[125] The vitamin deficiency can be corrected by large weekly injections of vitamin B_{12}, but simpler oral regimens using pharmacological dosages may also be effective.[126]

In atrophic gastritis, a chronic inflammation of the stomach mucosa, there is loss of gastric glandular cells with resultant reduced secretion of hydrochloric acid, pepsin, and IF, which in turn lowers the absorption of vitamin B_{12} from food proteins. The gastric levels may also result in an overgrowth of bacteria in the stomach and small intestine that bind the vitamin for their own use. Atrophic gastritis is the major cause of vitamin B_{12} deficiency among the 10%–15% of people over the age of 60 years.[127]

The neurological symptoms associated with vitamin B_{12} deficiency

may include tingling and numbness sensations in the extremities, motor disturbances, and cognitive changes such as loss of concentration and dementia. In extreme cases, the degeneration of peripheral nerves may progress to irreversible paralysis. Neurological complications may in some cases present without hematological symptoms. In fact, there is evidence of an inverse association between the prominence of neurological complications and the severity of anemia.

GI impairments due to vitamin B_{12} deficiency include atrophic glossitis, papillary atrophy of the tongue, appetite loss, flatulence, and constipation. Among children, severe vitamin B_{12} deficiency is related to irritability, failure to thrive, apathy, anorexia, and developmental regression and can often be reversed with supplementation.[128]

There is no evidence of adverse effects of vitamin B_{12} obtained from foods or supplements. As a result, ULs have not been defined for vitamin B_{12}.[118]

BIOTIN

History, Structure, and Metabolism

Using feeding studies with dried eggwhite, Boas described in 1927 that mammals require the water-soluble vitamin biotin. Koegl and Toennis isolated the vitamin from egg yolk in 1932 and the chemical structure was elucidated by du Vigneaud and colleagues in 1942. One year later, Harris and colleagues first synthesized biotin.

Biotin is an imidazole derivative that is widely distributed in foods and generally bound to protein. Digestion of dietary protein releases biotinyl peptides and intestinal biotinidases further hydrolyze biotinyl peptides to biotin. At low luminal concentrations, the vitamin is absorbed via the SMVT, which binds biotin, pantothenic acid, and lipoic acid with similar affinity. At high luminal concentrations, a nonsaturable process dominates. The bioavailability of biotin from food and supplements is nearly complete, but is impaired by the heat labile protein avidin contained in raw eggs. Biotin is bound non-specifically by albumin, α-globulin, and β-globulin and taken up into the liver and peripheral tissues via SMVT. The vitamin is stored in the liver and is excreted mainly in urine and to a lesser degree in bile.

Functions

Biotin serves as a coenzyme for carboxylases active in energy metabolism, fatty acid synthesis, and amino acid metabolism. It also binds to DNA binding proteins called histones and functions in cell signaling pathways.

Biotin is attached to the active site of five mammalian carboxylases: Acetyl-CoA carboxylases I and II catalyze the binding of bicarbonate to acetyl-CoA to form malonyl-CoA, required for the synthesis of fatty acids; pyruvate carboxylase is a key enzyme in gluconeogenesis; propionyl-CoA carboxylase catalyzes essential steps in the metabolism of several amino acids, cholesterol, and odd chain fatty acids; and methylcrotonyl-CoA carboxylase catalyzes an essential step in leucine catabolism.

Histones are essential for the folding of DNA into chromatin. The five major classes of histones H1, H2A, H2B, H3, and H4 have been identified in mammals. The covalent binding of biotin to lysine residues in histones, mediated by biotinidase and holocarboxylase synthetase, regulates posttranslational modifications of histones important for processes such as transcription, replication, and repair of DNA.[129]

Biotin functions in the expression of genes for enzymes involved in biotin utilization in human cells. In these reactions, biotin is transformed into biotinyl-5'-AMP by holocarboxylase synthetase and activated by soluble guanylate cyclase and a cGMP-dependent protein kinase.[130]

Even though symptomatic human biotin deficiency is rare, it has been manifested among persons on biotin-free total parenteral nutrition and among persons who consume large amounts of raw eggs over extended periods of time. The first manifestation of biotin deficiency is a depressed biotin level in urine. Signs of frank biotin deficiency include hair loss and a scaly red rash around the eyes, nose, mouth, and genital area. Neurologic symptoms in adults have included hallucination, depression, lethargy, and numbness and tingling of the extremities. Among infants, deficiency symptoms occur after three to six months on biotin-free total parenteral nutrition, which is earlier than among adults, possibly because of increased biotin requirements due to growth. Biotin deficiency has also been related to impaired cellular and humoral immunity, cell proliferation, and fatty acid metabolism.

Biotin deficiency may also be due to genetic defects. Infants with a genetic defect in the enzyme biotidinase fail to recycle biotin and lose large amounts of biotin metabolites in urine. Symptoms generally occur between 1 week and 1 year of age and include poor feeding, vomiting, rash around eyes, nose, and mouth, and neurological symptoms such as muscle pain, lethargy, and numbness. Genetic defects may also induce deficiencies in individual and multiple carboxylases or biotin transporters.

Pharmacological biotin supplementation generally improves symptoms related to these genetic defects and must be maintained throughout the lifetime.

There are no reports of adverse effects due to high doses of biotin. Even doses as high as 200 mg given orally and 20 mg given intravenously are not related to toxicity symptoms.[118]

PANTOTHENIC ACID

History, Structure, and Metabolism

Pantothenic acid was discovered as a growth factor for yeast and certain bacteria by Williams in 1933. Elvehjem and Jukes showed in 1939 that pantothenic acid was required for growth and prevented dermatitis in chicken. Williams established the chemical structure of the vitamin in 1940.

Pantothenic acid consists of a β-alanine molecule and a dihydroxy acid called pantoic acid joined by an amide link. There is little data on the pantothenic acid content of food, but the vitamin is thought to be widely distributed in both plant and animal based foods. Major known sources include chicken, beef, egg yolk, milk, potatoes, oat cereals, broccoli, and avocado. However, up to 75% of the vitamin may be lost during food processing, such as freezing and canning and refining of whole grains. In food, the vitamin is present as CoA or as 4´-phosphopantetheine, and dephosphorylation and hydrolysis reactions produce pantothenic acid, which is readily absorbable. The vitamin is taken up into the enterocyte at low concentrations by the SMVT, and passive transport at higher concentrations. After absorption, the vitamin is taken up from the plasma into the brain by facilitated diffusions and into other organs by the SMVT. The vitamin is excreted in urine, primarily as pantothenic acid.

Functions

Pantothenic acid serves as the substrate for the synthesis of CoA and Acyl Carrier Protein (ACP), which play important roles in the metabolism of carbohydrates, proteins, and fats. There are five enzymatic steps in the conversion of pantothenic acid to CoA, which then serves as the 4´-phosphopantetheine substrate in the formation of ACP (Figure 10-7).

As part of CoA, pantothenic acid functions in the transfer of acetyl and acyl groups in many vital reactions. These include reactions in oxidative metabolism and catabolism that generate energy from carbohydrates,

lipids, and proteins. In carbohydrate metabolism, CoA functions in reactions such as in the citric acid cycle and the acetylation of sugars; in lipid metabolism, CoA is required to synthesize essential fats, cholesterol, and steroid hormones. ACP is central to the synthesis of fatty acids through its role as a carrier of acyl intermediates.[131]

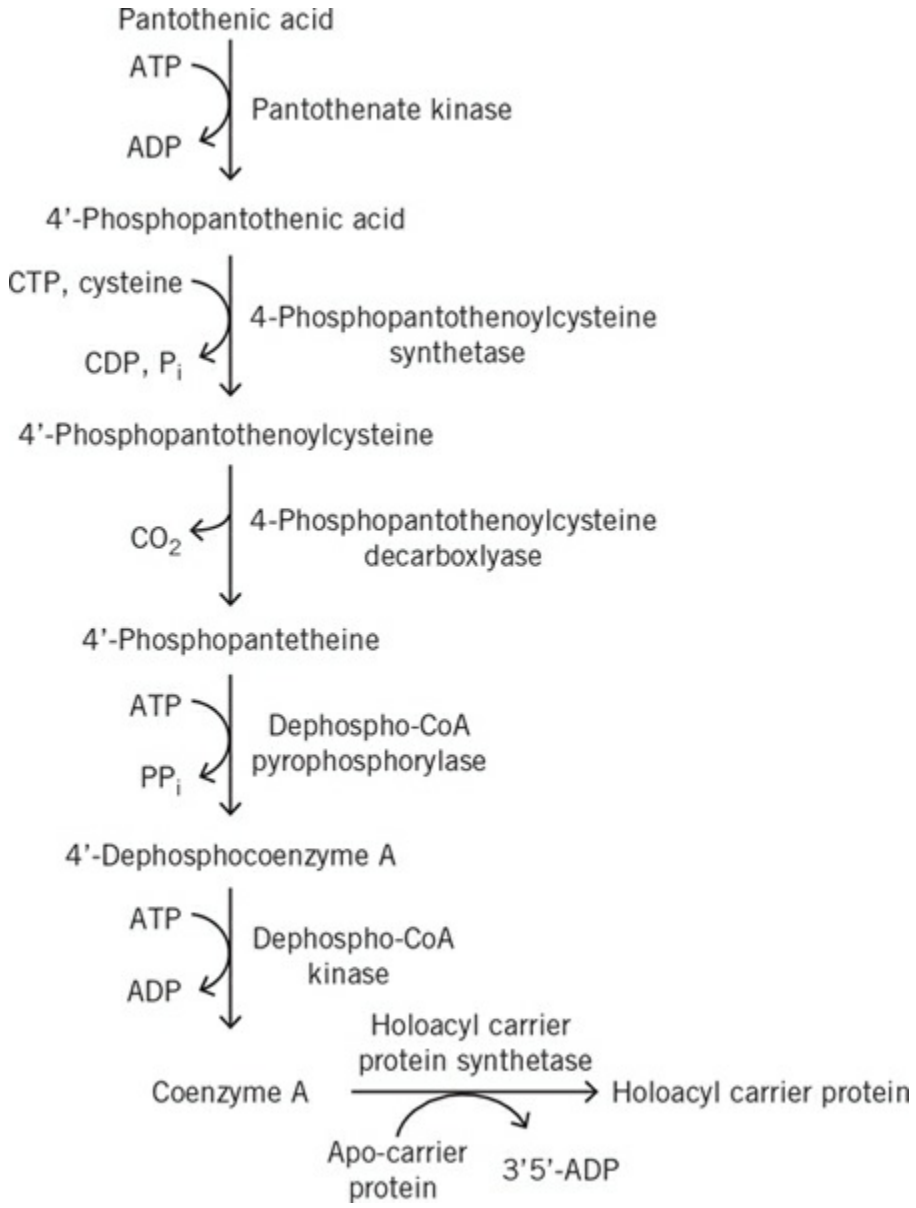


Figure 10-7 Conversion of pantothenic acid to Coenzyme A.

Pantothenic acid deficiency in humans is rare. Among severely malnourished prisoners of war during World War II, suffering from multiple deficiency diseases, the "burning feet" syndrome, a new condition

of paresthesia and severe pain in the feet and toes, was thought to be due to pantothenic acid deficiency. In experimental states of pantothenic acid deficiency induced by feeding volunteers synthetic diets devoid of pantothenic acid or by administering pantothenic acid antagonists, neuromotor disorders, mental depression, GI symptoms, and increased insulin sensitivity have been noted.

A variety of health claims have been made for pantothenic acid and its derivatives.

Pantethine, a metabolite of pantothenic acid, may have a hypocholesterolemic effect and raise high-density lipoprotein levels in individuals with dyslipidemia and hypercholesterolemia.[132] Oral pantothenic acid has been implicated in improved wound healing, but epidemiologic studies do not support this claim.[133] High dosages of the vitamin may have therapeutic benefits during rheumatoid arthritis[134] and lupus erythematosus[135] and are not related to adverse effects.

SUMMARY

We have reviewed the history, structure, function, and metabolism of the 13 water- and fat-soluble vitamins. Although the “golden age” of vitamin discovery and biochemical identification was 100 years ago, modern medical and public health advances continue to be made in the field, and both population and genetic approaches to determine optimal vitamin intake are being undertaken.

REFERENCES

1. Kennedy E, Meyers L. Dietary reference intakes: development and uses for assessment of micronutrient status of women – a global perspective. *Am J Clin Nutr*. 2005;81:1194S–1197S.
2. Health Canada. *Do Canadian Children Meet Their Nutrient Requirements Through Food Intake Alone?* Ottawa, Ontario, Canada: Health Canada Publications; 2009
3. Troesch B, Hoeft B, McBurney M, et al. Dietary surveys indicate vitamin intakes below recommendations are common in representative Western countries. *Br J Nutr*. 2012;108:692–698.
4. Troesch B, Eggersdorfer M, Weber P. 100 years of vitamins: adequate intake in the elderly is still a matter of concern. *J Nutr*. 2012;142:979–980.
5. World Health Organization. *Global Prevalence of Vitamin A Deficiency in Populations at Risk 1995–2005*. WHO Global Database on Vitamin A Deficiency. Geneva, Switzerland: World Health Organization; 2009.
6. Black RE, Allen LH, Bhutta ZA, et al. Maternal and child undernutrition: global and regional exposures and health consequences. *Lancet*. 2008;371:243–260.

7. Napoli JL. A gene knockout corroborates the integral function of cellular retinol-binding protein in retinol metabolism. *Nutr Rev*. 2000;50:230–235.
8. Leung WC, Hessel S, Meplan C, et al. Two common single nucleotide polymorphisms in the gene encoding beta-carotene 15,15'-monoxygenase alter beta-carotene metabolism in female volunteers. *FASEB J*. 2009;23:1041–1053.
9. Otten JJ, Pitzi Hellwig J, et al, eds. *Dietary Reference Intakes: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006.
10. Underwood BA. Maternal vitamin A status and its importance in infancy and early childhood. *Am J Clin Nutr*. 1994;59:517S–522S; discussion 22S–24S.
11. Subar AF, Krebs-Smith SM, Cook A, Kahle LL. Dietary sources of nutrients among US children, 1989–1991. *Pediatrics*. 1998;102:913–923.
12. Palmer AC, West KP, Dalmiya N, Schultink W. The use and interpretation of serum retinol distributions in evaluating the public health impact of vitamin A programmes. *Public Health Nutr*. 2012;15(7):1201–1215.
13. van den Broek N, Dou L, Othman M, et al. Vitamin A supplementation during pregnancy for maternal and newborn outcomes. *Cochrane Database Syst Rev*. 2010;(11):CD008666.
14. Gogia S, Sachdev HS. Maternal postpartum vitamin A supplementation for the prevention of mortality and morbidity in infancy: a systematic review of randomized controlled trials. *Int J Epidemiol* 2010;39:1217–1226.
15. Gogia S, Sachdev HS. Vitamin A supplementation for the prevention of morbidity and mortality in infants six months of age or less. *Cochrane Database Syst Rev*. 2011;(10):CD007480.
16. Haider BA, Bhutta ZA. Neonatal vitamin A supplementation for the prevention of mortality and morbidity in term neonates in developing countries. *Cochrane Database Syst Rev*. 2011;(10):CD006980.
17. Kirkwood B, Humphrey J, Moulton L, Martines J. Neonatal vitamin A supplementation and infant survival. *Lancet*. 2010;376:1643–1644.
18. World Health Organization. Vitamin A supplementation in infants and children 6–59 months of age. Geneva, Switzerland: World Health Organization; 2011.
19. Fawzi W, Chalmers T, Herrera M, Mosteller F. Vitamin A supplementation and child mortality: a meta-analysis. *JAMA*. 1993;269:898–903.
20. Rosales F, Kjolhede C, Goodman S. Efficacy of a single oral dose of 200,000 IU of oil-soluble vitamin A in measles-associated morbidity. *Am J Epidemiol*. 1996;143:413–422.
21. Ogaro F, Orinda V, Onyango F, Black R. Effect of vitamin A on diarrheal and respiratory complications of measles. *Trop Geograph Med*. 1993; 45:283–286.
22. American Academy of Pediatrics Committee on Infectious Diseases: Vitamin A treatment of measles. *Pediatrics*. 1993;91:1014–1015.
23. Villamor E, Mbise R, Spiegelman D, et al. Vitamin A supplements ameliorate the adverse effect of HIV-1, malaria, and diarrheal infections on child growth. *Pediatrics*. 2002;109:E6
24. Klemm RD, West KP, Jr, Palmer AC, et al. Vitamin A fortification of wheat flour: considerations and current recommendations. *Food Nutr Bull*. 2010;31:S47–S61.
25. Darnton-Hill I, Nalubola R. Fortification strategies to meet micronutrient needs: successes and failures. *Proc Nutr Soc*. 2002;61:231–241.
26. Dewey KG, Adu-Afarwuah S. Systematic review of the efficacy and effectiveness of complementary feeding interventions in developing countries. *Matern Child Nutr*. 2008;4 Suppl 1:24–85.
27. Tanumihardjo SA. Vitamin A: biomarkers of nutrition for development. *Am J Clin*

Nutr. 2011;94:658S–665S.
28. Zlotkin S. A critical assessment of the upper intake levels for infants and children. *J Nutr*. 2006;136:502S–506S.
29. Norman AW. Minireview: vitamin D receptor: new assignments for an already busy receptor. *Endocrinology*. 2006;147:5542–5548.
30. Haussler MR, Whitfield GK, Haussler CA, et al. The nuclear vitamin D receptor: biological and molecular regulatory properties revealed. *J Bone Miner Res*. 1998;13:325–349.
31. Harris S. Can vitamin D supplementation in infancy prevent type 1 diabetes? *Nutr Rev*. 2002;60:118–121.
32. Thornton KA, Marin C, Mora-Plazas M, Villamor E. Vitamin D deficiency associated with increased incidence of gastrointestinal and ear infections in school-age children. *Pediatr Infect Dis J*. 2013;32(6):585–593
33. Urashima M, Segawa T, Okazaki M, et al. Randomized trial of vitamin D supplementation to prevent seasonal influenza A in schoolchildren. *Am J Clin Nutr*. 2010;91:1255–1260.
34. Camargo CA, Jr., Ganmaa D, Frazier AL, et al. Randomized trial of vitamin D supplementation and risk of acute respiratory infection in Mongolia. *Pediatrics*. 2012;130:e561–e567.
35. Baeke F, Takiishi T, Korf H, et al. Vitamin D: modulator of the immune system. *Curr Opin Pharmacol*. 2010;10:482–496.
36. Liu PT, Stenger S, Li H, et al. Toll-like receptor triggering of a vitamin D-mediated human antimicrobial response. *Science*. 2006;311:1770–1773.
37. Villamor E, Marin C, Mora-Plazas M, Baylin A. Vitamin D deficiency and age at menarche: a prospective study. *Am J Clin Nutr*. 2011;94:1020–1025.
38. Gilbert-Diamond D, Baylin A, Mora-Plazas M, et al. Vitamin D deficiency and anthropometric indicators of adiposity in school-age children: a prospective study. *Am J Clin Nutr*. 2010;92:1446–1451.
39. Brehm JM, Celedon JC, Soto-Quiros ME, et al. Serum vitamin D levels and markers of severity of childhood asthma in Costa Rica. *Am J Respir Crit Care Med*. 2009;179:765–771.
40. Mansbach JM, Ginde AA, Camargo CA, Jr. Serum 25-hydroxyvitamin D levels among US children aged 1 to 11 years: do children need more vitamin D? *Pediatrics*. 2009;124:1404–1410.
41. Wagner CL, Greer FR. Prevention of rickets and vitamin D deficiency in infants, children, and adolescents. *Pediatrics*. 2008;122:1142–1152.
42. Institute of Medicine. *Dietary Reference Intakes for Calcium and Vitamin D*. Washington, DC: The National Acadamies Press; 2010.
43. Field CJ, Johnson IR, Schley PD. Nutrients and their role in host resistance to infection. *J Leukoc Biol*. 2002;71:16–32.
44. Meydani SN, Leka LS, Fine BC, et al. Vitamin E and respiratory tract infections in elderly nursing home residents: a randomized controlled trial. *JAMA*. 2004;292:828–836.
45. Myung SK, Ju W, Cho B, et al. Efficacy of vitamin and antioxidant supplements in prevention of cardiovascular disease: systematic review and meta-analysis of randomised controlled trials. *BMJ*. 2013;346:f10.
46. Farina N, Isaac MG, Clark AR, et al. Vitamin E for Alzheimer's dementia and mild cognitive impairment. *Cochrane Database Syst Rev*. 2012;(11):CD002854.
47. Klein EA, Thompson IM, Jr., Tangen CM, et al. Vitamin E and the risk of prostate cancer: the Selenium and Vitamin E Cancer Prevention Trial (SELECT). *JAMA*. 2011;306:1549–1556.

48. Greenough A, Shaheen SO, Shennan A, et al. Respiratory outcomes in early childhood following antenatal vitamin C and E supplementation. *Thorax*. 2010;65:998–1003.
49. Brion LP, Bell EF, Raghuveer TS. Vitamin E supplementation for prevention of morbidity and mortality in preterm infants. *Cochrane Database Syst Rev*. 2003; (4):CD003665.
50. American Academy of Pediatrics. *Pediatric Nutrition Handbook*. Elk Grove Village, IL: American Academy of Pediatrics; 2004.
51. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Vitamin C, Vitamin E, Selenium, and Carotenoids*. Washington, DC: National Academy Press; 2000.
52. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc*. Washington, DC: National Academy Press; 2001.
53. Shearer MJ. Vitamin K deficiency bleeding (VKDB) in early infancy. *Blood Rev*. 2009;23:49–59.
54. Kalkwarf HJ, Khoury JC, Bean J, Elliot JG. Vitamin K, bone turnover, and bone mass in girls. *Am J Clin Nutr*. 2004;80:1075–1080.
55. Shiraki M, Shiraki Y, Aoki C, Miura M. Vitamin K2 (menatetrenone) effectively prevents fractures and sustains lumbar bone mineral density in osteoporosis. *J Bone Miner Res*. 2000;15:515–521.
56. Shea MK, Holden RM. Vitamin K status and vascular calcification: evidence from observational and clinical studies. *Adv Nutr*. 2012;3:158–165.
57. Shearer MJ. The roles of vitamins D and K in bone health and osteoporosis prevention. *Proc Nutr Soc*. 1997;56:915–937.
58. Bellido-Martin L, de Frutos PG. Vitamin K-dependent actions of Gas6. *Vitam Horm*. 2008;78:185–209.
59. Vos M, Esposito G, Edirisinghe JN, et al. Vitamin K2 is a mitochondrial electron carrier that rescues pink1 deficiency. *Science*. 2012;336:1306–1310.
60. Nimptsch K, Rohrmann S, Kaaks R, Linseisen J. Dietary vitamin K intake in relation to cancer incidence and mortality: results from the Heidelberg cohort of the European Prospective Investigation into Cancer and Nutrition (EPIC-Heidelberg). *Am J Clin Nutr*. 2010;91:1348–1358.
61. Rees K, Guraewal S, Wong YL, et al. Is vitamin K consumption associated with cardio-metabolic disorders? A systematic review. *Maturitas*. 2010;67:121–128.
62. Cockayne S, Adamson J, Lanham-New S, et al. Vitamin K and the prevention of fractures: systematic review and meta-analysis of randomized controlled trials. *Arch Intern Med*. 2006;166:1256–1261.
63. Controversies concerning vitamin K and the newborn. American Academy of Pediatrics Committee on Fetus and Newborn. *Pediatrics*. 2003;112:191–192.
64. Robitaille L, Mamer OA, Miller WH, Jr., et al. Oxalic acid excretion after intravenous ascorbic acid administration. *Metabolism*. 2009;58:263–269.
65. Johnston CS. Vitamin C. In: Erdman, Jr. JW, Macdonald IA, Zeisel SH, eds. *Present Knowledge in Nutrition*. 10th ed. Hoboken, NJ: Wiley-Blackwell; 2012.
66. Gaziano JM, Glynn RJ, Christen WG, et al. Vitamins E and C in the prevention of prostate and total cancer in men: the Physicians' Health Study II randomized controlled trial. *JAMA*. 2009;301:52–62.
67. Knekt P, Ritz J, Pereira MA, et al. Antioxidant vitamins and coronary heart disease risk: a pooled analysis of 9 cohorts. *Am J Clin Nutr*. 2004;80:1508–1520.
68. Willcox BJ, Curb JD, Rodriguez BL. Antioxidants in cardiovascular health and

disease: key lessons from epidemiologic studies. *Am J Cardiol*. 2008;101:75D-86D.
69. Douglas RM, Hemila H, Chalker E, Treacy B. Vitamin C for preventing and treating the common cold. *Cochrane Database Syst Rev*. 2007;(3):CD000980.
70. Diaz M, Rosado JL, Allen LH, et al. The efficacy of a local ascorbic acid-rich food in improving iron absorption from Mexican diets: a field study using stable isotopes. *Am J Clin Nutr*. 2003;78:436–440.
71. Bettendorff L. Thiamin. In: Erdman Jr JW, Macdonald IA, Zeisel SH, eds. *Present Knowledge of Nutrition*. 10th ed. Hoboken, NJ: Wiley-Blackwell; 2012:261–279.
72. Trostler N, Sklan D. Lipogenesis in the brain of thiamine-deficient rat pups. *J Nutr Sci Vitaminol (Tokyo)*. 1978;24:105–111.
73. Fournier H, Butterworth RF. Effects of maternal thiamine deficiency on the development of thiamine-dependent enzymes in regions of the rat brain. *Neurochem Int*. 1989;15:439–444.
74. Heinze T, Weber W. Determination of thiamine (vitamin B1) in maternal blood during normal pregnancies and pregnancies with intrauterine growth retardation. *Z Ernahrungswiss*. 1990;29:39–46.
75. Krapels IP, van Rooij IA, Ocke MC, et al. Maternal dietary B vitamin intake, other than folate, and the association with orofacial cleft in the offspring. *Eur J Nutr*. 2004;43:7–14.
76. Lu'o'ng K, Nguyen LT. Role of thiamine in Alzheimer's disease. *Am J Alzheimers Dis Other Demen*. 2011;26:588–598.
77. Manzanares W, Hardy G. Thiamine supplementation in the critically ill. *Curr Opin Clin Nutr Metab Care*. 2011;14:610–617.
78. Fattal-Valevski A, Kesler A, Sela BA, et al. Outbreak of life-threatening thiamine deficiency in infants in Israel caused by a defective soy-based formula. *Pediatrics*. 2005;115:e233–e238.
79. Shumyantseva VV, Bulko TV, Schmid RD, Archakov AI. Photochemical properties of a riboflavins/cytochrome P450 2B4 complex. *Biosens Bioelectron*. 2002;17:233–238.
80. Fujimura M, Yamamoto S, Murata T, et al. Functional characteristics of the human ortholog of riboflavin transporter 2 and riboflavin-responsive expression of its rat ortholog in the small intestine indicate its involvement in riboflavin absorption. *J Nutr*. 2010;140:1722–1727.
81. Yonezawa A, Masuda S, Katsura T, Inui K. Identification and functional characterization of a novel human and rat riboflavin transporter, RFT1. *Am J Physiol Cell Physiol*. 2008;295:C632–C641.
82. Yazdanpanah B, Wiegmann K, Tchikov V, et al. Riboflavin kinase couples TNF receptor 1 to NADPH oxidase. *Nature*. 2009;460:1159–1163.
83. Gariballa S, Forster S, Powers H. Riboflavin status in acutely ill patients and response to dietary supplements. JPEN J Parenter Enteral Nutr 2009;33:656–61.
84. Allen LH. B vitamins in breast milk: relative importance of maternal status and intake, and effects on infant status and function. *Adv Nutr*. 2012;3:362–369.
85. Er TK, Liang WC, Chang JG, Jong YJ. High resolution melting analysis facilitates mutation screening of ETFDH gene: applications in riboflavin-responsive multiple acyl-CoA dehydrogenase deficiency. *Clin Chim Acta*. 2010;411:690–699.
86. McNulty H, Strain JJ, Pentieva K, Ward M. C(1) metabolism and CVD outcomes in older adults. *Proc Nutr Soc*. 2012;71:213–221.
87. Takata Y, Cai Q, Beeghly-Fadiel A, et al. Dietary B vitamin and methionine intakes and lung cancer risk among female never smokers in China. *Cancer Causes Control*. 2012;23:1965–1975.

88. Condo M, Posar A, Arbizzani A, Parmeggiani A. Riboflavin prophylaxis in pediatric and adolescent migraine. *J Headache Pain*. 2009;10:361–365.
89. Park YK, Sempos CT, Barton CN, et al. Effectiveness of food fortification in the United States: the case of pellagra. *Am J Public Health*. 2000;90:727–738.
90. Horwitt MK, Harper AE, Henderson LM. Niacin–tryptophan relationships for evaluating niacin equivalents. *Am J Clin Nutr*. 1981;34:423–427.
91. Dali-Youcef N, Lagouge M, Froelich S, et al. Sirtuins: the 'magnificent seven', function, metabolism and longevity. *Ann Med*. 2007;39:335–345.
92. Rose DP, Braidman IP. Excretion of tryptophan metabolites as affected by pregnancy, contraceptive steroids, and steroid hormones. *Am J Clin Nutr*. 1971;24:673–683.
93. Kamanna VS, Kashyap ML. Mechanism of action of niacin. *Am J Cardiol*. 2008;101:20B-26B.
94. Crouse JR, 3rd. New developments in the use of niacin for treatment of hyperlipidemia: new considerations in the use of an old drug. *Coron Artery Dis*. 1996;7:321–326.
95. Benavente CA, Jacobson MK, Jacobson EL. NAD in skin: therapeutic approaches for niacin. *Curr Pharm Des*. 2009;15:29–38.
96. Thangaraju M, Cresci GA, Liu K, et al. GPR109A is a G-protein-coupled receptor for the bacterial fermentation product butyrate and functions as a tumor suppressor in colon. *Cancer Res*. 2009;69:2826–2832.
97. Morris MC, Evans DA, Bienias JL, et al. Dietary niacin and the risk of incident Alzheimer's disease and of cognitive decline. *J Neurol Neurosurg Psychiatry*. 2004;75:1093–1099.
98. Penberthy WT, Tsunoda I. The importance of NAD in multiple sclerosis. *Curr Pharm Des*. 2009;15:64–99.
99. Said ZM, Subramanian VS, Vaziri ND, Said HM. Pyridoxine uptake by colonocytes: a specific and regulated carrier-mediated process. *Am J Physiol Cell Physiol*. 2008;294:C1192-C1197.
100. Coursin DB. Convulsive seizures in infants with pyridoxine-deficient diet. *J Am Med Assoc*. 1954;154:406–408.
101. Larsson SC, Orsini N, Wolk A. Vitamin B6 and risk of colorectal cancer: a meta-analysis of prospective studies. *JAMA*. 2010;303:1077–1083.
102. Johansson M, Relton C, Ueland PM, et al. Serum B vitamin levels and risk of lung cancer. *JAMA*. 2010;303:2377–2385.
103. Friso S, Girelli D, Martinelli N, et al. Low plasma vitamin B-6 concentrations and modulation of coronary artery disease risk. *Am J Clin Nutr*. 2004;79:992–998.
104. Kelly PJ, Shih VE, Kistler JP, et al. Low vitamin B6 but not homocyst(e)ine is associated with increased risk of stroke and transient ischemic attack in the era of folic acid grain fortification. *Stroke*. 2003;34:e51–e54.
105. Albert CM, Cook NR, Gaziano JM, et al. Effect of folic acid and B vitamins on risk of cardiovascular events and total mortality among women at high risk for cardiovascular disease: a randomized trial. *JAMA*. 2008;299:2027–2036.
106. Ebbing M, Bleie O, Ueland PM, et al. Mortality and cardiovascular events in patients treated with homocysteine-lowering B vitamins after coronary angiography: a randomized controlled trial. *JAMA*. 2008;300:795–804.
107. Morris MS, Picciano MF, Jacques PF, Selhub J. Plasma pyridoxal 5'-phosphate in the US population: the National Health and Nutrition Examination Survey, 2003–2004. *Am J Clin Nutr*. 2008;87:1446–1454.
108. Clayton PT. B6-responsive disorders: a model of vitamin dependency. *J Inherit Metab Dis*. 2006;29:317–326.

109. Flour Fortification Initiative. *FFI Newsletter December 2012*. Atlanta, GA: Flour Fortification Initiative; 2012.
110. Stover PJ. Polymorphisms in 1-carbon metabolism, epigenetics and folate-related pathologies. *J Nutrigenet Nutrigenomics*. 2011;4:293–305.
111. Zhou YH, Tang JY, Wu MJ, et al. Effect of folic acid supplementation on cardiovascular outcomes: a systematic review and meta-analysis. *PLoS One*. 2011;6:e25142.
112. Kennedy DA, Stern SJ, Moretti M, et al. Folate intake and the risk of colorectal cancer: a systematic review and meta-analysis. *Cancer Epidemiol*. 2011;35:2–10.
113. Czeizel AE, Dudas I, Paput L, Banhidy F. Prevention of neural-tube defects with periconceptional folic acid, methylfolate, or multivitamins? *Ann Nutr Metab*. 2011;58:263–71.
114. Blue GM, Kirk EP, Sholler GF, et al. Congenital heart disease: current knowledge about causes and inheritance. *Med J Aust*. 2012;197:155–159.
115. FDA. Food standards; amendment of standards of identity for enriched grain products to require addition of folic acid, final rule. *Fed Regist*. 1996;61:8781–8797.
116. Pfeiffer CM, Johnson CL, Jain RB, et al. Trends in blood folate and vitamin B-12 concentrations in the United States, 1988 2004. *Am J Clin Nutr*. 2007;86:718–727.
117. National Birth Defects Prevention Network. *Neural Tube Defect Ascertainment Project 2010*. Houston, TX: National Birth Defects Prevention Network; 2010.
118. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline*. Washington, DC: National Academy Press; 1998.
119. Watanabe F. Vitamin B12 sources and bioavailability. *Exp Biol Med (Maywood)*. 2007;232:1266–1274.
120. Quadros EV. Advances in the understanding of cobalamin assimilation and metabolism. *Br J Haematol*. 2010;148:195–204.
121. Matthews RG, Koutmos M, Datta S. Cobalamin-dependent and cobamide-dependent methyltransferases. *Curr Opin Struct Biol*. 2008;18:658–666.
122. Healton EB, Savage DG, Brust JC, et al. Neurologic aspects of cobalamin deficiency. *Medicine (Baltimore)*. 1991;70:229–245.
123. McLean E, de Benoist B, Allen LH. Review of the magnitude of folate and vitamin B12 deficiencies worldwide. *Food Nutr Bull*. 2008;29:S38-S51.
124. Stabler SP, Allen RH. Vitamin B12 deficiency as a worldwide problem. *Annu Rev Nutr*. 2004;24:299–326.
125. Carmel R. Prevalence of undiagnosed pernicious anemia in the elderly. *Arch Intern Med* 1996;156:1097–1100.
126. Andres E, Fothergill H, Mecili M. Efficacy of oral cobalamin (vitamin B12) therapy. *Expert Opin Pharmacother*. 2010;11:249–256.
127. Baik HW, Russell RM. Vitamin B12 deficiency in the elderly. *Annu Rev Nutr* 1999;19:357–377.
128. Dror DK, Allen LH. Effect of vitamin B12 deficiency on neurodevelopment in infants: current knowledge and possible mechanisms. *Nutr Rev*. 2008;66:250–255.
129. Zempleni J, Wijeratne SS, Hassan YI. Biotin. *Biofactors*. 2009;35:36–46.
130. Leon-Del-Rio A. Biotin-dependent regulation of gene expression in human cells. *J Nutr Biochem*. 2005;16:432–434.
131. Chan DI, Vogel HJ. Current understanding of fatty acid biosynthesis and the acyl carrier protein. *Biochem J*. 2010;430:1–19.
132. Gaddi A, Descovich GC, Noseda G, et al. Controlled evaluation of pantethine, a natural hypolipidemic compound, in patients with different forms of

hyperlipoproteinemia. *Atherosclerosis*. 1984;50:73–83.
133. Vaxman F, Olender S, Lambert A, et al. Effect of pantothenic acid and ascorbic acid supplementation on human skin wound healing process. A double-blind, prospective and randomized trial. *Eur Surg Res*. 1995;27:158–166.
134. Calcium pantothenate in arthritic conditions. A report from the General Practitioner Research Group. *Practitioner*. 1980;224:208–211.
135. Leung LH. Systemic lupus erythematosus: a combined deficiency disease. *Med Hypotheses*. 2004;62:922–924.

Dietary Reference Intakes in the United States

11

Alice H. Lichtenstein, DSc and Stephen R. Daniels, MD, PhD

INTRODUCTION

The dietary reference intakes (DRIs) are a set of recommendations intended to provide guidance in evaluating nutrient intakes and planning meals on the basis of nutrient adequacy.[1] In contrast to their predecessor, recommended dietary allowances (RDAs) last published in 1989, the DRIs differ in two ways: the emphasis has shifted from preventing nutrient deficiencies to reducing chronic disease risk and in addition to providing guidance for daily intakes, it also includes recommendations for maximum upper levels at which no adverse health outcomes would be expected, termed tolerable upper intake levels (ULs).

The DRIs were established during the mid-1990s through the mid-2000s with a series of committees that included both American and Canadian members under the auspices of the Food and Nutrition Board, Institute of Medicine of the National Academies of Science, in collaboration with Heath Canada. Separate committees were established for the following subcategories of nutrients: calcium, phosphorus, magnesium, vitamin D, and fluoride[2]; thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, pantothenic acid, biotin, and choline[3]; vitamin C, vitamin E, selenium, and carotenoids[4]; vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel,

silicon, vanadium, and zinc[5]; energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein, and amino acids[1]; and water, potassium, sodium, chloride, and sulfate.[6]

Depending on the available data for each nutrient, their DRIs are comprised of estimated average requirements (EARs), RDA or adequate intakes (AIs), and UL value.[7] In the case of macronutrients, there is an alternate category of recommendations, the acceptable macronutrient distribution ranges (AMDRs).[1] This latter category was established because evidence indicated that for energy containing nutrients optimal health outcomes were observed over a range of intakes.

When it appears an adequate body of new data is available for a specific nutrient or group of nutrients to warrant a reassessment of the DRI values, the data are reviewed and updated if necessary. Most recently, in 2010 a committee was convened to review the data for vitamin D and calcium. To facilitate this process, a systematic review was commissioned by the IOM for their use.[8] On the basis of the committee's deliberations, a modification was made to the DRIs for these nutrients.[9]

Table 11-1 provides summary definitions for these terms as established by the Food and Nutrition Board, including EAR, RDA, AI, UL, and AMDR.[10]

TABLE 11-1 Dietary Reference Intakes

Estimated Average Requirements (EAR): the average daily nutrient intake level estimated to meet the requirements of half the healthy individuals in a particular life stage and gender group.

Recommended Dietary Allowance (RDA): the average daily nutrient intake level sufficient to meet the nutrient requirements of nearly all (97–98%) healthy individuals in a particular life stage and gender group.

Adequate Intake (AI): a recommended average daily nutrient intake level based on observed or experimentally determined approximations or estimates of nutrient intake by a group (or groups) of apparently healthy people that are assumed to be adequate—used when an RDA cannot be determined.

Tolerable Upper Intake Level (UL): the highest average daily nutrient intake level likely to pose no risk of adverse health effects to almost all individuals in the general population. As intake increases above the UL, the potential risk of adverse effects increases.

Acceptable Macronutrient Distribution Ranges (AMDR): the intakes that are associated with reduced risk of chronic disease, intakes at which essential dietary nutrients can be consumed at sufficient levels and intakes based on adequate energy intake and physical activity to maintain energy balance.

Source: Adapted from IOM.[10]

BASIS FOR ESTABLISHING DRI VALUES

In formulating the DRIs, data were drawn primarily from peer reviewed publications. In some cases, the basis on which the reference values were derived was scanty, particularly for the younger age categories. In those cases, with the exception of birth through 1 year of age, the reference values were extrapolated from adult data. The actual criteria on which nutrient adequacy was based varies among nutrients and age groups. This information is documented, in detail, in each of the DRI committee reports.[1–6]

The DRI values are presented for the following age categories: 0 through 6 months, 7 through 12 months, 1 through 3 years, 4 through 8 years, 9 through 13 years, 14 through 18 years, 19 through 30 years, 31 through 50 years, and greater than 70 years. This chapter will be restricted to recommendations for categories up to and including age 18 years.

In general, the recommendations for infants aged 0 through 6 months are based on average consumption of human milk. Recommendations for individuals 7 through 12 months are based on average consumption of human milk and complementary foods. For subsequent age groups, sources of data, depending on the specific nutrient, include mean national intake data (Continuing Survey of Food Intake by Individuals) and amounts shown to provide the greatest protection against a specified chronic disease or deficiency state. Further taken into consideration was whether the recommendations provided for adequate levels of energy meet expenditure plus energy deposition and maintenance of nitrogen equilibrium plus protein deposition. The criteria used to assess adequacy can differ for individuals at different stages in the life cycle.

DIETARY REFERENCE INTAKES

Estimated Average Requirement

EAR is the daily value that is estimated to meet the requirement, defined by a predetermined indicator or criterion of adequacy, of half the healthy population of individuals represented by each age and sex group. The underlying assumption is that the distribution of actual requirements in a population is normally distributed. The EAR value is dependent on the indicator used to assess nutrient adequacy. The primary use of EAR values is to establish RDA values.

Recommended Dietary Allowance

RDA is the daily value that is estimated to meet the requirement, defined by a predetermined indicator or criterion of adequacy, by 97%–98% or nearly all of the healthy population of individuals represented by each age and sex group. By definition, the RDA value is calculated by summing the EAR and two standard deviations of the EAR if there are adequate data on which to calculate the variance. In cases where there are inadequate data with which to calculate the standard deviation of the EAR, the variance is assumed to be 10% of the EAR and the RDA is calculated by summing the EAR and two times 10% of the EAR. The primary use of RDA values is as a daily intake goal. For nutrients, where there were inadequate data on which to calculate EAR and RDA values for children, the numbers were extrapolations from adult data.

Adequate Intake

AI is the daily value that is estimated from the actual intake data of healthy people when there are inadequate data with which to calculate an EAR. The AI values are expected to meet or exceed the amounts needed to maintain a defined nutritional state or criterion of adequacy in essentially all members of a specific age and sex group. Hence, the AI values may exceed the EAR values in the range of individuals covered. The AI values are intended to be used similarly to the RDAs. Nonetheless, the level of certainty for AI values is less than that for RDA values. Both the RDA and AI values are used as goals for nutrient adequacy.

Acceptable Macronutrient Distribution Range

AMDR values were established for the macronutrients (fat, carbohydrate and protein) and *n*-6 and *n*-3 fatty acids (Table 11-2). The subcategory for fatty acids was created because they are essential nutrients—humans cannot make them in adequate amounts to support growth, development, and maintenance. The AMDR values provide a recommended range of intakes that are associated with optimal health outcomes and fulfill the RDAs for essential nutrients based on adequate energy intake and physical activity to maintain energy balance. Ranges were established after reviewing the available data and superimposing information about the interrelated metabolic fates of the macronutrients. The recommendations are expressed as percent of dietary energy because their absolute intakes are dependent on total energy needs. For the most part, the criteria used to establish AMDRs are the levels associated with low chronic disease risk and levels necessary to ensure sufficient intakes of other essential

nutrients. In some cases, for subclasses of the macronutrients, for which specific, unique functions were identified, additional criteria were used to establish either ERA or AI values. Examples include omega-3 fatty acids and indispensable amino acids.

Tolerable Upper Limit

The UL is the daily value that is estimated to be the highest level likely to pose no risk of adverse health effects for most people. The word "tolerable" was chosen as a qualifier for "upper levels" specifically to avoid implying that the upper levels are preferable or beneficial, especially compared to the RDAs or AIs. UL values were determined using a risk assessment methodology. ULs are a new category of recommendations and were added because of the concern that widespread fortification of foods and common use of dietary supplements can result in intake levels considerably higher than that attainable with mixed diets and may result in adverse health outcomes.

TABLE 11-2 Uses of DRIs for Assessing Intakes by Individuals and Groups

For an Individual	For a Group
EAR: Use to examine the probability that usual intake is inadequate.	**EAR:** Use to estimate the prevalence of inadequate intakes within a group.
RDA: Usual intake this level has a low probability of inadequacy.	**RDA:** Do not use to assess intakes of groups.
AI: Usual intake at or above this level has a low probability of inadequacy.	**AI:** Mean usual intake at or above this level implies a low prevalence of inadequate intakes.[a]
UL: Usual intake above this level may place an individual at risk of adverse effects from excessive nutrient intake.	**UL:** Use to estimate the percentage of the population at potential risk of adverse effects from excessive nutrient intake.

Abbreviations: AI, adequate intake; EAR, estimated average requirement; RDA, recommended dietary allowance; UL, tolerable upper intake level.

[a]When the AI for a nutrient is not based on mean intakes of healthy populations, this assessment is made with less confidence.

Source: Adapted from IOM.[10]

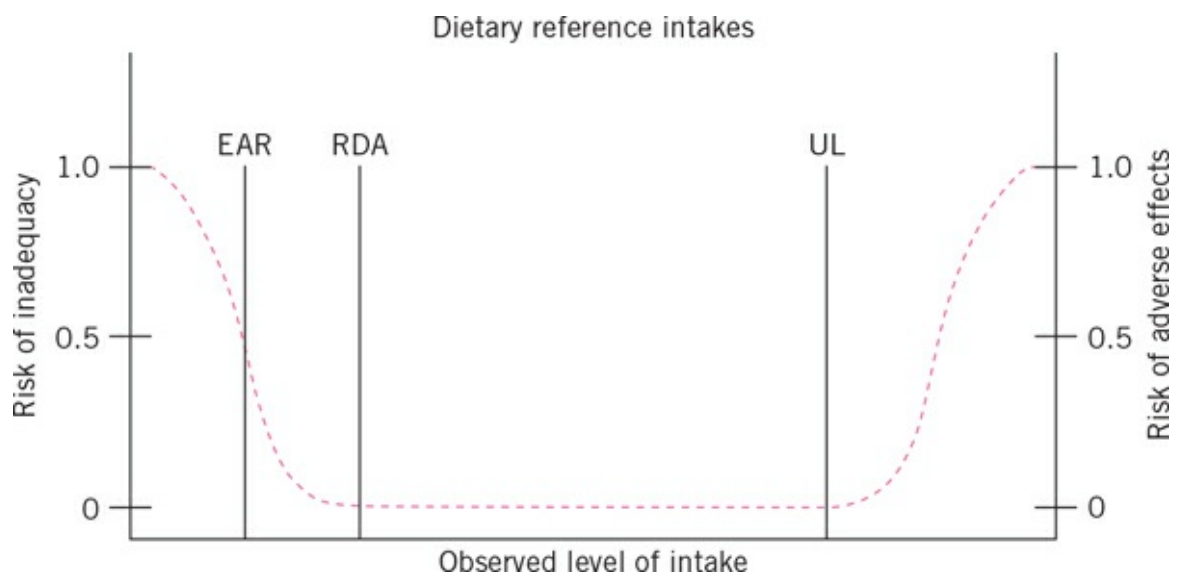


Figure 11-1 The relationship between the estimated average requirement (EAR), recommended dietary allowance (RDA), and tolerable upper intake level (UL). The risk of inadequate intake is 50% at the EAR, 2%–3% of the RDA, and close to 0% at the UL. Risks of adverse effects are close to 0% at the UL but increase with increasing intake. (Reproduced with permission from the Institute of Medicine.[2])

The relationship among the EAR, RDA, and UL values is depicted in Figure 11-1. Intakes between the RDA and UL values are assumed to be optimal because the risk of inadequacy and excess are both estimated to be close to zero.

INTENDED USES OF THE DRI

The major uses of DRIs as defined by the Food and Nutrition Board include determining the nutrient adequacy and for the purpose of planning diets for both individuals and groups of individuals (Table 11-3). They serve as the basis for all federal feeding programs, food assistance programs, and nutrition education programs, and are used to set Dietary Values (DV) for Food and Drug Administration's mandatory Nutrient Facts Panel. The DRIs, Dietary Guidelines for Americans and MyPlate are internally consistent.

DRI LIFE STAGE GROUPS RELEVANT TO THE

PEDIATRIC POPULATION

Infancy

DRI values are provided for two 6-month age groups during the first year of life, 0 through 6 months and 7 months through 1 year. Recommendations are based on the assumption that the infants are full-term, born to healthy and well nourished women, and exclusively fed human milk. Ethical considerations preclude the experimental determination of EAR values for infants. For this reason all the recommended values for these age categories are AIs. They are derived from the mean composition of an average volume of milk consumed by the specific age group. The recommendations are based on AI values, hence, may exceed the EAR values. It is important to note the decision of the Food and Nutrition Board committees to base the AIs on human milk is consistent with the recommendation of the American Academy of Pediatrics and the Canadian Paediatric Society, both of which state that exclusive breastfeeding is the preferred method of feeding for normal full-term infants for the first 4–6 months of life.

TABLE 11-3 Dietary Reference Intakes (DRIs): Acceptable Macronutrient Distribution Ranges[a]

	Range (Percent of Energy)	
Macronutrient	**Children, 1–3 y**	**Children, 4–18 y**
Fat	30–40	25–35
n-6 polyunsaturated fatty acidsb (linoleic acid)[a,b]	5–10	5–10
n-3 polyunsaturated fatty acidsb (α-linoleic acid)	0.6–1.2	0.6–1.2
Carbohydrate	45–65	45–65
Protein	5–20	10–30

[a]Food and Nutrition Board, Institute of Medicine, National Academies

[b]Approximately 10% of the total can come from longer-chain n-3 or n-6 fatty acids.

0 Through 6 Months

For the first 6 months of life, food intake relative to body weight is for the most part constant. As an infant grows, their food intake will increase. The mean intake of a nutrient is calculated based on the average concentration

of the nutrient from 2 through 6 months of lactation and an average volume of milk (780 mL/day). It is assumed that infants will have adequate access to human milk and can thus increase their intake to meet their requirements for growth and maintenance.

7 Through 12 Months

For the second 6 months of life, growth velocity slows and daily nutrient needs, on a body weight basis, are lower than during the first 6 months. This 6-month period is characterized by a gradual weaning from milk and transitioning to a mixed diet including solid foods. It was assumed on the basis of available data that nutrient needs have not changed markedly during this time period.

Toddlers: Age 1 Through 3 Years

For ages 1 through 3 years, growth velocity in height has been greater than other age periods after infancy. From a practical perspective the Food and Nutrition Board committees also took into consideration that after age 4 years children enter into daycare and school systems and that hence, the source and exposure to food frequently changes. It is noteworthy that data for individual nutrients are relatively scarce for this age category and many values were extrapolated.

Early Childhood: Ages 4 Through 8 Years

For ages 4 through 8 years there are continued changes in growth velocity and endocrine status. These changes separate this group from the younger and older age categories. Although limited, the Food and Nutrition Board committees determined there were more data to define EAR's and AIs for this than the prior age categories. The DRIs for ages 4 through 8 years form the basis of many school feeding programs.

Puberty/Adolescence: Ages 9 Through 13 Years and Ages 14 Through 18 Years

The distinction between ages 9 through 13 years and 14 through 18 years was made on the basis of multiple developmental criteria. For girls, the criteria were increased estrogen secretion physically marked by breast development and onset of the growth spurt. For boys, the criteria were initiation of testicular development and onset of the growth spurt. In general, the growth spurt in boys begins about 2 years later than in girls. For this reason sex specific recommendations are made starting with these

age categories.

TYPES OF DATA USED TO FORMULATE THE DRIs

The type of data used to formulate the DRIs varies from age group to age group and nutrient to nutrient, especially between energy containing and non-energy containing nutrients. Nevertheless, there are certain common features. The primary source of the data used to formulate the DRI values is clinical, dose–response, balance, depletion–repletion, and observational and case-controlled studies.

On the basis of a review of the available data, the possible criteria of nutrient adequacy were identified by each committee for each individual nutrient. The collective scientific judgment of the committee members was then used to select the indicator criterion to determine whether the available data would allow for the establishment of an EAR or AI value. The criterion differed among nutrients and in many cases differed by life stage group. The evaluation and decision were documented for each nutrient by the committee members and was included in the individual reports. Factors that went into the decisions were the strength of the data, consistency and preponderance of the data, and the degree of concordance among epidemiological, and clinical and laboratory findings.

The following factors were taken into consideration when assessing the available data: strength of association (usually expressed as relative risk), dose–response relationship, temporally correct association (with exposure preceding the onset of disease), consistency of association; specificity of association, and biological plausibility.

RDA's AND AI's FOR INFANTS, CHILDREN, AND ADOLESCENTS

The current RDA, AI, and AMDR values for infants, children, and adolescents can be found on the Food and Nutrition Board's website.[11] Noteworthy, these numbers should be used as targets on a daily basis. Actual intakes should be averaged over a number of days to assess adequacy.

For individuals below 18 years, there are few age-specific categories of nutrients that have AMDR values than have RDAs and IAs values; the categories include children aged 1 through 3 years and 4 through 18 years.

The collapsing of categories was necessary due to limited data for the potentially unique needs of each of the original age grouping.

Summarized in Table 11-4 are additional macronutrient recommendations for non-essential dietary components: cholesterol, *trans* fatty acids, saturated fatty acids, and added sugars. These non-essential dietary components are ubiquitous in the food supply and intakes, specifically with respect to the lipid soluble compounds, and have been associated with increased risk of chronic diseases.[12] These are general recommendations across all age and sex categories.

TABLE 11-4 Dietary Reference Intakes (DRIs): Additional Macronutrient Recommendations[a]

Macronutrient	Recommendation
Dietary cholesterol	As low as possible while consuming a nutritionally adequate diet
Trans fatty acids	As low as possible while consuming a nutritionally adequate diet
Saturated fatty acids	As low as possible while consuming a nutritionally adequate diet
Added sugars	Limit to no more than 25% of total energy

[a]Food and Nutrition Board, Institute of Medicine, National Academies.

Source: Adapted from IOM.[1]

TOLERABLE UPPER INTAKE LEVELS

Current ULs for those nutrients for which values have been established can be found on the Food and Nutrition Board's website.[11] The dearth of data for some nutrients precluded establishing UL values and does not indicate that intake at any level is without risk. It is important to note that UL values can only be used to assess the likelihood that an individual may be at risk for adverse effects, not that they actually will be at risk. As indicated, these values represent upper limits at which no adverse effects are likely to occur, not optimal intakes.

SELECTED NUTRIENT FUNCTIONS AND MAJOR FOOD SOURCES FOR VITAMINS AND

ELEMENTS

Summarized in Table 11-5 are nutrient functions and their major sources in the U.S. food supply. The wide range of foods noted among the nutrients is reflective of both natural occurrence and mandatory federal guidelines for fortification. For example, there are a limited number of foods that are good sources of vitamin D. For this reason, the federal government has mandated that all fluid milk be fortified with vitamin D. Fluid milk was chosen because of the unique needs of growing children and adolescents for this nutrient and historically the habitual food intake patterns of these age groups. As the intake of milk declines in these age groups, alternate approaches may be needed to ensure AI of this essential nutrient.[13]

TABLE 11-5 Dietary Reference Intakes: Macronutrients (Continued)

Nutrient	Function	Selected Food Sources
Carbohydrate-total digestible	RDA based on its role as the primary energy source for the brain; AMDR based on its role as a source of kilocalories to maintain body weight.	Starch and sugar are the major types of carbohydrates. Grains and vegetables (corn, pasta, rice, potatoes, breads) are sources of starch. Natural sugars are found in fruits and juices. Sources of added sugars are soft drinks, candy, fruit drinks, and desserts.
Total fiber	Improves laxation, reduces risk of coronary heart disease, assists in maintaining normal blood glucose level.	Includes dietary fiber naturally present in grains (such as found in oats, wheat, or unmilled rice) and functional fiber synthesized or isolated from plants or animals and shown to be of benefit to health.
Total Fat	Energy source and when found in foods, is a source of *n*-6 and *n*-3 polyunsaturated fatty acids. Its presence in the diet increases absorption of fat soluble vitamins and precursors	Butter, margarine, vegetable oils, whole milk, visible fat on meat and poultry products, invisible fat in fish, shellfish, some plant

	such as vitamin A and pro-vitamin A carotenoids	products such as seeds and nuts, and bakery products.
n-6 polyunsaturated fatty acids (linoleic acid)	An essential component of structural membrane lipids, involved with cell signaling, and precursor of eicosanoids. Required for normal skin function.	Nuts, seeds, and vegetable oils such as soybean, safflower, and corn oil.
n-3 polyunsaturated fatty acids (α-linoleic acid)	Involved with neurological development and growth. Precursor of eicosanoids.	Vegetable oils such as soybean, canola, and flax seed oil, fish oils, fatty fish, with smaller amounts in meats and eggs.
Saturated and *trans* fatty acids, and cholesterol	No required role for these nutrients other than as energy sources was identified; the body can synthesize its needs for saturated fatty acids and cholesterol from other sources.	Saturated fatty acids are present in animal fats (meat fats and butter fat), and coconut and palm kernel oils. Sources of cholesterol include liver, eggs, and foods that contain eggs such as cheesecake and custard pies. Sources of *trans* fatty acids include sticking margarines and foods containing hydrogenated or partially hydrogenated vegetable shortenings.
Protein and amino acids	Serves as the major structural component of all cells in the body, and functions as enzymes, in membranes, as transport carriers, and as some hormones. During digestions and absorption dietary proteins are broken down to amino acids, which become the building blocks of these structural compounds. Nine of the amino acids must be provided in the diet; these are termed indispensable amino acids. The body can make the other amino acids needed to synthesize structures from other amino acids.	Proteins from animal sources, such as meat, poultry, fish, eggs, milk, cheese, and yogurt, provide all nine indispensable amino acids in adequate amounts, and for this reason are considered “complete proteins”. Proteins from plants, legumes, grains, nuts, seeds, and vegetables tend to be deficient in one or more of the indispensable amino acids and are called “incomplete proteins”. Vegan diets adequate in

	total protein content can be “complete” by combining sources of incomplete proteins which lack different indispensable amino acids.

Source: Dietary Reference Intakes for Energy, Carbohydrates, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids (2002/2005). This report may be accessed via www.nap.edu. Adapted from IOM.[10]

With the increasing trend for widespread fortification of the food supply, issues related to potential intakes approaching the UL values need to be monitored.[14] In some cases, the consumption of fortified foods can be passive and in some cases intentional. In both cases, non-traditional sources of vitamins and elements will alter total intake. This general issue came to light when it was reported that long-term intake of a diet high in retinol, independent of dietary supplements, was associated with the risk of osteoporotic hip fractures in women.[15] Recent concern has been raised about the potential for high intakes of folate and risk of developing colon cancer.[16] Better nationwide monitoring tools, especially for children, are necessary.

DIETARY GUIDELINES FOR AMERICANS

In addition to the DRIs, the U.S. Departments of Health and Human Services and Agriculture jointly issue Dietary Guidelines for Americans.[17] “Key Recommendations” are provided to summarize and focus the information in terms of actionable items. The U.S. Dietary Guidelines are reviewed and revised on the basis of the most recent scientific evidence every five years.

The intent of the Dietary Guidelines is to summarize and synthesize current knowledge in the area of nutrition with the intent of providing straight forward food based recommendations that promote health and decrease chronic disease risk. The Dietary Guidelines are intended for the entire U.S. population. The current recommendations are summarized in Table 11-6.

PRACTICAL ASPECTS OF ADEQUATE DIETARY INTAKE IN CHILDREN AND ADOLESCENTS

The DRI values are a helpful tool in evaluating whether individuals or groups are taking in adequate levels of nutrients to ensure appropriate growth and development throughout childhood and adolescence. An understanding of DRI values and the Dietary Guidelines can help pediatricians and other pediatric health professionals guide families in developing appropriate dietary patterns. Unfortunately, this may not be enough as parents, caregivers, and children are making dietary choices in a challenging food environment where some types of foods are highly promoted and advertised while other foods more consistent with the guidance provided by the Dietary Guidelines are not. Additional concerns include the cost of more nutrient dense foods relative to less dense foods,[18] the availability of foods in schools and after school programs, and difficulty with portion sizes and packaging which are not designed to meet dietary requirements of children.

The DRI values and Dietary Guidelines were designed in part to recognize issues with regard to growth and development as well as the contribution of diet to the development of chronic disease risk. This relationship is increasingly true in the pediatric age range where the prevalence of obesity is increasing along with the more frequent development of type 2 diabetes and other risk factors for future cardiovascular disease.[19] Thus, the practical implementation of developing dietary patterns in children that will meet the age appropriate DRI intake levels without exceeding energy requirements is critical. There are age-specific issues regarding the implementation of DRIs that must be considered.

Newborns

The American Academy of Pediatrics has recommended breastfeeding as the best strategy for feeding newborns. Human breast milk is the reference against which other forms of infant feedings must be judged.[20] Human milk is relatively high in saturated fat and cholesterol and is low in sodium. The American Academy of Pediatrics has encouraged breastfeeding exclusively, if possible, for the first 4 to 6 months of life.[21] Human milk appears to be uniquely tailored to meet nutritional needs over the first 6 months of life. The composition of human milk changes over time to meet these needs.[20] Babies appear to digest human milk more easily than formula[22] and babies who were breast fed appear to adapt more readily to new foods based on the diet of the mother when solids are introduced.[23] Nevertheless, some mothers find exclusive breastfeeding for

the first 6 months difficult. For these mothers, commercial formula or a combination of breast milk and the gradual introduction of solid foods provides an appropriate alternative.

Age 1–6 Years

This age range is when children adopt eating patterns that are more similar to adults. The transition period toward solid foods provides a unique and important opportunity for parents to develop healthful eating patterns in their children. However, in the later part of this period, it is also a time when children begin to be influenced by other aspects of the food environment such as eating patterns in peers, food advertising, and the availability of foods at home. Food consumption data indicate that infants and young children are exposed to a wide variety of foods that are high in saturated fat and added sugars.[24] These foods and beverages include fruit juice, sugar-sweetened drinks, snack foods, and French fries. For children 1–4 years of age, current intakes may exceed calorie and other requirements by 30% or more.[25] The excess calorie intake is especially true for children who are sedentary.

TABLE 11-6 US Dietary Guidelines for Americans, 2010—Key Recommendations

Balancing Calories to Manage Weight

- Prevent and/or reduce overweight and obesity through improved eating and physical activity behaviors.
- Control total calorie intake to manage body weight. For people who are overweight or obese, this will mean consuming fewer calories from foods and beverages.
- Increase physical activity and reduce time spent in sedentary behaviors.
- Maintain appropriate calorie balance during each stage of life—childhood, adolescence, adulthood, pregnancy and breastfeeding, and older age.

Foods and Food Components to Reduce

- Reduce daily sodium intake to less than 2300 mg and further reduce intake to 1500 mg among persons who are 51 and older and those of any age who are African American or have hypertension, diabetes, or chronic kidney disease. The 1500 mg recommendation applies to about half of the U.S. population, including children, and the majority of adults.
- Consume less than 10% of calories from saturated fatty acids by

replacing them with monounsaturated and polyunsaturated fatty acids.
- Consume less than 300 mg per day of dietary cholesterol.
- Keep *trans* fatty acid consumption as low as possible by limiting foods that contain synthetic sources of *trans* fats, such as partially hydrogenated oils, and by limiting other solid fats.
- Reduce the intake of calories from solid fats and added sugars.
- Limit the consumption of foods that contain refined grains, especially refined grain foods that contain solid fats, added sugars, and sodium.
- If alcohol is consumed, it should be consumed in moderation—up to one drink per day for women and two drinks per day for men—and only by adults of legal drinking age.

Foods and Nutrients to Increase

Individuals should meet the following recommendations as part of a healthy eating pattern while staying within their calorie needs.

- Increase vegetable and fruit intake.
- Eat a variety of vegetables, especially dark-green and red and orange vegetables and beans and peas
- Consume at least half of all grains as whole grains. Increase whole-grain intake by replacing refined grains with whole grains.
- Increase intake of fat-free or low-fat milk and milk products, such as milk, yogurt, cheese, or fortified soy beverages.6
- Choose a variety of protein foods, which include seafood, lean meat and poultry, eggs, beans and peas, soy products, and unsalted nuts and seeds.
- Increase the amount and variety of seafood consumed by choosing seafood in place of some meat and poultry.
- Replace protein foods that are higher in solid fats with choices that are lower in solid fats and calories and/or are sources of oils.
- Use oils to replace solid fats where possible.
- Choose foods that provide more potassium, dietary fiber, calcium, and vitamin D, which are nutrients of concern in American diets. These foods include vegetables, fruits, whole grains, and milk and milk products.

Building Healthy Eating Patterns

- Select an eating pattern that meets nutrient needs over time at an appropriate calorie level.
- Account for all foods and beverages consumed and assess how they fit within a total healthy eating pattern.
- Follow food safety recommendations when preparing and eating foods to reduce the risk of foodborne illnesses.

It is important that parents introduce more healthful foods during this age period. This includes the introduction of fruits and vegetables, whole grain products, fish and after age 2 years, low-fat dairy products. Parents should avoid introducing foods with lower nutritional value and should recognize that new foods may need to be introduced repeatedly before their ultimate acceptance by young children. This may require as many as 10 repetitions to establish acceptance and taste preference.[26,27] During this period of time, as with older children, parents should be mindful of their attitudes and behavior toward food and eating because in many cases they will be reflected in their children.[28,29]

Parents should use this time to establish healthy eating patterns and as much as possible should choose and maintain on a regular basis consistent time for meals. Self-regulation of the amount of food a child consumes serves to allow them to develop appropriate cues for satiety, a behavior that should be encouraged.[30] Parents can help to manage this by providing appropriate portion sizes for the age of the child. Likewise, it is important for parents to recognize that the amount of food eaten at any one time will vary from meal to meal and day to day, and to allow their children to adapt to internal clues. In addition, parents should to aware of and, when appropriate, limit snacking and other food intake that may be related to boredom or other cues rather than hunger.[28] It is important to establish healthy eating patterns early in young children.

Age 7–12 Years

During this age range eating outside the home, at school, aftercare programs, friends‘ houses, and quick-serve establishments, becomes a greater part of the daily pattern. Schools and other venues have become an increasingly difficult part of the food environment for children due to the availability of competitive foods and drinks which are high in calories and have low nutrient diversity. Fortunately, there are strong and consistent efforts to improve school menus, especially those available to children.

Although establishing good habits for food choices prior to the onset of school can be challenging, maintaining these habits as children get older and more independent is ever more complex due to peer pressure, advertising and other factors. Other issues during this age range include the introduction of fad diets.

Parents must serve as positive role models and continue to equip children and early adolescents with strategies to maintain healthful dietary

patterns. Pediatricians and other pediatric health professionals can also help by emphasizing these issues as part of routine health maintenance. Parents and health professionals can also be important advocates for children and for appropriate diet at their schools and other organized programs (e.g., sports teams and after school activities). Parents can work with those involved in providing meals and snacks to assess and improve the food environment. They can also encourage school systems to include more age-appropriate educational programs regarding food and nutrition for children. The objective should be to make more healthful foods and drinks available and to adopt local marketing strategies to encourage students to make more appropriate food choices.

Adolescents

Adolescence may be a particularly vulnerable period with regard to food choices. Because of accelerated growth and maturation, nutritional needs change during adolescence. The increased caloric need may stimulate appetite. These factors combined with a difficult food environment and peer factors may lead to less healthful choices.

In this age range, food choices and purchases are increasingly made outside of parental control. Current data suggest that adolescents have increased intake of fast food including pizza, hamburgers, and French fries, as well as increased consumption of sugar-sweetened beverages.[31] Of primary concern these foods and beverages can lead to excessive energy intake and weight gain. In addition, they tend to displace the recommended fruits, vegetables, low fat dairy, whole grains, lean meats, and fish. This dietary pattern leads to excess intake of saturated fat, *trans* fatty acids, sugar and insufficient intake of some micronutrients such as calcium, iron, zinc and potassium. In addition, intake of vitamins A, D, C and folic acid may be low.[31,32]

Parents and pediatric health professionals can have an important impact on the dietary intake of adolescents. Interactions with adolescents must take into account their increasing independence and must recognize that they will ultimately be making their food choices with less and less outside influence. Recommendations and counseling must be individualized to account for different cultural, household, and lifestyle situations. In addition, the food environment at home and the role-modeling of parents' choices is critical. The family remains an important influence for many adolescents. Pediatric health professionals should also discuss the potential impact of food marketing and provide behavioral tools to minimize the influence of marketing on food choices of

adolescents.

It is clear that many factors influence the food selection of children and their families. These issues must be recognized and considered in attempts to get children and adolescents to make appropriate food choices which then result in meeting the DRIs for energy, macronutrients, and micronutrients. Better strategies need to be developed to accomplish this goal.

CONCLUSIONS

The DRIs are a set of recommendations intended to provide guidance in evaluating nutrient intakes and planning meals on the basis of nutrient adequacy. They are nutrient, not food based recommendations. The emphasis in the development of the most recent DRI values was on reducing chronic disease risk. Depending on the available data for each nutrient, their DRIs are comprised of the EAR and RDA or when inadequate data are available AI and UL. In the case of macronutrients, there is an additional category of recommendations, AMDR. Within the category of infants to adolescents, DRI values have been established for the following age categories; 0 through 6 months, 7 through 12 months, 1 through 3 years, 4 through 8 years, 9 through 13 years, and 14 through 18 years. When assessing the available data the following were taken into consideration: strength of association (usually expressed as relative risk), dose–response relationship, temporally correct association (with exposure preceding the onset of disease), consistency of association, specificity of association, and biological plausibility. For full-term infants born to healthy mothers, it is expected that all nutrient needs will be met with exclusive breastfeeding for the first 4 to 6 months of life. For the second 6 months of life, the expectation is that nutrient needs will be met by a transition from breast milk to a mixed diet which includes solid foods. As children get older, the assumption was made that an increasing amount of food will be consumed outside the home, and as the child becomes more independent, that they will be determined more and more of these food choices. Inherent in the ability of children and adults to meet their nutrient requirements is the need to make wise food choices. It is important for parents, health care professionals, daycare providers and schools to educate children in the area of food and nutrition. This includes making healthy food choices available and encouraging children to take advantage of these options. In addition, it is important for adults to serve as positive role models for children in terms of behavior and attitudes about food and

nut.

REFERENCES

1. IOM. *Dietary Reference Intakes. Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein and Amino Acids.* Washington, DC: National Academy of Sciences; 2002.
2. IOM. *Dietary Reference Intakes. Calcium, Phosphorus, Magnesium, Vitamin D and Fluoride.* Washington, DC: National Academy of Sciences; 1997.
3. IOM. *Dietary Reference Intakes. Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin and Choline.* Washington, DC: National Academy of Sciences; 1998.
4. IOM. *Dietary Reference Intakes. Vitamin C, Vitamin E, Selenium and Carotenoids.* Washington, DC: National Academy of Sciences; 2000.
5. IOM. *Dietary Reference Intakes. Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium and Zinc.* Washington, DC: National Academy of Sciences; 2001.
6. IOM. *Dietary Reference Intakes. Water, Potassium, Sodium, Chloride and Sulfate.* Washington, DC: National Academy of Sciences; 2005.
7. IOM. *How Should the Recommended Dietary Allowances Be Revised?* Washington, DC: National Academy of Sciences; 1994.
8. Chung M, Balk EM, Brendel M, Ip S, Lau J, Lee J, Lichtenstein AH, Patel K, Raman G, Tatsioni A, Terasawa T, Trikalinos TA. Vitamin D and calcium: Systematic review of health outcomes. Evidence Report/Technology Assessment No. 183. (Prepared by Tufts Evidence-based Practice Center under Contract No. 290-2007-10055-I). AHRQ Publication No. 09-E015, Rockville, MD: Agency for Healthcare Research and Quality. August 2009.
9. IOM. *Dietary Reference Intakes for Calcium and Vitamin D. http://www.iom.edu/Reports/2010/Dietary-Reference-Intakes-for-Calcium-and-Vitamin-D.aspx.* 2010 (accessed May 15, 2014).
10. IOM. *Dieary Reference Intakes. The Essentail Guide to Nutrient Requirements.* Washington, DC: National Academy of Sciences; 2006.
11. RDA. *Dietary Reference Intake Reports*. http://www.iom.edu/?id=21377 (accessed May 15, 2014).
12. Lichtenstein AH, Appel LJ, Brands M, et al. *Diet and lifestyle recommendations revision 2006: a scientific statement from the American Heart Association Nutrition Committee. Circulation. 2006;114:82–96.*
13. Nielsen SJ, *Popkin BM. Changes in beverage intake between 1977 and 2001. Am J Prev Med*. 2004;27:205–210 [erratum appears in *Am J Prev Med*. 2005 May;28(4):413].
14. Lichtenstein AH, Russell RM. Essential nutrients: food or supplements? Where should the emphasis be? *JAMA*. 2005;294:351–358.
15. Feskanich D, Singh V, Willett WC, Colditz GA. Vitamin A intake and hip fractures among postmenopausal women. *JAMA*. 2002;287:47–54.
16. Mason JB. Folate consumption and cancer risk: a confirmation and some reassurance, but we're not out of the woods quite yet. *Am J Clin Nutr.* 2011;94:965–966.
17. Dietary Guidelines for Americans. http://www.cnpp.usda.gov/DGAs2010-DGACReport.htm. 2010 (accessed May 15, 2014).
18. Andrieu E, Darmon N, Drewnowski A. Low-cost diets: more energy, fewer

nutrients. *Eur J Clin Nutr*. 2006;60:434–436.
19. IOM. *Preventing Childhood Obesity: Health in the Balance.* 2005, http://www.iom.edu/reports/2004/preventing-childhood-obesity-health-in-the-balance.aspx (accessed May 15, 2014).
20. Gartner LM, Morton J, Lawrence RA, et al; American Academy of Pediatrics Section on B. Breastfeeding and the use of human milk. *Pediatrics*. 2005;115:496–506.
21. Kramer MS, Kakuma R. The optimal duration of exclusive breastfeeding: a systematic review. *Adv Exp Med Biol.* 2004;554:63–77.
22. Hamosh M. The milky way: from mammary gland to milk to newborn–Macy-Gyorgy Award presentation (1999). *Adv Exp Med Biol*. 2002;503:17–25.
23. Mennella JA, Griffin CE, Beauchamp GK. Flavor programming during infancy. *Pediatrics.* 2004;113:840–845.
24. Fox MK, Pac S, Devaney B, Jankowski L. *Feeding infants and toddlers study: what foods are infants and toddlers eating? J Am Diet Assoc.* 2004;104:s22–s30.
25. Devaney B, Ziegler P, Pac S, et al. Nutrient intakes of infants and toddlers. *J Am Diet Assoc.* 2004;104:s14–s21.
26. Addessi E, Galloway AT, Visalberghi E, Birch LL. *Specific social influences on the acceptance of novel foods in 2–5-year-old children. Appetite.* 2005;45:264–271.
27. Sullivan SA, Birch LL. Infant dietary experience and acceptance of solid foods. *Pediatrics.* 1994;93:271–277.
28. Francis LA, Birch LL. *Does eating during television viewing affect preschool children's intake? J Am Diet Assoc.* 2006;106:598–600.
29. Faith MS, Scanlon KS, Birch LL, et al. Parent–child feeding strategies and their relationships to child eating and weight status. *Obes Res.* 2004;12:1711–1722.
30. Birch LL, Fisher JO. Development of eating behaviors among children and adolescents. *Pediatrics*. 1998;101:539–549.
31. Putnam J, Allshouse J, Kantor LS. U.S. per capita food supply trends: more calories, refined carbohydrates, and fats. *Food Rev.* 2002;25:2–15.
32. Bowman SA, Gortmaker SL, Ebbeling CB, et al. Effects of fast-food consumption on energy intake and diet quality among children in a national household survey. [see comment]. *pediatrics.* 2004;113:112–118.

12

Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals

Mandana Amir Shaghaghi, MSc, RD, Peter J.H. Jones, PhD, and Philip M. Sherman, MD, FRCPC

PROBIOTICS AND PREBIOTICS

Introduction

The intestinal **epithelial surface** comes into contact with a continuous flux of a vast variety of antigens. The primary function of the intestinal mucosa is the absorption of nutrients. This function necessitates the transport of molecules across a single layer of epithelial cells (enterocytes in the small bowel and colonocytes in the large intestine) and distinguishes the lining of the gut mucosa functionally from that of the respiratory or genitourinary

tracts.

Epithelial cells lining the surface of the body form a protective physical barrier against the external environment. The intestinal epithelium, however, must simultaneously be able to selectively absorb nutrients. Therefore, the epithelium in the alimentary tract (with the exception of the oral cavity, esophagus, and rectum) consists of a single layer of cells. The majority of the cells in the epithelial monolayer consist of polarized absorptive cells referred to as enterocytes. The apical surface of enterocytes is covered with microvilli, with the lipid bilayer of the microvillus membrane and an overlying glycocalyx formed by membrane-bound glycoproteins constituting a significant barrier to antigen entry. Paracellular antigen penetration is prevented by apical junctional complexes (including intercellular tight junctions) creating a physical barrier between enterocytes. The permeability of tight junctions can be modified by cytokines, such as tumor necrosis factor (TNF)-α and transforming growth factor (TGF)-β, and by substances elaborated by microbial pathogens.

The intestinal **immune system** must be able to recognize potential pathogens and inhibit their adherence and invasion by protective immune exclusion or inflammatory responsiveness while still allowing for the uptake and transport of dietary nutrients and certain intact macromolecules, such as growth factors and maternal immune globulins present in breast milk, without inappropriate and detrimental immune reactions.[1] The intestinal immune system has unique structural and functional characteristics which have evolved to fulfill the dual task of immune defense and tolerance (Table 12-1).

TABLE 12-1 Components of the Intestinal Mucosal Barrier

1. External Barrier
   - Gastric `acid
   - Digestive enzymes
   - Non-specific antimicrobial factors (e.g. lysozyme, lactoferrin, peroxidase, defensins)
   - Secretory IgA
   - Intestinal mucus
   - Peristalsis
2. Epithelial Layer
   - Microvilli
   - Plasma membrane
   - Tight junctions

- Innate immune receptors
- Intraepithelial immune cells

3. Intestinal Lymphoid Tissue
   - Uptake and presentation of antigens
   - Induction of secretory IgA production
   - Local and systemic inflammatory responses
   - Maintenance of tolerance
4. Indigenous Intestinal Microbiota
   - Degradation of luminal antigens
   - Inhibition of colonization and adherence of pathogens
   - Stimulation of the intestinal immune system

The indigenous **intestinal microbiota** is established during the first years of life, with commensal bacteria having a profound impact on the structural and functional maturation of the gut.[2] Indeed, the intestinal microbiota is considered an essential component of the intestinal immune system (Figure 12-1). Intestinal epithelial cells express a number of pattern recognition receptors (PRRs), which recognize microorganism-associated molecular patterns (MAMPs). MAMPs, present on both commensal and pathogenic microbes, are required for their survival and, therefore, are evolutionarily highly conserved.[3] These conserved microbial structures include unmethylated CpG motifs characteristic of bacterial DNA, virus-derived single-stranded RNA, bacterial cell wall compounds such as lipoteichoic acid, peptidoglycan, and lipopolysaccharide (LPS, present on Gram-negative bacteria) and bacterial flagellin.[4] Intestinal PRRs and their ligands are summarized in Table 12-2.

Normally, the human intestinal tract harbors an enormous quantity and diversity of indigenous microbes. The number of prokaryotic cells in the intestine is ~10^{14}, the vast majority of which are found in the lumen and outer mucus layers of the colon. In health, the microorganisms are not adherent to the inner mucus layer or to surface epithelial cells,[5] but intimately adherent organisms are seen with the presence of pathogenic infections (such as enteropathogenic *Escherichia coli*) and adherent commensal organisms in active Crohn's disease involving the large intestine.

With the emergence of culture-independent molecular identification techniques, the number of microbial species constituting the healthy gut microbiota is between 500 and 1000.[6] Colonization of a sterile intestinal tract commences at birth and proceeds in a step-wise and systematic fashion. Maternal intestinal and vaginal microbes provide the first bacterial

inoculum to the newborn. Consequently, the microbiota composition of infants born by cesarean section differs significantly from that of vaginally born infants.[7] Early nutrition also has a profound impact on the composition of the infant gut microbiota. In particular, breastfeeding promotes the growth of bifidobacteria and lactobacilli, which become the predominant microbiota species in breast-fed infants. After weaning, and by about 1–2 years of age, the gut microbiome composition comes to resemble that seen in older children and adults.[8]

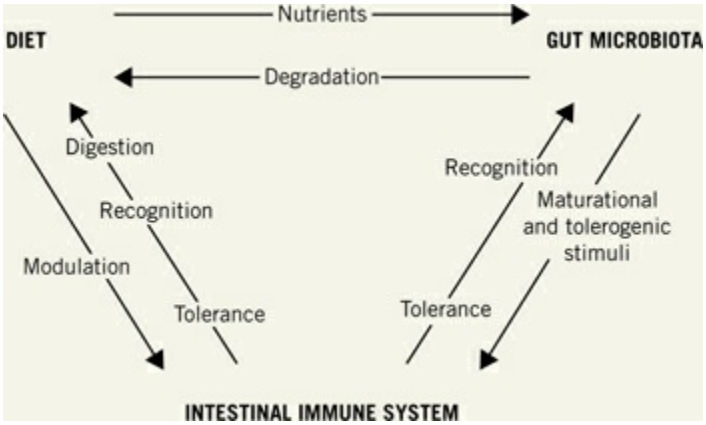


Figure 12-1 Potential for interactions between the indigenous intestinal microbiota, and the intestinal immune system.

TABLE 12-2 Intestinal Pattern Recognition Receptors and Their Known Ligands

Receptor	Ligand
TLR1	Lipopeptides
TLR2	Lipopeptides and lipoteichoic acid
TLR3	Viral dsRNA and viral sense ssRNA
TLR4	Lipopolysaccharide
TLR5	Bacterial flagellin
TLR6	Lipoteichoic acid
TLR8	Viral antisense ssRNA
TLR9	CpG DNA
TLR11 (mice, not humans)	Bacterial flagellin
NLR1 (Nod1)	Diaminopimelic acid of bacterial peptidoglycan
NLR2 (Nod2)	Muramyl dipeptide of bacterial peptidoglycan

Abbreviations: CpGDNA, unmethylated cytosine-guanine rich deoxyribonucleic acid; dsRNA, double-

stranded ribonucleic acid; NOD, nucleotide-binding oligomerization domain; ssRNA, single-stranded ribonucleic acid; TLR, toll-like receptor.

The composition of the diet appears to impact the composition and functions of the gut microbiome. For instance, there are differences described in the fecal microbial composition of children living in Florence, Italy and those growing up in a rural village in Burkino Faso.[9] Gut microbial communities of herbivores differs from carniverous mammals, with adult humans having an intestinal microbial composition that is characteristic of omniverous primates.[10] In murine studies, feeding of high saturated fats drive changes in the composition of the intestinal microbiome which impacts on disease susceptibility, using the development of colitis in genetically susceptible (IL-10 gene knockout) mice as the measured outcome variable.[11]

Intestinal microbes are necessary for normal morphological and immunological maturation of the gut.[12] For instance, germ-free mice exhibit impaired sIgA production. Moreover, specific strains of the intestinal microbiota modulate the expression of host genes involved in a variety of intestinal functions, including nutrient absorption, metabolism, mucosal barrier function, and intestinal maturation. Defective immune maturation in germ-free mice can be corrected by colonization with *Bacteroides fragilis*, a member of murine indigenous intestinal microbiota.[13]

Indigenous microbes contribute to intestinal barrier function by degrading intraluminal antigens and by inhibiting the adherence and subsequent internalization of pathogenic microbes. Antigen transport across the intestinal epithelium is increased in the absence of intestinal microbes in experimental animals.

The host is tolerant to its indigenous gut microbiota.[14] Abrogation of oral tolerance to dietary antigens results in food allergy, whereas inappropriate inflammatory immune response against the gut microbiota is described in chronic inflammatory bowel disease (IBD). Several studies indicate that oral tolerance cannot be induced in germ-free animals. Reconstitution of the intestinal microbiota, with bifidobacteria, in such animals restores the ability to establish oral tolerance in the neonatal period, but interestingly not at a later age.

The importance of enteral nutrition and antigenic stimulation to both intestinal microbiota and gut immunophysiology is demonstrated by studies showing that total parenteral nutrition results in a reduction of both aerobic and anaerobic gut bacteria and decreased intestinal concentrations

of polymeric IgA and anti-inflammatory cytokines, such as interleukin (IL)-4 and IL-10. Studies in both mice and humans indicate that three way cross-talk between the gut luminal microbiota, intestinal epithelial cells, and the adaptive immune system (in particular, B cells and the production of sIgA) affects nutritional function as measured by dietary fat absorption.[15]

In a variety of disease states (necrotizing enterocolitis (NEC), irritable bowel syndrome (IBS), IBDs, celiac disease, obesity, and diabetes), there is a loss of the diversity of the gut microbiome. The studies to date in humans are associative in nature, but experimental models indicate the potential for a cause-and-effect relationship.[16]

Probiotics are live micro-organisms, which when administered in adequate amounts confer a health benefit to the host. Probiotics taken orally transiently alter the composition of the gut microbiota, especially in settings with reduced diversity.[17] They are marketed in both single-bacteria and multiple-bacteria preparations, with some of the more common strains being *Lactobacillus casei*, *Lactobacillus acidophilus*, and *Bifidobacterium bifidum.* In older children and adults, intestinal colonization of probiotics is transient unless taken on a regular basis, because shedding of the orally ingested microbe in the feces stops soon after cessation of the supplementation.

In children, multiple meta-analyses of randomized controlled trials report that a variety of probiotic strains taken by pre-schoolers living in the developed world shortens the duration of acute infectious diarrhea by about 1 day and reduces the duration of virus shedding in rotavirus-infected children. Systematic reviews also demonstrate that probiotics can reduce the frequency of *Clostridium difficile*- and antibiotic-associated diarrhea in a strain- and dose-dependent fashion both in children and in adults.[18] There is also evidence that probiotics reduce dietary lactose intolerance when live bacterial cultures are present in fermented dairy products, resulting in decreased abdominal cramping and bloating as well as a decrease in concentrations of exhaled hydrogen. Probiotics have also been reviewed for their roles in immune modulation, lowering cholesterol levels, reducing eczema and atopy in high risk infants, eradication of *Helicobacter pylori* infection, and maintaining chronic IBDs in remission.[19] Data on the beneficial effects of probiotics with regards to either upper or lower respiratory tract infections are more limited.

Probiotic effects include degradation of dietary antigens, promotion of mucosal barrier functions, inhibition of mucosal pathogen adherence and

interaction with the innate and adaptive immune systems of the host.[20] Alterations in the intestinal microbiota composition in infancy precede the development of atopic disease, suggesting a causal link between indigenous gut bacteria and the atopic phenotype. Some probiotic bacterial strains have immunological effects that could contribute to reducing the risk of allergic diseases. In experimental studies, probiotics suppress IgE responses, inhibit Th2 (allergy promoting) cytokine production and induce the production and activity of regulatory (anti-inflammatory) T cells.[21]

Supplementation with *Lactobacillus rhamnosus* GG during late pregnancy and lactation induces anti-inflammatory mucosal immune responses, as indicated by an increase in the concentration of TGF-β2 in breast milk, and reduces the frequency of atopic eczema for up to 5 years in at least some study populations.[22] *Lactobacillus* GG has shown promising potential in reducing the risk of atopic dermatitis in infants at high hereditary risk for atopic disease. In a double-blind, placebo-controlled clinical trial of 159 pregnant women *Lactobacillus* GG was administered 2–4 weeks prior to expected delivery and the probiotics subsequently administered to either the breastfeeding mother or the infant until the age of 6 months. Probiotic supplementation reduced the risk of atopic dermatitis in these high risk infants from 46% to 23% during the first two years of life and this protective effect has subsequently been reported, at least in some studies, to extend out to the age of 4 years.

The pathogenesis of NEC in premature newborns, albeit not yet completely clarified, involves increased gut permeability combined with enteral feeding in the setting of an altered intestinal microbiota characterized by reducing gut diversity.[23] Consequently, probiotics have been used in prospective randomized controlled trials. The findings suggest a reduced risk of developing NEC in premature infants entered into the probiotic arm of the study trials compared to the babies given placebo. The probiotics may act to reduce NEC by promoting gut barrier function, by inhibiting colonization of potentially pathogenic bacteria, or by controlling innate immune responses in immature gut epithelia.[24] Subsequently, multiple randomized, placebo-controlled trials using different probiotic combinations and the results of several meta-analyses have also described significantly reduced incidence of NEC compared to placebo.[25] The findings now have to be extended to other settings around the world and the active ingredient(s) mediating the biological effects should be identified and well characterized, since regulators in some parts of the world remain reluctant to provide high doses of viable microbes to

vulnerable premature babies.[26]

IBS is increasingly recognized to be a long-term complication of acute infectious enteritis. Reduced microbial diversity has been described in the fecal microbiota of both adults and children with IBS.[27] Visceral hyperalgesia is a feature of post-infectious IBS and treatment with selected probiotic strains reduces clinical symptoms in a dose-dependent manner. In a meta-analysis of studies undertaken in adults, the number needed to treat was just 4; equivalent to or even better than any other described therapies in IBS which have also been subjected to the rigors of a randomized controlled trial. What is needed now are head-to-head comparisons of various treatment options in well defined populations employing clearly define and widely accepted outcome parameters.[28]

The chronic IBDs, Crohn's disease and ulcerative colitis, are characterized by mucosal inflammation as a result of detrimental immune reactivity towards indigenous intestinal microbes. Thus, modulating either the composition of intestinal microbiota or mucosal immune responses by probiotics provides a means to either treat or maintain remission of IBD symptoms.[29] *Lactobacillus* GG enhances intestinal IgA production in patients with Crohn's disease. A probiotic *E. coli*, strain Nissle 1917 is effective in maintaining remission in patients with ulcerative colitis. The probiotic combination VSL#3 (containing *Lactobacillus casei*, *Lactobacillus plantarum*, *Lactobacillus. acidophilus*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Bifidobacterium longum*, *Bifidobacterium breve*, *Bifidobacterium infantis*, and *Streptococcus salivarius* subsp. *thermophilus*) induces remission in mild to moderate ulcerative colitis. The multi-strain probiotic mixture is also effective in preventing acute pouchitis, a non-specific inflammation of an ileal reservoir surgically fashioned following sub-total colectomy, and in reducing the risk of recurrence of chronic pouchitis. These beneficial effects might be the result of inducing tolerogenic immune responses by increasing levels of the anti-inflammatory cytokine IL-10.

Prebiotics are defined as non-digestible food ingredients (generally complex, non-digestible, but fermentable galacto-oligosaccharides (GOS) and fructo-oligosaccharides (FOS)) that beneficially affect the host by selectively stimulating the growth or activity of intestinal bacteria and thereby promote host health.[30] The commensal microbiome (in particular, Bacteroides species) contains carbohydrate-active enzymes that serve to extract energy from complex polysaccharides and provide short-chain fatty acids as substrate for colonocytes.[31] Prebiotics are preferentially utilized as

substrates by Bifidobacteria, that also generate butyrate, and by certain lactic acid-producing bacteria.

Breast milk contains substantial amounts (5–8 g/L) of GOS, a complex group of substances derived from lactose. These oligosaccharides share similarities with epithelial cell surface carbohydrates and adhesion molecules. They exert a number of important functions, from blocking the adhesion of bacteria to intestinal cell surfaces to being responsible for the predominance of the Bifidobacteria and lactic acid-producing bacteria (Lactobacilli) in the fecal microflora of the breast-fed infant.[32] The complex milk oligosaccharides are only minimally digested in the upper gastrointestinal tract and then undergo fermentation in the colon, thereby accounting for much of the hydrogen gas that is exhaled by breast-fed infants.

Breast milk provides the newborn infant with optimal nutrition for growth and development and immune protection during a critical period when neonatal immune defenses are still immature. Infectious disease is a major threat to the neonate and a leading cause of infant mortality in many areas of the world. It is well-established that breastfeeding protects the infant from infectious diseases. This protective effect is mediated by a number of passive antimicrobial factors, including lysozyme, lactoferrin, peroxidase, defensins, cathelicidin, sIgA, TLRs with soluble CD14 and oligosaccharides.

The protection afforded by breast milk is particularly crucial in developing countries, where breastfeeding reduces the need for infants to consume potentially contaminated water. In addition to passive immune protection, breast milk contains factors which contribute to immune maturation by stimulating the intestinal immune system either directly or via modulating the indigenous intestinal microbiota.33 The role of breast milk in reducing the risk of NEC in premature neonates is well-established. This effect may be mediated, at least in part by TGF-β, which is present in colostrum in large amounts. TGF-β attenuates intestinal epithelial cell inflammatory responses associated with the development of NEC. Increased gut permeability during the first days of life is associated with an increase in the risk of developing NEC. Breastfeeding improves the integrity of the intestinal barrier during the neonatal period, as assessed by the absorption of macromolecules in a rabbit model. TGF-β is known to enhance intestinal repair and promote barrier integrity.34.[34]

After weaning, the most common prebiotics include lactulose, inulin, FOS, and GOS. The effects of prebiotic oligosaccharides in promoting bifidobacteria in the intestinal microbiota of infants have been

demonstrated in a number of double-blind, placebo-controlled trials.[35] Prebiotics may also modulate micronutrient intake, as demonstrated by a study in which administration of an inulin-type fructan significantly increased calcium absorption and bone mineralization in adolescents.

Given an epidemiological link between intestinal microbiota composition and the development of atopic disease, prebiotics have been tested in reducing the risk of developing atopic disorders. In a randomized, placebo-controlled trial of 259 infants a hypoallergic formula supplemented with a GOS/FOS mixture reduced the frequency of atopic dermatitis in at-risk infants.[36]

Conclusions

Nutrients affect mucosal and systemic immune functions and the gut microbiota. General malnutrition or lack of certain nutrients may result in immunodeficiency predisposing to infectious disease. Indigenous intestinal microbes process dietary antigens and contribute to gut immune homeostasis by excluding pathogens and providing maturational and modulatory stimuli. The intestinal immune system constantly monitors intestinal antigens. Protective immune defenses are launched against potential pathogens whilst a number of active processes ensure tolerance towards dietary antigens and the microbiota.

Recent advances in understanding the mechanistic basis of these interactions provide the means to design specific interventions to restore intestinal homeostasis by supplementation of the diet with probiotics, prebiotics, immunomodulatory micronutrients and selected phytochemicals. Such novel dietary constituents are considered to promote overall health and are now being evaluated both as therapeutic and preventive modalities. The evidence required for changing current clinical practices will be defined by advances in basic research related to more a detailed understanding of underlying mechanisms and then applied to the clinical setting by coordinated and rigorously conducted clinical trials in a variety of settings including across the life-span continuum.

NUTRACEUTICALS WITH PEDIATRIC CONSIDERATIONS

Introduction

Nutraceuticals are foods or food products which provide health benefits and/or prevent chronic disease. Nutraceuticals range from isolated

nutrients and dietary supplements to herbal products. More individuals in developed countries than ever are now using dietary supplements, expecting to either enhance their overall health and well being or improve their physical and mental performance.

Supplement use is common across all population segments, including young children and adolescents. It is estimated that approximately half of children living in the United States receive some form of vitamin and mineral supplements.[37,38] There are a myriad of reasons for supplement use by children. Parents give dietary supplements to their children to enhance health, as a substitute for good nutrition, to be a good parent, because of personal positive experiences, or at the recommendation of alternative medicine health specialists.[39] Indeed, children whose parents use dietary supplements themselves are more likely to give them to their offspring.[40] In one birth cohort, nearly a third of participating two-year-old children receive nutrient supplements.[41] Many older children and adolescents also continue to obtain vitamin and minerals from dietary supplements.[42] Teenagers often use additional supplements that are touted as appearance, energy and athletic performance enhancers.

Many children also ingest dietary supplements with herbal and other non-nutrient ingredients either occasionally or on a regular basis. Use of both nutrient and herbal supplements is most common in children with long-term health problems.[43] Given insufficient safety data on the potential impact of these supplements on the developing fetus, they cannot be recommended for use by pregnant women. Supplement use has been studied specifically in children with chronic diseases, such as IBDs,[44] cancers,[45,46] and in some conditions, such as cystic fibrosis, that limit nutrient absorption from foods or increase requirements, which makes nutrient supplementation a critical component of treatment.[47]

Appropriately dosed dietary supplements are an important source of essential nutrients for some children and adolescents, ensuring adequate intakes for optimal growth and development. Dietary supplements that provide higher than adequate amounts of nutrients, or contain non-essential ingredients, on the other hand, can cause concern because of potential health risks and questionable benefits, particularly during pregnancy. Interactions of supplement ingredients with prescription or over-the-counter medications also must be considered. The growth of the industry and a lack of product regulation raise concerns for the health and safety of children. Because so many children use them, each visit to the pediatric health care provider should include a review of current

supplement use. A careful evaluation of individual dietary patterns and specific medical circumstances should then guide the selection of dietary supplements for use, if any are needed.

NON-PHYTOCHEMICAL SOURCES

Long-Chain *n*-3 Polyunsaturated Fatty Acids

Dietary changes are concerned at the recent increase in prevalence of chronic disease, such as obesity and type 2 diabetes. Indeed, increased intake of highly processed modern diets in developed countries has resulted in declining intake of a number of essential nutrients required for optimizing physical health and brain function in children. Included are decreases in the intake of two crucial long chain *n*-3 polyunsaturated fatty acids (PUFAs) eicosapentaenoic acid and decosahexaenoic acid (DHA) (derived mainly from oily fish), which serve critical roles in membrane fluidity, neurotransmission, neurogenesis, signal transduction, and ion channel activation and protection.[48] As a result, *n*-3 PUFA supplementation during infancy and childhood has received a great deal of interest for its potential protective effects on diseases ranging from allergy to learning and behavioral problems as well as for beneficial effects on optimal growth velocity, visual acuity and neurodevelopment. There may also be reduced rates of prematurity when PUFAs are taken as supplements during pregnancy.

Infants, including premature infants, have the ability to synthesize DHA from precursors, but the synthetic capacity appears insufficient to keep up with high demands required for growth. DHA is needed for incorporation into the cortical gray matter and the retina. The long-chain *n*-3 PUFA, DHA, is highly concentrated in gray brain matter (15%–20% of all lipids in the brain) and plays an essential role in normal brain structure and function.[49] In infants fed formulas devoid of DHA concentrations in the gray matter and retina are substantially lower than the levels observed in breast fed infants. The same is true for DHA concentrations present in plasma and erythrocyte membrane lipids.[50]

Infants fed DHA-free formulas show evidence of impaired retinal function during the early months of life.[50] However, this effect appears to be transient and may not be demonstrable in older infants. The presence of DHA in infant formula is considered desirable, because these fatty acids are present in breast milk and their absence from the diet produces low concentrations in the brain, retina, erythrocytes and plasma of human

infants.

There is evidence for beneficial effects of supplementation of *n*-3 PUFAs in subgroups of children with behavioral problems, learning difficulties, and developmental disorders.[51–53] In clinical studies, the protective effect of supplementation of *n*-3 PUFA on subsequent disease outcomes, including attention deficit hyperactivity disorder (ADHD) and autism spectrum disorders (ASD), has been assessed. Existing studies evaluating the effects of supplementation of *n*-3 PUFAs on children with ADHD have not shown conclusive results.[54] Moreover, to date there is no high quality evidence indicating that supplementation of omega-3 fatty acids improves core and associated symptoms in children with ASD including social interactions, communication and stereotypes.[55]

Fish oil supplementation during pregnancy exclusively[56–58] and postnatally alters cellular immune responses and function in the offspring, including a decrease in allergen-specific Th2 responses, an increase in polyclonal Th1 responses[57,58] and potential allergy-protection by modestly reducing the risk of sensitization in the infant.[56,57,59] Fish oil supplementation after 6 months of age, however, has not proven effective in reducing allergies later during childhood.[60,61]

Supplementation of *n*-3 PUFA also appears to have beneficial effects for primary and secondary prevention of cardiovascular disease. Effects of *n*-3 PUFA on cardiovascular health results from a multifaceted synergistic process involving anti-inflammation, reduction of very-low-density lipoprotein, reduction of triglycerides (TG), regulation of transcription factors and gene expression and both anti-arrhythmic and anti-thrombotic effects.[62]

Supplementation of DHA in children with hyperlipidemia is associated with restoration of brachial artery endothelial function, a decrease in serum TG level and a shift in lipoprotein subclasses towards large, buoyant low-density lipoprotein particles.[63–65]

PHYTOCHEMICAL SOURCES

Phytosterols

Plant sterols and stanols (collectively referred to as phytosterols) represent a cluster of vegetable sterols which are chemically analogous to cholesterol. This cluster includes a series of structurally specific compounds, including sitosterol, campesterol, and stigmasterol as the major plant sterols. Plant stanols, which are a hydrogenated form of plant

sterols, are found much less abundantly than plant sterols. Plant sterols and stanols occur naturally in the diet at various levels, generally below 450 mg/dL, although both vegetarian diets and our ancestors' food intake patterns suggest that intakes were considerable higher in less industrialized lifestyles. In general, only 5%–10% of the ingested major plant sterols are absorbed, while cholesterol absorption can exceed 50%.

For over 40 years, phytosterols have been known to have cholesterol lowering properties. Along with rising interest in functional foods and nutraceuticals during the last two decades, phytosterols have gained much attention and are increasingly being used for the prevention and management of hypercholesterolemia. Numerous studies have examined the cholesterol lowering effects and mechanism of actions of phytosterols. As a result, scientific and regulatory bodies[66–68] support the efficacy of phytosterols and recommend a typical intake of 2 g/d of phytosterols to optimize blood lipid concentrations in adult populations. Cholesterol lowering efficacy of phytosterols has also been examined in children with familial hypercholesterolemia.[69,70] However, the numbers of studies evaluating children still are quite limited.

Increases in serum plant sterol concentrations (from 19 to 30 μM) are described in people taking plant sterols either as functional foods or supplements.[71] High serum phytosterols concentrations are also reported in children consuming diets high in phytosterols.[72] Considering the possible atherogenic effect of phytosterols, concerns have been raised with regards to the safety of increased serum phytosterols concentrations from the high intake of phytosterols in functional foods and supplements.

Herbal Supplements

Herbal products exist as both crude (roots, leaves, seeds, or teas) and commercial (tablets or capsules) preparations. While crude herbal products are more often ingested in less developed countries, commercial herbal products are more commonly used in developed countries.[73] Children and adolescents are increasingly using herbal supplements, often reflecting the use patterns of their parents and other family members. Very little, if any, information is available about age-specific pharmacodynamics, clinical benefits, or potential risks.

Echinacea (*Echinacea purpurea*)

Fresh or dried root of *Echinacea purpurea* and its preparations contain caffeic acid derivatives, alkamides, polyacetylene derivatives,

polysaccharides, glycoproteins, and essential oils. The German Commission E has approved Echinacea for the treatment of symptoms of upper respiratory infections associated with colds.

Ephedra/Ma huang (*Ephedra sinica*)

Dried, young branchlets, harvested in the fall, of *Ephedra* sinica or equivalent *Ephedra* species contains alkaloids (ephedrine and pseudoephedrine). In the United States, ephedra is regulated under the Dietary Supplement Health and Education Act (DSHEA) in aqueous infusion, alcoholic tincture, and dry extract available as either capsules or tablets. The alkaloids of ephedra (ephedrine and pseudoephedrine) are approved by the Food and Drug Administration (FDA) as over-the-counter ingredients in remedies used for managing the common cold, flu and allergies.

The use of ephedra in products for weight loss, athletic performance enhancement, and central nervous system stimulation has raised concerns, with questions about the safety of these products. As a result, its use in dietary supplements is now banned. The FDA has received reports of 80 deaths and 1400 adverse effects associated with ephedra in dietary supplements; ephedra has been linked to heart attack, stroke, seizures, and psychosis.

St. John's Wort (*Hypericum perforatum*)

Dried parts above ground are gathered during the flowering season of the *H. perforatum*. The German Commission E approved the use of St. John's Wort internally for the treatment of psycho-vegetative disturbances, depressive moods, anxiety, and nervous unrest. Oily preparations are approved for external use in the treatment of acute and contused injuries, myalgia and first-degree burns.

Ginkgo Biloba (*Ginkgo biloba*)

Dry extract, which is pharmaceutically prepared (in a 35–67:1 ratio of dried leaves to final extract), contains flavonone glycosides (quercetin and kaempferol), terpene lactones (ginkgolides A, B and C), and ginkgolic acids. Ginkgo Biloba is approved by the German Commission E for symptomatic treatment of disturbed performance in organic brain syndrome within a therapeutic regimen, improvement in pain free walking in peripheral arterial occlusive disease, vertigo and tinnitus.

Garlic (*Allium sativum*)

Fresh or dried compound bulbs of *Allium sativum* with no less than 0.5% alliin have active compounds that include thiosulfinates (allicin). In clinical trials, garlic has been effective in lowering cholesterol in patients with hyperlipidemia and in treating hypertension. Garlic has antibacterial, antimycotic, lipid-lowering, inhibiting of platelet aggregation, prolonging of bleeding and clotting time, tumor inhibition and fibrinolytic activity enhancing properties. Commission E approved the use of garlic as a support to dietary measures for lowering elevated levels of blood lipids and in the prevention of age-dependent vascular changes. However, there are only limited results related to the use of garlic for pediatric patients with hyperlipidemia.

Chamomile (*Chamomilla recutita*)

Fresh or dried flower-head of *Matricaria recutita* is a popular ingredient in herbal teas. It is prepared for topical use in rinses, creams, ointments, and vapor baths. In Germany, chamomile is widely used in caring for sensitive skin in infants and children and for inflammatory skin conditions. The Commission E has approved chamomile for intestinal spasms and inflammatory diseases of the gut.

Ginger (*Zingiber officinale*)

Peeled, fresh or dried rhizome of *Zingiber officinale* contains oleoresin, volatile oil, carbohydrates, lipids, vitamin A and niacin, minerals, and amino acids. Ginger is used alone and as a component in digestive, anti-nausea, and cold and flu dietary supplements in the United States. The Commission E has approved ginger for dyspepsia and the prevention of motion sickness.

Valerian Root (*Valeriana officinalis*)

Fresh underground plant parts or carefully dried parts of *V. officinalis* and its preparations in effective dosages contain essential oil with monoterpenes and sesquiterpenes. The Commission E approves the product for restlessness and sleeping disorders based on nervous conditions. Clinical data support its action as a mild sedative and sleep-promoting agent. However, the U.S. Pharmacopoeia does not support the use of valerian due to inadequate evidence provided by high quality clinical studies.

It should be noted that many herbal products used as food supplements can be associated with a spectrum of liver injury ranging from asymptomatic elevations in serum liver enzyme tests to fulminant liver

failure and death.[73] Since children are more susceptible for hepatotoxicity, special considerations are needed for herbal supplements in this population.

Other Plant-derived Bioactives

Phytoestrogens are not nutritionally important, but they have biologically active effects, exerting estrogenic as well as anti-estrogenic activities. Three types of dietary phytoestrogens include isoflavones, lignans, and coumestans. Isoflavones, the major class of dietary phytoestrogens, are abundant in soy products and have a similar structure and function to endogenous estrogen.[74] Lignans are present in flaxseeds, grains, nuts, fruits, vegetables, tea, and coffee. Coumestans contribute very minimally to dietary phytoestrogen intake.[75] Soy protein-based infant formula provides phytoestrogens that are not present in human breast milk. Phytoestrogens are readily absorbed by the infant gut, but produce no clinically recognizable effects. However, when followed into adulthood, young women who had been exposed to soy formula as infants have greater menstrual flow and experience more discomfort with menstruation.[76] These effects are biologically significant, because they demonstrate that even a relatively brief exposure during a susceptible period of the life-span have effects that can last for decades. Higher prepubertal phytoestrogen intake in girls is associated with later breast development and later pubertal onset.[74,77] Several experimental studies show that intake of soy and isoflavones can have protective effects against hormone-related cancers. Higher exposure to phytoestrogens (both isoflavones and lignans) during childhood and adolescence is associated with a reduced risk of breast cancer.[78] Isoflavone and lignan intake in adolescence may decline breast cancer risk through a number of biologic mechanisms including: decreasing the sensitivity of developing adolescent breast tissue and mammary glands to chemical carcinogens and decreases tumor incidence, promoting mammary cell differentiation through increasing mammary duct branching and decreasing the number of highly proliferative terminal end buds which are more susceptible to carcinogens in adulthood, influencing the expression of proteins in breast tissue that are involved in malignant tumor progression including estrogen and progesterone receptors, and eliminating carcinogenic free radicals through the anti-oxidant properties of isoflavones.[72]

Special Considerations in Pediatrics

Much of the knowledge about important aspects of dietary supplements comes from studies in adults that have then been extrapolated to children. Considerable uncertainty still exists with regard to estimates about appropriate dosing, clinical effectiveness and potential risks for pregnant women, newborns, infants and young children. Growing tissues, including the brain and bones, may be more susceptible to the effects of a supplement than is the case in adults, but firm evidence in this regard is lacking. Infants under 4 months of age have not yet fully developed the hepatic drug metabolizing enzymes and, as a result, are likely to be more sensitive to herbal phytochemical exposures. Thus, caution is warranted in recommending these products for use in young children.

When assessing the safety and efficacy of a dietary supplement for a child, several considerations must be made. For example, a high dose of phytosterols can compete for absorption of cholesterol and fat soluble vitamins and make it potentially dangerous. A further reason to regulate the preparation of dietary supplements, nutraceuticals, and functional foods is that some plants contain a wide variety of toxic chemicals that promote survival in their environmental niche and defend against bacteria, insects, and herbivores. Manufacturers may inadvertently include toxic constituents during the manufacturing process. Natural ingredients stored improperly can be substrates for molds that make highly dangerous mycotoxins. For instance, mold on peanuts forms the potent carcinogen aflatoxin.[79] Taken together, there is as much reason for having oversight of natural supplements as there is for oversight of synthetic drugs and foods. On the other hand, the increased review and regulation of dietary supplements could decrease access of the public to some products. For supplements administered at doses that are found in foods, the adoption of Good Manufacturing Practices should not alter availability to the consumer. For nutraceuticals that expose humans to ingredients at the doses they would normally not be exposed to, demonstration of safety should be mandated, even if this means a more prolonged period of time to introduce a new product to the marketplace. On balance, this seems a reasonable cost to protect the public health.

ACKNOWLEDGMENTS

The authors thank the authors of chapters from the 4th Edition of this textbook portion of which have been incorporated into this chapter (Chapter 22: Drs. Samuli Rautuva & W. Allan Walker; Chapter 35: Drs. Ekhard E. Ziegler, Samuel Fomon, & Susan J. Carlson; and Chapter 70:

Drs. Steven H. Zeisel & Martin Kohlmeier). Work in the authors' laboratories is supported by operating grants from the Canadian Institutes of Health Research. PJHJ is the recipient of a Canada Research Chair in Nutrition and Functional Foods and PMS the recipient of a Canada Research Chair in Gastrointestinal Disease.

REFERENCES

1. Kau AL, Ahern PP, Griffin NW, et al.Human nutrition, the gut microbiome and the immune system. *Nature.* 2011;474:327–336.
2. Backhed F, Fraser CM, Ringel Y, et al.Defining a healthy human gut microbiome: current concepts, future directions, and clinical applications. *Cell Host Microbe.* 2012;12:611–622.
3. Bosco-Drayon V, Poidevin M, Boneca IG, et al. Peptidoglycan sensing by the receptor PGRP-LE in the Drosophila gut induces immune responses to infectious bacteria and tolerance to microbiota. *Cell Host Microbe.* 2012;12:153–165.
4. Carvalho FA, Koren O, Goodrich JK, et al. Transient inability to manage proteobacteria promotes chronic gut inflammation in TLR5-deficient mice. *Cell Host Microbe.* 2012;12:139–152.
5. Johansson ME, Larsson JM, Hansson GC. The two mucus layers of colon are organized by the MUC2 mucin, whereas the outer layer is a legislator of host–microbial interactions. *Proc Natl Acad Sci USA.* 2011;108 Suppl 1:4659–4665.
6. Flint HJ, Scott KP, Louis P, Duncan SH. The role of the gut microbiota in nutrition and health. *Nat Rev Gastroenterol Hepatol.* 2012;9:577–589.
7. Dominguez-Bello MG, Costello EK, Contreras M, et al. Delivery mode shapes the acquisition and structure of the initial microbiota across multiple body habitats in newborns. *Proc Natl Acad Sci USA.* 2010;107:11971–11975.
8. Johnson CL, Versalovic J. The human gut microbiome and its potential importance to pediatrics. *Pediatrics.* 2012;129:950–960.
9. De Filippo C, Cavalieri D, Di Paola M, et al. Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proc Natl Acad Sci USA.* 2010;107:14691–14696.
10. Ley RE, Hamady M, Lozupone C, et al. Evolution of mammals and their gut microbes. *Science.* 2008;320:1647–1651.
11. Devkota S, Wang Y, Musch MW, et al. Dietary-fat-induced taurocholic acid promotes pathobiont expansion and colitis in Il10-/- mice. *Nature.* 2012;487:104–108.
12. Chung H, Pamp SJ, Hill JA, et al. Gut immune maturation depends on colonization with a host-specific microbiota. *Cell.* 2012;149:1578–1593.
13. Reading NC, Kasper DL. The starting lineup: key microbial players in intestinal immunity and homeostasis. *Front Microbiol.* 2011;2:148.
14. Hooper LV, Macpherson AJ. Immune adaptations that maintain homeostasis with the intestinal microbiota. *Nat Rev Immunol.* 2010; 10:159–169.
15. Shulzhenko N, Morgun A, Hsiao W, et al. Crosstalk between B lymphocytes, microbiota and the intestinal epithelium governs immunity versus metabolism in the gut. *Nat Med.* 2011;17:1585–1593.
16. Gareau MG, Sherman PM, Walker WA. Probiotics and the gut microbiota in intestinal health and disease. *Nat Rev Gastroenterol Hepatol.* 2010;7:503–514.
17. Shanahan F. The gut microbiota-a clinical perspective on lessons learned. *Nat Rev*

Gastroenterol Hepatol. 2012;9:609–614.
18. Johnston BC, Ma SS, Goldenberg JZ, et al. Probiotics for the prevention of Clostridium difficile-associated diarrhea: a systematic review and meta-analysis. *Ann Intern Med.* 2012;157:878–888.
19. Michail S, Sherman PM, Eds. *Probiotics in Pediatric Medicine*. Totowa, NJ: Humana Press; 2009.
20. Sherman PM, Ossa JC, Johnson-Henry K. Unraveling mechanisms of action of probiotics. *Nutr Clin Pract.* 2009;24:10–14.
21. Schiavi E, Barletta B, Butteroni C, et al. Oral therapeutic administration of a probiotic mixture suppresses established Th2 responses and systemic anaphylaxis in a murine model of food allergy. *Allergy.* 2011;66:499–508.
22. Rautava S, Kainonen E, Salminen S, Isolauri E. Maternal probiotic supplementation during pregnancy and breast-feeding reduces the risk of eczema in the infant. *J Allergy Clin Immunol.* 2012;130:1355–1360.
23. Neu J, Walker WA. Necrotizing enterocolitis. *N Engl J Med.* 2011;364:255–264.
24. Ganguli K, Meng D, Rautava S, et al. Probiotics prevent necrotizing enterocolitis by modulating enterocyte genes that regulate innate immune-mediated inflammation. *Am J Physiol Gastrointest Liver Physiol.* 2013;304:G132–G141.
25. Deshpande G, Rao S, Patole S, Bulsara M. Updated meta-analysis of probiotics for preventing necrotizing enterocolitis in preterm neonates. *Pediatrics.* 2010;125:921–930.
26. Katan MB. Why the European Food Safety Authority was right to reject health claims for probiotics. *Benef Microbes.* 2012;3:85–89.
27. Saulnier DM, Riehle K, Mistretta TA, et al. Gastrointestinal microbiome signatures of pediatric patients with irritable bowel syndrome. *Gastroenterology.* 2011;141:1782–1791.
28. Marchand V. Using probiotics in the pediatric population. Canadian Paediatric Society Nutrition and Gastroenterology Committee. Abridged version: *Paediatr Child Health*. 2012;17(10):575.
29. Manichanh C, Borruel N, Casellas F, Guarner F. The gut microbiota in IBD. *Nat Rev Gastroenterol Hepatol.* 2012;9:599–608.
30. Sherman PM, Cabana M, Gibson GR, et al. Potential roles and clinical utility of prebiotics in newborns, infants, and children: proceedings from a global prebiotic summit meeting, New York City, June 27–28, 2008. *J Pediatr.* 2009;155:S61-S70.
31. Hehemann JH, Kelly AG, Pudlo NA, et al. Bacteria of the human gut microbiome catabolize red seaweed glycans with carbohydrate-active enzyme updates from extrinsic microbes. *Proc Natl Acad Sci USA.* 2012;109:19786–19791.
32. Donovan SM, Wang M, Li M, et al. Host–microbe interactions in the neonatal intestine: role of human milk oligosaccharides. *Adv Nutr.* 2012;3:450S–455S.
33. Iyengar SR, Walker WA. Immune factors in breast milk and the development of atopic disease. *J Pediatr Gastroenterol Nutr.* 2012;55:641–647.
34. Howe KL, Reardon C, Wang A, et al. Transforming growth factor-beta regulation of epithelial tight junction proteins enhances barrier function and blocks enterohemorrhagic Escherichia coli O157:H7-induced increased permeability. *Am J Pathol.* 2005;167:1587–1597.
35. Roberfroid M, Gibson GR, Hoyles L, et al. Prebiotic effects: metabolic and health benefits. *Br J Nutr.* 2010;104 Suppl 2:S1–S63.
36. Moro G, Arslanoglu S, Stahl B, et al. A mixture of prebiotic oligosaccharides reduces the incidence of atopic dermatitis during the first six months of age. *Arch Dis Child.* 2006;91:814–819.

37. Yu SM, Kogan MD, Gergen P. Vitamin-mineral supplement use among preschool children in the United States. *Pediatrics.* 1997;100:E4.
38. US Department of Agriculture. Agricultural Research Service: Date tables: Results from USDA's 1994–1996 *Continuing Survey of Food Intakes by Individuals and 1994–1996:* Washington, DC: Diet and Health Knowledge Survey; 1997.
39. Baker SS. Unregulated dietary supplements: bitter medicine for children. *J Pediatr Gastroenterol Nutr.* 2001;33:439–441.
40. Lee Y, Mitchell DC, Smiciklas-Wright H, Birch LL. Maternal influences on 5- to 7-year-old girls' intake of multivitamin-mineral supplements. *Pediatrics.* 2002;109:E46.
41. Eichenberger Gilmore JM, Hong L, Broffitt B, Levy SM. Longitudinal patterns of vitamin and mineral supplement use in young white children. *J Am Diet Assoc.* 2005;105:763–772; quiz 773–764.
42. Dwyer JT, Garcea AO, Evans M, et al. Do adolescent vitamin-mineral supplement users have better nutrient intakes than nonusers? Observations from the CATCH tracking study. *J Am Diet Assoc.* 2001;101:1340–1346.
43. Ball SD, Kertesz D, Moyer-Mileur LJ. Dietary supplement use is prevalent among children with a chronic illness. *J Am Diet Assoc.* 2005;105:78–84.
44. Heuschkel R, Afzal N, Wuerth A, et al. Complementary medicine use in children and young adults with inflammatory bowel disease. *Am J Gastroenterol.* 2002;97:382–388.
45. Neuhouser ML, Patterson RE, Schwartz SM, et al. Use of alternative medicine by children with cancer in Washington state. *Prev Med.* 2001;33:347–354.
46. Kelly KM, Jacobson JS, Kennedy DD, et al. Use of unconventional therapies by children with cancer at an urban medical center. *J Pediatr Hematol Oncol.* 2000;22:412–416.
47. Hayek KM. Medical nutrition therapy for cystic fibrosis: beyond pancreatic enzyme replacement therapy. *J Am Diet Assoc.* 2006;106:1186–1188.
48. Innis SM. Dietary (n-3) fatty acids and brain development. *J Nutr.* 2007;137:855–859.
49. McNamara RK, Carlson SE. Role of omega-3 fatty acids in brain development and function: potential implications for the pathogenesis and prevention of psychopathology. *Prostaglandins Leukot Essent Fatty Acids.* 2006;75:329–349.
50. Hoffman DR, Birch EE, Birch DG, et al. Impact of early dietary intake and blood lipid composition of long-chain polyunsaturated fatty acids on later visual development. *J Pediatr Gastroenterol Nutr.* 2000;31:540–553.
51. Amminger GP, Berger GE, Schafer MR, et al. Omega-3 fatty acids supplementation in children with autism: a double-blind randomized, placebo-controlled pilot study. *Biol Psychiatry.* 2007;61:551–553.
52. Johnson M, Ostlund S, Fransson G, et al. Omega-3/omega-6 fatty acids for attention deficit hyperactivity disorder: a randomized placebo-controlled trial in children and adolescents. *J Atten Disord.* 2009;12:394–401.
53. Richardson AJ, Montgomery P. The Oxford-Durham study: a randomized, controlled trial of dietary supplementation with fatty acids in children with developmental coordination disorder. *Pediatrics.* 2005;115:1360–1366.
54. Gillies D, Sinn J, Lad SS, et al. Polyunsaturated fatty acids (PUFA) for attention deficit hyperactivity disorder (ADHD) in children and adolescents. *Cochrane Database Syst Rev.* 2012;7:CD007986.
55. James S, Montgomery P, Williams K. Omega-3 fatty acids supplementation for autism spectrum disorders (ASD). *Cochrane Database Syst Rev.* 2011;

(11):CD007992.
56. Palmer DJ, Sullivan T, Gold MS, et al. Effect of n-3 long chain polyunsaturated fatty acid supplementation in pregnancy on infants' allergies in first year of life: randomised controlled trial. *BMJ*. 2012;344:e184.
57. Dunstan JA, Mori TA, Barden A, et al. Fish oil supplementation in pregnancy modifies neonatal allergen-specific immune responses and clinical outcomes in infants at high risk of atopy: a randomized, controlled trial. *J Allergy Clin Immunol.* 2003;112:1178–1184.
58. Krauss-Etschmann S, Hartl D, Rzehak P, et al. Decreased cord blood IL-4, IL-13, and CCR4 and increased TGF-beta levels after fish oil supplementation of pregnant women. *J Allergy Clin Immunol.* 2008; 121:464–470.e466.
59. D'Vaz N, Meldrum SJ, Dunstan JA, et al. Fish oil supplementation in early infancy modulates developing infant immune responses. *Clin Exp Allergy*. 2012;42:1206–1216.
60. Mihrshahi S, Peat JK, Marks GB, et al; Childhood Asthma Prevention, S. Eighteen-month outcomes of house dust mite avoidance and dietary fatty acid modification in the Childhood Asthma Prevention Study (CAPS). *J Allergy Clin Immunol.* 2003;111:162–168.
61. Peat JK, Mihrshahi S, Kemp AS, et al. Three-year outcomes of dietary fatty acid modification and house dust mite reduction in the Childhood Asthma Prevention Study. *J Allergy Clin Immunol.* 2004;114:807–813.
62. Adkins Y, Kelley DS. Mechanisms underlying the cardioprotective effects of omega-3 polyunsaturated fatty acids. *J Nutr Biochem.* 2010;21:781–792.
63. Engler MM, Engler MB, Malloy M, et al. Docosahexaenoic acid restores endothelial function in children with hyperlipidemia: results from the EARLY study. *Int J Clin Pharmacol Ther*. 2004;42:672–679.
64. Engler MM, Engler MB, Malloy MJ, et al. Effect of docosahexaenoic acid on lipoprotein subclasses in hyperlipidemic children (the EARLY study). *Am J Cardiol.* 2005;95:869–871.
65. Garaiova I, Muchova J, Nagyova Z, et al. Effect of a plant sterol, fish oil and B vitamin combination on cardiovascular risk factors in hypercholesterolemic children and adolescents: a pilot study. *Nutr J* 2013;12:7.
66. NCEP. Executive Summary of The Third Report of The National Cholesterol Education Program Expert Panel on Detection, Evaluation, And Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). *JAMA.* 2001;285:2486–2497.
67. Canada H. *Plant Sterols and Blood Cholesterol Lowering, Health Canada, Food Directorate, Health Products and Food Branch, Bureau of Nutritional Sciences*. Ottawa Ontario; Health Canada—Bureau of Nutritional Sciences: Food Directorate, Health Products and Food Branch, 2010.
68. A.A.D. Nutrition recommendations and interventions for diabetes A position statement of the American Diabetes Association. *Diabetes Care*. 2008;31:S61–S78.
69. Amundsen AL, Ose L, Nenseter MS, Ntanios FY. Plant sterol ester-enriched spread lowers plasma total and LDL cholesterol in children with familial hypercholesterolemia. *Am J Clin Nutr.* 2002;76:338–344.
70. De Jongh S, Vissers MN, Rol P, et al. Plant sterols lower LDL cholesterol without improving endothelial function in prepubertal children with familial hypercholesterolaemia. *J Inherit Metab Dis.* 2003;26:343–351.
71. Plat J, Mackay D, Baumgartner S, et al. Progress and prospective of plant sterol and plant stanol research: report of the Maastricht meeting. *Atherosclerosis.*

2012;225:521–533.
72. Tammi A, Ronnemaa T, Valsta L, et al. Dietary plant sterols alter the serum plant sterol concentration but not the cholesterol precursor sterol concentrations in young children (the STRIP Study). Special Turku Coronary Risk Factor Intervention Project. *J Nutr*. 2001;131:1942–1945.
73. Bunchorntavakul C, Reddy KR. Review article: herbal and dietary supplement hepatotoxicity. *Aliment Pharmacol Ther*. 2013;37:3–17.
74. Cheng G, Remer T, Prinz-Langenohl R, et al. Relation of isoflavones and fiber intake in childhood to the timing of puberty. *Am J Clin Nutr*. 2010;92:556–564.
75. Thanos J, Cotterchio M, Boucher BA, et al. Adolescent dietary phytoestrogen intake and breast cancer risk (Canada). *Cancer Causes Control.* 2006;17:1253–1261.
76. Strom BL, Schinnar R, Ziegler EE, et al. Exposure to soy-based formula in infancy and endocrinological and reproductive outcomes in young adulthood. *JAMA.* 2001;286:807–814.
77. Wolff MS, Britton JA, Boguski L, et al. Environmental exposures and puberty in inner-city girls. *Environ Res.* 2008;107:393–400.
78. Messinaab M, Hilakivi-Clarkec L. Early intake appears to be the key to the proposed protective effects of soy intake against breast cancer. *Nutr Cancer.* 2009;61:792–798.
79. Wogan GN, Kensler TW, Groopman JD. Present and future directions of translational research on aflatoxin and hepatocellular carcinoma. A review. *Food Addit Contam Part A Chem Anal Control Expo Risk Assess.* 2012;29:249–257.

Energy Requirements in Health and Disease

13

Jean L. Bresson, MD, PhD

The interpretation of the changes in metabolic rate resulting from either malnutrition or disease finds its roots in the physiologic determinants of daily energy expenditure. The technical aspects of the measurement of energy expenditure and its obligate correlate, body composition, extend beyond the scope of this chapter and have been thoroughly reviewed in the literature.[1,2] Once raw data are available, many factors may confuse cross-sectional and, even more, age-related comparisons. The influence of body components, whether fat-free mass (FFM), fat mass (FM), or organ weight, should be considered first in an attempt to account for the wide differences in metabolic rate among infants, children, and adults. This is also relevant to a better understanding of the responses to underfeeding and malnutrition. Conversely, recent advances in the field of minor contributors to energy expenditure, such as the thermic effect of feeding or the thermic response to cold, may help us to understand how physiologic mechanisms may be turned into energy-losing machinery.

One of the most common findings in the sick child is a reduced food intake, which is widely accepted as the main, if not the only, cause of malnutrition. However, evidence accumulates suggesting that metabolic dysregulations may also participate and, in some instances, be primarily responsible for the nutritional deficit. This may be particularly true for proteins which, unless fat, are not stored but make up functional or structural tissues. Nitrogen balance in the adult or net protein deposition in the growing child results from protein synthesis rates equal to or higher

than protein breakdown, respectively. This equilibrium may be disrupted by the overproduction of peptides, otherwise playing a pivotal role in the regulation of the immune system. It results in a protracted increase in protein breakdown over protein synthesis. This mechanism is found in acute as well as in chronic inflammatory syndromes and may lead to severe protein malnutrition, despite apparently normal nutrient intakes. Consequently, underfeeding is clearly not the only factor in malnutrition. Furthermore, these protein losses are not easily controlled by classic dietary treatment and represent a pressing case for developing a specific pharmacologic approach.

PHYSIOLOGIC DETERMINANTS OF ENERGY EXPENDITURE

Basal Metabolic Rate

Comparisons between the rates of energy expended, or heat produced, by individual subjects require that the measurements should be free of the effects of any factor known to increase it. Failure to control for the effects of activity, feeding, and physical environment on metabolic rate would obviously result in spurious interindividual differences. Benedict proposed a set of standard conditions required to perform accurate measurements of the basal metabolic rate (BMR) in adults[3]: the subject should be awake, but in complete muscular repose both before and during the measurement, ideally performed in the morning before the patient rises; the subject should be in the postabsorptive state, normally accepted as 12 or more hours after the last meal, and should have received prior to the measurement a maintenance diet ensuring the stability of body weight; the subject should be in emotional repose and familiar with the measurement devices; in women, the measurement should not be made immediately before or during the menses; ambient temperature should be set in the range of the subject's thermal neutrality to avoid any thermoregulatory effect on heat production.

Obviously, these requirements may be impractical in infants and young children or in clinical settings. Metabolic rate measurement not performed in the postabsorptive state, but complying with the remaining conditions, and usually termed resting metabolic rate (RMR), is about 15% higher than true BMR.[4] It should be pointed out that, even in adults, the conventional period of 12 hours after the last meal may not be a sufficient interval to guarantee that the continuing effects of its metabolism are

negligible.[5] Hence, BMR should not be viewed as a measure of the absolute minimal metabolism. Indeed, energy expenditure is about 8%–12% lower during sleep than under basal conditions. As it is difficult to quantify the amount of work performed during spontaneous physical activity,[4] measurement of the sleeping metabolic rate (SMR) may be a convenient way to minimize its consequences on energy expenditure in young children. Thus, rather than defining a specific physiologic event, the set of standardized conditions chosen for metabolic rate measurements is primarily aimed at reducing the variance related to uncontrolled factors, to increase the power of interindividual or intergroup comparisons. When the rigorous conditions listed by Benedict are fulfilled, repeated measures display a remarkably low level of intraindividual variations. For example, the coefficient of variation (σ/m) of replicate indirect calorimetry measurements amounts to about 2%–4%, of which 2% could be accounted for by the variability related to the technique.[4]

The reproducibility of such measurements is high, even over a prolonged investigation. Thus, the range of intraindividual variations in BMR becomes much narrower than the range of interindividual variations that one may observe in cross-sectional studies of normal adult populations. A threefold difference is usually found between the lower and higher BMR individual values, expressed in kcal or kJ/d.[6] Repeated measures confirm that these are true between-subject differences. Furthermore, BMR is about 15% lower in adult females than in adult males. Much of these interindividual differences may be related to body build, as suggested by the significant correlations between BMR and body weight or height, or Quételet index (P/T^2).[7] Multiple regression analysis indicates that FFM accounts for about 65%–75% of BMR variance between subjects[8] and that FM, age, and sex together account for another 5%.[8] It is noteworthy that—exclusive of pregnancy—the contribution of FM to BMR variance is limited,[8,9] or even negligible.[4] This may be related to the low metabolic rate of adipose tissue, estimated to 3–4.5 kcal/kg fat/d. Thus, large differences in FM would not be expected to contribute greatly to differences in basal energy expenditure. Indeed, BMR becomes comparable in males and females after adjusting for FFM. Thus, FFM, FM, age, and sex account for about 80% of the interindividual variability in BMR, and nearly all the effect of these covariates is attributable to FFM (Figure 13-1).[10] However, differences between lower and higher individual metabolic rates still amount to about 600 kcal/d even after adjustment for these factors.[8] Further studies showed that family

membership is another significant determinant of BMR.[11] In the Pima Indians, metabolic rate among families may vary by nearly 600 kcal/d, whereas the mean variation within families is only about 60 kcal/d.[10] In this population, FFM, age, sex, and family membership account for 94% of the variability in metabolic rate, family membership contributing about 11%. Actually, a metabolic rate lower than that predicted by FFM, age, and sex at the beginning of the study turned out to be a major risk factor for body weight gain during the subsequent follow-up,[8] implying that obesity may also result from an economy in energy expenses. These results emphasize the necessity to express metabolic rate in relation to FFM, its major contributor, rather than in relation to any other parameter such as crude body weight or body area.

However, this mode of expression may still hamper interindividual comparisons in the pediatric population. During growth, RMR expressed either as kcal/kg body weight/d[12] or as kcal/kg FFM/d[13] follows a nonlinear increase, with slopes substantially higher in infants and young children than in adolescents and adults (Figure 13-2). After a steep increase in the days following birth,[14] RMR falls disproportionately to the rise in body weight[15] or FFM,[13] being about 67 kcal/kg FFM/d at 3 months, 46 kcal/kg FFM/d at 11 years, and 37 kcal/kg FFM/d at 13 years, 15 as compared to adult values of 28 kcal/kg FFM/d (Figure 13-3). This suggests that the metabolic activity of FFM is changing as a whole[16] or that the relative contribution of tissues with different metabolic rates to the composition of FFM is changing,[17] or both.

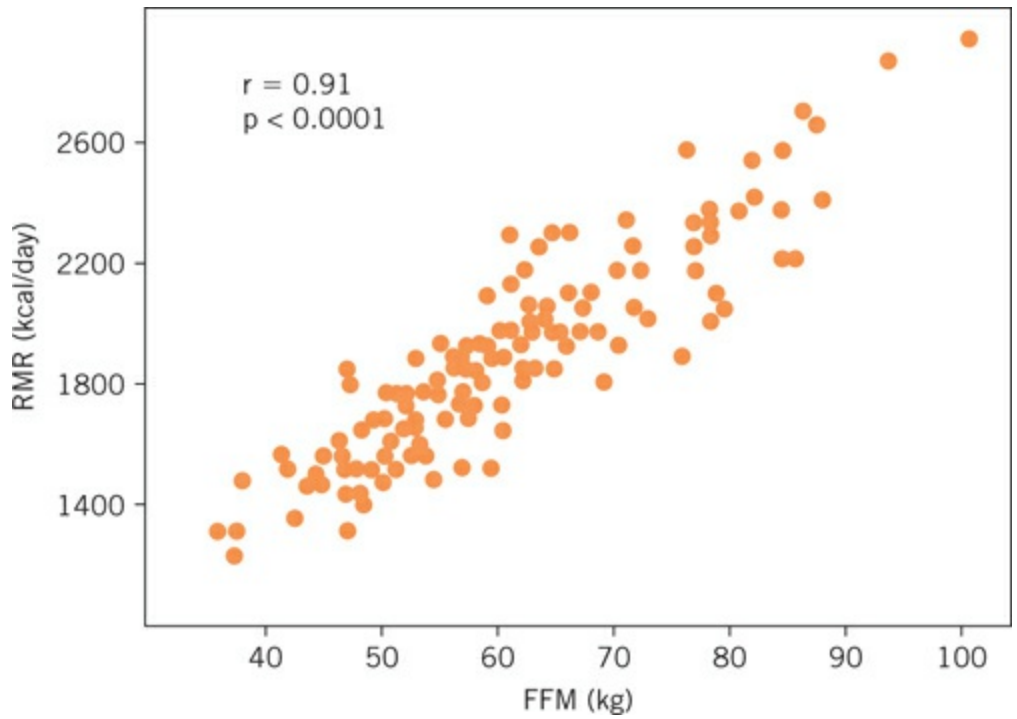


Figure 13-1 Relationship between resting metabolic rate (RMR) and fat-free mass (FFM) in 130 subjects. (From Bogardus et al.[10] With permission from the *New England Journal of Medicine*.)

Holliday pointed out that the brain, heart, liver, and kidney together make up only 5%–6% of the body weight, but account for 60%–70% of the BMR in the adult. Conversely, muscle mass, which makes up about 44% of the body weight, accounts for only 15%–30% of BMR.[12] In contrast, the FFM of infants and young children comprises about 30% muscle and 20% other organs, and that of adolescents averages 42% muscle and 8% other organs.[12] Therefore, the higher metabolic rate of infants and children in relation to FFM could be ascribed to a greater proportion of metabolically active non-muscle organ mass and to a lesser proportion of less metabolically active muscle mass than in adults (Table 13-1).[12,13] This is exemplified by experiments carried out on pigs with different nutritional background.[18] A first group was fed to gain steadily 14 kg over 70 days. A second group was fed to gain 19 kg during the first 35 days (high-energy period) and to lose 5 kg during the second 35 days (low-energy period; group HL), and this pattern was reversed (low-energy period first) in a third group (LH). At the end of the 70-day experiment, all animals had achieved the same body weight, but the fasting metabolic rate

of the LH group was 25%–50% higher than that of the HL group and 13%–40% higher than that of the first one. Furthermore, there was a significant correlation between fasting metabolic rate and organ weights.[18] The consequences of these observations are twofold. First, comparing the BMR/FFM ratio between two groups of subjects may be misleading, when values of FFM are markedly different,[13] mainly because the ratio of the weight of organs (heart, liver, brain, spleen, and kidneys) to total FFM decreases as FFM increases.[19] Second, this raises the possibility that abnormal metabolic rates in disease states may be related to a change in the proportion of metabolically active organs to less active muscles. For example, a reduction in muscle mass could account for the inverse relationship between RMR per kilogram of body weight and per cent ideal body weight observed in patients with Crohn's disease.[20]

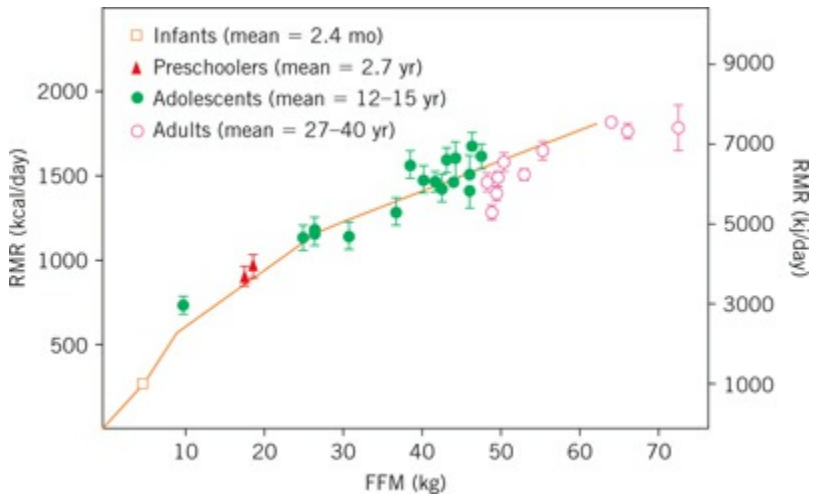


Figure 13-2 Relationship between resting metabolic rate (RMR) and fat-free mass (FFM) among different age groups. (Adapted from Weinsier et al.[13] With permission from the American Society for Clinical Nutrition.)

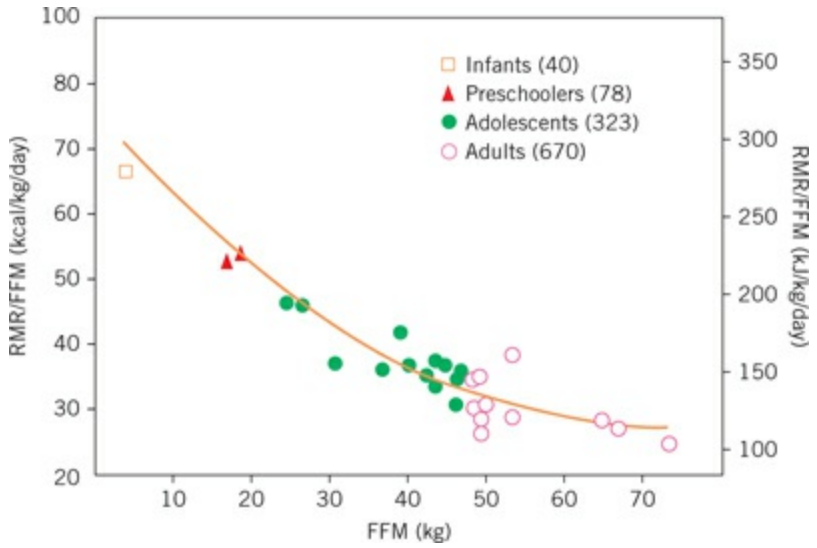


Figure 13-3 Relationship between resting metabolic rate (RMR) per unit fat-free mass (FFM) (RMR/FFM) to FFM among different age groups. (Adapted from Weinsier et al.[13] With permission from the American Society for Clinical Nutrition.)

Differences in metabolic rate between young and mature animals have also been found in other mammalian species. These weight-related differences further extend to comparisons between mature animals of different species. For example, mouse metabolic rate is about 5 times greater than man's and about 10 times that of the elephant.[21] Kleiber, and Brody and Proctor proposed a generalization according to which RMR follows a metabolic power function corresponding to weight 0.75.[21,22] However, the mass coefficient is not the same in various species, ranging from 0.48 to 0.91, the mean value being 0.67 ± 0.03. As there is no definite value for either the intraspecific or interspecific mass exponents, any attempt to explain the Brody–Kleiber empiric relationship by a general physical principle is most likely to fail.[22] Actually, this loose statistical relationship may obscure rather than reveal biologically important changes. The relative proportion of metabolically active tissue may contribute to these differences as large mammals have a greater proportion of tissues with low metabolic activity, whereas smaller ones have a greater proportion of tissues with higher metabolic activity. However, Krebs showed that the rates of respiration of homologous tissues are affected by the size of the animal, although there is no strict parallelism between measured tissue oxygen consumption and the BMR of the animals.[23] A

comparison between homeo- and poikilotherms might give a hint about a possible cause for this difference. The standard metabolic rate is sevenfold greater in the rat (a typical mammal) than in an ectothermic vertebrate (bearded dragon) of the same body size and at the same body temperature.[24]

TABLE 13-1 Relation Between Basal Metabolic Rate (BMR) and Muscle and Organ Metabolic Rates with Age

Height (cm)	Weight (kg)	BMR (kcal/d)	Muscle MR (kcal/d)	Organ MR (kcal/d)	B/A (%)
		A	B	C	
60	5.5	300	21	279	7
80	11.0	590	45	545	7.6
110	19.0	830	117	713	14
140	31.0	1160	204	956	17.6
160	50.0	1480	373	1107	25.2
180	70.0	1800	500	1300	27.7

Source: From Blaxter.[21]

This large difference in metabolism is also manifest at the cellular level and may be related to membrane properties.[25] Rat hepatocytes respire fourfold faster than do hepatocytes from the lizard. The inner membrane of isolated rat liver mitochondria has a permeability that is four- to fivefold greater than the proton permeability of the lizard liver mitochondrial membrane.[24] Thus, the greater proton permeability of the mitochondrial inner membrane contributes to the greater metabolic rate of mammals. Actually, this is a major source of heat production (see below), the variations of which might impinge on lifespan.[26] Similarly, plasma membrane permeability to Na^+ and K^+ is higher in mammals than in ectothermic vertebrates, increasing the amount of energy devoted to active Na^+–K^+ exchange.[27] Thus, the activity of the Na^+–K^+-ATPase accounts for about 20% of the total in vitro energy expenditure by mammalian tissues.[28] The use of ouabain to inhibit Na^+–K^+-ATPase in vivo leads to a 20%–30% inhibition of resting oxygen consumption in guinea pigs.[29] and to a 20%–25% inhibition in mice,[30] supporting the idea that active Na^+–K^+ exchange is a significant determinant of resting energy expenditure. In

humans, Na^+–K^+-ATPase activity may contribute 20%–25% to BMR.[31] As in animals, Na^+–K^+-ATPase is subject to chronic control by thyroid hormones, and slight interindividual variations in free T_3 index among healthy adults would account for a between-subject difference in SMR of 142 kcal/d.[32] Furthermore, the magnitude of Na^+–K^+-ATPase-dependent energy expenditure in tissues appears to be related to the physiologic status, and particularly to age. Muscle preparations from 7- to 21-day-old calves expend 26% more total aerobic energy in support of Na^+–K^+ transport than do the same muscles from 7-month-old calves.[28] Similarly, the Na^+–K^+-ATPase-dependent respiration of skeletal muscle preparations from 4-week-old lambs accounts for a 48% greater amount of energy than similar muscles from adult ewes. Also, energy consumption in support of active Na^+–K^+ exchange by the intercostal muscle from young lambs is 119% greater than for similar preparations taken from adults.[33] Comparable differences exist between hepatocytes obtained from 1- to 8-week-old lambs and from mature sheep.[34] In this latter case, the lower oxygen consumption of hepatocytes isolated from mature sheep is largely attributable (70%–90%) to the decrease in the Na^+–K^+-ATPase-dependent component of respiration. Thus, membrane-associated processes appear to be significant components of BMR.

It is striking that in quiescent-cultured animal cells exposed to mitogens, one of the first events to take place is a rapid, very large increase in the rate of Na^+ flow into the cells,[35] which in turn induces increased Na^+–K^+ pumping. Activation of Na^+–K^+-ATPase appears to be an early growth-related event, and it is tempting to speculate that increased energy expended on active Na^+–K^+ transport by young mammals is also related to their higher growth rate.[33] Active ion transport is probably not the sole cause of this difference. Measurement of whole-body protein synthesis shows that the smaller mammals display the higher protein synthesis rates in relation to their body weight. Within-species comparisons also indicate that immature animals have higher protein turnover rates than adults. For example, protein synthesis amounts to 6.7 ± 1.1 g/kg/d in term infants as compared to 3.5 ± 0.6 g/kg/d in adult humans. Simultaneously, basal energy expenditure is 45.1 ± 4.6 and 23.6 ± 3.8 kcal/kg/d in newborns and adults, respectively.[36] Thus, a similar ratio of BMR to whole-body protein synthesis (6.7–6.9 kcal/g) applies to both healthy newborn and young adults, suggesting that the turnover of cellular constituents, including proteins, and energy expenditure change in parallel with maturation.

Therefore, differences in BMR in relation to body weight or FFM during growth is not merely a matter of proportion between tissues with high or low metabolic rates, but is also related to differences in the amount of work done by the cells to ensure the turnover of their constituents and maintain the membrane electrochemical gradients.[37]

Energy Cost of Growth

An estimate of the energy cost of growth can be obtained in groups of rapidly growing infants from the relationship between growth rate and metabolizable energy intake.[38] The slope of this linear relationship provides an estimate of the total amount of energy stored and expended for synthesis, and its intercept at zero growth indicates the amount of energy required for maintenance. It should be pointed out that maintenance requirement may be higher than BMR by 10% in premature infants[39] and 50% in children,[40] as maintenance includes expenses for limited activity and feeding.[41] The energy cost of growth can also be estimated from the independent measurement of stored energy and energy expended for synthesis.[39] An indirect method for calculating the cost of tissue synthesis is to relate weight gain to energy expenditure, assuming that an increase in expenditure is directly related to the increase in cost of synthesis. Unfortunately, the result may be obscured by activity. A direct measurement of the cost of synthesis rests on the assumption that the thermic effect of feeding (i.e., the increase in metabolic rate after the meal over the resting value in the postabsorptive state; thermic effect of feeding [TEF]) is largely accounted for by synthesis. This is supported by the positive correlation observed between weight gain and TEF in rapidly growing individuals.[41] However, the TEF may be grossly underestimated unless the interval between feeds is long enough to allow metabolic rate to return to postabsorptive levels.

Various estimates of the total cost of growth are in the range of 3–6 kcal/g of new tissue. A widely accepted figure in normal infants is 5 kcal/g.[39,40] Metabolic balance and body composition studies indicate that the average composition of weight gain is 28% fat and 16% protein during the first year of life.[42] Assuming crude energy contents of 9.25 and 5.65 kcal/g for fat and protein, respectively, it is possible to estimate the amount of energy stored to about 3.5 kcal/g of body weight gain. In contrast, the cost of synthesis is about 1.5 kcal/g of body weight gain. Direct estimates of the amount of energy expended for synthesis usually provide figures in the range of 0.55–0.7 kcal/g.[39,43] This discrepancy may be related to an

underestimation of the TEF, as it is measured only two to three hours after the last meal. The so-called fasting metabolic rate used as a baseline is likely to be falsely high due to the lasting effect of feeding. As the energy stored in weight gain represents about 70% of the total energy cost of growth, one can expect wide differences according to the composition of tissues laid down. For example, it is possible to calculate that the total amount of energy required to deposit 1 g of protein is about 13.4 kcal/g and is 10.8 kcal/g for fat.[44] Thus, a weight gain comprising 16% protein and no fat would cost about 2.2 kcal/g. Conversely, a weight gain consisting of fat with no protein would cost about 9.7 kcal/g, assuming that adipocytes may contain up to 90% lipids. These differences may account for the high cost of weight gain found in overfed adults. In these conditions, weight gain consists of about 70% fat and 30% fat-free tissue,[45] the theoretic cost of growth then amounting to approximately 7 kcal/g. Similarly, the average cost of weight gain in patients with anorexia nervosa ranges from 5 to more than 7 kcal/g, suggesting either important changes in the composition of body weight gain during rehabilitation,[46] or an important confounding effect of activity.[47]

The daily energy cost of growth will depend obviously on growth rate as well as on the composition of new tissue. Growth rate is highest during the first three months of life, reaching 30–35 g/d between birth and 8 weeks of age, or a fractional growth rate of about 1% per day.[42] It decreases rapidly thereafter to 7–10 g/d at 1 year of age, or 0.07% per day to 0.09% per day, and to about 5 g/d (less than 0.03% per day) by 5 years of age. Thus, the cost of growth represents a significant part of daily energy needs (approximately 33%) from birth to 4 months of age. Later on, it decreases to about 7% between the ages of 4 and 12 months, 1.6% between 12 and 24 months, and 1% between 24 and 36 months. Thereafter, it is negligible compared to daily energy expenditure. Thus, on the basis of nutrient partition between growth and nongrowth, the 1- to 2-year-old child appears more like the adult than the small, rapidly growing infant.[48]

Thermic Effect of Feeding

It was recognized long ago that fed animals have substantially higher metabolic rates than fasted ones.[49] The effect of specific nutrients was investigated during the 19th century showing that the ingestion of proteins induces a greater increase in energy expenditure than carbohydrates or fat. The ingestion of a mixed meal also results in an increased metabolic rate

above BMR. Energy expenditure may increase by up to 30% over BMR in response to large energy challenges, a sizable change equivalent to 6% to 10% of the energy intake.[50] Unlike BMR, the TEF appears to be independent of weight, FFM, FM, and Quételet index.[50] In contrast, its duration and magnitude are linearly related to the energy intake,[50] which explains the greater effect of large meals.[51] Duration and magnitude of TEF do not appear to be significantly different when the same meal is ingested or directly delivered into the stomach.[52] However, the magnitude of the thermic effect is significantly greater if the same energy load is delivered in a short time rather than over several hours.[51,53] The route of administration of the nutrients, either intragastric or intravenous, does not seem to affect the magnitude of the response,[54] suggesting that the energy expended on luminal digestion, fermentation, mechanical mixing, and nutrient absorption makes only a modest contribution to the TEF in normal subjects. Energy expenditure may still be significantly higher than postabsorptive levels by five to eight hours after a large energy challenge.[50] Furthermore, the effect of the antecedent diet may significantly affect both SMR and RMRs. For example, RMR remains elevated by 12% 14 hours after the last meal during short-term overfeeding.[5] In these conditions, the TEF lasts not only for the whole day, but also throughout the night until the following morning, when the effect is still apparent. There is also strong indication that the TEF of one meal merges into the thermic effect of the next meal. Thus, the duration of the TEF should be carefully considered before attempting to determine either BMR or the effect of feeding, particularly in infants.

The composition of the meal bears some importance. In accordance with earlier work, the thermic response to a high-protein meal is greater than that to a high-carbohydrate one.[55] Similarly, the TEF is greater after a high-carbohydrate meal when compared to a high-fat[56] or a mixed diet.[57] Thus, high-fat meals could be a risk factor for obesity in children.[58] However, the ingestion of a mixed meal elicits many simultaneous and complex events, and an experimental account of the thermic response to feeding could only come from a simplified model. The euglycemic-hyperinsulinemic clamp offers several advantages in this regard.[59] It involves a single nutrient and bypasses absorption, allowing one to know precisely the rate of glucose appearance within the milieu intérieur at any time. Combined with indirect calorimetry, it allows one to assess the effect of glucose infusion on glucose oxidation and storage as well as on energy expenditure. When different plasma insulin concentrations are achieved,

together with glucose infusion rates appropriate for maintaining euglycemia, it is readily apparent that the increase in energy expenditure secondary to glucose administration is not related to an increase in its oxidation rate. In contrast, the increase in energy expenditure is closely related to the rate of glucose storage.[60] This is supported by studies in insulin-resistant patients showing that their blunted thermic response to glucose infusion is restored when glucose storage rate is matched to controls by further increasing insulin concentration.[61] Thus, in agreement with Flatt's proposal,[62] the TEF is primarily related to the cost of storage of the nutrients. According to the stoichiometry of these reactions, it is possible to calculate that the synthesis of glycogen costs about 4% of the energy of the glucose stored (0.15 kcal/g), and that this cost increases to 28% (1.05 kcal/g) when glucose is converted to fat. Conversely, the cost of triglyceride synthesis from absorbed fats amounts to about 2% of their energy content. Because of the many ATP-consuming steps involved in the synthesis and breakdown of proteins, the cost of amino acid incorporation into proteins is estimated to about 25%, but might be as high as 55% of their energy content.[62] Indeed, comparing the thermic effect of high-protein and high-carbohydrate diets shows that the former results in a greater thermic response (+45%) than the later. Simultaneously, the increase in protein turnover is twice as much with the high-protein diet as with the high-carbohydrate diet. The metabolic cost of protein synthesis would thus accounts for about 68% and 36% of the thermic effect of each diet, respectively, and the combined effect of increased protein synthesis and nitrogen excretion would account for 96% of the thermic effect of the high-protein diet.[55]

The cost of storage may not be the only component of the TEF. Indeed, the increase in energy expenditure related to glucose infusion is substantially higher than predicted from the amount of glucose stored.[60] The fact that overfeeding or the ingestion of a glucose-rich meal elicits an increase in sympathetic nervous system (SNS) outflow,[63] or in plasma noradrenaline concentration and turnover,[64] a known thermogenic hormone, suggests that the stimulation of the SNS is involved in the thermic response to feeding. This is supported by the blunting of the glucose-induced increase in energy expenditure by β-adrenergic blockade during insulin clamps. As a result, the residual variation in metabolic rate becomes almost identical to the value predicted from the amount of glucose stored.[65] This implies that the TEF has two components. One is an obligatory component directly related to the storage of nutrients and

accounts for about 60% of the response to feeding. The other is related to the stimulation of the SNS and represents about 30% of the increase in energy expenditure.[66] This partition is confirmed by the use of clonidine, a centrally acting α_2-adrenergic receptor agonist that inhibits SNS outflow. In association with the decreased SNS response to a carbohydrate-rich meal, TEF is blunted by 33%, a value similar to that obtained from peripheral β-adrenergic blockade during insulin clamps.[67] The effect of SNS activity on energy expenditure is not restricted to the hours following meals. Its inhibition by clonidine also decreases RMR. Although the reduction in RMR does not reach significance because of the small number of subjects, it should be stressed that a 6% decrease in baseline metabolic rate over a whole day might account for a greater overall diminution in energy expenditure than the larger 33% blunting of TEF over a few hours.[67] The contribution of SNS activity to energy expenditure in the postabsorptive state is further confirmed by the 8% reduction in BMR observed in response to a β-adrenergic antagonist that does not affect T_3 production, nadolol.[68] Furthermore, urinary noradrenaline excretion and muscle sympathetic nerve activity are significantly correlated to RMR adjusted for FFM, FM, and age in normal adults, implying that SNS activity is a physiologic determinant of energy expenditure.[69,70] A similar correlation between urinary noradrenaline excretion and oxygen consumption is observed in infants,[71] suggesting that the effects of SNS activity on energy expenditure extend to this age.

Therefore, the thermic effect of feeding involves a short-term meal-related component lasting for several hours, and is accounted for by both nutrient storage (obligatory) and acute stimulation of the SNS (facultative). In rapidly growing infants, this short-term effect also reflects the cost of synthesis of new tissues, as its magnitude is significantly correlated to growth rate.[41] However, the response to feeding also involves a longer term component, which may affect BMR and SMR.

Thermogenesis

Heat production by living organisms is associated with respiration and concomitant oxidation of nutrients. According to the chemiosmotic theory,[72] the enthalpy of oxidation is transduced by the respiratory chain of the mitochondria into an electrochemical gradient (or proton-motive force), which powers the ATP synthase. However, the efficiency of the system, characterized by the ratio of the amount of energy recovered as ATP to that liberated during substrate oxidation, is far from one, even if it

is higher than previously estimated from the standard-free energy of ATP, ΔG^0.[1] Attempts to directly evaluate this efficiency in man indicate that about 65% of the energy liberated may be recovered under the form of ATP.[73] Thus, a minimum of 35% is lost as heat, and oxidation at the level of mitochondrial respiration appears as the primary heat-releasing process. This is consistent with the significant relationship existing between body temperature and BMR or SMR.[74] However, basal heat production may be insufficient to maintain the core temperature within a narrow range of variation, particularly when heat exchange is increased by a low ambient temperature. Extra heat production can be generated by recruiting metabolic processes that do not produce any net work. Isometric muscular contraction, as it may occur during malignant hyperthermia, is a major heat-generating process. Similarly, shivering increases oxygen consumption,[75] and because no external work is performed, the energy appears as heat.

There is some evidence in adult humans that heat production may be increased by other means. For example, BMR or 24-hour energy expenditure (24hEE) increases significantly (6%–11%) in response to an ambient temperature drop from 28°C to 20°C–22°C, without any overt sign of shivering or any correlation with activity.[76] Similarly, the metabolic rate of full-term infants increases in response to an ambient temperature lowering from 32°C to 23°C. This increase is not correlated with activity and occurs in the absence of shivering. In these conditions, the infants' response is dramatic, amounting to a 100% increase in energy expenditure,[77,78] which suggests that a nonshivering thermogenesis is operative. In the rat, the early stage of cold adaptation is associated with shivering. However, shivering disappears with acclimatization, and a nonshivering heat source eventually takes over all the duties of heat production. It is now well established that cold exposure results in a marked increase in sympathetic activity, largely responsible for the elevated metabolic rate of the rats.[79] The dominant role in nonshivering thermogenesis has been ascribed to brown adipose tissue (BAT), even in adult animals.[80] BAT stimulation is achieved through β-adrenergic receptors, among which is a new type of receptor with pharmacologic properties differing from either β_1 or β_2, thus termed β_3.[81] BAT's prominent characteristic is the synthesis of a specific, 32,000 Mr protein, known as uncoupling protein-1 (UCP-1; review in Himss-Hagen[82]), located in the inner mitochondrial membrane and acting as a proton conductance pathway. UCP-1 dissipates the proton gradient created by

respiration, thus bypassing ATP synthase. In turn, the dissipation of the proton gradient causes a dramatic increase in the mitochondrial respiratory rate. Upon β-adrenergic stimulation, BAT substantially increases its respiration. As mitochondrial respiration and phosphorylation are uncoupled, ATP generation accounts for much less than 10% of oxygen consumption, and the enthalpy of oxidation of the nutrients is essentially dissipated as heat, making BAT a tissue specialized in thermogenesis. Acute cold exposure leads to a rapid and pronounced elevation of the level of the mRNA coding for UCP-1, and UCP-1 synthesis is evident within 15 minutes.[83] In contrast, hypothyroid rats are unable to maintain a normal body temperature when placed at 4°C, despite increased noradrenaline turnover rates. The response of UCP mRNA to adrenergic stimulation in hypothyroid brown adipocytes is readily restored by T_4 through mechanisms requiring its intracellular conversion to T_3. For a full UCP-1 response to cold, near saturation of the nuclear T_3 receptor is required. BAT contains a type II 5'deiodinase (5'D-II), which locally generates T_3 and allows it to reach a very high level of receptor occupancy despite the low physiologic plasma T_3 levels. Indeed, T_4 appears to be a much more efficient source of T_3 for BAT than plasma T_3 when the 5'D-II is activated. Conversely, iopanoic acid, an inhibitor of 5'D-II, abolishes the T_4-induced restoration of both UCP-1 response and normothermia in hypothyroid rats. In turn, 5'D-II activity is stimulated by SNS, and nuclear T_3 receptors are nearly saturated within four hours of cold exposure. Thus, SNS, through cAMP generation, and thyroid hormones act synergistically on BAT. T_3 affects the UCP-1 gene expression at a transcriptional and posttranscriptional level.[84] However, this effect of T_3 is not sufficient to elevate UCP-1 mRNA cellular content in the absence of significant levels of transcription, explaining why T_3 cannot increase UCP-1 mRNA in vivo in the absence of adrenergic stimulation.[85] Thus, T_3 amplifies the effects of the adrenergic stimulation on UCP-1 gene expression, allowing BAT to reach its full thermogenic capacity. Conversely, pretreatment of rats with corticosterone abolishes the UCP-1 mRNA response to noradrenaline and cold by inhibiting UCP-1 gene expression.[86] Interestingly, this effect is antagonized by the action of the corticotropin-releasing hormone (CRH; under feedback control by glucocorticoid) on the central nervous system, stimulating SNS outflow and leading to an increased expression of UCP-1.[87] The simultaneous expression of several proteins (UCP, 5'D-II, and β_3-

adrenoreceptors) appears to characterize functionally active BAT, although only UCP-1 is tissue specific.

The search for BAT in humans implied the characterization of UCP-1. This protein was first reported in fat depots from patients with pheochromocytoma.[88] It was subsequently isolated from newborn fat.[89] Later on, UCP-1 was found in humans at all ages, but its content was highest in children.[90] Since then, human UCP-1 gene has been cloned. Its sequence is 80% homologous to rat UCP-1, both at the nucleotidic and amino acid levels. Both UCP-1 and UCP-1 mRNA are present in infants and in adult patients, UCP-1 mRNA being detected in the periadrenal adipose tissue of all the subjects studied.[91] UCP-1 transcripts are abundant in an infant's perirenal fat, but are in low quantity in subcutaneous depots.[92] Full-term newborn interscapular UCP-1 content is equal to, or even higher than, the level found in 3-week-old mice acclimatized at 20°C (a temperature below thermal neutrality). In contrast, ATPase content is low in infant BAT, similar to that found in animal BAT mitochondria, compared with heart and liver mitochondria.[93] The specific content of UCP-1 in infant BAT increases twofold between 25 and 32 weeks of gestation and remains nearly constant thereafter.[93] Infant BAT also expresses a 5'D-II. Strikingly, 5'D-II content follows the same pattern as UCP-1 and achieves levels comparable to 5'D-II activities found in animals stimulated during the neonatal period by exposure to cold or increased cAMP levels.[93] Moreover, β_3-adrenoreceptor mRNA is expressed in children and adults.[92] This coexpression of UCP-1, 5'D-II, and β_3-adrenoreceptor suggests that functional BAT exists in humans from birth to old age. Measurements performed in vitro on isolated cell preparations as well as in vivo suggest that the heat-dissipating capacity of BAT is in the range of 300 W/kg of tissue.[94] Thus, 16 g of BAT would be sufficient to account for the 25–30 W/m^2 increase in energy expenditure observed in newborns exposed at 23°C.[78] This would represent about 3% of the adipose tissue mass of newborns, or a figure similar to the amount of BAT found in adult rats acclimated to cold.[80] It is radiologists, not physiologists, who rediscovered active BAT in human adults. Indeed, the infusion of 2-deoxy-2-[^{18}F]fluoro-d-glucose to cancer patients, to identify metastases by positron emission tomography, resulted in many "false positive" signals (particularly in the neck, supra-, subclavicular, and paravertebral areas) which could be completely quenched by one-hour exposure to a warm environment.[95] It was soon confirmed that this highly

active, cold-sensitive glucose uptake was due to fully functional BAT,[96] which contributes to energy expenditure and to the control of adiposity.[97,98]

Adrenaline-mediated thermogenesis also occurs in human muscle,[99] even if its mechanism is still uncertain. Adrenaline infusion causes a 25% increase in whole-body energy expenditure and stimulates forearm muscle oxygen consumption by as much as 90%. Assuming that forearm muscle is representative of total body musculature, skeletal muscle would then account for up to 40% of the adrenaline-infusion-induced thermogenesis in humans.[100] More recently, it was shown that T_3 administration to normal adults results in a significant increase in the activity of muscle tricarboxylic acid (TCA) cycle, without any change in the rate of ATP synthesis.[101] The disproportionate increase in TCA cycle flux compared with ATP synthesis suggests that T_3 induces an increase in muscle thermogenesis by promoting mitochondrial energy uncoupling. Indeed, human muscle has the intrinsic capacity for cold-induced adaptative thermogenesis via mitochondrial uncoupling under physiologic conditions.[102] In this regard, it is noteworthy that intraindividual variations in BMR are tightly correlated to small, spontaneous changes in thyroid hormone concentrations in healthy adults,[32,103] a mechanism which could contribute to cold adaptation.

Besides mitochondrial uncoupling, the thermogenic potential for Ca^{2+} cycling is exemplified by the pathological syndrome of malignant hyperthermia, a dominant pharmacogenetic myopathy that occurs in humans and several other mammalian species. Ca^{2+} cycling-mediated thermogenesis in skeletal muscle cells is a well-characterized mechanism for nonshivering thermogenesis in fish and recently in birds. It could prove to be an important source of heat in mammals lacking large amounts of BAT. In any cases, the biochemical properties of muscle (i.e., fast-twitch versus slow oxidative fibers) do contribute to the interindividual variability in SMR and 24hEE in the adult.[104] However, the respective contributions of BAT and muscle to the overall thermogenic response remain difficult to ascertain. Some studies in adults suggest that approximately 700 g BAT would contribute 25% to adrenaline-induced thermogenesis, whereas the approximately 34 kg muscle mass would account for 50% of the thermic response.[105] Despite the uncertainties about the respective contribution of its effectors, the reality of nonshivering thermogenesis in humans is now well established and its impact on energy expenditure and body FM recognized.

24-Hour Energy Expenditure

The measurement of 24hEE in free-living conditions over relatively long time intervals can be assessed by determining the turnover rates of isotopes of hydrogen (^{2}H) and oxygen (^{18}O) in total body water.[106] Despite its cost and its methodological limits, the doubly labeled water technique is quite suited for studies in infants and children. Actually, it led to recognize that recommended dietary allowances overestimated energy requirements by a significant extent in these age groups.[107]

BMR represents a major determinant of 24hEE. For example, it amounts to about 60% of 24hEE in sedentary adults.[108] The difference between BMR and 24hEE comprises thermogenesis, the thermic effect of feeding and the amount of energy expended on physical activity, be it exercise or nonexercise activity. Obviously, this is the most variable component of total energy expenditure.

In 1985, the FAO/WHO/UNU expert committee on energy requirements expressed the energy needs as multiple of BMR. The 24hEE to BMR ratio was subsequently referred to as physical activity level (PAL). Although not entirely satisfactory, PAL offers a convenient means of assessing and comparing physical activity, which is its main source of variance. Extreme PAL range from about 1.2 in the completely chair- or bed-bound subjects to about 4.7 in Tour de France cyclists.[109] PAL > 4 represents levels of expenditure which are not sustainable on a permanent way of life. On the contrary, PAL range of 1.2–2.5 corresponds to sustainable lifestyles, where 2.5 represents a very physically active one. Actually, a PAL of 1.35 is now considered as the lowest value compatible with long-term maintenance in normal adults and a PAL range of 1.55–1.65 represents the average of the so-called sedentary lifestyle.[110] Data suggest that activities do not have to be obviously strenuous for relatively high PAL values to be achieved. In adults, the time spent at activities with low and moderate intensity determines the activity level, whereas high-intensity activities do not have much impact.[111] In other words, the cumulative impact of low to moderate activities of prolonged duration is of greater energetic significance than short bursts of high-intensity activities. In children, PAL show an inverse relation with the time spent on low-intensity activities[112] and, as in adults, engagement in vigorous-intensity activities does not influence significantly 24hEE.[113] It is noteworthy that the fraction of time spent on low-intensity activities appears lower in 8-year-old children than in young adults and elderly persons.[112] PAL values vary from about 1.4–1.5 to 1.7–1.9 in boys and girls between 1 year of age

and adolescence.[114] Indirect estimates suggest that the energy cost of spontaneous activity represents only 5%–10% of 24hEE in premature infants,[43] but amounts to 20%–26% of 24hEE between 1 and 4 months of age[115] and up to 50% in 8- to 12-year-old children. A mean PAL value of 1.5 was reported in 10- to 13- year-old boys and girls. In this latter case, the variability of the 24hEE to BMR ratio is quite large, reflecting important individual differences in the amount of energy expended for activity (1.20–1.87).[116]

Of note, PAL is negatively associated with body fat in 5- to 10.5-year-old[117] or 6- to 8-year-old children.[118] This suggests that the amount of energy expended on physical activity may significantly affect energy balance, a potential asset in the prevention and treatment of obesity. Although most cross-sectional studies support a relationship between body fat and activity in children,[119] the results of interventions aimed at preventing childhood obesity through the implementation of physical activity programs remain inconclusive.[120] This is intriguing, as obesity may develop from a remarkably small daily net imbalance between energy intake and expenditure (median of estimated energy accumulation: 15 kcal/d),[121] a difference well within the range of moderate-intensity physical activity expenses. However, this discrepancy could be accounted for by the lack of effect of physical activity interventions on the overall activity level of children.[122]

On the contrary, it is not quite unexpected that most studies in obese adults demonstrate that participating in routine endurance exercise activities for up to a year does not induce a significant weight loss in the absence of an energy-restricted diet. Obesity represents the storage of a large energy surplus (over 100 Mcal in adults) and any attempt to reduce this imbalance, even by a small amount, would require a very significant increase in energy expenditure. Indeed, diminishing such reserves by 1% would imply a net negative balance of ≥1000 kcal. For a 100 kg-overweight individual, this would translate into more than 2 hours of brisk walking (4 mph) or light jogging every day to expend about 700 kcal/d.[123] Thus, a negative outcome with exercise alone is likely related to a failure to achieve an adequate level of physical activity.

Nonexercise, spontaneous physical activity, also known as nonexercise activity thermogenesis (NEAT), encompasses the daily expenditure of living activities (mainly ambulation) and the energy cost of all nonvolitional muscle activity, such as muscle tone, posture maintenance, and fidgeting. NEAT may play a significant role in the maintenance of

energy balance. Spontaneous physical activity measured within the confines of a respiratory chamber accounts for an average 348 kcal/d, with individual extremes of 100–800 kcal/d.[4] NEAT appears to be a familial trait, amounting to as much as 13% of 24hEE. Thus, NEAT may be—independently of volitional exercise—an important mediator of resistance to fat gain. Changes in NEAT account for differences in fat storage between individuals and failure to activate NEAT in the face of excess energy may lead to weight gain.[124] The comparison of mildly obese and control adults suggests that obese individuals may save up to 352 kcal/d (269–477 kcal/d) by staying seated 164 minutes longer per day than their lean counterparts, an economy not matched by a similar reduction in energy intake.[125] The contribution of NEAT to 24hEE in childhood is unknown, but might be quite significant. The regulation of NEAT is also unknown. However, familial resemblance in twin pairs' physical activity is predominantly explained by shared environmental factors rather than by genetic variability.[126]

Thus, components other than BMR and the energy cost of activity contribute modestly to the 24hEE of normal infants and children. However, an increase in the energy expended by these minor components (SNS activity and thermogenesis) may significantly alter energy balance in disease states. Conversely, the activation of nonshivering thermogenesis by activation/recruitment of BAT could have a positive effect: increasing energy expenditure would result in a decrease in body fat, an exciting prospect for the prevention of obesity.[127]

EFFECTS OF UNDERFEEDING AND MALNUTRITION

Effects on Energy Expenditure

Underfeeding healthy or obese adults result in a consistent 20%–30% reduction in BMR and 24hEE expressed either in absolute terms or related to body weight, body surface area, body weight to the 0.73 power,[128] or FFM.[129] It should be pointed out that this effect has important implications for the clinical management of obesity, as this compensatory change opposes the maintenance of a body weight that is different from the usual weight.[129] The decrease in BMR is rapid during the first weeks, then proceeds at a slower rate.[128,130] Thus, the initial, rapid decrease in BMR is not entirely accounted for by a corresponding loss of metabolically

active tissue.[128,131] (See also Refs. 142 and 145.) An analysis of fasting and semi-starvation studies suggests that about 25%–30% of the short-term decrease in BMR may be independent of the loss in lean body mass, indicating either a change in the composition of FFM or an alteration of cellular metabolism.[130] The dramatic effect of refeeding on BMR, which returns to normal within a few days without a parallel increase in body weight,[128] further supports the hypothesis that the decrease in metabolic rate during acute dietary restriction is of larger magnitude than what could be expected from tissue losses. A downregulation of BMR might be related to the decrease in noradrenaline appearance rate,[64,132] and in thyroid hormones[64,133,134] in response to fasting or semi-starvation. This possibility is supported by the prevention or the reversal of the metabolic response to acute energy restriction by replacement with either levodopa or T_3.[133,134] The enhancement of the thermic response to adrenaline infusion by prior starvation is consistent with a reduction in SNS activity.[135] A decrease in SNS outflow might also explain the significant reduction in the energy cost of glucose storage observed after underfeeding normal adults for seven days.[136] Similarly, it might account for the absence of any increase in the metabolic rate of undernourished patients exposed to mild cold. As their thermic response to adrenaline is conserved, the defect in cold-induced thermogenesis following weight loss cannot be ascribed to a decrease in tissue responsiveness. In these patients, the thermic response to mild cold is fully restored after weight gain.[137] Similar functional defects are apparent in children. Malnourished infants switched from 28°C to 24°C respond with a progressive fall in central temperature. When recovered, these infants increase their BMR by 20% in response to temperature lowering, and are able to maintain their body temperature within the normal range without shivering.[138] A decrease in skeletal muscle respiration proportional to the change in BMR in malnourished infants further supports a decrease in cellular metabolism.[139] However, metabolic rate measurements in chronically undernourished children may provide results that apparently conflict with those observed during acute energy restriction in adults.

Surprisingly, BMR may be approximately normal relative to body weight, and may even appear higher in those infants who are the most wasted.[140] Similar results are obtained in chronically undernourished adults. BMR related to body weight appears higher in the undernourished than in the controls, although in absolute terms it is significantly lower.[141] Total body cell mass (BCM) is reduced even in moderate deficiency, but

muscle mass seems to decrease linearly with the increasing severity of undernutrition, whereas visceral cell mass shows limited changes. For example, body fat and BCM are reduced by 29%, whereas muscle mass is decreased by 41% in severe undernutrition.[142] This is in general agreement with animal experiments, although a larger range in the weight change of some organs in response to nutritional manipulations may be observed.[143] Thus, taking into account the respective metabolic rate of tissues,[14] it is not surprising that BMR related to BCM or FFM may appear falsely high in some cases of severe malnutrition. This increase in metabolic rate related to FFM or body weight does not preclude an overall depression of tissue metabolism.[140,144] In infants, protein turnover, synthesis, and breakdown rates are significantly lower (approximately 40%) before than after recovery from malnutrition. Diminished substrate cycling is observed in adults during short-term underfeeding, and the resulting decrease in both protein turnover and glucose cycling may account for about 35% of the reduction in 24hEE.[145] Conversely, the malnourished infants' increase in energy expenditure in response to dietary treatment is an early event,[140] concurrent with a dramatic increase in growth rate[41] and in protein turnover,[146] suggesting that it is not entirely accounted for by the change in body composition. Fasting metabolic rate (related to body weight) during rapid growth is equal to, or even higher than,[140,144] the recovered values, a fact that further strengthens this possibility. Another way to look at the reality of metabolic rate downregulation would be to measure the cost of growth, which might be decreased by a higher metabolic efficiency. Although an overall increase in food efficiency may exist during the early period of catch-up growth,[41] the cost of growth during recovery from malnutrition generally appears similar to that of the normal infant.[146] However, longitudinal studies may easily overlook any enhancement in metabolic efficiency if it is short lived.[147]

Effects on Substrate Use

The classic studies of starvation showed that endogenous glucose production amounts to about 180 g/d after a few days' fast, of which 80% is oxidized by the brain.[148] The bulk of gluconeogenic precursor is provided by body proteins.[148] After five or six weeks of starvation, ketone bodies have replaced glucose as the predominant fuel for brain metabolism,[149] and glucose production has decreased to about 86 g/d, of which about 50% comes from recycled lactate and pyruvate.[150] Thus,

fasting endogenous glucose production relies on gluconeogenesis much earlier than previously expected[151] and both decrease as peripheral glucose uptake diminishes. The reduced demand in gluconeogenic precursors is paralleled by a significant reduction in protein breakdown, which favors sparing of tissue proteins.[152] In parallel, there is a state of insulin resistance at both the hepatic and peripheral levels, and normal insulin sensitivity is restored after refeeding.[153] From the beginning, fat is the predominant fuel, the respiratory quotient falling to 0.70 by the fourth day of fasting.[148] Observations made in malnourished children are consistent with these results. Protein contributes little to total energy expenses.[154] The average daily amount of glucose oxidized is also lower, and glucose oxidation decreases more rapidly in response to fasting before recovery than after recovery,[154] which does not demonstrate, but is suggestive of, a reduction in glucose production.[148] It is striking that the reduction in urinary nitrogen output, which occurs in lean, starving adults, is proportionate to the simultaneous decrease in metabolic rate, suggesting that protein sparing might result from the general reduction in fuel use rather than from a specific fat-related mechanism.[155] However, the protein-conserving effect of growth hormone under fasting conditions has been ascribed to the stimulation of lipolysis,[156] thus demonstrating that free fatty acids are important protein-sparing agents.

Therefore, underfeeding results in a significant decrease in metabolic rate related to tissue loss and, to a lesser extent, to a reduction in the activity of the remaining tissues. This reduction in metabolic rate can be viewed as an attempt to save energy in the face of reduced intakes. However, such an economy goes far beyond the physiologic range of adaptation, as it is obtained at the expense of growth and body composition. Furthermore, its only flexibility depends on the use of functional tissues as a source of substrates. Thus, any interfering increase in energy demand may entirely jeopardize this precarious balance.

EFFECTS OF DISEASE STATES

The compromise achieved by malnourished infants may be imperiled in two obvious ways. The first one would be to increase energy expenditure, for example, through fever. The other one would be to disrupt the adaptation of protein metabolism by a selective increase in protein breakdown, leading to a rapid net loss of nitrogen.

Effects on Protein Metabolism

It has been known for a long time that urinary nitrogen output is dramatically increased during infection and trauma, in sharp contrast with what is observed during malnutrition. In vivo studies reveal that this is accounted for by a disproportionate increase in protein breakdown over synthesis. However, infusion of counterregulatory hormones to healthy adults falls short of reproducing the massive 20- to 40-g/d nitrogen losses observed after trauma or thermal injury, although it does induce plasma hormone elevations typical of severe stress.[157] In contrast, peptides isolated from the plasma of patients with sepsis or trauma are able to specifically induce a dramatic increase in protein breakdown, without affecting synthesis.[158,159] This specific effect on protein breakdown is reproduced in vitro by incubating muscle biopsies with the culture medium of activated macrophages, suggesting that cytokines are involved. Cytokines are a large family of cellular regulators that include interleukins (ILs), tumor necrosis factors (TNFs), interferons (IFNs), and others. They play a pivotal role in the immune system, but their effects extend far beyond the immune cells. Each cytokine may interact with different cell types, thus exhibiting pleiotropic functions in different target cells.[160] Conversely, many different cytokines may exert the same biological actions—a property known as redundancy. Administration of recombinant tumor necrosis factor (rTNF) or recombinant interleukin-1α (rIL-1α) to animals results in accelerated muscle proteolysis.[161] Chronic administration of either rTNF or rIL-1α induces muscle wasting.[161,162] Furthermore, there is a synergistic effect between TNF and IL-1 on the stimulation of proteolysis.[161] In contrast with earlier expectations, the effect of cytokines on protein breakdown is only partially abolished by inhibitors of prostaglandin (PG) E_2 synthesis.[163] Moreover, proteolysis in response to rIL-1α is not mediated by glucocorticoids. In humans, administration of rTNF also results in a dose-dependent rise in the amino acid efflux from muscle,[164] confirming the central role of cytokines in the protein losses associated with various acute and chronic diseases. In muscle, the underlying mechanism involves the induction of E3 ubiquitin ligases gene (Atrogin-1 and muscle-specific ring finger protein 1 or MuRF1) leading to muscle atrophy through the selective action of the ubiquitin-dependent proteasome on muscle cell gene products. The induction of these ubiquitin ligases can be down regulated by insulin-like growth factor I (IGF-I).[165] As in adults, the rates of protein synthesis and breakdown increase markedly in response to infection in children.

However, breakdown increases more than synthesis, leading to a net loss of nitrogen.[166] Although malnutrition blunts the response to infection, infected malnourished children have protein turnover rates twice as high as uninfected ones, the relative increase in protein breakdown resulting in a negative protein balance.[166] Increased protein breakdown was first noted in sepsis or trauma patients, but a significant change may also be observed in response to much less severe conditions, such as vaccinations,[167,168] suggesting that even mild infections are likely to offset the fragile equilibrium of undernourished children.

It should be pointed out that protein metabolism imbalance per se (i.e., protein breakdown rates greater than synthesis) results in a net protein loss and thus may lead to protein deficiency. Growth failure in Crohn's disease might be accounted for by such a mechanism. Growth failure is often associated with Crohn's disease and may even precede symptoms. These patients have a short stature, but in most cases their body weight is close to the expected weight for height, but their FFM is significantly lower than in age-matched controls. As growth hormone (GH) secretion is normal,[169] a search for nutritional deficiencies was undertaken. A reduction in energy intakes is noted in some reports, but not all, whereas protein intakes appear consistently within the ranges of recommended dietary allowances. Furthermore, energy losses in the feces are not different from those of normal children, and intestinal protein losses are only slightly increased over control values. When measured, energy expenditure related to FFM is comparable to results in matched controls[170] and the preserved FM argues against a significant energy deficit.[171] Despite the lack of evidence in favor of nutritional deficiencies, energy supplements do result in a significant increase in growth rate, suggesting that a nutritional deficiency is indeed responsible for growth failure in Crohn's disease. That such a deficiency could not be ascribed to an overt imbalance between intakes and needs suggests that it might result from an abnormal utilization of available substrates. Indeed, abnormal protein turnover rates are consistently found in these patients.[172,173] Moreover, alterations in protein metabolism are correlated with the intensity of the inflammatory syndrome[172,173] and are consistent with an increased production of TNF-α and IL-1.[174] Besides the dramatic effect of energy supplements, another line of evidence points toward a nutritional deficiency in Crohn's disease. The plasma concentration of IGF-I is significantly decreased in these patients and further correlates with growth rates.[175] Reduced plasma IGF-I concentrations, despite normal or increased circulating GH levels, is a trait

of malnutrition and measurements of IGF-I levels have been proposed as a highly efficient way of monitoring nutritional rehabilitation.[176] Of particular interest is the fact that plasma IGF-I concentration is closely correlated with protein balance in both animal and man.[176,177] In the rat, IGF-I level is also correlated with protein intake.[178] Furthermore, low fasting IGF-I concentrations cannot be restored to normal levels by refeeding protein-free diets.[177] This is related to the direct modulation of IGF-I expression by essential amino acid availability, independently of insulin and glucocorticoid contributions.[179] Likewise, IGF-I levels in infected malnourished children are sensitive to dietary protein alterations.[180] High-protein diets result in higher IGF-I levels and in improved growth rates, 180 despite the possible direct effect of some cytokines on IGF-1 expression.[181] Therefore, the decrease in IGF-I levels in Crohn's disease pinpoints the critical importance of protein metabolism in growth failure. Indeed, nutritional supplementation results in elevated IGF-I concentrations and increased growth rates.[175] Food supplements induce a significant increase in protein synthesis which results in an increase in protein balance. In contrast, nutritional support does not seem to affect the elevated breakdown rates.[172] The favorable effect of the nutritional supplements could then be accounted for by the specific induction of protein synthesis by amino acids. Therefore, growth failure in Crohn's disease may be related to an abnormal utilization of substrates, due to an imbalance between protein synthesis and breakdown rates, the latter being specifically increased by the associated inflammatory syndrome. This may be exaggerated by direct effects of cytokines on the GH-IGF-1 axis,[182] not to mention the deleterious effects of steroid treatment on protein metabolism. Reduction in food intake would only exaggerate the protein debt by further reducing the synthesis-to-breakdown ratio, whereas nutritional support compensates for the increased breakdown rates by promoting protein synthesis.

Findings in rheumatoid arthritis (RA) are also consistent with a dominant effect of inflammatory syndromes on protein metabolism. RA is associated with a chronic inflammatory syndrome involving local cytokine production. The immunologic response occurs in affected joints and can also be observed and monitored in peripheral blood. The circulating level of some cytokines (IL-6) is significantly correlated with the commonly used markers of inflammatory syndromes, indicating that it is related to disease activity.[183] The measurement of ^{40}K and total body water in RA patients shows that their BCM is decreased, as compared to controls. In

addition, there is a strong dose–response between RA severity and the amount of BCM adjusted for stature: the more severe the RA, the lower the BCM.[184] BCM is also inversely associated with TNF-α production.[185] This decrease in BCM is important in two regards: it amounts to about one-third of what is thought to be a survivable loss and it occurs in patients with energy and protein intakes comparable to those of controls. Surprisingly, the FM measured by dual-energy x-ray absorptiometry is not very different in RA adult patients and in controls.[186] This last observation suggests that the covert nutritional deficiency responsible for the decrease in BCM in adults is not primarily related to energy imbalance. Alternatively, the increase in protein breakdown rates observed in these situations might account for the preferential loss in FFM,[187] particularly in muscle.[188]

A comparable increase in protein breakdown may be induced by ammonium chloride infusion to healthy volunteers, an effect also found in chronic renal failure (CRF). For example, proteolysis may be increased in children with CRF by up to threefold in relation to metabolic acidosis.[189] In contrast with cytokine-mediated protein breakdown, net proteolysis is related to changes in plasma cortisol levels, suggesting that it plays an important role in protein metabolism during metabolic acidosis. It should be pointed out that the acidosis-induced increase in net proteolysis may be quite deleterious to infants and children with inherited diseases of amino acid metabolism.

Therefore, the disruption of protein metabolism regulation in favor of breakdown, by cytokines overproduction during chronic inflammatory syndromes or by some glucocorticoid-mediated mechanisms during chronic renal failure, might well constitute an entry into severe malnutrition, independent of decreased food consumption.

Effects on Energy Expenditure and Energy Substrate Utilization

Fever is a common event in children. It is also an efficient way to increase energy expenditure. DuBois emphasized the striking similarity between fevers of greatly different origins by showing that metabolic rate has a much closer relationship with body temperature than with the nature of diseases. This relationship indicates that metabolic rate increases by an average 13% for each 1°C above normal temperature.[75] A comparable relationship is found in children, resting energy expenditure increasing by about 12% for each 1°C above normal temperature.[190] For example,

during acute malaria the mean increment in RMR is about 30%.[191] This would amount to 1200 kcal/d expenditure in a 5-year-old child, or to a 250–300 g/d weight loss, should the child not eat.[190] It is noteworthy that the increase in RMR for each degree of increase in temperature is positively correlated to the ratio of actual weight to the expected weight for height, suggesting that undernutrition blunts the RMR response to fever.[190] Although the rise in metabolic rate was first related to shivering,[75] there is some evidence that other mechanisms are operative. An endotoxin bolus delivered to healthy volunteers induces a significant pyrogenic response (+2°C in body temperature) within 90 minutes, maximal at 180 minutes, and sustained for more than six hours. Total body oxygen consumption is increased by 28% at two hours, when body temperature is about half-maximal, and remains elevated at six hours. Coordinately, splanchnic oxygen consumption is increased by 64% at two hours and is still increased by 47% at four hours.[192] The increase in splanchnic oxygen extraction represents approximately 80% of the increase in whole-body oxygen consumption at two hours, suggesting that shivering is not the major source of heat production at this time, although body temperature is still rising. This interpretation is consistent with the absence of increase in leg oxygen extraction at two hours.[192]

Both the pharmacological approach and the gene knockout technology stress the importance of cytokines in the development of this thermic response. However, some discrepancies persist, which may be related to cytokine redundancy and to the models used. For example, IL-1β and IL-1 type-1-receptor knockout mice challenged with low or high dose of lipopolysaccharide (LPS) display a thermic response similar to wild-type mice, whereas fever, anorexia, and weight loss resulting from turpentine injection are completely abolished.[193] Similarly, the significant increase in plasma IL-6 levels observed in wild-type mice injected with turpentine is virtually absent in the IL-1β knockout mice.[194] Injection of turpentine into IL-6 knockout mice also fails to induce fever, anorexia, and weight loss.[195] All these data support the direct implication of IL-1β and IL-6, but also TNF-α, in the induction of fever and sickness behaviors. The thermic response to LPS is dependent upon PG synthesis, as it is blocked by indometacin, as well as by cyclooxygenase 2 (COX2) or PG receptor gene disruption. PG receptor (subtype EP3) knockout mice fail to mount a febrile response to LPS, IL-1β, and PGE2. The pyrogenic effect of IL-1β may also be inhibited by pretreatment with either a receptor antagonist or a monoclonal antibody to CRH.

In parallel to their effects on energy expenditure, cytokines affect food intake in both animals[196] and humans.[197] Chronic administration of IL-1 to rats causes persistent inhibition of feeding.[196] Strikingly, this effect is not obtained in fish oil–fed rats.[198] Anorexia induced by IL-1 is mediated by CRF,[198] melanocortin-3/4 receptors[199] and, to a lesser extent, by PGs.[200] Other cytokines including IL-2, IL-6, and IFN-α also stimulate hypothalamic CRF expression and/or release.[201] Thus, CRF seems to modulate both aspects of energy balance regulation in the rat, that is, food intake and energy expenditure. CRF effect on energy expenditure is mediated by a stimulation of SNS outflow, as reflected by a dramatic increase in the secretion of both adrenaline and noradrenaline,[99,202] explaining that the increase in body temperature and oxygen consumption induced by experimental inflammatory syndromes may be significantly reduced by β-adrenoreceptor blockade.[203,204] The chronic administration of IL-1β induces a sustained rise in body temperature and a reduction in food intake. The simultaneous increase in energy expenditure and decrease in energy intake result in a significant reduction in body weight gain of the experimental rats compared to controls, indicating that low levels of IL-1β release, maintained over periods of several days, could be responsible for changes in energy balance during chronic infection or inflammation.[205]

Chronic cytokine overproduction also affects energy expenditure in humans. In RA, for example, RMR adjusted for BCM is about 12% higher than in adult controls. Furthermore, there is a positive correlation between cytokine production and the increase in RMR, cytokine production and BCM accounting for about 20% and 50% of RMR variability, respectively.[184] RMR adjusted for FFM is increased by about 20% in children with systemic juvenile rheumatoid arthritis (JRA) as compared to controls. As a result, they are stunted and their FFM is reduced. Oligo and polyarticular JRA children still display an 8% increase in RMR over controls.[206] Similarly, RMR is increased in stable, cystic fibrosis (CF) patients by 10%–25% above control values,[207,208] a change which is not accounted for by the increase in the oxygen cost of breathing.[207] There is a positive relationship between RMR and TNF-α levels,[209] and adrenaline levels, TNF-α accounting for about 27% of RMR variability. Contrary to RA patients who have a significant reduction in FFM but a relatively conserved FM, both FFM and FM are significantly reduced in CF patients.[210] Thus, besides the dramatic thermic response to infectious disease or injury, cytokines may also mediate lower-magnitude increases in metabolic rate, which might lead to a progressive depletion of energy

stores if sustained for weeks or month.

Infection also induces important changes in the production or utilization of energy substrates. Infected patients present glucose intolerance, irrespective of the severity of the infection and of the infective agent.[211] There is a significant increase in endogenous glucose production, which is not suppressible by glucose infusion and partially accounts for the increase in plasma glucose concentration. The stimulation of hepatic glucose production is principally mediated by increased levels of catecholamines and an increased glucagon-to-insulin ratio.[212] Although whole-body glucose turnover is increased, insulin ability to stimulate glucose uptake is reduced. Indeed, peripheral glucose utilization is impaired when compared to controls.[213] It is noteworthy that marked insulin resistance is present in previously healthy individuals with uncomplicated viral infections.[214] Insulin resistance is readily apparent four hours after the administration of LPS to healthy volunteers. The increase in glucose turnover and the peripheral insulin resistance may be partially accounted for by a sustained β_2 stimulation[215] and by the increase in free fatty acid appearance rate in infected humans and animals,[204,212] an increase related to adrenergic stimulation.[212] Cytokines, particularly TNF-α, also contribute to insulin resistance by a direct effect on the insulin signaling cascade[216] and by increasing free fatty acid appearance rate by a stimulation of adipose tissue lipolysis.[217] Similarly, IL-6 is a potent modulator of fat metabolism in humans, increasing lipolysis and fatty acid oxidation.[218] This might be related to the suppression of adipocyte glyceroneogenesis, through the inhibition of cytosolic phosphoenolpyruvate carboxykinase (PEPCK-C) gene expression, which results in a dramatic increase in free fatty acid release by adipose tissue.[219]

When compared to healthy controls, fat oxidation is increased in septic patients and falls by a much lesser percentage in response to glucose and insulin.[220] Indeed, at the end of an insulin clamp, fat oxidation still contributes about 58% to metabolic rate in the septic patients, whereas it amounts to only 23% in controls.[220] Moreover, fat oxidation rate appears to be positively related to the severity of sepsis. Despite an increase in glucose turnover, fat thus appears as a preferred calorie source during sepsis.[221] Endotoxin administration to humans induces an increase in adrenaline and cortisol levels, whereas glucagon and insulin do not change.[192] Simultaneously, there is a significant rise in free fatty acid levels. Splanchnic glucose output is also increased and a progressive

hyperglycemia ensues. The reduction in glucose utilization, like that observed in sepsis, is related to impaired nonoxidative glucose disposal and not to abnormal glucose oxidation. Increases in plasma glucose, free fatty acid turnover (126%), and fat oxidation (60%), and similar variations in hormone concentrations follow the injection of rTNF to human volunteers, further supporting the role of cytokines in the development of the metabolic response to acute infection or injury.[222] It is interesting that an increase in both free fatty acid and glycerol concentrations is also observed in CF patients despite a reduction in FM, suggesting that chronic inflammatory syndromes may result in changes in the pattern of substrate utilization qualitatively comparable to those observed during acute infection.

ENERGY REQUIREMENTS IN DISEASE

Energy

The Food and Agricultural Organization of the World Health Organization (FAO/WHO) has published detailed estimates for energy requirements in infants and children of various ages.[223] In the adult, baseline energy needs, that is, energy needed to cover the metabolic response to food and the energy cost of minor activities, are assumed to be equal to 1.35 × BMR, and the time spent sleeping equal to BMR. This is considered as a survival requirement and is of practical value only for estimating the short-term needs of inactive and dependent individuals. Otherwise, estimates should include the energy cost of various occupational activities. However, it is not possible to specify with any confidence the allowance that should be made for a desirable level of physical activity in infants and children. Thus, energy requirements from birth to 10 years have been derived from the observed intakes of healthy children growing normally. Beyond 10 years, calculations are based on published BMR measurements, incremented for the cost of growth and for a cost of activity assumed to be about 1.6 × BMR. Free-living measurements of 24hEE corrected for the energy deposited during growth show that estimated energy requirements are substantially higher than total energy expenditure.[224] Furthermore, calculated energy expenditure also exceeds the energy intakes of 10- to 17-year-old children and adolescents.[224] Clearly, the difficulties encountered in achieving an accurate prediction of the energy needs in healthy infants and children would make unrealistic any such attempt in sick children, as the issue is further confused by the opposing effects of underfeeding and

disease on energy expenditure. Then the only option left is to try to identify some agreement about the range of variations in metabolic rate induced by diseases.

Studies in 13–60 month children with acute measles-associated underfeeding suggest that negative energy balance is related to a dramatic reduction in gross energy intake concomitant with a sustained level of energy expenditure. Metabolic rate after recovery is not significantly different from the values obtained during acute infection, although an average 20% increase would have been expected from the presence of fever.[225] The surprising stability of metabolic rate during acute infection might be accounted for by the balanced effects of underfeeding and infection. In any case, maintenance energy requirement (MER) determined at zero energy balance is 64 kcal/kg/d,[225] a figure similar to the MER of thriving infants.[42] In these infants, the safe level of energy intake is estimated to be between 91 and 100 kcal/kg/d,[225] as compared to the 81–87 kcal/kg/d free-living 24hEE of normal infants, 224 or the average 103 kcal/kg/d estimated energy requirements in the toddler group. It is further possible to evaluate the consequences of sepsis and trauma on energy expenditure. Earlier measurements in adults indicated 20%–50% increases in metabolic rate in response to severe infection or multiple trauma. Studies performed in critically ill patients without malnutrition indicate that the mean increase in fasting metabolic rate ranges between 5%[226] and 14% to 18% over predicted values.[227] This is consistent with reports comparing septic patients to matched controls and showing a 15%–19% increase in fasting metabolic rate over normal values.[228] Comparable estimates are obtained in traumatized children. RMR peaks at three days after injury, representing an 8% increase over predicted values or a 14% increase over replicate measurements obtained after discharge.[229] After surgery, the mean increase in RMR over baseline reaches about 15%. In these conditions, RMR peaks at four hours postoperatively and returns to baseline levels by 12 hours, thus resulting in a modest overall change in metabolic rate. Furthermore, the increase in RMR is related to the nature of the surgical procedure, the maximal rise being about 5% for minor surgery and 19% for major operations.[230] A similar trend toward decreasing estimates is observed regarding the energy expenses following burn injury. RMR in burned infants is independent of burn size and of the time elapsed between burn injury and measurements. Indeed, these two factors only account for 24% and 21% of the variation in RMR above predicted values, which is primarily dependent on body size.[231]

Furthermore, β-adrenergic blockade induces a substantial decrease in RMR in severely burned children.[231] As a result, the amount of energy required to achieve energy balance may be much lower than previously estimated. Actually, caloric delivery in excess of 1.2 × RMR might only result in increased FM without changes in lean body mass.[232] A comparable reevaluation of the change in metabolic rate resulting from severe head injury suggests that patients who are properly taken care of have a lower than expected increase in RMR.[233] Therefore, it is likely that an energy intake equivalent to 1.5 × BMR would meet the requirements of most septic, traumatized, or burned infants and children. That increases in RMR after trauma, sepsis, or burn injuries are somewhat comparable is consistent with the existence of common mediators. Although the increase in energy expenditure resulting from disease appears to be substantially lower than previously estimated, applying equations that were originally developed for healthy, nonhospitalized individuals to predict the energy requirements of critically ill children could result in significant errors and may lead to provision of inappropriate nutritional support.[234]

Energy Substrates and Amino Acids

Energy intake should provide glucose and fat in amounts adapted to insulin resistance and to the preferential oxidation of lipids. Physiologic considerations also suggest that the amount of glucose infused to malnourished, uninfected patients be limited. In adults, glucose oxidation levels off at about 4 mg/kg/min in response to increasing glucose infusion rates.[235] A similar limitation exists in 6-month-old infants, with a maximal oxidative glucose disposal rate of 12 mg/kg/min.[15] Furthermore, this maximal oxidative disposal decreases in parallel to energy expenditure with age, amounting to about 9 mg/kg/min at 6 years and about 6 mg/kg/min at 10 years.[15] In the uninfected infant, glucose infusion in excess of the maximal oxidative glucose disposal rate only results in inhibiting fat oxidation whether of endogenous or exogenous origin,[15] and may adversely affect protein metabolism and net protein deposition.[236] Thus, expecting some benefit from high glucose intakes in infected infants seems hazardous. On the contrary, glucose intakes should be lower because of the glucose intolerance. Glucose intakes in the range of the physiologic endogenous glucose production of the corresponding age range[237] are sufficient to meet the needs of glucose-requiring tissues. In these conditions, the clearance of intravenous emulsions as well as their oxidation are enhanced in septic patients. The other goal is to equilibrate

the increased proteolysis by promoting protein synthesis. In the healthy adult, protein synthesis is primarily regulated by amino acid inflow. Similarly, amino acid infusion decreases net nitrogen losses in septic patients; the higher the amino acid intake, the more rapid the effect. In burned infants and toddlers, amino acid intakes in the range of 2.5–3 g/kg/d efficiently support recovery.[238]

The Inflammatory Syndrome: A Case for a Pharmacologic Approach

Providing nutrients may not be enough in many of these situations. Nutritional support should be aimed at avoiding unnecessary increases in metabolic rate or, better, at controlling the disease-induced increase in energy expenditure. Obviously, attention should be given to ambient temperature and humidity, particularly in infants and burn patients. Furthermore, the use of inhibitors of either cytokine expression or COX, as well as of blockers of SNS peripheral effects, should be considered. As glucocorticoids suppress the production and the action of several cytokines as well as the production of PGs, it appeared warranted to test their effects in vivo in septic patients. The results are disappointing and do not support their use in sepsis, probably because corticoids could be administered only after cytokine release.[239] The use of molecules (antibodies or receptor analogs) directed against cytokines[240] or modulating inflammation[241] offers very interesting possibilities. Another way to curb the inflammatory response is to interfere with signal transduction in target cells through inhibitors of the COX. Indeed, pretreatment with ibuprofen results in the complete suppression of fever, tachycardia, and increase in stress hormones after endotoxin injection, as compared to the subjects who were not pretreated, despite an identical rise in TNF in both groups.[242] Similarly, the use of β-adrenergic receptor antagonists may substantially diminish energy expenditure.[204] If these dispositions may reduce metabolic rate, in many cases they will have little effect on the accelerated protein breakdown.

A pharmacologic intervention would be even more pressing when nutrition support fails to prevent protein losses, as it is the case in sepsis or during cancer cachexia. Animal models confirm that protein wasting is not accounted for by the effects of underfeeding in these situations,[182] thus requiring an approach other than dietary therapy. Besides their effect boosting proteolysis, glucocorticoids have an inhibitory effect on protein synthesis, which could further exaggerate cytokine-induced protein losses.

Unfortunately, COX inhibitors seem to have only a partial and transient effect.[182] One remaining possibility is to directly inhibit cytokine production by new agents. Pretreatment with pentoxifylline inhibits TNF secretion and partially suppresses anorexia, weight loss, and muscle protein wasting in septic rats,[243] although fever, myalgia, and increased cortisol levels are unaffected in pretreated, endotoxin-injected humans.[244] However, these molecules are effective when administered prior to endotoxin or TNF injection, but little is known about their efficacy as therapeutic agents.

The other possibility is to antagonize the effects of cytokines on protein metabolism. Recombinant growth hormone (rGH) administered to postoperative patients induces a dramatic change in nitrogen balance, despite low energy intakes. Furthermore, rGH treatment is able to reverse net whole-body and skeletal muscle protein catabolism in cancer patients.[245] This positive effect has been confirmed in severe sepsis and in patients with HIV-related wasting. Similarly, recombinant insulin-like growth factor I (rIGF-I) infusion to rTNF-treated animals preserves its anabolic action and induces a significant reduction in net protein losses in the face of high TNF levels.[246] It is noteworthy that decreased IGF-I levels, a common finding in acutely ill patients, may enhance protein catabolism. The protein anabolic effects of both rGH and rIGF-I may be substantially increased by using both agents simultaneously,[247] a synergy that might prevent GH resistance. Although recombinant hormones are not the only agents favoring net protein synthesis, available evidence suggests that they are the best candidates to date for improving the nutritional therapy of protein-wasting inflammatory syndromes through direct pharmacologic intervention. Indeed, data suggest that rGH treatment improves growth and clinical conditions of CF patients,[248] in children with chronic renal failure[249] or juvenile arthritis,[250] increases linear growth and lean body mass in patients with Crohn's disease[251] and, by and large, has substantial anabolic effects on whole-body protein and bone metabolisms in children on chronic steroid therapy.[252]

REFERENCES

1. McLean JA, Tobin G. *Animal and Human Calorimetry*. Cambridge, UK: Cambridge University Press; 1987.
2. Lukaski HC. Methods for the assessment of human body composition: traditional and new. *Am J Clin Nutr*. 1987;46:537–556.
3. Benedict FG. *Vital Energetics. A Study in Comparative Basal Metabolism.*

Publication No 503. Washington, DC: Carnegie Institute; 1938.
4. Ravussin E, Lillioja S, Anderson TE, et al. Determinants of 24-hour energy expenditure in man. Methods and results using a respiratory chamber. *J Clin Invest*. 1986;78:1568–1578.
5. Dauncey MJ. Metabolic effects of altering the 24h energy intake in man, using indirect and direct calorimetry. *Br J Nutr*. 1980;43:257–269.
5a. Ferraro R, Lillioja S, Fontvieille AM, et al. Lower sedentary metabolic rate in women compared with men. *J Clin Invest*. 1992;90:780–784.
5b. Brebbia DR, Altshuler KZ. Oxygen consumption rate and electroencephalographic stage of sleep. *Science*. 1985;150:1621–1623.
5c. Henry CJK, Hayter J, Rees DG. The constancy of basal metabolic rate in free-living male subjects. *Eur J Clin Nutr*. 1989;43:727–731.
6. Ravussin E, Swinburn BA. Energy metabolism. In: Stunkard AJ, Wadden TA, eds. *Obesity: Theory and Therapy*. New York: Raven Press; 1993:97–123.
7. Schofield WN. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr*. 1985;39(suppl 1):5–41.
8. Ravussin E, Lillioja S, Knowler WC, et al. Reduced rate of energy expenditure as a risk factor for body weight gain. *N Engl J Med*. 1988;318:467–472.
9. Garby L, Garrow JS, Jørgensen B, et al. Relation between energy expenditure and body composition in man: specific energy expenditure in vivo of fat and fat-free tissue. *Eur J Clin Nutr*. 1988;42:301–305.
10. Bogardus C, Lillioja S, Ravussin E, et al. Familial dependence of the resting metabolic rate. *N Engl J Med*. 1986;315:96–100.
11. Toubro S, Sorensen TI, Ronn B, et al. Twenty-four-hour energy expenditure: the role of body composition, thyroid status, sympathetic activity, and family membership. *J Clin Endocrinol Metab*. 1996;81:2670–2674.
12. Holliday MA. Metabolic rate and organ size during growth from infancy to maturity and during late gestation and early infancy. *Pediatrics*. 1971;47:169–179.
13. Weinsier RL, Schutz Y, Bracco D. Reexamination of the relationship of resting metabolic rate to fat-free mass and to metabolically active components of fat-free mass in humans. *Am J Clin Nutr*. 1992;55:790–794.
14. Chessex P, Reichman BL, Verellen G, et al. Influence of postnatal age, energy intake, and weight gain on energy metabolism in the very-low-birth-weight infant. *J Pediatr*. 1981;99:761–766.
15. Bresson JL, Narcy P, Putet G, et al. Energy substrate utilization in infants receiving total parenteral nutrition with different glucose to fat ratios. *Pediatr Res*. 1989;25:645–648.
16. Sparti A, DeLany JP, de la Bretonne JA, et al. Relationship between resting metabolic rate and the composition of the fat-free mass. *Metabolism*. 1997;46:1225–1230.
17. Heymsfield SB, Gallagher D, Kotler DP, et al, Body-size dependence of resting energy expenditure can be attributed to nonenergetic homogeneity of fat-free mass. *Am J Physiol*. 2002;282:E132–E138.
18. Koong LJ, Nienaber JA, Mersmann HJ. Effects of plane of nutrition on organ size and fasting heat production in genetically obese and lean pigs. *J Nutr*. 1983;113:1626–1631.
19. Garby L, Lammert O. An explanation for the non-linearity of the relation between energy expenditure and fat-free mass. *Eur J Clin Nutr*. 1992;46:235–236.
20. Chan ATH, Fleming CR, O'Fallon WM, Huitzenga KA. Estimated versus measured basal energy requirements in patients with Crohn's disease. *Gastroenterology*. 1986;91:75–78.

21. Blaxter K. *Energy Metabolism in Animals and Man*. Cambridge: Cambridge University Press; 1989.
22. Heusner AA. What does the power function reveal about structure and function in animals of different body size? *Ann Rev Physiol*. 1987;49:121–133.
23. Krebs HA. Body size and tissue respiration. *Biochim Biophys Acta*. 1950;4:249–269.
24. Brand MD, Couture P, Else PL, Withers KW, Hulbert AJ. Evolution of energy metabolism. Proton permeability of the inner membrane of liver mitochondria is greater in a mammal than in a reptile. *Biochem J*. 1991;275:81–86.
25. Hulbert AJ, Else PL. Membranes and the setting of energy demand. *J Exp Biol*. 2005;208:1593–1599.
26. Speakman JR. Body size, energy metabolism and lifespan. *J Exp Biol*. 2005;208:1717–1730.
27. Else PL, Hulbert AJ. Evolution of mammalian endothermic metabolism: "leaky" membranes as a source of heat. *Am J Physiol*. 1987;253:R1–R7.
28. Gregg VA, Milligan JP. In vitro energy cost of Na^+, K^+-ATPase activity and protein synthesis in muscle from calves differing in age and breed. *Br J Nutr*. 1982;48:65–71.
29. Swaminathan R, Chan ELP, Sin LY, et al. The effect of ouabain on metabolic rate in guinea-pigs: estimation of energy cost of sodium pump activity. *Br J Nutr*. 1989;61:467–473.
30. Lin MH, Romsos DR, Akera T, Leveille GA. Functional correlates of Na^+, K^+-ATPase in lean and obese (ob/ob) mice. *Metabolism*. 1981;30:431–438.
31. Poehlman ET, Toth MJ, Webb GD. Sodium-potassium pump activity contributes to the age-related decline in resting metabolic rate. *J Clin Endocrinol Metab*. 1993;76:1054–1057.
32. Astrup A, Buemann B, Christensen NJ, et al. The contribution of body composition, substrates, and hormones to the variability in energy expenditure and substrate utilization in premenopausal women. *J Clin Endocrinol Metab*. 1992;74:279–286.
33. Milligan LP, McBride BW. Shifts in animal energy requirements across physiological and alimentational states. Energy costs of ion pumping by animal tissues. *J Nutr*. 1985;115:1374–1382.
34. McBride BW, Milligan LP. Magnitude of ouabain-sensitive respiration in the liver of growing, lactating and starved sheep. *Br J Nutr*. 1985;54:293–303.
35. Rosengurt E. Stimulation of Na influx, Na-K pump activity and DNA synthesis in quiescent cultured cells. *Adv Enzyme Regul*. 1981;19:61–85.
36. Denne SC, Kalhan SC. Leucine metabolism in human newborn. *Am J Physiol*. 1987;253:E608–E615.
37. Gill M, France J, Summers M, et al. Simulation of the energy costs associated with protein turnover and the Na^+, K^+-transport in growing lambs. *J Nutr*. 1989;119:1287–1299.
38. Brooke OG, Alvear J, Arnold M. Energy retention, energy expenditure, and growth in healthy immature infants. *Pediatr Res*. 1979;13:215–220.
39. Reichman BL, Chessex P, Putet G, et al. Partition of energy metabolism and energy cost of growth in the very low-birth-weight infant. *Pediatrics*. 1982;69:446–451.
40. Payne PR, Waterlow JC. Relative energy requirement for maintenance, growth, and physical activity. *Lancet*. 1971;2:210–211.
41. Ashworth A. Metabolic rates during recovery from protein-calorie malnutrition: the specific need for a new concept of specific dynamic action. *Nature*.

1969;223:407–409.
42. Fomon SJ, Haschke F, Ziegler EE, Nelson SE. Body composition of reference children from birth to age 10 years. *Am J Clin Nutr*. 1982;35:1169–1175.
43. Heim T, Verellen G, Chessex P, et al. Partition of energy metabolism in the very-low-birth-weight infant. In: Stern L, Bard H, Friis-Hansen B, eds. *Intensive Care of the Newborn*. New York: Masson Publishing Inc; 1983:169–181.
44. Roberts SB, Young VR. Energy costs of fat and protein deposition in the human infant. *Am J Clin Nutr*. 1988;48:951–955.
45. Forbes GB, Brown MR, Welle SL, Lipinski BA. Deliberate overfeeding in women and men: energy cost and composition of the weight gain. *Br J Nutr*. 1986;56:1–9.
46. Walker J, Roberts SL, Halmi KA, Goldberg SC. Caloric requirements for weight gain in anorexia nervosa. *Am J Clin Nutr*. 1979;32:1396–1400.
47. Kaye WH, Gwirtsman HE, Obarzanek E, George DT. Relative importance of calorie intake needed to gain weight and level of physical activity in anorexia nervosa. *Am J Clin Nutr*. 1988;47:989–994.
48. Fomon SJ. Infant Nutrition. Philadelphia: WB Saunders Company, 1974.
49. Séguin A, de Lavoisier AL. Premier mémoire sur la respiration des animaux. *Mémoires de l'Académie des Sciences*, 1789.
50. D'Alessio DA, Kavie EC, Mozzoli MA, et al. Thermic effect of food in lean and obese men. *J Clin Invest*. 1988;81:1781–1789.
51. Tai MM, Castillo P, Pi-Sunyer FX. Meal size and frequency: effect on the thermic effect of food. *Am J Clin Nutr*. 1991;54:783–787.
52. Hill JO, DiGirolamo M, Heymsfield SB. Thermic effect of food after ingested versus tube-delivered meals. *Am J Physiol*. 1985;248:E370–E374.
53. Nacht CA, Schutz Y, Vernet O, et al. Continuous versus single bolus enteral nutrition: comparison of energy metabolism in humans. *Am J Physiol*. 1986;251:E524–E529.
54. Vernet O, Christin L, Schutz Y, et al. Enteral versus parenteral nutrition: comparison of energy metabolism in lean and moderately obese women. *Am J Clin Nutr*. 1986;43:194–209.
55. Robinson SM, Jaccard C, Persaud C, et al. Protein turnover and thermogenesis in response to high-protein and high-carbohydrate feeding in men. *Am J Clin Nutr*. 1990;52:72–80.
56. Schwartz RS, Ravussin E, Massari M, et al. The thermic effect of carbohydrate versus fat feeding in man. *Metabolism*. 1985;34:285–293.
57. Hurni M, Burnand B, Pittet P, Jéquier E. Metabolic effects of a mixed and a high-carbohydrate low-fat diet in man, measured over 24h in a respiration chamber. *Br J Nutr*. 1982;47:33–42.
58. Maffeis C, Schutz Y, Grezzani A, et al. Meal-induced thermogenesis and obesity: is a fat meal a risk factor for fat gain in children? *J Clin Endocrinol Metab*. 2001;86:214–219.
59. DeFronzo RA, Tobin JD, Andres R. Glucose clamp technique: a method for quantifying insulin secretion and resistance. *Am J Physiol*. 1979;237:E214–E223.
60. Thiebaud D, Schutz Y, Acheson K, et al. Energy cost of glucose storage in human subjects during glucose-insulin infusions. *Am J Physiol*. 1983;244:E216–E221.
61. Ravussin E, Acheson KJ, Vernet O, et al. Evidence that insulin resistance is responsible for the decreased thermic effect of glucose in human obesity. *J Clin Invest*. 1985;76:1268–1273.
62. Flatt JP. The biochemistry of energy expenditure. In: Bray G, ed. *Recent Advances in Obesity Research. II*. London: Newman Publishing Ltd; 1978:211–228.
63. Berne C, Fagius J, Niklasson F. Sympathetic response to oral carbohydrate

administration. Evidence from microelectrode nerve recording. *J Clin Invest*. 1989;84:1403–1409.
64. O'Dea K, Esler M, Leonard P, et al. Noradrenaline turnover during under- and overeating in normal weight subjects. *Metabolism*. 1982;31:896–899.
65. Acheson K, Jéquier E, Wahren J. Influence of β-adrenergic blockade on glucose-induced thermogenesis in man. *J Clin Invest*. 1983;72:981–986.
66. Acheson KJ, Ravussin E, Wahren J, Jéquier E. Thermic effect of glucose in man. Obligatory and facultative thermogenesis. *J Clin Invest*. 1984;74:1572–1580.
67. Schwartz RS, Jaeger LF, Veith RC. Effect of clonidine on the thermic effect of feeding in humans. *Am J Physiol*. 1988;254:R90–R94.
68. Welle S, Schwartz RG, Statt M. Reduced metabolic rate during β-adrenergic blockade in humans. *Metabolism*. 1991;40:619–622.
69. Spraul M, Ravussin E, Fontvieille AM, et al. Reduced sympathetic nervous activity. A potential mechanism predisposing to body weight. *J Clin Invest*. 1993;92:1730–1735.
70. Monroe MB, Seals DR, Shapiro LF, et al. Direct evidence for tonic sympathetic support of resting metabolic rate in healthy adult humans. *Am J Physiol Endocrinol Metab*. 2001;280:E740–E744.
71. Weinstein MR, Bell EF, Oh W. Energy intake, norepinephrine excretion, and oxygen consumption in low birthweight infants. *J Pediatr Gastroenterol Nutr*. 1985;4:774–777.
72. Mitchell P. Vectorial chemistry and the molecular mechanics of chemiosmotic coupling: power transmission by proticity. *Biochem Soc Trans*. 1976;4:399–430.
73. Flatt JP, Pahud P, Ravussin E, Jéquier E. An estimate of the P:O ratio in man. *Trends Biochem Sci*. 1984;9:466–468.
74. Rising R, Keys A, Ravussin E, Bogardus C. Concomitant interindividual variation in body temperature and metabolic rate. *Am J Physiol*. 1992;263:E730–E734.
75. DuBois EF. The basal metabolic rate in fever. *JAMA*. 1921;77:352–355.
76. Warwick PM, Busby R. Influence of mild cold on 24h energy expenditure in "normally" clothed adults. *Br J Nutr*. 1990;63:481–488.
77. Brück K. Temperature regulation in the newborn infant. *Biol Neonat*. 1961;3:65–119.
78. Hey E. Thermal neutrality. *Br Med Bull*. 1975;31:69–74.
79. Himms-Hagen J. Cellular thermogenesis. *Annu Rev Physiol*. 1976;38:315–351.
80. Foster DO, Frydman ML. Nonshivering thermogenesis in the rat. II. Measurements of blood flow with microspheres point to brown adipose tissue as the dominant site for the calorigenesis induced by noradrenaline. *Can J Physiol Pharmacol*. 1978;56:110–122.
81. Arch JRS, Ainsworth AT, Cawthorne MA, et al. Atypical β-adrenoreceptor on brown adipocytes as target for anti-obesity drugs. *Nature*. 1984;309:163–165.
82. Pecqueur C, Couplan E, Bouillaud F, Riquier D. Genetic and physiological analysis of the role of uncoupling proteins in human energy homeostasis. *J Mol Med*. 2001;79:48–56.
83. Ricquier D, Bouillaud F, Toumelin P, et al. Expression of uncoupling protein mRNA in thermogenic or weakly thermogenic brown adipose tissue. Evidence for a rapid β-adrenoreceptor-mediated and transcriptionally regulated step during activation of thermogenesis. *J Biol Chem*. 1986;261:13905–13910.
84. Rehnmark S, Bianco AC, Kieffer JD, Silva JE. Transcriptional and posttranscriptional mechanisms in uncoupling protein mRNA response to cold. *Am J Physiol*. 1992;262:E58–E67.
85. Silva JE. Full expression of uncoupling protein gene requires the concurrence of

norepinephrine and triiodothyronine. *Mol Endocrinol*. 1988;2:706–713.
86. Moriscot A, Rabello R, Bianco AC. Corticosterone inhibits uncoupling protein gene expression in brown adipose tissue. *Am J Physiol*. 1993;265:E81–E87.
87. Arase K, York DA, Shimizu H, et al. Effects of corticotropin-releasing factor on food intake and brown adipose tissue thermogenesis in rats. *Am J Physiol*. 1988;255:E255–E259.
88. Ricquier D, Nechad M, Mory G. Ultrastructural and biochemical characterization of human brown adipose tissue in pheochromocytoma. *J Clin Endocrinol Metab*. 1982;54:803–807.
89. Lean MEJ, James WPT. Uncoupling protein in human brown adipose tissue mitochondria: isolation and detection by specific antiserum. *FEBS Lett*. 1983;163:235–240.
90. Lean MEJ, James WPT, Jennings G, Trayhurn P. Brown adipose tissue uncoupling protein content in human infants, children and adults. *Clin Sci*. 1986;71:291–297.
91. Garutti G, Ricquier D. Analysis of uncoupling protein and its mRNA in adipose tissue deposits of adult humans. *Int J Obes Relat Metab Disord*. 1992;16:383–390.
92. Michel MC, Ochodnicky P, Summers RJ. Tissue functions mediated by β_3-adrenoreceptors-findings and challenges. *Naunyn-Schmied Arch Pharmacol*. 2010;382:103–108.
93. Houstek, J, Vizek K, Pavelka S, et al. Type II iodothyronine 5′-deiodase and uncoupling protein in brown adipose tissue of human newborns. *J Clin Endocrinol Metab*. 1993;77:382–387.
94. Girardier L. Brown fat: an energy dissipating tissue. In: Girardier L, Stock MJ, eds. *Mammalian Thermogenesis*. London: Chapman and Hall; 1983:50–98.
95. Nedergaard J, Bengtsson T, Cannon B. Unexpected evidence for active brown adipose tissue in adult humans. *Am J Physiol*. 2007;293:E444–E452.
96. Virtanen KA, Lidell ME, Orava J, et al. Functional brown adipose tissue in humans. *N Engl J Med*. 2009;360:1518–1525.
97. Saito M. Brown adipose tissue as a regulator of energy expenditure and body fat in humans. *Diabetes Metab J*. 2013;37:22–29.
98. Matsushita M, Yoneshiro T, Aita S, et al. Impact of brown adipose tissue on body fatness and glucose metabolism in humans. *Int J Obes*. 2013; doi:10.1038/ijp.2013.206.
99. Astrup A, Bülow J, Christensen NJ, et al. Facultative thermogenesis induced by carbohydrate: a skeletal muscle component mediated by epinephrine. *Am J Physiol*. 1986;250:E226–E229.
100. Simonsen L, Bulow J, Madsen J, Christensen NJ. Thermogenic response to epinephrine in the forearm and abdominal subcutaneous adipose tissue. *Am J Physiol*. 1992;263:E850–E855.
101. Lebon V, Dufour S, Falk Petersen K, et al. Effect of triiodothyronine on mitochondrial energy coupling in human skeletal muscle. *J Clin Invest*. 2001;108:733–737.
102. Wijers SLJ, Schrauwen P, Saris WHM, van Marken Lichtenbelt WD. Human skeletal muscle mitochondrial uncoupling is associated with cold induced adaptive thermogenesis. *Plos One*. 2008;3: e1777. doi:10.1371.
103. Boivin M, Camirand A, Carli F, et al. Uncoupling protein-2 and -3 messenger ribonucleic acids in adipose tissue and skeletal muscle of healthy males: variability, factors affecting expression, and relation to measures of metabolic rate. *J Clin Endocrinol Metab*. 2000;85:1975–1983.
104. Zurlo F, Larson K, Bogardus C, Ravussin E. Skeletal muscle metabolism is a

major determinant of resting energy expenditure. *J Clin Invest*. 1990;86:1423–1427.
105. Astrup A, Bülow J, Madsen J, Christensen NJ. Contribution of BAT and skeletal muscle to thermogenesis induced by ephedrine in man. *Am J Physiol*. 1985;248:E507–E515.
106. Lifson N, McClintock R. Theory of use of the turnover rates of body water for measuring energy and material balance. *J Theor Biol*. 1966;12:46–74.
107. Prentice AM, Lucas A, Vasquez-Velasquez L, et al. Are current dietary guidelines for young children a prescription for overfeeding? *Lancet*. 1988;2:1066–1069.
108. Zurlo F, Ferraro RT, Fontvieille AM, et al. Spontaneous physical activity and obesity: cross-sectional and longitudinal studies in Pima Indians. *Am J Physiol*. 1992;263:E296–E300.
109. Black AE, Coward WA, Prentice AM. Human energy expenditure in affluent societies : an analysis of 574 doubly-labelled water measurements. *Eur J Clin Nutr*. 1996;50:72–92.
110. EFSA. Scientific opinion on dietary reference values for energy. *EFSA J*. 2013;11:3005.
111. Westerterp KR. Pattern and intensity of physical activity. *Nature*. 2001;410:539.
112. Hoos MB, Kuipers H, Gerver WJ, Westerterp KR. Physical activity pattern of children assessed by triaxial accelerometry. *Eur J Clin Nutr*. 2004;58:1425–1428.
113. Montgomery C, Reilly JJ, Jackson DM, et al. Relation between physical activity and energy expenditure in a representative sample of young children. *Am J Clin Nutr*. 2004;80:591–596.
114. Torun B, Davies PSW, Livingstone MBE et al. Energy requirements and dietary energy recommendations for children and adolescents 1 to 18 years old. *Eur J Clin Nutr*. 1996;50(suppl 1):S37–S81.
115. Butte NF, Wong WW, Ferlic L, et al. Energy expenditure and deposition of breast-fed and formula-fed infants during early infancy. *Pediatr Res*. 1990;28:631–640.
116. Henry CJ, Webster-Gandy JD, Elia M. Physical activity levels in a sample of Oxford school children aged 10-13 years. *Eur J Clin Nutr*. 1999;53:840–843.
117. Abbott RA, Davies PS. Habitual physical activity and physical activity intensity: their relation to body composition in 5.0-10.5-y-old children. *Eur J Clin Nutr*. 2004;58:285–291.
118. Rennie KL, Livingstone MBE, Wells JCK, et al. Association of physical activity with body-composition indexes in children aged 6–8 y at varied risk of obesity. *Am J Clin Nutr*. 2005;82:13–20.
119. Rowlands AV, Ingledew DK, Eston RG. The effect of type of physical activity measure on the relationship between fatness and habitual physical activity in children: a meta-analysis. *Ann Hum Biol*. 2000;27:479–497.
120. Summerbell CD, Waters E, Edmunds LD, et al. Interventions for preventing obesity in children. *Cochrane Database Syst Rev*. 2005;CD001871.
121. Hill JO, Wyatt HR, Read GW, Peters JC. Obesity and the environment: where do we go from here. *Science*. 2003;299:853–855.
122. Metcalf B, Henley W, Wilkin T. Effectiveness of intervention on physical activity of children: systematic review and meta-analysis of controlled trials with objectively measured outcomes. *Br Med J*. 2012;345:e5888
123. Ross R, Dagnonne D, Jones P et al. Reduction in obesity and related comorbid conditions after diet-induced weight loss or exercise-induced weight loss in men: a randomized, controlled trial. *Ann Intern Med*. 2000;133:92–103.
124. Levine JA, Eberhardt NL, Jensen MD. Role of nonexercise activity thermogenesis

in resistance to fat gain in humans. *Science*. 1999;283:212–214.
125. Levine JA, Lanningham-Foster LM, McCrady S et al. Interindividual variation in posture allocation: possible role in human obesity. *Science*. 2005;307:584–586.
126. Franks PW, Ravussin E, Hanson RL et al. Habitual physical activity in children: the roles of genes and the environment. *Am J Clin Nutr*. 2005;82:901–908.
127. Yoneshiro T, Saito M. Transient receptor potential activated brown fat thermogenesis as a target of food ingredients for obesity management. *Curr Opin Nutr Metab Care*. 2013;16:625–631.
128. Grande F, Anderson JT, Keys A. Changes in basal metabolic rate in man in semistarvation and refeeding. *J Appl Physiol*. 1958;12:230–238.
129. Leibel RL, Rosenbaum M, Hirsch J. Changes in energy expenditure resulting from altered body weight. *N Engl J Med*. 1995;332:621–628.
130. Luke A, Schoeller DA. Basal metabolic rate, fat-free mass, and body cell mass during energy restriction. *Metabolism*. 1992;41:450–456.
131. Heilbronn LK, de Jonge L, Frisard MI et al. Effect of 6-moth calorie restriction on biomarkers of longevity, metabolic adaptation, and oxidative stress in overweight individuals. *JAMA*. 2006;295:1539–1548.
132. Landsberg L, Young JB. Fasting, feeding and the regulation of the sympathetic nervous system. *N Engl J Med*. 1978;298:1295–1301.
133. Shetty PS, Jung RT, James WPT. Effect of catecholamine replacement with levodopa on the metabolic response to semistarvation. *Lancet*. 1979;1:77–79.
134. Welle SL, Campbell RG. Decrease in resting metabolic rate during rapid weight loss is reversed by low doses of thyroid hormones. *Metabolism*. 1986;35:289–291.
135. Mansell PI, Fellows IW, MacDonald IA. Enhanced thermogenic response to epinephrine after 48-h starvation. *Am J Physiol*. 1990;258:R87–R93.
136. Gallen IW, MacDonald IA. The effects of underfeeding for 7d on the thermogenic and physiological response to glucose and insulin infusion (hyperinsulinaemic euglycaemic clamp). *Br J Nutr*. 1990;64:427–437.
137. Mansell PI, Fellows IW, MacDonald IA, Allison SP. Defect in thermoregulation in malnutrition reversed by weight gain. Physiological mechanisms and clinical importance. *Q J Med*. 1990;76:817–829.
138. Brooke OG, Harris M, Salvosa CB. The response of malnourished babies to cold. *J Physiol (Lond)*. 1973;233:75–91.
139. Nichols BL, Barnes DJ, Ashworth A, et al. Relationship between total body and muscle respiratory rates in infants with malnutrition. *Nature*. 1968;217:475–476.
140. Montgomery RD. Changes in the basal metabolic rate of the malnourished infant and their relation to body composition. *J Clin Invest*. 1962;41:1653–1663.
141. Soares MJ, Shetty PS. Basal metabolic rates and metabolic economy in chronic undernutrition. *Eur J Clin Nutr*. 1991;45:363–373.
142. Barac-Nieto M, Spurr GB, Lotero H, Maksud MG. Body composition in chronic undernutrition. *Am J Clin Nutr*. 1978;31:23–40.
143. Koong LJ, Ferrell CL. Effects of short term nutritional manipulation on organ size and fasting heat production. *Eur J Clin Nutr*. 1990;44(suppl 1):73–77.
144. Brooke OG, Cocks T. Resting metabolic rate in malnourished babies in relation to total body potassium. *Acta Paediatr Scand*. 1974;63:817–825.
145. Stein TP, Rumpler WV, Leskiw MJ, et al. Effect of reduced dietary intake on energy expenditure, protein turnover, and glucose cycling in man. *Metabolism*. 1991;40:478–483.
146. Spady DW, Payne PR, Picou D, Waterlow JC. Energy balance during recovery from malnutrition. *Am J Clin Nutr*. 1976;29:1073–1078.
147. Dulloo AG, Girardier L. Adaptative role of energy expenditure in modulating

body fat and protein deposition during catch-up growth after early undernutrition. *Am J Clin Nutr*. 1993;58:614–621.
148. Cahill GF, Herrera MG, Morgan AP, et al. Hormone-fuel interrelationships during fasting. *J Clin Invest*. 1966;45:1751–1769.
149. Owen OE, Morgan AP, Kemp HG, et al. Brain metabolism during fasting. *J Clin Invest*. 1967;46:1589–1595.
150. Owen OE, Felig P, Morgan AP, et al. Liver and kidney metabolism during prolonged starvation. *J Clin Invest*. 1969;48:574–583.
151. Rothman DL, Magnusson I, Katz LD, Shulman RG, Shulman GI. Quantitation of hepatic glycogenolysis and gluconeogenesis in fasting humans with ^{13}C NMR. *Science*. 1991;254:573–576.
152. Felig P, Owen OE, Wahren J, Cahill GF. Amino acid metabolism during prolonged starvation. *J Clin Invest*. 1969;48:584–594.
153. Pénicaud L, Kandé J, Le Magnen J, Girard JR. Insulin action during fasting and refeeding in rat determined by euglycemic clamp. *Am J Physiol*. 1985;249:E514–E518.
154. Kerr DS, Stevens MCG, Robinson HM. Fasting metabolism in infants. I. Effect of severe undernutrition on energy and protein utilization. *Metabolism*. 1978;27:411–435.
155. Henry CJK, Rivers JPW, Payne PR. Protein and energy metabolism in starvation reconsidered. *Eur J Clin Nutr*. 1988;42:543–549.
156. Nørrelund H, Nair KS, Nielsen S et al. The decisive role of free fatty acids for protein conservation during fasting in humans with and without growth hormone. *J Clin Endocrinol Metab*. 2003;88:4371–4378.
157. Gelfand RA, Matthews DE, Bier DM, Sherwin RS. Role of counterregulatory hormones in the catabolic response to stress. *J Clin Invest*. 1984;74:2238–2248.
158. Clowes GHA Jr., George BC, Villee CA Jr., Saravis CA. Muscle proteolysis induced by a circulating peptide in patients with sepsis or trauma. *N Engl J Med*. 1983;308:545–552.
159. Baracos V, Rodeman HP, Dinarello CA, Goldberg AL. Stimulation of muscle protein degradation and prostaglandin E_2 release by leukocytic pyrogen (interleukin-1). *N Engl J Med*. 1983;308:553–558.
160. Kitamura T, Ogorochi T, Miyajima A. Multimeric cytokine receptors. *Trends Endocrinol Metab*. 1994;5:8–14.
161. Fong Y, Moldawer LL, Marano M, et al. Cachectin/TNF or IL-1α induces cachexia with redistribution of body proteins. *Am J Physiol*. 1989;256:R659–R665.
162. Hoshino E, Pichard C, Greenwood CE, et al. Body composition and metabolic rate in rat during a continuous infusion of cachectin. *Am J Physiol*. 1991;260:E27–E36.
163. Tessitore L, Costelli P, Baccino FM. Pharmacological interference with tissue hypercatabolism in tumor-bearing rats. *Biochem J*. 1994;299:71–78.
164. Starnes HF Jr., Warren RS, Jeevanadam M, et al. Tumor necrosis factor and the acute metabolic response to tissue injury in man. *J Clin Invest*. 1988;82:1321–1325.
165. Lang CH, Huber D, Frost RA. Burn-induced increase in atrogin-1 and MuRF-1 in skeletal muscle is glucocorticoid-independent but down regulated by IGF-I. *Am J Physiol*. 2007;292:R328–336.
166. Tomkins AM, Garlick PJ, Schofield WN, Waterlow JC. The combined effects of infection and malnutrition on protein metabolism in children. *Clin Sci*. 1983;65:313–324.

167. Garlick PJ, McNurlan MA, Fern E, et al. Stimulation of protein synthesis and breakdown by vaccination. *Br Med J*. 1980;190:263–265.
168. Cayol M, Tauveron I, Rambourdin F, et al. Whole-body protein turnover and hepatic protein synthesis are increased by vaccination in man. *Clin Sci*. 1995;89:389–396.
169. Gotlin RW, Dubois RS. Nyctohemeral growth hormone levels in children with growth retardation and inflammatory bowel disease. *Gut*. 1973;14:191–195.
170. Kushner RF, Schoeller DA. Resting and total energy expenditure in patients with inflammatory bowel disease. *Am J Clin Nutr*. 1991;53:161–165.
171. Burnham JM, Schults J, Semeao E, et al. Body-composition alterations consistent with cachexia in children and young adults with Crohn's disease. *Am J Clin Nutr*. 2005;82:413–420.
172. Powell-Tuck J, Garlick PJ, Lennard-Jones JE, Waterlow JC. Rates of whole body protein synthesis and breakdown increase with the severity of inflammatory bowel disease. *Gut*. 1984;25:469–474.
173. Thomas AG, Miller V, Taylor F, et al. Whole body protein turnover in childhood Crohn's disease. *Gut*. 1992;33:675–677.
174. Murch SH, Lamkin VA, Savage MO, et al. Serum concentrations of tumor necrosis factor alpha in childhood chronic inflammatory bowel disease. *Gut*. 1991;32:913–917.
175. Kirschner BS, Sutton MM. Somatomedin-C levels in growth-impaired children and adolescents with chronic inflammatory bowel disease. *Gastroenterology*. 1986;91:166–176.
176. Donahue SP, Phillips LS. Response of IGF-I to nutritional support in malnourished hospital patients: a possible indicator of short-term changes in nutritional status. *Am J Clin Nutr*. 1989;50:962–969.
177. Phillips LS, Orawski AT, Belosky DC. Somatomedin and nutrition. IV. Regulation of somatomedin activity and growth cartilage activity by quantity and composition of diet in rats. *Endocrinology*. 1978;103:121–127.
178. Dardevet D, Manin M, Balage M, et al. Influence of low- and high-protein diets on insulin and insulin-like growth factor-1 binding to skeletal muscle and liver in the growing rat. *Br J Nutr*. 1991;65:47–60.
179. Harp JB, Goldstein S, Phillips LS. Nutrition and somatomedin. XXIII. Molecular regulation of IGF-I by amino acid availability in cultured hepatocytes. *Diabetes*. 1991;40:95–101.
180. Pucilowska JB, Davenport ML, Kabir I, et al. The effect of dietary protein supplementation on insulin-like growth factors (IGFs) and IGF-binding proteins in children with shigellosis. *J Clin Endocrinol Metab*. 1993;77:1516–1521.
181. Thissen JP, Verniers J. Inhibition by interleukin-1β and tumor necrosis factor-alpha on the insulin-like growth factor 1 messenger ribonucleic acid in response to growth hormone in rat hepatocyte primary culture. *Endocrinology*. 1997;138:1078–1084.
182. Walters TD, Griffiths AM. Mechanisms of growth impairment in pediatric Crohn's disease. *Nat Rev Gastroenterol Hepatol*. 2009;6:513–523.
183. Houssiau FA, Devogelaer JP, Van Damme J, et al. Interleukin-6 in synovial fluid and serum of patients with rheumatoid arthritis and other inflammatory arthritis. *Arthritis Rheum*. 1988;31:784–788.
184. Roubenoff R, Roubenoff RA, Cannon JG, et al. Rheumatoid cachexia: cytokine-driven hypermetabolism accompanying reduced body cell mass in chronic inflammation. *J Clin Invest*. 1994;93:2379–2386.
185. Walsmith J, Abad L, Kehavias J, Roubenoff R. Tumor necrosis factor-alpha

production is associated with less body cell mass in women with rheumatoid arthritis. *J Rheumatol*. 2004;31:23–29.
186. Roubenoff R. Sarcopenic obesity: does muscle loss cause fat gain? *Ann N Y Acad Sci*. 2000;904:553–557.
187. Rall LC, Rosen CJ, Dolnikowski G, et al. Protein metabolism in rheumatoid arthritis and aging. Effects of muscle strength and tumor necrosis factor alpha. *Arthritis Rheum*. 1996;39:1115–1124.
188. Gibson JNA, Poyser NL, Morrison WL, et al. Muscle protein synthesis in patients with rheumatoid arthritis: effect of chronic corticosteroid therapy on prostaglandin F_{2a} availability. *Eur J Clin Invest*. 1991;21:406–412.
189. Boirie Y, Broyer M, Gaganadoux MF, et al. Alterations in protein metabolism by metabolic acidosis in children with chronic renal failure. *Kidney Int*. 2000;58:236–241.
190. Stettler N, Schutz Y, Whitehead R, Jéquier E. Effect of malaria and fever on energy metabolism in Gambian children. *Pediatr Res*. 1992;31:102–106.
191. Berclaz PY, Benedek C, Jéquier E, Schutz Y. Changes in protein turnover and resting energy expenditure after treatment of malaria in Gambian children. *Pediatr Res*. 1996;39:401–409.
192. Fong Y, Marano MA, Moldawer LL, et al. The acute splanchnic and peripheral tissue metabolic response to endotoxin in humans. *J Clin Invest*. 1990;85:1896–1904.
193. Leon LR. Cytokine regulation of fever: studies using gene knockout mice. *J Appl Physiol*. 2002;92:2648–2655.
194. Zheng H, Fletcher D, Kozak W, et al. Resistance to fever induction and impaired acute-phase response in interleikin-1β deficient mice. *Immunity*. 1995;3:9–19.
195. Kozak W, Kluger MJ, Soszynski D, et al. IL6 and IL-1β in fever. Studies using cytokine-deficient (knockout) mice. *Ann NY Acad Sci*. 1998;856:33–47.
196. Hellerstein MK, Meydani SN, Meydani M, et al. Interleukin-1-induced anorexia in the rat. Influence of prostaglandins. *J Clin Invest*. 1989;84:228–235.
197. Reichenberg A, Kraus T, Haack M, et al. Endotoxin-induced changes in food consumption in healthy volunteers are associated with TNF-α and IL-6 secretion. *Psychoneuroendocrinol*. 2002;27:945–956.
198. Uehara A, Sekika C, Takasugi Y, et al. Anorexia induced by interleukin-1: involvement of corticotropin-releasing factor. *Am J Physiol*. 1989;257:R613–R617.
199. Lawrence CB, Rothwell NJ. Anorexic but not pyrogenic actions of interleukin-1 are modulated by central melanocortin-3/4 receptors in the rat. *J Neuroendocrinol*. 2001;13:490–495.
200. Johnson PM, Vogt SK, Burney MW, Muglia LJ. COX-2 inhibition attenuates anorexia during systemic inflammation without impairing cytokine production. *Am J Physiol*. 2002;282:E650–E656.
201. Plata-Salaman CR. Central nervous system mechanisms contributing to the cachexia-anorexia syndrome. *Nutrition*. 2000;16:1009–1012.
202. De Fanti BA, Martinez JA. Central urocortin activation of sympathetic-regulated energy metabolism in Wistar rats. *Brain Res*. 2002;930:37–41.
203. Cooper AL, Rothwell NJ. Mechanisms of early and late hypermetabolism and fever after localized tissue injury in rats. *Am J Physiol*. 1991;261:E698–E705.
204. Herndon DN, Hart DW, Wolf SE, et al. Reversal of catabolism by beta-blockade after severe burns. *N Engl J Med*. 2001;345:1223–1229.
205. Busbridge NJ, Dascombe MJ, Rothwell NJ. Chronic effects of interleukin-1β on fever, oxygen consumption and food intake in the rat. *Horm Metab Res*.

1993;25:222–227.
206. Knops N, Wulfraat N, Lodder S, et al. Resting energy expenditure and nutritional status in children with juvenile rheumatoid arthritis. *J Rheumatol*. 1999;26:2039–2043.
207. Bell SC, Saunders MJ, Elborn JS, Shale DJ. Resting energy expenditure and oxygen cost of breathing in patients with cystic fibrosis. *Thorax*. 1996;51:126–131.
208. McCloskey M, Redmond AO, Pyper S, et al. Total energy expenditure in stable patients with cystic fibrosis. *Clin Nutr*. 2001;20:235–241.
209. Burdet L, Hugli O, Aubert JD, et al. Effect of elective antibiotic therapy on resting energy expenditure and inflammation in cystic fibrosis. *Eur J Pediatr*. 1999;158:711–716.
210. Stettler N, Kawchak DA, Boyle LL, et al. Prospective evaluation of growth, nutritional status, and body composition in children with cystic fibrosis. *Am J Clin Nutr*. 2000;72:407–413.
211. Sammalkorpi K. Glucose intolerance in acute infections. *J Intern Med*. 1989;225:15–19.
212. Wolfe RR, Shaw JHF. Glucose and FFA kinetics during sepsis: role of glucagon and sympathetic nervous system activity. *Am J Physiol*. 1985;248:E236–E243.
213. White RH, Frayn KN, Little RA, et al. Hormonal and metabolic responses to glucose infusion in sepsis studied by the hyperglycemic glucose clamp technique. *J Parenter Enteral Nutr*. 1987;11:345–353.
214. Henderson AA, Frayn KN, Galasko CSB, Little RA. Dose-response relationships for the effects of insulin on glucose and fat metabolism in injured patients and control subjects. *Clin Sci*. 1991;80:25–32.
215. Yki-Järvinen H, Sammalkorpi K, Koivisto VA, Nikkila EA. Severity, duration, and mechanisms of insulin resistance during acute infections. *J Clin Endocrinol Metab*. 1989;69:317–323.
216. Lang CH. Sepsis-induced insulin resistance in rats is mediated by a β-adrenergic mechanism. *Am J Physiol*. 1992;263:E703–E711.
217. Rosenzweig T, Braiman L, Bak A, et al. Differential effects of tumor necrosis factor-alpha on protein kinase C isoforms alpha and delta mediates inhibition of insulin receptor signaling. *Diabetes*. 2002;51:1921–1930.
218. Gasic S, Tian B, Green A. Tumor necrosis factor alpha stimulated lipolysis in adipocytes by decreasing Gi protein concentrations. *J Biol Chem*. 1999;274:6770–6775.
219. Van Hall G, Steenberg A, Saccheti M, et al. Interleukin-6 stimulates lipolysis and fat oxidation in humans. *J Clin Endocrinol Metab*. 2003;88:3005–3010.
220. Hanson RW, Reshef L. Glyceroneogenesis revisited. *Biochimie*. 2003;85:1199–1205.
221. Levinson MR, Groeger JS, Jeevanandam M, Brennan MF. Free fatty acid turnover and lipolysis in septic mechanically ventilated cancer-bearing patients. *Metabolism*. 1988;37:618–625.
222. Van der Poll T, Romijn JA, Endert E, et al. Tumor necrosis factor mimics the metabolic response to acute infection in healthy humans. *Am J Physiol*. 1991;261:E457–E465.
223. FAO/WHO/UNU. *Energy and Protein Requirements*. Geneva, Switzerland: WHO Technical Report Series 724; 1985.
224. Butte NF, Henry CJK, Torun B. Report on the working group on energy requirements of infants, children and adolescents. *Eur J Clin Nutr*. 1996;50:S188–S189.

225. Duggan MB, Milner RDG. The maintenance energy requirement for children: an estimate based on a study of children with infection associated underfeeding. *Am J Clin Nutr*. 1986;43:870–878.
226. Baker JP, Detski AS, Stewart S, et al. Randomized trial of total parenteral nutrition in critically ill patients: metabolic effects of varying glucose-lipid ratios as the energy source. *Gastroenterology*. 1984;87:53–59.
227. Roulet M, Detsky AS, Marliss EB, et al. A controlled trial of the effect of parenteral nutritional support on patients with respiratory failure and sepsis. *Clin Nutr*. 1983;2:97–105.
228. Arnold J, Leinhardt D, Carlson G, et al. Thermogenic and hormonal responses to amino acid infusion in septic humans. *Am J Physiol*. 1992;263:E129–E135.
229. Wintrop AL, Wesson DE, Pencharz PB, et al. Injury severity, whole body protein turnover and energy expenditure in pediatric trauma. *J Pediatr Surg*. 1987;22:534–537.
230. Jones MO, Pierro A, Hammond P, Lloyd DA. The metabolic response to operative stress in infants. *J Pediatr Surg*. 1993;28:1258–1263.
231. Goran MI, Broemeling L, Herndon DH, et al. Estimating energy requirements in burned children: a new approach derived from measurements of energy expenditure. *Am J Clin Nutr*. 1991;54:35–40.
232. Hart DW, Wolf SE, Herndon DN, et al. Energy expenditure and caloric balance after burn: increased feeding leads to fat rather than lean mass accretion. *Ann Surg*. 2002;235:152–161.
233. Matthews DS, Bullock RE, Matthews JN, et al. Temperature response to severe head injury and the effect on body energy expenditure and cerebral oxygen consumption. *Arch Dis Child*. 1995;72:507–515.
234. Fung EB. Estimating energy expenditure in critically ill adults and children. *AACN Clin Issues*. 2000;11:480–497.
235. Wolfe RR, O'Donnell TF Jr., Stone MD, et al. Investigation of factors determining the optimal glucose infusion rate in total parenteral nutrition. *Metabolism*. 1980;29:892–900.
236. Bresson JL, Bader B, Rocchiccioli F, et al. Protein-metabolism kinetics and energy-substrate utilization in infants fed parenteral solutions with different glucose-fat ratios. *Am J Clin Nutr*. 1991;54:370–376.
237. Bier DM, Leake RD, Haymond MW, et al. Measurement of "true" glucose production rates in infancy and childhood with 6,6-dideuteroglucose. *Diabetes*. 1977;26:1016–1023.
238. Cunningham JJ, Lydon MK, Russell WE. Calorie and protein provision for recovery from severe burns in infants and young children. *Am J Clin Nutr*. 1990;51:553–557.
239. Bone RC, Fisher CJ, Clemmer TP, et al. A controlled clinical trial of high-dose methylprednisolone in the treatment of severe sepsis and septic shock. *N Engl J Med*. 1987;317:653–658.
240. Cooney R, Kimball SR, Eckman R, et al. TNF-binding protein ameliorates inhibition of skeletal muscle protein synthesis during sepsis. *Am J Physiol*. 1999;276:E611–E619.
241. Bernard GR, Vincent JL, Laterre PF, et al. Efficacy and safety of recombinant activated protein C for severe sepsis. *N Engl J Med*. 2001;344:699–709.
242. Michie HR, Manogue KR, Spriggs DR, et al. Detection of circulating tumor necrosis factor after endotoxin administration. *N Engl J Med*. 1988;318:1481–1486.
243. Breuille D, Farge MC, Rose F, et al. Pentoxifylline decreases body weight loss

and muscle protein wasting characteristics of sepsis. *Am J Physiol*. 1993;265:E660–E666.
244. Zabel P, Wolter DT, Schönharting MM, Schade UF. Oxpentifylline in endotoxaemia. *Lancet*. 1989;2:1474–1477.
245. Wolf RF, Pearlstone DB, Newman E, et al. Growth hormone and insulin reverse net whole body and skeletal muscle protein catabolism in cancer patients. *Ann Surg*. 1992;216:280–290.
246. Douglas RG, Gluckman PD, Breier BH, et al. Effects of recombinant IGF-I on protein and glucose metabolism in rTNF-infused lambs. *Am J Physiol*. 1991;261:E606–E612.
247. Kupfer SR, Underwood LE, Baxter RC, Clemmons DR. Enhancement of the anabolic effects of growth hormone and insulin-like growth factor I by use of both agents simultaneously. *J Clin Invest*. 1993;91:391–396.
248. Hardin DS, Adams-Huet B, Brown D, et al. Growth hormone treatment improves growth and clinical status in prepubertal children with cystic fibrosis – Results of a multicenter randomized controlled trial. *J Clin Endocrinol Metab*. 2006;91:4925–4929.
249. Haffner D, Schaefer F, Nissel R, et al. Effect of growth hormone treatment on the adult height of children with chronic renal failure. *N Engl J Med*. 2000;343:923–930.
250. Bechtold S, Ripperger P, Bonfig W, et al. Growth hormone changes bone geometry and body composition in patients with juvenile idiopathic arthritis requiring glucocorticoid treatment: a controlled study using peripheral quantitative computed tomography. *J Clin Endocrinol Metab*. 2005;90:3168–3173.
251. Heyman MB, Garnett EA, Wojcicki J et al. Growth hormone treatment for growth failure in pediatric patients with Crohn's disease. *J Pediatr*. 2008;153:651–658.
252. Savage MO, Simon D, Czernichow PC. Growth hormone treatment in children on chronic glucocorticoid therapy. *Endocr Dev*. 2011;20:194–201.

Severe Acute Malnutrition: 14 Pathophysiology, Clinical Consequences, and Treatment

Mary E. Penny, MBChB

The number of children affected and long-term consequences of malnutrition place it among the greatest public health problems facing the world today. An estimated 19.3 million children (3.5%) under 5 years in developing countries are severely wasted, that is, they are less than 3 SD below the median of weight-for-height.[1] The majority, 12.9 million, live in Asia and 5.6 million in Africa. These children are at an increased risk of dying. Even mild degrees of malnutrition increase the risk of mortality by 50%, but this risk greatly increases with the severity of wasting.[2] An analysis of eight low-income countries reported that children with severe wasting had nine times higher risk of dying of any cause (OR 9.4 [95% CI 5.3–16.8] with the greatest risk for pneumonia deaths (OR 8.7 [95% 4.-15.6]).[4,3,4]

Severe wasting or severe acute malnutrition (SAM), which used to be called protein energy malnutrition, is eminently treatable, but sadly the treatment of SAM is an area where the gap between existing knowledge of

successful practice and the reality of the management of most malnourished children is abysmal. Several reports document how the adoption of improved treatment following the World Health Organization (WHO) guidelines[5] can greatly reduce mortality even in resource-restricted circumstances (and with optimum management mortality can be as low as 5%[6–9], but the reality in many areas is mortality rates of 20% or more, with even higher rates for children with kwashiorkor.[4,10–12]

The global importance of malnutrition and its long-term consequences are dealt with in Chapters 15, 40, and 41. The public health significance, as discussed in Chapters 15 and 16, emphasizes the necessity for the combination of policies designed to prevent malnutrition with programs to treat children with malnutrition. Malnutrition is not only a problem of developing countries. Failure to recognize malnutrition may also contribute to increased morbidity and mortality in other conditions such as congenital heart disease and cystic fibrosis in developed countries.[13,14] This chapter deals with the clinical management of severe malnutrition and attempts to show how an understanding of the pathophysiology provides a basis for the treatment of the severely malnourished children; it is principally concerned with severe SAM as a clinical entity because the failure to recognize the special needs of these children contributes to high mortality.[5]

DEFINITIONS AND DIAGNOSIS OF SEVERE ACUTE MALNUTRITION

Malnutrition has many causes. In any individual, inadequate food intake, infections, psychosocial deprivation, the environment, and perhaps genetic variability contribute. The clinical manifestations in the child depend on the duration and degree of shortfall in dietary intake, the quality of the diet, host factors such as ageand the interplay with infection. Recently, the role of the microflora (microbiome) of the intestine has been demonstrated.[15]

The first step in the diagnosis of SAM is nutrition assessment. This is an essential part of any clinical pediatric evaluation and is discussed in detail in Chapter 2, “Clinical Assessment of Nutritional Status.” Repeated nutritional assessments of all ill children are important to prevent the insidious onset of malnutrition, which may hinder recovery. Adequate growth is an indication of health and recovery. For an individual child, the most important information is gained from serial measurements of the

child‘s growth, and this provides essential information for diagnosing cause and guiding treatment. The importance of using this often available, but overlooked information cannot be overstated.

Although measurements of height (or length in children under 2 years) and weight are the usual mainstays of diagnosis, the mid-upper arm circumference (MUAC) has also been shown to be useful in identifying children with SAM who are at increased risk of dying and need treatment in hospital or specialized facility care.[5] Nutritional survey data from 31 countries show that applying a cutoff of less than –3 SD weight-for-height (WHO standards) and a MUAC cutoff of less than 115 mm identifies a similar number of children, 3.22% and 3.27%, respectively.[16] This new cut off identifies more children at a greatly increased risk, about nine times the risk of dying. The former cut off criteria of –3SD weight for height using the NCHS reference standards only identified 1.48% and a similar percentage, 1.48%, were identified by a MUAC cut off of 110 mm.[6]

The two criteria identify different children so WHO has recommended that both criteria be used, together with the presence of edema to diagnose SAM (WHO combined statement) (Table 14-1).[6] MUAC is more sensitive for younger aged children, an age when malnutrition is particularly critical for future development.[17] It is an easy and cheap test and does not require looking up values in chart. Sole reliance on weight and length or height or MUAC can lead to failure to recognize kwashiorkor and an under estimation of the severity of the malnutrition in these children. The scheme recommended by WHO Table 14-1. has the merit of distinguishing these two entities. Some authorities report that MUAC <115 mm is better than weight-for-height at identifying children at high risk of mortality and recommend using only MUAC[18] plus an examination for edema.

Biochemical and hematologic tests are useful for specific diagnoses and for monitoring progress but are rarely necessary for starting treatment. SAM is often associated with micronutrient deficiencies, and examination should include a specific search for Bitot‘s spots or xerophthalmia, manifestations of vitamin A deficiency and examination for signs of other micronutrient deficiencies.[19]

(Figure 14-1) illustrates a child with marasmus and shows the characteristic muscle wasting particularly evident in the buttocks, the pinched face, and anxious demeanor. (Figure 14-2) shows the same child on recovery.

Figure 14-3 illustrates a child with kwashiorkor. Symmetric edema is evident, especially in the lower limbs. The hair is sparse, and the flaky

paint rash is seen on the buttocks, legs, and arms. Figure 14-4 shows the typical bilateral symmetric edema of the legs.

Marasmus is generally believed to be the result of a cumulative, usually slow, inadequate energy, and protein intake. The child's metabolism has adapted, producing changes that prolong survival and protect essential visceral and brain function. This is the type of SAM traditionally seen in famine, food restriction, or anorexia. Kwashiorkor is also a severe form of malnutrition. The etiology is not yet well understood, but the long-held view that protein deficiency in the face of adequate energy intake causes kwashiorkor is now untenable. Observed findings are more in keeping with an interaction between nutritional deficits and the response to injury, infection, and oxidative stress.[20,21] The intestinal microflora appears to play an important role.[15] Marasmus and kwashiorkor commonly coexist, and although a simple unified approach to clinical management can be applied successfully to both conditions,[5] both are clinically distinct entities. When the conditions coexist, the child should be treated as having kwashiorkor.

TABLE 14-1 Criteria for Diagnosis of Severe Acute Malnutrition

Result (Indicator)	Measure	Cut-off for Diagnosis
Severe wasting (marasmus)	Weight-for-height[a]	<–3SD
Severe wasting (marasmus)	MUAC	<115 mm
Bilateral Edema (kwashiorkor)	Clinical examination	Any edema

[a]Based on WHO Standards (www.who.int/childgrowth/standards).

Source: Adapted from World Health Organization.[6]



Figure 14-1 Child with marasmus



Figure 14-2 Same child as shown in Figure 14-1, on recovery.



Figure 14-3 Child with kwashiorkor



Figure 14-4 Lower limbs of a child with kwashiorkor

PATHOPHYSIOLOGY

Metabolic Responses to Inadequate Energy Intake

SAM is the result of a chronic and cumulative failure to meet physiologic energy and nutrient requirements.[22] The manifestations of this process depend on different factors: age, concomitant infection, the intestinal microflora, prior nutritional state, and the nature of the dietary restriction. Both the early classic experimental studies of food energy deprivation and starvation in animals and humans and studies of severely malnourished children on admission and during recovery have contributed to our understanding of the physiological responses to severe undernutrition.[23,24]

In the absence of infection, fasting results in an initial depletion of fat stores and then glycogen stores mediated by metabolic and endocrine changes that have the common goal of preserving vital functions, allowing the animal or human to survive until dietary energy can be restored. Early changes include a reduction in activity that conserves energy expenditure.[25] Growth is slowed, reducing the energy need to maintain this, and changes occur in body composition. Metabolic rate expressed in relation to height or lean body mass decreases.[23] Brain and viscera are relatively preserved Figure 14-5, resulting in the body composition typical of marasmic children.[26–28] There is an increase in total-body water, which

is mainly extracellular but may also be intracellular.[28]

These metabolic adjustments to starvation are mediated, at least in part, by hormones. Cortisol concentration rises but remains responsive to stress.[27] Insulin secretion falls, and there is a reduction in plasma insulin levels, a reduced response to glucose, and peripheral insulin resistance.[29,30] Growth hormone is generally high, and the normal suppression by glucose load is lost, although exceptions occur in marasmus.[31] There is low activity of insulin-like growth factor, the metabolic effector of the growth-promoting effect of growth hormone.[27] The net effects of these hormonal changes are mobilization of fat, degradation of muscle protein, and reduction in basal metabolic rate. Increased aldosterone contributes to potassium loss already compromised by the effects of energy restriction and reduced adenosine triphosphate synthesis on the sodium pump.[32]

Adaptation to Reduced Protein Intake

During energy and protein deprivation, especially in marasmus, skeletal muscle is lost as structural protein is recycled to conserve essential enzymes and provide energy for metabolic processes.[27] There is both a fall in muscle protein synthesis and an increase in breakdown, which provides essential amino acids to the liver for protein synthesis and gluconeogenesis.[27]

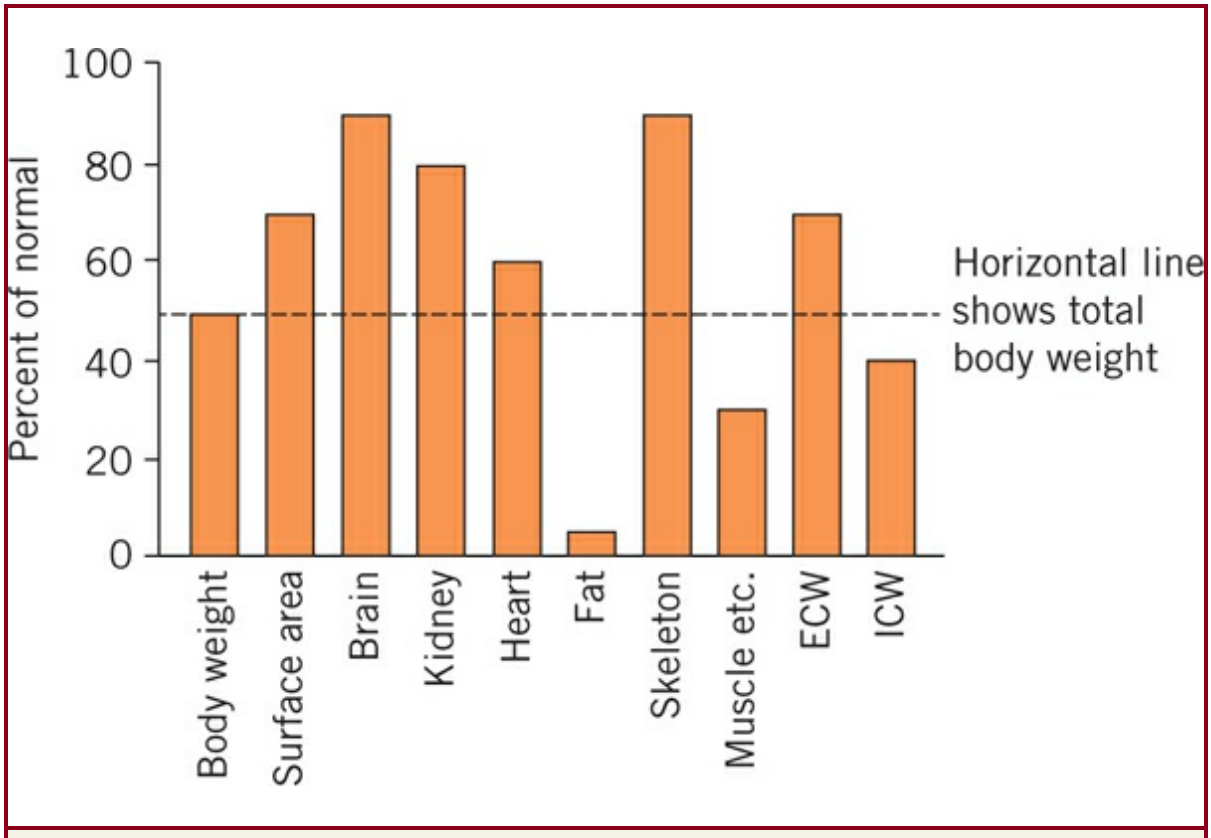


Figure 14-5 Weights of organs at death in atrophic children as percentages of normal age controls. ECW, extracellular water; ICW, intracellular water. (Adapted from Kerpal-Fronius and Frank)[16]

In kwashiorkor, whole-body protein breakdown is slower than in marasmus.[33] Within the liver, there is a shift in the rates of synthesis of different proteins; synthesis of albumin, transferrin, and apolipoprotein B are decreased, but synthesis of other proteins is maintained.[34]

Loss of skeletal muscle tissue in SAM may result in a relative shift in protein turnover from skeletal muscle to organs such as liver which have a different amino acid composition. This together with inadequate intake of sulfur amino acids may lead to a reduced availability of the essential amino acid methionine. It is suggested that one effect of these metabolic changes is a deficiency of methionine, which is manifested in the skin and hair changes of kwashiorkor.[35] Methionine is the sulfur donor for cysteine synthesis. All children with SAM have reduced cysteine synthesis, but this is much more marked in kwashiorkor.[35] Reduced availability of cysteine means that GSH synthesis is reduced and there is less glutathione and thus decreased antioxidant activity, a consistent finding in kwashiorkor.[36–38] This explanation is supported by studies showed faster reduction in edema

and improved outcome in children with kwashiorkor who received N-acetylcysteine supplements.[39] These findings need to be considered in establishing guidelines for dietary rehabilitation of children with SAM, especially kwashiorkor.[40]

Of relevance, cysteine is essential for maintenance of intestinal structure and function. Children with SAM and especially those with kwashiorkor typically have enteropathy[41] with intestinal mucosal damage, and in these circumstances, there is a high requirement for cysteine. The potentially important role of sulfur amino acids in kwashiorkor may provide the key to understanding the mechanisms where by certain patterns of gut microbiome predispose to kwashiorkor in the presence of diets that are characteristically deficient in cysteine and methionine since mice transplanted with fecal material from kwashiorkor children had lower concentrations of methionine and cysteine in their feces compared with mice transplanted with feces of healthy children.

Changes in Electrolytes

Changes in chemical composition of the body that occur during malnutrition have important consequences for treatment. Sodium, potassium, and phosphate merit special mention, but changes in other electrolytes, such as magnesium and calcium, are also important.[20]

The vast proportion of total-body potassium is intracellular, in contrast to sodium, which is actively removed from cells by the sodium pump. In both marasmus and kwashiorkor, sodium retention occurs, resulting in an increase in whole-body sodium, even though serum levels may be low, reflecting an increase in extracellular water. There is a decrease in total-body potassium, although serum potassium may remain normal.[42] Alleyne and colleagues described how potassium kinetics are altered during recovery.[43] There was an initial acute repletion of potassium when hypokalemia and whole-body deficiency improved, followed by a longer phase of cell membrane recovery and reestablishment of the normal Na/K gradients. A third phase occurred when rapid growth of skeletal muscle increased potassium requirements.[44,45] Nichols and colleagues calculated from balance studies that 7.0 mmol/kg of potassium was needed in the first week or acute phase of recovery, an amount similar to the level of supplement giving best results in a clinical trial in Malawi.[46,47]

The inconsistencies between serum and total-body potassium are explained by events at the cell membrane. In marasmus, there is a decrease in the activity of the ouabain-sensitive energy-dependent sodium pump,

resulting in increased intracellular sodium and reduced potassium.[48] These cell membrane changes are associated with intracellular changes, with relatively low intracellular K and relatively high sodium.

Different mechanisms operate in kwashiorkor, in which there is an increase in cell membrane leakiness. Sodium, responding to the electrolyte gradient, enters the cell, stimulating increased activity of the sodium pump but not enough to prevent Na increase and K loss.[48,49] Similar differences between marasmus and kwashiorkor were reported by Kaplay in India.[50] Forrester and colleagues found that these electrolyte changes could be reproduced in the in vitro erythrocytes if glutathione was artificially reduced, resembling the situation in kwashiorkor.[40] Increased intracellular sodium is accompanied by increased cell water, which may be one of the explanations for edema in kwashiorkor.[51] Hypoinsulinemia is implicated in this process.[52] These electrolyte changes are the basis for the recommendations of potassium supplementation in all severely malnourished children; dietary restriction of sodium, and the use of oral rehydration solution (ORS) with less sodium than the standard 75 mmol/L solution. As will be discussed, a balance is needed to avoid too little sodium that can result in severe hyponatremia.[41]

Hypophosphatemia has been shown to occur in malnourished children and is associated with high mortality. In a South African study, 10 of 60 patients died, all but one with very low serum phosphate.[53] The lowest levels of serum phosphate were associated with diarrhea and dehydration. A study from Malawi supports the association of hypophosphatemia and increased mortality.[54] However, it is difficult to distinguish the effects of low phosphate from hypokalemia, which also causes hypotonia and sudden death. In Jamaica, serum phosphate levels were generally higher than those recorded in Africa and although these correlated with edema, hypophosphatemia was not associated with mortality.[55] Repletion, even on a milk diet, took two to three weeks. The advantage of milk-based diets is relatively high phosphate content.

Interaction With Infection

Infection and nutrition are mutually interlocked in a vicious cycle that operates at many levels, from social cultural interrelations to intracellular metabolism. Environmental conditions that give rise to inadequate energy and protein intakes are also associated with conditions in which bacterial and other microbial contamination are often rife[56] affecting the microbial milieu of the intestine. In addition, in famine or poverty, children‘s diets

tend to consist largely of carbohydrate staples with little or no animal products or fat.[57] Animal products such as red meat, poultry, offal, fish, milk, and eggs are important bio available sources of minerals and other micronutrients that have an essential role in combating infections.[58] Fat, needed to provide essential fatty acid (EFA) and facilitate the absorption of fat-soluble vitamins such as E, D, and A, which also protect against infections, is also often low in these diets.

During infection, there are metabolic changes that concentrate body energy resources on production of acute-phase proteins in the liver and are often the opposite of those seen in starvation.[59] The production of acute-phase proteins and the metabolic consequences of infection are mediated by protein cytokines, lipid-derived factors that include prostaglandins, leukotrienes, and platelet activation factor. Endocrine changes also play a role; the concentration of catabolic hormones such as glucocorticoids, glucagons, and epinephrine increases. The cytokine interleukin (IL) 6 increases norepinephrine, cortisol, and glucagon and is the main stimulus for the mobilization of acute-phase proteins in the liver. Cytokines also enhance the effect of stress-related hormones on the production of acute-phase proteins.[60] Because of the interaction between food restriction and infection in the pathogenesis of malnutrition, any integrated approach to explaining the pathophysiology has to take both into account. The differences between kwashiorkor and marasmus may be partially explained by an increased shift toward the metabolic consequences of infection in children with kwashiorkor. In addition, preceding nutritional status may modify the metabolic effects of infection. A review of children presenting with kwashiorkor indicated that they had higher birth weights than those presenting with marasmus supporting the concept that intrauterine factors and preceding nutritional status influence the development of kwashiorkor.[61] Recent findings that the pattern of the gut microbiome is a determinate of kwashiorkor also support this concept.[15]

CYTOKINES

Cytokines and Response to Infection

Because of the fundamental role of cytokines in infection and the interaction with malnutrition, a brief summary of their action is necessary. For more detail, readers are referred to more complete reviews.[60,62] Cytokines are small, nonstructural proteins that can be produced by nearly all nucleated cells in very small amounts, have local and systemic effects,

and are involved in the primary response to infection. Synthesis of cytokines is rapidly induced in response to infection, trauma, ischemia, and other events. Cytokines are also involved in mediating the changes in protein metabolism and muscle function that accompany infections, fasting, and cancer cachexia. The family of tumor necrosis factors (TNFs), IL-1, and IL-6 are all pro-inflammatory cytokines. The pro-inflammatory cytokines mediate the local inflammatory response, which includes local heat, redness, pain, swelling, and systemic effects such as fever and anorexia. To avoid self-injury during the inflammatory response, the induction and synthesis of cytokines are controlled in a complex interactive process full of checks and balances. This tightly controlled system is disturbed in malnutrition. Clinically, children with severe malnutrition often have a reduced inflammatory reaction and blunted febrile response,[63] and some studies have reported reduced levels of in vitro production of IL-1 and TNF by circulating monocytes in malnutrition.[64,65] Other studies suggest that, in a similar way to well-nourished children, the immune cells of marasmic children are able to mount a cytokine response involving IL-6 and TNF?, and this occurs in response to acute infection.[63,66,67] It is less clear to what extent this occurs in vivo and how to interpret these results in the context of the malnourished child‘s reduced acute-phase response and the blunted clinical expression of the inflammatory response.

Acute-Phase Proteins

Cytokines modulate the production of acute-phase proteins. C-reactive protein, α-1-antitrypsin, x03B1-macroglobulin, and fibrinogen are examples of positive acute-phase proteins, so-called because their synthesis by the liver increases in response to stresses, including infection.[68] Serum concentration of the so-called negative acute-phase proteins (albumin, prealbumin, fibronectin, retinol-binding protein) is reduced in children with malnutrition.[65,68] Low levels of fibronectin have been reported to be a useful early indicator of malnutrion.[69] Positive acute-phase proteins are an essential part of the defense against infections, but uncontrolled inflammation has been linked to adverse consequences, for instance, in inflammatory bowel disease, rheumatoid arthritis, and perhaps kwashiorkor. Studies are complicated by the coexistence of infection and malnutrition and the limitations of interpreting serum levels, which represent a balance between synthesis and catabolism. Studies in Ghana on children without sign of infection found raised concentrations of C-

reactive protein in kwashiorkor and to a lesser extent in marasmus.[70] α-1 Acid glycoprotein levels were raised in kwashiorkor in Thai children.[68] On the other hand, Ekanem and colleagues,[71] studying children, 75% of whom had kwashiorkor, documented only slight increases in C-reactive protein in children without infection and large increases in infected children, indicating apparent preservation of the acute-phase response to infection in this population of children. This is in contrast to findings in Jamaica of normal levels of C-reactive protein and serum amyloid A with a blunted response in the expected acute-phase response to diphtheria-pertussis-tetanus vaccination.[72] Elegant studies in Jamaica and in Malawi using stable isotope labeled leucine, in marasmic children, with infections showed that there was an increase in some acute-phase proteins (C-reactive protein, haptoglobin, α-1 acid glycoprotein, α-1 antitrypsin) in response to infection but this response was attenuated despite preservation of the cytokine response.[73] The increase in plasma levels of acute-phase proteins was attributable to a reduction in catabolism rather than an increase in the synthesis of proteins, in contrast to the situation in normally nourished infected children.[73] Plasma fibrinogen, which is normally increased in response to infection, showed no response in these children. A study from Malawi suggests that the reduced acute-phase protein response might be modulated by the protein content of the diet. A diet with higher protein from egg white and additional trytophan, the rate limiting amino acid for acute-phase protein synthesis, resulted in an increase in serum concentrations of six acute-phase proteins.[74]

The consequence of a reduced inflammatory response to infections has important clinical repercussions both because the usual manifestations of infection may be absent in SAM making diagnosis difficult and because the mortality from infection is greatly increased. Further studies are needed to understand the different changes that occur in response to infection and most importantly the way in which dietary management may counteract these changes.

Kwashiorkor

Kwashiorkor has historically been attributed to a diet lacking in protein, and the edema was believed to be explained by low albumin. In consequence, children were treated with high-protein diets. However, several observations have long cast doubt on this hypothesis.[75] Kwashiorkor often follows infections such as measles and dysentery and has been reported in breast-fed babies, who presumably are ensured

adequate protein intake, and children in the same community on very similar diets may develop kwashiorkor or marasmus. Edema has been shown to improve independently of albumin. Golden demonstrated that recovery was possible with relatively low-protein diets and was dependent on energy intake and not on protein.[76] The latest findings of the importance of prior gut microbiome confirm that kwashiorkor is not due to overall protein deficiency although the amino acid composition of the diet might possibly have a role in the interplay between diet, infection, and factors that determine what is going on in the child‘s gut.[15]

Free Radicals

While progress is being made in understanding the causes of kwashiorkor, full understanding of the pathophysiology will need to account for the well–established, decreased antioxidant response that allows unabated free radical damage to cells and explains some of the clinical findings in kwashiorkor.[36]

The acute inflammatory cells that form the first line of defense against infections respond with a respiratory burst during which free radicals are produced. This response to infection or cell injury relies on these free radicals for the destruction of bacteria. The free radicals are characterized by one or more unpaired electrons, which make them highly reactive and unstable. Reactive oxygen species include oxygen radicals such as singlet oxygen and nonradicals such as hydrogen peroxide; reactive nitrogen species include nitric oxide. The most potent free radicals include the hydroxyl radical and iron free radicals, which occur when unbound iron is present during free radical production. Free radicals are produced in normal metabolism, and a certain level of these radicals is required for normal functioning. However, this amount is kept to a minimum by a number of scavenging mechanisms, which detoxify the radicals. Free radical production is increased by stresses, which include inflammation, infections, and environmental stresses.[77] Oxidative stress is a process in which the normal balance between pro-oxidants and antioxidants is shifted toward the oxidant side, resulting in an increase in free radicals that cause biologic damage.[78] Increased oxidative stress has been demonstrated in kwashiorkor.[79] The damage caused by free radicals includes disrupting enzymes and nucleic acids and the peroxidation of lipoproteins and EFAs in cell membranes by hydrogen peroxide. Dietary antioxidants protect from oxidant damage either directly by scavenging free radicals or because they are required for enzymes that neutralize free radicals. Examples

include the copper and zinc-dependent enzyme superoxide dismutase, the selenium-dependent enzyme glutathione peroxidaseand catalase. The sulfur amino acids methionine and cysteine are precursors involved in the production of glutathione, which reduces oxidative stress and protects against free radicals damage.

Many studies show low concentrations of antioxidants in the serum and red cells of children with malnutrition, especially kwashiorkor.[40,80] Blood cell concentrations of glutathione, vitamin E, and the vitamin E-to-cholesterol ratio are reduced, resulting in diminished resistance to oxidative stress.[66] It seems that the balance between production and quenching of free radicals is perturbed in kwashiorkor, resulting in oxidative stress. This has been attributed to low levels of dietary antioxidants, with increased free radicals as a consequence of the disturbance in the acute-phase response to infections and other environmental stresses.

In keeping with this concept, a randomized trial of a supplement of N-acetyl cysteine given to children with kwashiorkor improved clearance of edema and shortened time to recovery in children.[39] However, a placebo controlled trial designed to test the dietary antioxidant deficiency hypothesis gave daily supplementation of an antioxidant mixture of riboflavin, vitamin E, selenium, and N-acetylcysteine for 20 weeks to children in Malawi but failed to prevent the occurrence of kwashiorkor.[81] EFAs were not included in the supplement. In this context, simply giving dietary supplements of these antioxidants was not enough to prevent kwashiorkor, but it may well be that this is only part of the story and we need to understand more about the interplay between diet and other determinants of kwashiorkor.

Lipid Metabolism, Polyunsaturated Fatty Acids, and Free Radicals

Recent research investigating the role of lipids in malnutrition lends support to the theory of oxidant stress as a cause of kwashiorkor. It has long been known that there are changes in the lipid profile of red cell membranes and plasma in malnutrition.[82–84] Recent interest has focused on these EFAs and their metabolites as these have protean functions, which include maintaining the integrity of cell membranes and involvement in the synthesis of immunologically active elements such as prostaglandins and leukotrienes.[85] A low proportion of arachidonic acid, one of the EFAs, in the lipid profile of cell membranes and low

concentrations in the plasma of children with severe malnutrition have been reported.[86] These changes in EFAs are not due to dietary deficiency alone and are more likely a result of shifts in the metabolism of these EFAs. In particular, desaturation, which is subject to endocrine influence, is reduced, and low desaturase activity has been reported in SAM.[87] There may also be peroxidative degradation of EFAs in cell membranes. Levels of antioxidants are consistently reduced in SAM, and this increases the peroxidation of membrane long-chain polyunsaturated fatty acids (PUFAs).[88,89]

Leukotrienes are a family of mediators involved in the inflammatory response. The EFA arachidonic acid mentioned above is a precursor of the leukotrienes. Glutathione is also needed for the synthesis of leukotrienes from arachidonic acids, and several studies have documented low levels of whole-blood glutathione and low urinary excretion in kwashiorkor and marasmus patients,[40,90,91] suggesting that synthesis of the leukotrienes might be compromised in severe malnutrition. However, it seems that different leukotrienes are affected in different ways because concentrations of the cysteinyl leukotrienes LTE4 and LTC4, which mediate vascular damage and edema, have been found to be increased in the red cells of children with kwashiorkor but not marasmus, whereas LTB4, which stimulates chemotaxis, was much reduced.[38] Cysteine supplementation improved the erythrocyte synthesis rate in children with kwashiorkor.[39]

The changes in sodium and potassium fluxes across cell membranes characteristic of kwashiorkor may also be explained in this model of reduced antioxidants. In an in vitro model, red cells depleted of glutathione showed increased leakiness to sodium and potassium, mimicking the findings seen in kwashiorkor.[40] The active transport of sodium out of the cell was increased but could not compensate for the increased permeability, resulting in the low K, high Na typical of malnutrition.

In summary, some of the pathology of kwashiorkor can be explained by low antioxidants and increased oxidative stress, resulting in changes of free radical damage, increased sodium/K flux across membranes, triglyceride accumulation in the liver, low albumin, changes in n-6 PUFAs, and altered leukotriene production. Until recently, the determinants of this interplay of nutrient deprivation and infectious or environmental stresses, leading to a disturbed balance in the normal physiologic response to these insults, were not understood.

Exciting results from recent studies in Malawi have been a major break through in understanding of the pathogenesis of Kwashiorkor.[15] These

studies have produced evidence that the gut microbiome in conjunction with a diet high in carbohydrates and deficient in other nutrients predisposes to kwashiorkor. The researchers followed 371 twin pairs from birth collecting monthly fecal samples. During the study,[19] same gender twin pairs became discordant for kwashiorkor and gut microbiome of serial fecal samples from 13 of these were examined. The age-related progression in one of the principal coordinates that describes variability in these fecal microbiomes was absent in the kwashiorkor twin compared to the healthy twin suggesting that some aspect of the microbiome might predispose to kwashiorkor.[15]

Fecal samples from healthy and kwashiorkor children were transplanted into gnotobiotic mice fed either regular chow or a staple-based nutrient deficient "Malawi type diet". The mice receiving the Malawi diet and the kwashiorkor transplant showed marked weight loss where as the other mice did not.[15] When these mice were fed "Ready to Feed Therapeutic food" (RUTF), there was marked weight gain, but the microbiome profile remained unchanged. This provided clear evidence that the combination of the deficient diet and the microbiome was responsible for the weight loss. Further work will no doubt clarify many of the mechanisms involved but of interest, given earlier findings of a possible role for methionine and cysteine, the mice harboring the kwashiorkor microbiomes had significantly lower methionine and cysteine concentrations in feces, amino acids that are deficient in the "Malawi-type diet."[15] They also described urinary metabolite changes suggesting a selective inhibition of some of the enzymes of the tricarboxylic acid cycle involved in energy metabolism and the authors suggest that children with this pattern of gut microbiome might have "difficulty with energy metabolism when exposed to a micro-and macronutrient-deficient low-calorie diet" such as the "Malawi-type" diet.[15]

Changes in Organs and Systems in Malnutrition

In addition to the myriad cellular manifestations of SAM, widespread alterations in organ and system function occur, many of which are relevant to clinical management.

Endocrine System

Endocrine changes mediate the metabolic adaptation to starvation and have been mentioned above. Pancreatic atrophy is a common finding in marasmus.[92] Glucose clearance is reduced in both marasmus and

kwashiorkor due to pancreatic α cell dysfunction rather than insulin resistance.[93] These changes have important consequences on the clinical management of severely malnourished child children are potentially at risk of hyperglycemia when fed high carbohydrate diets or if given intravenous infusions of glucose especially as a bolus[93] yet at the same time vulnerable to hypoglycemia if subjected to periods without food.

Studies in malnutrition have generally reported raised concentrations of growth hormone on admission,[31] but these concentrations reduce in those gaining weight on dietary treatment. Cortisol concentrations rise, especially in kwashiorkor, and the circadian rhythm is abolished, but the response to adrenocorticotropic hormone is preserved.[22] Cortisol levels also rise with infection; 80% of children with kwashiorkor showed a rise in cortisol in response to infection compared with only 50% of those with marasmus[22] This reduced response in marasmus may explain why these children are so susceptible to hypoglycemia. High levels of adrenaline are reported early in hospitalization, especially in kwashiorkor.[22] Both the plasma concentration and the secretion rate of aldosterone are high.[32]

Thyroid gland function is altered in malnutrition.[94,95] During nutrition deprivation, at first thyroxine (T4) increases, but as malnutrition becomes more severe, and especially if kwashiorkor develops, total T4 decreases. There is also a decrease in thyroid binding proteins, but this does not account for all of the reduction in T4 and suggests a primary effect on synthesis.[96] Increased thyroid stimulating hormone secretion heralds recovery.[97] There is also a reduction in deiodination of T4 to triiodothyronine (T3), resulting in reduced T3.

Immune System

Children with severe malnutrition are very susceptible to infection, especially with gram-negative organisms, and deaths are often attributable to sepsis. The subject has been extensively reviewed[70,98,99] and is it is well documented that there are profound changes in cell-mediated immunity, the complement system, and function of polymorphonu clear cells and fewer reported effects on humoral immunity.

Cell-Mediated Immunity

Cell-mediated immunity is altered in severe malnutrition.[70] The thymus, necessary for normal differentiation of T cells, is consistently reduced in size,[100] and the production of thymic hormones is reduced.[98,99] Evidence of T-cell functional impairment also comes from the observation of

reduced cutaneous tests to the recalled antigens, for instance, the tuberculin test[101]; however, studies differ in reports of numbers of circulating T cells and the ratio and effect on T-cell subsets. In general, it appears that T4 cells are relatively reduced, and T8 cells are affected to a lesser degree.

Humoral Immunity

Immune globulin production by B cells has been reported to be normal or raised70 and immunoglobulin (Ig)G seroconversion in response to several vaccines is preserved.[102] Increased IgA has been reported but children with severe malnutrition in Brazil were found to have lower secretory IgA in urine, which increased on rehabilitation.[103] The complement system shows consistent abnormalities with decreased C3 level.[104] Some studies report particularly low concentrations of C3 in kwashiorkor. However, Ekanem and colleagues found that C3 levels, although low, rose significantly in the presence of infection.[71] C4 levels seem to be increased or unaffected by nutritional status. The consequence of these various effects is a reduced capacity of the malnourished child to respond to and combat infections, leading to an increased risk of sepsis, one of the main reasons for mortality in malnourished children.

Liver

In the liver, there is a shift away from the production of carrier proteins and a relative increase in the production of acute inflammatory proteins, indices of response to injury or infection. This is particularly marked in kwashiorkor. The liver in kwashiorkor is often enlarged and shows a fatty infiltrate owing to accumulation of triglycerides.[105] These changes improve on clinical recovery, and there is no evidence that kwashiorkor per se results in long-term liver damage or cirrhosis.[106]

Cardiac System

Cardiac output is reduced in children with acute SAM compared with cardiac output on recovery.[23] There is a reduction in left ventricular mass and impaired function.[107] Sinus bradycardia may be present, as well as mild hypotension. Concomitant deficiencies such as hypokalemia, anemia, and vitamin deficiencies may affect the heart. Pericardial effusion may be present in edematous malnutrition. During recovery, the heart size increases rapidly owing to chamber enlargement, and left ventricle musculature increases in proportion to increasing weight.[108] Heart failure

is a risk in these children and fluid management of children with SAM requires care.[107]

Unnecessary intravenous transfusions including blood transfusions are associated with higher mortality. Bachou analyzed deaths of children admitted to a Ugandan teaching hospital and reported mortality rate of 24%, despite following WHO management criteria. Most deaths occurred in the first week (70%) and 29% in the first 48 hours. Mortality was not associated with HIV status or signs of infection. Mortality was increased almost five times (OR 4.8 95% CI 1.5–7.2) in children with hemoglobin >5g/dL and three times (OR 3.2 95% CI 1.6–4.3) if they received an intravenous infusion in the absence of signs of severe dehydration.[10] Deaths were attributed to fluid overload. These findings led to retraining of hospital staff and a comparison of mortality before and after adherence to the WHO guidelines for IV infusions and blood transfusions. They documented a reduction in inappropriate transfusions and IV infusions and significant reduction in the mortality of children who were transfused and/or infused although overall case mortality in this setting was not reduced.[109]

WHO guidelines advise blood transfusions only when hemoglobin is <5 g/dL and intravenous infusion for children with severe acute dehydration. Under these circumstances, intravenous fluids are essential and should not be withheld from children with severe acute dehydration. Brewster recommends delayed capillary refill time as a useful indicator for dehydration and the need for IV fluids.[110]

Respiratory System

The reduction in muscle mass that occurs in severe malnutrition affects respiratory muscle, including the diaphragm; this leads to reduced muscular function, which influences vital capacity and maximal inspiration and inspiratory pressures.[111] This weakness may be exacerbated by electrolyte abnormalities such as low phosphate[112]and hypokalemia. The ventilatory response to hypoxia is blunted, but not the response to hypercapnia.[113] Despite these alterations, tachypnea and sub costal retraction remain useful signs in diagnosing pneumonia in malnutrition.[114]

Gastrointestinal Tract

Diarrhea and malnutrition often occur together and in circumstances where there is a lack of hygiene and constant exposure to bacterial contamination

of food and water. Children and adults living in tropical unhygienic areas tend to have abnormal intestinal mucosal structure and function, tropical enteropathy, which may or may not be asymptomatic.[41,115] Small bowel bacterial overgrowth may also occur and it is estimated that lack of hygiene contributes to overall malnutrition.[3] The interaction among the environment, microflora of the intestine, nutrition, and growth is not limited to the small intestine. Remarkable advances in the ability to study the pattern of gut microflora, the microbiome and its metabolic activity based on fecal samples suggest that there are complex interactions between the environment, host genome, and diet that determine nutritional outcomes from malnutrition to obesity.[15]

Malnutrition increases the risk of more prolonged episodes of diarrhea,[116] and the combination of persistent diarrhea (more than 14 days of diarrhea) and malnutrition carries an especially high mortality.[117] Any concurrent diarrhea is associated with increased mortality in hospitalized children with SAM, 21% of children with diarrhea on admission to hospital in Kenya with SAM died compared with 12% of those with no diarrhea.[118]

Diarrhea and malnutrition so often coexist that it is difficult to determine the extent to which each contributes to pathology. Nevertheless, both animal and human studies suggest that severe malnutrition per se affects the intestinal tract with reduced gastric acid production, thinning of the small intestinal mucosa, and flattening or disappearance of the villi with relative sparing of the crypts.[23,119] There is increased cellularity of the lamina propria, especially in children with kwashiorkor.[119] This enteropathy is reflected in impaired mucosal function, increased permeability as demonstrated by the double sugar technique, and malabsorption as shown by nutrient absorption studies.[120–122] The mucosa seems to be particularly affected in kwashiorkor. In children with SAM in Malawi, kwashiorkor was associated with abnormal intestinal permeability results, and persistent diarrhea was not an independent risk factor for altered permeability.[121] Loss of the villous architecture of the small intestine for any reason reduces disaccharide activity because the disaccharides, especially lactase, are found at the villous tips. Thus, lactose malabsorption is a common finding in SAM.[41]

Fat malabsorption is also seen in both persistent diarrhea[123] and SAM[124] as well as the mucosal changes; bacterial overgrowth in the upper small intestine has been described in SAM and may lead to bile salt deconjugation, further impairing fat absorption[125], but in contrast to

carbohydrate, fractional fat absorption remains constant over a wide range of intake,[126] so more fat in the diet results in more fat absorbed, and in practice, the increased fat loss is of little clinical consequence. This is of considerable practical importance because the high energy density of fat means that more calories can be given in smaller volume, which is essential when feeding SAM children, who are usually anorexic. RUTF, which is a fat-based food, has been successfully used for the treatment of uncomplicated SAM.[127] Certain nutrients including amino acids, zinc and vitamin A are especially important for mucosal health.[128,129] and additional advantage of RUTF is the ability to sustain fortification with different micronutrients including the fat soluble vitamins.

However, while RUTF represents a significant advance in feeding options for uncomplicated SAM, there is still controversy over the use of F75 and F100 liquid diets especially in children with SAM and diarrhea,[41] and further research is needed to design and test fluid diets that maximize nutrients for gut health, avoid carbohydrate intolerance and address the need for adequate phosphate, magnesium, zinc, and other micronutrients in practical and palatable combinations.

Diets including F75 and F100 that are milk based and fluid provide quality protein and essential elements such as phosphate and magnesium as well as the added micronutrients but the presence of lactose may exacerbate diarrhea.[41] Carbohydrates are well absorbed in the small intestine until disaccharidase capacity is overwhelmed, at which point, the unabsorbed sugars cause osmotic diarrhea and the risk of dehydration. Therefore presenting a mixture of different carbohydrates in small frequent feeds in presentations with relatively slow gastric emptying can help allow even lactose to be tolerated.[130] Studies in children with persistent diarrhea, many of whom were malnourished, demonstrated that locally prepared diets with less than 3 g/kg body weight/day as lactose were well tolerated and led to prompt recovery.[131] Nevertheless in some situations, for instance in Southern Africa where HIV is common, milk-containing foods even in small frequent doses is a problem produce clinically lactose intolerance.[41] As lactose intolerance can cause life-threatening severe diarrhea and dehydration, any suggestion of worsening diarrhea with fecal reducing substances should be taken seriously and the diet changed. It is important not to discontinue food but to seek a low osmolar lactose-free alternative.

Hematology: Anemia

Anemia is common in severe malnutrition and may be attributable either to iron deficiency and/or reduced red cell production in adaptation to a smaller lean body mass.[132] Despite the fact that most children will have been consuming a diet deficient in bioavailable iron, in kwashiorkor, there is elevated hepatic iron, and bone marrow iron shows stainable iron in half of children. This is an unusual finding in marasmus.[132] Iron is normally tightly bound to proteins during storage and transport to prevent free iron forming the extremely damaging hydroxyl radical. In kwashiorkor, the reduced plasma levels of transferrin mean that iron-binding capacity is reduced and the concentration of free iron may increase.[133] Low serum transferrin has been associated with an increased risk of mortality in hospitalized children with SAM.[134] It has been suggested that free iron causes or contributes to the etiology of kwashiorkor,[134] although not all children with kwashiorkor have raised indices of free iron. The homeostatic control of intestinal iron absorption is maintained in children with kwashiorkor.[135] Those with high serum ferritin and percent saturation of transferrin were found to absorb less oral iron than those with low serum ferritins.[135] The iron issue is important not only because of its possible role in the etiology of kwashiorkor but also because iron supplements might actually increase mortality in acute SAM. Smith and colleagues, in a small non randomized study in Nigeria, found higher mortality; 10 of 31 children given iron supplements at admission or day 3 died compared with 2 of 12 non supplemented children.[136]

In contrast, additional iron is required during recovery, as the accompanying increase in lean body and red cell mass quickly depletes iron stores and classic iron deficiency features can occur.[137] Iron supplements should be given during this stage.

Little is known about the relative role of other micronutrient deficiencies in contributing to the anemia of malnutrition. In early studies in South Africa and Egypt, there was no evidence of vitamin B12 deficiency, but serum folic acid levels were low.[138,139] In Cairo, low serum folate levels correlated with megaloblastic findings on bone marrow and with improvement in these parameters following treatment with injected folic acid, but in South Africa, serum folate was unrelated to hemoglobin levels, which improved without folic acid supplements. Copper, riboflavin, and vitamin E deficiencies may contribute to the anemia of SAM.

Skin, Hair, and Teeth

In marasmus, the dry wrinkled loose skin is a result of almost total loss of

subcutaneous fat. This leads to a relative increase in surface area, reduced protection from ambient temperature, and therefore increased susceptibility to hypothermia. Hair is thin, grows slowly, and falls out readily, and on microscopy, there is a lack of hair root bulbs and a shift to the resting state of hair growth.[140] In kwashiorkor, skin changes may be florid and show the characteristic signs described earlier, or they may be absent. Some changes are similar to those seen in acrodermatitis enteropathica, and improvement may occur with zinc ointments, suggesting that concomitant zinc deficiency may be implicated.[141] Other nutrient deficiencies, such as EFA deficiency, the B vitamins, and amino acid deficiencies, especially methionine, also cause skin changes and may contribute. Hair is also affected; depigmentation is a classic sign. Hair follicles show fewer and abnormal growing bulbs, showing atrophy and shaft constriction with depletion of pigment.[142] Even a single episode of prolonged malnutrition in the first year of life delays primary dentition and is associated with higher prevalence of primary dentition caries and possibly also permanent dentition caries.[143,144]

Brain Function and Development

Most child malnutrition occurs at a time of rapid development, so any insult at this time may have far-reaching consequences and SAM has been shown to prejudice the long-term development of children.[145] In addition to primary effects on neuronal tissue, there may be secondary effects owing to anemia, micronutrient deficiencies, apathy, and reduced activity. The environment in which most malnourished children live often fails to provide the stimulation and support that the developing child needs to realize his/her potential. The clinically recovered child usually returns to this same environment, thus making it more difficult to determine the impact of malnutrition per se on long-term development. Early studies comparing the outcome of children recovered from severe malnutrition who either returned home or were adopted into families with more resources suggested that environmental influences outweighed the sequelae of the malnutrition.[146] The randomized trials conducted by Grantham-McGregor in Jamaica[147], and in Bangladesh[148] and others in India[149] have contributed greatly to our understanding of the importance of stimulation independent of dietary therapy in mitigating the effect of severe malnutrition on developmental delay. The benefits of such early stimulation last into adulthood.[150] This important subject is further addressed in Chapter 25, “Neuropsychological Development, Nutrition

and Behavior".

Bones

Children with severe malnutrition often remain stunted after recovery. Branca and colleagues used pyridinoline and deoxypyridinoline, cross-linking amino acids of collagen that are excreted in the urine in proportion to skeletal turnover, to study bone turnover in severe malnutrition.[151] They found low rates of bone turnover during the acute phase of malnutrition and much higher levels during recovery. Age, severity of wasting on admission, and crosslink excretion accounted for 44% of the variability in height velocity during recovery.[151–153]. Bone demineralization has also been reported and may be attributable to phosphate deficiency.[154] Bone changes typical of copper deficiency may also be present, as well as scurvy caused by vitamin C deficiency. Vitamin D deficiency which causes rickets and osteomalacia, deserves special mention. Once a common condition, rickets almost disappeared with the widespread consumption of vitamin D fortified milk, but it is now being reported more frequently and subclinical vitamin D deficiency is thought to be common[155] and Vitamin D is known to be essential for B and T cell function, especially phagocytosis.[156,157] Children with rickets are at greater risk of pneumonia[158] and studies have shown an association between asymptomatic low concentrations of serum 25-hydroxy vitamin D3 (25OHD3) and lower respiratory tract infections.[159] Low concentrations of 25OHD3 have also reported to increase risk of tuberculosis especially in those with certain polymorphisms of the Vitamin D receptor.[160] In addition, there is evidence that vitamin D deficiency in childhood is associated with increased risk of chronic diseases such as type 1 diabetes and some cancers in adults.[155] Although little studied, malnourished children may be at increased risk of vitamin D deficiency because of low dietary calcium and vitamin D intakes, including insufficient vitamin D in breastmilk, low dietary fat and perhaps less sun exposure if they are frequently ill and more likely to be indoors.

CLINICAL MANAGEMENT

In 1999, the WHO published "Guidelines for the Management of Severe Malnutrition: A Manual for Physicians and Other Senior Health Workers".[5] These guidelines were a seminal step forward in the management of this important life-threatening condition. The guidelines

were drawn up by international experts and based on the experience of centers of excellence across the world. The guidelines were simplified into ten steps to facilitate teaching and implementation Figure 14-6.[161]

At that time, there was little evidence available and virtually no randomized clinical trials of treatment options so current knowledge of pathophysiology and best practice was the basis for the recommendations. Although the guidelines were slow to be adopted[162] and are still not used in many areas, considerable improvements in mortality have been demonstrated when they have been implemented[163–167] although the results have been less favorable in some areas notable Southern Africa.[109,110] Over the last few years, there have been some clinical trials of aspects of management but for many other steps there has been no evaluation.[162] The guidelines did not contemplate the contribution of HIV-AIDS to the profile of SAM although this has radically increased the prevalence of severe malnutrition in Southern Africa where mortality rates for SAM remain high.[110] In recent years, there have been major advances in our understanding especially in the pathophysiology of kwashiorkor. In the light of these new findings; a review of the available evidence[162]; the now extensive experience of applying the guidelines in different countries[41,110,20]; and new opportunities offered by simpler diagnostic measures and RUTF the guidelines are currently being reviewed. Nevertheless, the basic principles and the value of standardized care remain relevant today and form the basis of the following discussion.

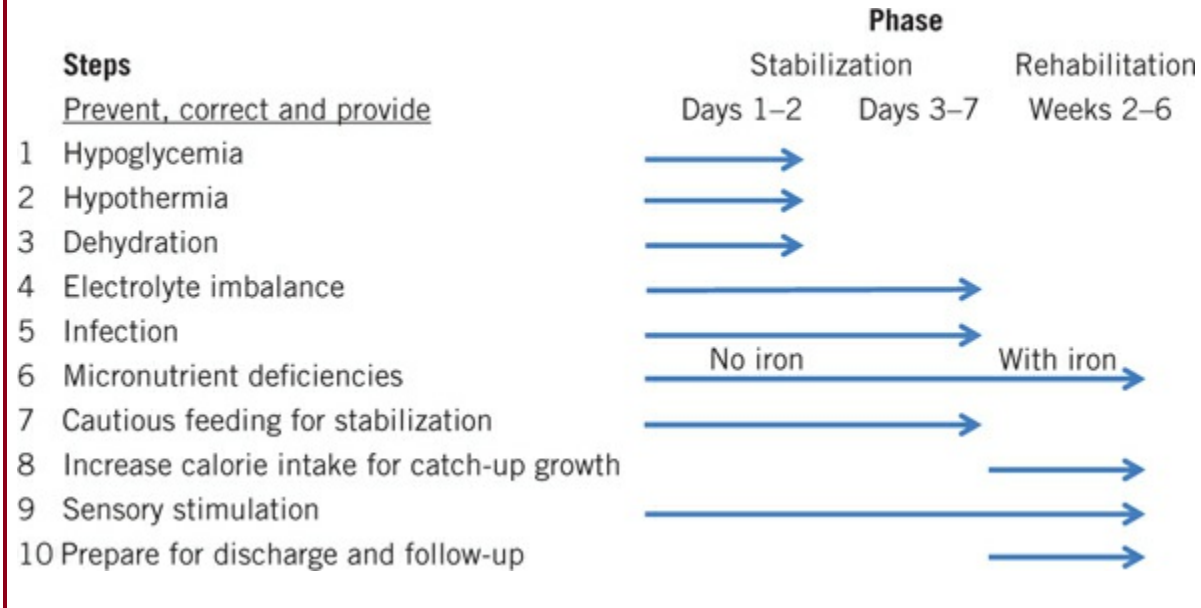


Figure 14-6 Ten steps for the Management of a child with

severe acute malnutrition[161]

CLINICAL ASSESSMENT AND TRIAGE-HOSPITAL VERSUS COMMUNITY REHABILITATION

The diagnosis of SAM has already been discussed and Table 14-1 shows the current recommended criteria.

When malnourished children first present, health personnel are usually faced with a question about whether hospitalization is necessary and advisable. This is often a tricky decision and deserves some discussion. If medical complications associated with increased mortality, for instance, pneumonia, severe diarrhea (more than six stools a day, dehydration), signs of sepsis, hypothermia, severe anemia, fever, electrolyte abnormalities present,[121,168,169] children should be admitted urgently to a hospital or specialized facility. Assessment of appetite is also critical, children with no appetite need admission. Very severe malnutrition and young age are also associated with increased risk of mortality. However, when children are screened either in community household surveys or at presentation in a refugee type setting, as many as 80% of children do not present with life-threatening complications and can be managed at home with domiciliary visits, or in outpatient community-based centers.[6] Table 14-2 lists factors to be taken into consideration when deciding on admission.

Two advances in the management of SAM have made this possible. First, the use of MUAC has simplified the diagnosis. This is easy to use, portable, simple, cheap, and does not require using a chart.[170,6] Recognizing malnutrition in the community, identifying uncomplicated cases, and starting treatment early avoids hospital admission.[6] Second, the availability of RUTF which is nutrient dense, micronutrient fortified, resistant to contamination, and palatable has revolutionized feeding of malnourished children[171–174,012] and has greatly facilitated home or community rehabilitation. Locally made versions of RUTF have also been successful[175] which is good news as the cost of imported RUTF is an issue.

Even when hospitalization is required for the severely ill child with SAM once the acute condition has been treated, and the child is improving, they may be referred to a rehabilitation centers or community care.

Improved appetite is a good sign of recovery. Increasingly, community care following a short stay in hospital is being shown to be feasible especially when ready to use therapeutic food (RUTF) is available. The importance of local support and intensive nutrition education was demonstrated in a study in Bangladesh where community nutrition promoters working within an integrated nutrition project were able to rehabilitate children with moderate malnutrition even without food supplements.[176]

USE OF PROTOCOLS

The child admitted with SAM should be treated according to a locally agreed protocol that takes into account the WHO guidelines. The use of such protocols has been shown to reduce mortality.[6,162,166] The management of malnutrition is a team activity, and all staff who comes into contact with the child or the child‘s caretaker in any way should be trained in the management plan.

An important aspect of these improvements is the organization of services with a view to preventing complications. Early deaths, within the first day, are often attributable to failure to recognize and treat infections adequately, poor management of dehydration and failure to correct electrolyte imbalance especially potassium depletion.[7] Later deaths may be owing to overwhelming infections, but sudden unexpected deaths after a few days may be caused by cardiac failure associated with the refeeding syndrome.[110]

The WHO guidelines were presented as 10 steps Figure 14-6 divided into three phases:

1. The initial or acute phase (2–10 days) when the child is being treated for complications such as dehydration, hypoglycemia, hypothermia, and infections. Dietary therapy starts.
2. The second phase of recovery or rehabilitation (two to six weeks) when the child will increase dietary intake and gain weight.
3. The follow-up phase (6–26 weeks), which may be after discharge.

Phase 1: Examination and Emergency Treatment

A full clinical history should be obtained; symptoms suggestive of infection and diarrhea are particularly relevant. Birth history is important

because low birth weight may explain the infant‘s low weight, and it is essential to review any growth data that may be available. A developmental history and past medical problems may reveal concomitant conditions that cause or contribute to the malnutrition. Most children will have been chronically ill, and information on medication and therapies, including traditional healers, may be relevant. It will be essential to have information on the child‘s diet history, including breast-feeding, family diet, food constraints, and access to food programs, as well as a full social and family history, but this information may be better obtained once the acute phase is completed and is part of building up a relationship with the family. A detailed clinical examination is essential, including length/height and weight and MUAC, but care should be taken not to stress the patient and to protect him/her from cold. Table 14-3 shows some signs and symptoms of particular importance and their management.

Hypoglycemia

As has been mentioned severely malnourished children are susceptible to hypoglycemia. Hypoglycemia is defined as a blood glucose less than 3 mmol/L. If hypoglycemia is diagnosed it may be treated with oral glucose or sucrose, WHO guidelines recommend 50 mL 10% glucose or sucrose orally[5], but this has been criticized because of the possibility of inducing hypoglycemic rebound and intracellular flux that exacerbates hypophosphatemia, hypokalemia, and hypomagnesemia early components of the refeeding syndrome.[110] If children can take orally, they should be started on small frequent feeds from admission rather than oral glucose. On the other hand, children in coma should be assumed to have hypoglycemia and given an infusion of 10% glucose (5 mL/kg) followed by oral glucose or diet. Children with hypoglycemia should be assumed to have infections and be treated with parenteral antibiotics.

TABLE 14-2 Factors to Consider When Admitting Children With SAM

Child Factors	
Severity of the malnutrition	The more severe the malnutrition, the greater the need for specialized refeeding, monitoring, and intervention to treat complications such as infection, electrolyte abnormalities, hypothermia, and hypoglycemia
Edema	Children with edematous malnutrition may not appear to be ill and do not induce the same shock horror response as a child with marasmus. Their edematous weight masks the severity of

	their condition.
Presence of infection	Malnourished children should be regarded as immunodeficient so that infections are more likely to need parenteral antibiotic treatment and close monitoring, which is difficult to do at home. Persistent diarrhea may present special problems and usually quickly improves with a clean diet and appropriate therapy in hospital.
Presence of other conditions	Malnutrition is not always a consequence of primary energy and protein deprivation. The presence of other conditions such as congenital heart disease, cystic fibrosis, and tuberculosis may determine where these children will be best managed. Therapeutic malnutrition units may not be best able to cope with these pediatric emergencies.
Family Circumstances	
Social circumstances	The ability of caregivers to follow treatment schedules at home, who looks after the child, conflicting work and other family constraints on outpatient attendance, nearness to health facilities, availability of backup services and nutritional support in the community are all factors to be taken into consideration.
Previous treatment	Previous attempts at treatment should be taken into account. A child who is worsening despite being in an outpatient or community care program should not have to wait to reach a specific degree of severity to be admitted once the alternatives have failed.
Home social circumstances	These include the nearness to health facilities able to deal with emergencies or monitor the child and the presence of community support with or without food supplements. Children treated in well-organized programs may avoid hospitalization, although there needs to be provision for those who deteriorate, and this may occur suddenly and catastrophically.
Economic considerations	Inevitably, economic considerations will influence decisions and sadly often are the deciding factor, although not in the interest of the child. However, it is very important that health professionals do not add to financial burden with unnecessary tests that do not change management or expensive versions of medicines or medicines of no proven efficacy that compete in the household budget for the food that the child needs or dissuade parents from seeking help for their children.
Hospital Facilities	
Infection risk	Children with severe malnutrition are vulnerable. If admission to hospital means being on a general ward with children with measles and diarrhea, then they may be safer at home, despite the severity of their condition. Obviously, the best option is to provide isolation facilities and hand washing, but

	this may not always be the case in practice.
Feeding arrangements	Children with malnutrition require frequent feeding. Emergency departments may not have provisions for feeding; even on the ward, provision of food may depend on parents, and this may present an additional complication for caretakers if their time is split between home and hospital. Families may have to buy or prepare the food for their child.

The main thing is to prevent hypoglycemia. Travel to the hospital and waits to be seen in the emergency or outpatient department can place the child at risk, and this may be exacerbated by hospital policies such as “nil by mouth” until the child is reviewed and reluctance to start any “treatment” until experts are consulted or test results are available. Mothers should be encouraged to breastfeed their babies while waiting, and a triage system that quickly identifies children with severe malnutrition and directs them into a protocol providing immediate feeding will reduce the risk of hypoglycemia and the attendant mortality. Frequent feeds every two to four hours are needed at first. Night time, when staffing levels are reduced, often provides a challenge, and health facilities should have a plan to cope with this. This might involve having prepared diets or ingredients always available in the emergency department or on the ward and using nasogastric tube feeds if staff or family is not available for the time-consuming process of encouraging the malnourished child to eat frequent feeds. Monitoring is very important. If the mother is accompanying the child, she should have access to refreshments; otherwise, she may be tempted to share the child‘s ration.

TABLE 14-3 Identification and Management of Complications of SAM

Sign or Symptom	Findings	Management
Edema	Usually symmetric and positional; pretibial edema may be evident only on pressing on the shin with a finger and looking for the indentation. The presence of edema also means that gross weight underestimates the child‘s degree of malnutrition.	Edema indicates kwashiorkor and is an indication of severity. Dietary guidelines for kwashiorkor should be applied.
Odd faces, palmar creases, heart murmurs,	Chromosomal and congenital disorders may contribute to malnutrition.	These children should receive the same treatment for their malnutrition, but concomitant conditions will affect outcome

cyanosis, signs of congenital abnormalities		and long-term management.
Developmental assessment	An adequate developmental assessment is not possible in a sick or malnourished child, but signs such as increased tone and contractures may point to cerebral palsy and degenerative neurologic disorders may be discernable.	
Infection	Malnourished children may not mount a full-blown inflammatory response to infection, so signs may be attenuated and diagnostic tests misleading. The febrile response is reduced.	WHO recommends starting all children with severe malnutrition on a broad-spectrum antibiotic such as cotrimoxazole (sulfamethoxazole 25 mg plus trimethoprim 5 mg) even in the absence of signs of infection.
Sepsis	Difficult to distinguish from dehydration. Recent apathy, reduced level of consciousness, jaundice, abdominal distention (babies), fever or hypothermia, hypoglycemia, petechiae, and poor peripheral perfusion are all indicators of possible sepsis.	If there is any doubt, the child should be treated as septic and intravenous antibiotics used. These should be broad-spectrum and cover gram-negative bacteria. Combinations of aminoglycosides such as gentamicin (7.5 mg/kg IM daily) with ampicillin (50 mg/kg 6 hourly) or amoxicillin or third-generation cephalosporins such as ceftriaxone are recommended but should be tailored to local resistance patterns.
Focal infections	Pneumonia is common, but cough may be absent. Tachypnea and subcostal recession are preserved. In otitis media, the tympanum may not appear red, and infections may present with perforation and otic discharge. Urine should be examined for urinary tract infections, but false positives owing to dehydration mean that it is better to obtain nonurgent samples after rehydration. The skin should be carefully examined all over.	Infections should be treated as in well-nourished children, using appropriate antibiotics preferably based on local sensitivity patterns. Because of the depressed immunity, disturbed gut function, which might reduce absorption of oral preparations,[162,163] and limited muscle bulk for frequent intramuscular injection, intravenous antibiotics should be used for all serious infections.

HIV/AIDS	In many parts of the world, a large proportion of children presenting with severe malnutrition will have HIV/AIDS	The same management principles apply and the malnutrition should be treated actively. Improving the malnutrition will improve the child‘s quality of life.
Tuberculosis	Clinical manifestations may be absent. Radiographic changes may be seen only when the child starts to recover from malnutrition. Tuberculin skin tests are usually negative. A positive family history should be sought.	Tuberculosis therapy should be started once tracheal aspirate or gastric cultures have been collected. A trial of therapy is sometimes needed in children in whom there is a suspicion of tuberculosis.
Diarrhea and dehydration	Diarrhea is common, and signs of dehydration should be sought (see Table 14-4)	Diarrhea is not a contraindication to starting diet. Dehydration should be managed as indicated below.
Electrolyte disorders	All malnourished children should be considered to have sodium retention and potassium depletion. Measurement of serum electrolytes is useful to guide management, but low serum sodium is not necessarily an indication for administering sodium. Children with kwashiorkor may urinate infrequently and in small volume; neither this nor normal serum potassium should deter the administration of potassium supplements. Hypotonia may be a manifestation of hypokalemia.	Severely malnourished children are best managed with relatively low sodium ORSs. The old standard WHO ORS (90 mmol sodium) is not recommended. A solution with 60 mmol sodium was found to be superior and to speed recovery. [164] WHO recommends ReSoMal (see Table 14-5), but if not available, the new standard 75 mmol ORS could be used. Potassium at 40 mmol/L is recommended. Treatment schedules are given below.
Hypoglycemia	Blood sugar < 54 mg/dL or 3 mmol/L. Floppiness, unresponsiveness, hypothermia, convulsions, and coma are signs of hypoglycemia. If the child is convulsing, hypoglycemia should be assumed and treatment started empirically even before procedures such as lumbar puncture to exclude meningitis.	If the child is conscious, oral glucose (50 mL of 10% solution) may be given, by nasogastric tube if necessary. Follow with milk or diet as soon as possible. If in coma, an intravenous infusion of 10% glucose (5 mL/kg) should be given and followed by oral glucose or diet. Children with hypoglycemia should be considered septic and treated with antibiotics.
Hypothermia	Rectal temperature is more accurate, and, ideally, a low reading thermometer should be	The child should be warmed gradually either by wrapping in blankets, warming the room, or

	available. Rectal temperature < 35.5°C is diagnostic. Axillary temperature < 35°C is suggestive.	using the skin to skin kangaroo technique. If the child is hypothermic, assume sepsis.
Anemia	Malnourished children are often anemic, but in nonmalarial areas, severe anemia, <8 g/dL, should not be assumed to be due to nutritional deficiency, and other reasons should be sought.	Intestinal parasites, especially hookworm, may be present and should be treated. Anemia is usually chronic and only needs immediate treatment if there are signs of cardiac failure or hemoglobin <5 g/dL. In this case, transfusion should be given with great care as discussed with intravenous therapy.

Hypothermia

Children with SAM, especially those with marasmus, have less insulating fat and a relatively large surface area, so that body temperature tends to reflect ambient temperature, and they are very vulnerable to hypothermia.[168,177] Hypothermia (core temperature <35.5°C) can only be detected with a low reading rectal thermometer, something that is often not available. Hypothermia should be treated with gentle warming for instance by wrapping in a warm environment. Skin–to-skin contact with the mother may be recommended if it is possible. Children with hypothermia should be assumed to have sepsis and treated accordingly.

Prevention is most important and requires planning to avoid having the child unclothed for long periods, for instance, while having radiographs or during medical examination, and providing an ambient temperature that is warm, 25°C-30°C, without draughts. Washing or bathing should be kept to a minimum, and the child should be dried and wrapped immediately. At night, temperatures often fall, so additional precautions may be necessary.

Management of Diarrhea, Dehydration, and Electrolyte Imbalance

The principles of management, rehydration, and continued feeding are not different in the malnourished child (see Chapter 59, “Diarrheal Diseases” for details). Signs of dehydration and their management are shown in Table 14-4. Particular attention has to be paid to the volume of fluid given and electrolyte management. Children with SAM are usually low in potassium and when given high carbohydrate foods hypokalemia can be exacerbated. Additional potassium (final concentration 40 mmol/L) should be added to rehydration fluids and to the diet.

Most children with dehydration can be managed with oral rehydration therapy. In moderate dehydration, 70–100 mL/kg of ORS should be given over 8–12 hours, starting frequently (5 mL/kg every 30 minutes). Picot et al. reviewed the available randomized trials of different ORSs. In one trial comparing standard WHO rehydration solution (3.5% NaCl, sodium 111 mmol/L, glucose 90mmol/L, osmolarity 311 mosmol/L), versus lower osmolarity ORS (H-ORS: 2.6% NaCl, sodium 75 mmol/L, glucose 75 mmol/L, osmolarity 245 mosmol/L), children receiving H-ORS had fewer diarrheal stool and more rapid recovery.[162,178] ReSoMal, a solution containing additional minerals, low sodium, and higher concentration of potassium was recommended in the original WHO guidelines. Only one trial has evaluated ReSoMal compared with standard WHO formula ORS and although ReSoMal resulted in less hypokalemia, it was associated with severe hyponatremia in some children with high purging rates.[162] There is no advantage of ReSoMal over standard low-osmolar ORS with added potassium. Rice-ORS was more effective than Glucose-ORS in cholera with less diarrhea, shorter duration and better weight gain.[162,179]

TABLE 14-4 Signs of Dehydration and Interpretation in SAM

Sign	Mild or Absent		Moderate	Severe (with shock)
	Plan A	Plan B	Plan C	Comment
Conscious level	Normal/alert	Agitated/irritable	Lethargic/comatose	Agitation is typical. Infection and hypoglycemia also give lethargy.
Eyes	Normal	Sunken	Very sunken	Ask mother about eyes
Tears	Present	Absent	Absent	Tears often absent
Buccal mucosa	Moist	Dry	Very dry	Mouth breathing may cause dryness
Skin pinch	Returns quickly	Returns slowly <2 s	Returns very slowly >2 s	More difficult in SAM
Thirst	Drinks normally	Drinks avidly	Unable to drink	If avid, useful

While children continue to have diarrhea, their clinical state should be monitored at least hourly. If vomiting occurs, then rehydration can be

temporarily held for 15–30 minutes and then resumed administering small amounts frequently. If the child refuses to drink or becomes very tired by the effort of drinking, a nasogastrictube should be used. If the child drinks avidly, then only small amounts should be offered by spoon, never by bottle, as taking a large amount at a time may provoke vomiting. One option if vomiting continues is to use continuous rehydration with a nasogastric tube but care should be taken to avoid unintentionally large volumes being administered. If the child does not show signs of recovery (improved general condition, passing urine), consider sepsis. If diarrhea is ongoing, estimate diarrhea losses and increase the rehydration fluid volume accordingly. A diarrheal stool in a child less than 2 years can be estimated as 50–100 mL and 100–200 mL in an older child or in very severe diarrhea. As soon as the child shows signs of clinically recovery and dehydration has improved, the child can be started on frequent small amounts of diet, even though the programmed amount of ORS has not yet been completed or diarrhea continues. Both ORS and diet should be provided in alternate fashion.

Intravenous Therapy

WHO guidelines advise intravenous infusion for children with severe acute dehydration with shock. In these cases, intravenous fluids are essential and lifesaving. Brewster[110] recommends delayed capillary refill time as a useful indicator for dehydration and the need for IV fluids.

In these circumstances, Ringer‘s lactate (RL) has been shown to be as effective as low osmolar fluids.[162] Potassium chloride should be added to provide 20 mmol/L. Concerned about fluid overload, the WHO guidelines recommended slow infusion rates at a maximum of 30 mL/kg/h; however, this may not be adequate to correct shock.[110] A study from Kenya[180] compared half-strength Darrows solution (HSD) at a maximum of 30 mL/kg/h with RL at 40 mL/kg both over two hours. Oliguria was more effectively treated by the RL but neither regime adequately treated shock, which was still present eight hours after starting the infusions in 68% of the HSD and 52% of the RL. Case fatality was unacceptably high at 58% (HSD) and 42% for RL and the trial had to be stopped. Thus, severely dehydrated children in shock, independent of their degree of malnutrition, need more aggressive initial rehydration (60–100 mL/kg rapidly), but in light of the danger of fluid overload frequent and careful hourly monitoring of hydration status, pulse, and respiratory rate for signs of heart failure is mandatory and infusions should be reduced as soon as there are signs of improved hydration[162]. Diuretics such as furosemide 1 mg/kg may

be given if there are signs of heart failure. If the child was dehydrated, improvement should be obvious; if not, septic shock is likely, and parenteral antibiotics should be started, if not already given.

While intravenous infusions should not be withheld when necessary, inappropriate use for instance because it is a “routine in all sick children” or “in case it is needed” intravenous infusions and IV lines should be avoided as they are themselves associated with increased mortality.[109] If antibiotics are to be given intravenously, a heparinized cannula, rather than a continuous infusion, is safer to avoid excess IV fluid. It may be difficult to establish venous access in malnourished children, especially those with edema, but prolonged attempts and “cut-downs” performed without anesthesia and with little attention to sterility place the malnourished child at risk of hypoglycemia, hypothermia, sepsis, and stress, which increase mortality.

Infection

Infections are common in malnourished children but because of a reduced acute-phase response clinical manifestations may be muted. The WHO guidelines advised treating all children with oral antibiotics on admission but at the time no trials had supported this empirical approach. Reluctance to follow this advice and delay in starting antibiotics resulting in deaths due to sepsis was identified as the main avoidable causes of mortality in South African hospitals when WHO guidelines were first being implemented.[164] Recently a randomized controlled trial in children in Malawi with uncomplicated SAM treated at home showed that a one week course of either amoxicillin or a cephalosporin, cefdinir significantly reduced mortality.[181]

Tuberculosis (TB)

Tuberculosis is often common in the same populations that have high rates of SAM. TB can be difficult to diagnose in these children. Clinical signs, Mantoux tests, and X-ray changes may be absent because of attenuated inflammatory responses. In one study in South Africa TB was diagnosed clinically in 24% of children but could only be confirmed in 4%.[162,182] Newer rapid diagnostic techniques are needed.

Human Immunodeficiency Virus

Human immunodeficiency virus (HIV) infection is often associated with severe malnutrition in adults and in some areas, especially in sub-Saharan Africa children with SAM are often HIV positive. For instance, HIV was

detected in 51% of children with SAM in one hospital in Saharan Africa[182] and a review of 4891 children with SAM in sub-Saharan Africa found that 29.2% were infected with HIV and these children had a higher mortality.[183] Malnourished children should be treated with highly active antiretroviral therapy (HAART) but otherwise the management of their malnutrition should be unchanged. Bachou reported that treatment outcomes were no worse in HIV infected children in Uganda[109] although experts agree that more studies are needed on the management of SAM in the context of HIV.[184]

Parasites and Gut Flora

Parasites may also be common and should be treated if identified in the stools. If *Giardia lamblia* is present, metronidazole is recommended. A lower than usual dose, 12.9 mg/kg/d, has been suggested on the basis of pharmacokinetic studies.[185] As mentioned previously, bacterial contamination of the small intestine has been described in children with SAM. Children admitted to the hospital from highly contaminated home environments were found to have bacterial contamination of duodenal fluid regardless of nutritional or diarrheal status.[186] The bacterial flora reduced within days of eating a clean diet and without antibiotic therapy.

Recent studies using modern genomic techniques that can differentiate distinct patterns of the bacterial flora (the gut microbiome) have now demonstrated the importance of this flora in the susceptibility to kwashiorkor.[15]

Dietary Management

Timely initiation of oral feeding is essential and one of the most important messages of the WHO guidelines. This requires planning to have food available, negotiation to avoid the routine “nil by mouth” and consideration of feeding at the same level of importance as treatment of infections, hypothermia, anemia and other more obvious clinical problems. Early initiation of oral feeding was one of the major management changes associated with the introduction of a specific protocol for SAM in Bangladesh that halved mortality. Specifically, deaths from hypoglycemia were reduced.[7]

Dietary therapy is best accomplished when a protocol is in place and specific dietary therapy should be prescribed when the child is first seen. The exact diet to be used will depend on local circumstances. In some cases, it may be the easiest to have infant formula available when the diet

kitchen or daytime nursing is not available, even though it is too expensive to continue. When families are expected to provide food for patients, a special diet will need to be prescribed and made available until family food can take over. This might be the WHO-recommended F75 and F100 (Table 14-5), which have been designed and tested in malnutrition treatment centers.[5,20] The F75 is designed for the initial phase and F100 for the rehabilitation phase, but many centers use F100 from the start, especially in marasmic children. Alternative local diets have been used successfully; an example used at the Instituto de Investigacion Nutricional, Lima, is given in Table 14-6. This diet is palatable and despite containing lactose was well tolerated in most children but requires addition of a vitamins and a mineral mix Table 14-7, but this can be difficult to obtain. Whatever diet is chosen, this should have an energy density of 75–100 kcal/100 mL, the osmolarity should be lower than 350–400 mOsmol/L,[169] and 6%–12% of the calories should come from protein, [187,188] and at least some of this from an animal source (milk, chicken meat, or egg).[41,20] Techniques such as fermentation and the combining complementary vegetable proteins can improve the protein quality and may add valuable vitamins.[189] However, although diets based on vegetable protein have been used successfully, nitrogen retention is usually less than that seen with milk diets.[190,191] When milk is used, it is important to give small frequent feeds and be alert to possible lactose intolerance. At least 50% or more of calories can come from fat; higher fat results in higher overall energy absorption.[126] Diets should be liquid if nasogastric tube feeding is to be used; they should not thicken on standing, a problem of higher proportions of carbohydrate, and they should be palatable. Amylase rich flour has been used successfully to reduce viscosity and increase dietary intakes and acceptance in malnourished children Bangladesh,[192] Malt is an alternative. Sodium should not exceed 2 mmol/kg/d, and potassium should be added to give 5–7 mmol/kg/d. Additional vitamins and minerals should be added as mentioned below.

Ready to feed therapeutic foods (RUTF) high-fat solid rehabilitation diets resistant to bacterial contamination and with excellent preservation of micronutrients[193,194,12] have changed the dietary management of malnourished children once they have overcome acute complications. The composition of RUTF is shown in Table 14-8. Children should receive 80–100 kcal/kg/d; <80 kcal/kg is insufficient to maintain metabolic needs, and >100 kcal/kg may put the child at risk of the refeeding syndrome if high carbohydrate diets are used. Feeds should be given every two to four hours

at first. The amount actually consumed should be monitored by direct observation. Continuing or developing diarrhea does not necessarily mean that the diet should be changed. Only if diarrhea becomes worse, for instance more than six voluminous liquid stools per day, or dehydration occurs, it is necessary to change to a low-lactose or lactose-free diet with the same calorie content. Oral rehydration fluid should be given between meals to compensate for stool losses. An algorithm illustrating the dietary management in the acute phase is shown in Figure 14-7.

TABLE 14-5 Preparation of F75 and F100 Diets

Ingredients	Energy	
	0.75 kcal/mL	1 kcal/mL
Dried skimmed milk (g)	25	80
Sugar (g)	70	50
Cereal flour (g)	35	—
Vegetable oil (g)	27	60
Mineral mix (mL)	20	20
Vitamin mix (mg)	140	140
Water to total volume (mL)	1000	1000

Source: Adapted from World Health Organization.[5] Modified after Schofield and Ashworth.[11]

TABLE 14-6 Preparation of Rice Milk Diet

Ingredients	Energy 1 kcal/mL		Household Measures for 100 mL
Full fat dried milk (g)	76	7.6	5 heaped spoonfuls
Rice (g)	74	7.4	5 heaped spoonfuls
Vegetable oil (cc)	33	3.3	1 oz or use syringe
Sugar (g)	27	2.7	2 heaped spoonfuls
KCl (cc)	9	0.9	Measure with syringe
Water to total volume (cc)	1000	1000	

Instituto de Investigación Nutricional

TABLE 14-7 Composition of Mineral Mix Solution

Substance	Amount
Potassium chloride	89.5 g
Tripotassium citrate	32.4 g

Magnesium chloride (MgCl2.6H2O)	30.5 g
Zinc acetate	3.3 g
Copper sulfate	0.56 g
Sodium selenate[a]	10 mg
Potassium iodide[a]	5 mg
Water to make	1,000mL

[a] If it is not possible to weigh very small amounts accurately, this substance may be omitted.

Source: Adapted from World Health Organization.[5]

TABLE 14-8 Composition of RUTF

Moisture content	2.5%	Maximum	Vitamin A	0.8-1.1	mg/100g
Energy	520-550	kcal/100 g	Vitamin D	15-20	µg/100 g
Proteins	10%-12%	total energy	Vitamin E	20	mg/100 g min
Lipids	45%-60%	total energy	Vitamin K	15-30	µg/100 g
Sodium	290	mg/100 g max	Vitamin B1	0.5	mg/100 g min
Potassium	290	mg/100 g max	Vitamin B2	1.6	mg/100 g min
Calcium	300-600	mg/100 g	Vitamin C	50	mg/100 g min
Phosphorus	300-600	mg/100 g	Vitamin B6	0.6	mg/100 g min
Magnesium	80-140	mg/100 g	Vitamin B12	1.6	µg/100 g min
Iron	10-14	mg/100 g	Folic acid	200	µg/100 g min
Zinc	11-14	mg/100 g	Niacin	5	mg/100 g min
Copper	1.4-1.8	mg/100 g	Pantothenic acid	3	mg/100 g min
Selenium	20-40	µg	Biotin	60	µg/100 g min
Iodine	70-140	µg/100 g	n-6 fatty acids	3%-10%	Total energy
			n-3 fatty acids	0.3%-2.5%	Total energy

Joint Statement WHO/WFP/UNSCN/UNICEF: Community-based management of severe acute malnutrition 2007.

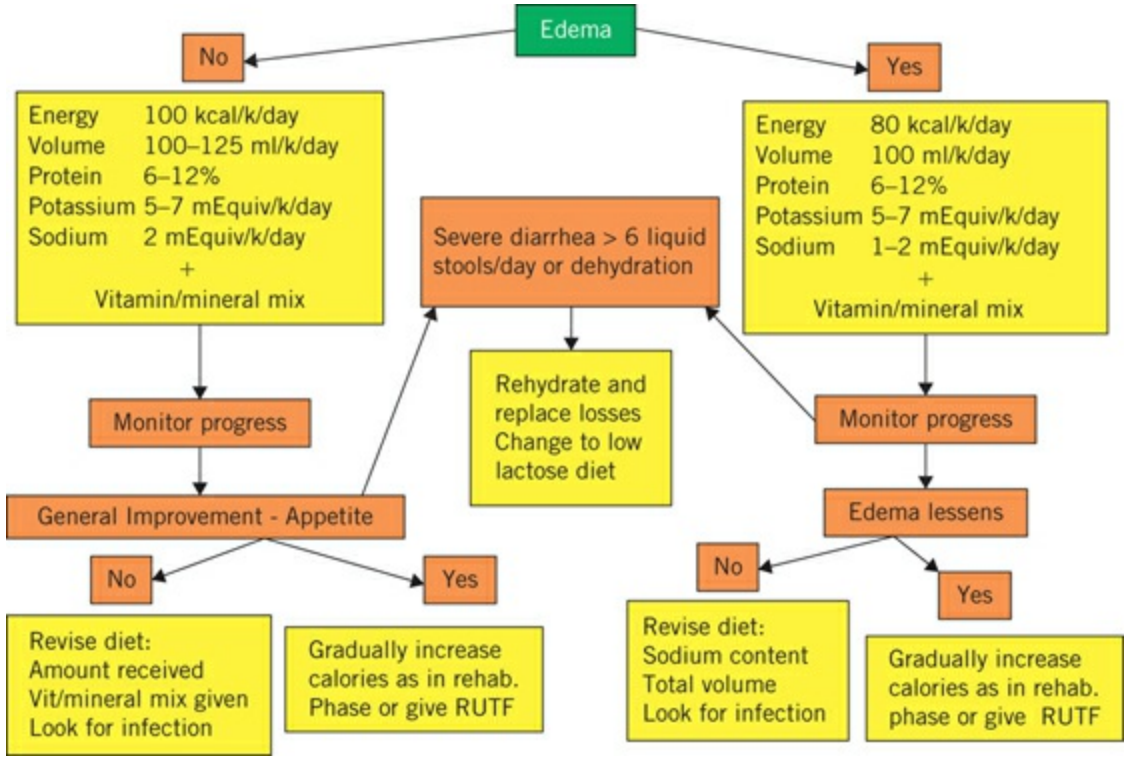


Figure 14-7 Dietary management

Breast-Fed Children

The mothers of breast-fed children should be encouraged to nurse as breastmilk continues to provide important nutritional support even through the second year of life.[57] Breastfeeding should be encouraged in the hospital whenever the child is strong enough to suck; if possible, breastmilk may be extracted to add to the diet, and this may help the mother feel involved and important.

Younger exclusively breast-fed babies may occasionally present with SAM. This may be a result of intrauterine growth retardation but in some cases, especially within the first month, poor weight gain is likely due to poor breastfeeding technique and correcting this may be enough to reverse the situation. These babies must be followed closely and if they do not start putting on weight additional food should be given. For babies under 4 months of age, a modified cow‘s milk formula is the recommended food; for infants older than 4 months, a cereal milk diet can be given. Relactation may be tried to increase breast milk supply once the baby is improving, to try and prevent the mother losing her milk supply and the baby losing the many advantages of breastmilk. The youngest babies present a problem when infant formulas are beyond the resources of parents or the hospital, but priority should be given to obtaining milk

formula for babies under 4 months. Complementing with another woman‘s breast milk is recommendedonly if a safe, sterile supply is available.

Mineral and Vitamin Supplements

Micronutrient deficiencies are common in malnutrition.[20] Prepared diets such as F75, F100, and RUTF are already fortified but for any locally prepared diet additional vitamins and minerals should be added to provide one to two times the daily requirement, sufficient zinc, potassium, magnesium, phosphorus, and thiamine are especially important.[20] Nicotinic acid supplements have been shown to improve weight gain.[195] A local vitamin supplement can be used, but it is often harder to find a mineral source, and WHO provides a recipe for a mineral mix that can be made in a local pharmacy or biochemistry department.[5]

Potassium should be added to the diet to ensure a total of at least 5 mmol/kg/d or more. Higher doses of potassium have been shown to be effective.[47]

Zinc supplementation of malnourished children has been shown to improve weight gain and improve immunologic outcomes, and 1 mL/kg/d of a 1.5% zinc acetate solution daily is recommended although a recent review[162] found the evidence of benefit in SAM inconclusive.[20,196,197] Zinc supplements generally appear to be less effective in children under 1 year[198] and care should be taken not to provide too much zinc as mortality was increased in Bangladeshi children receiving 6 mg/kg/d.[199] Zinc supplements (40 mg/d) given with oral rehydration therapy reduced duration of diarrhea.[200]

Copper deficiency has been reported in malnourished children[201] and copper sulfate should be added to the diet if not included in the mineral mix.

Magnesium is needed for potassium to enter cells, and magnesium supplements should be added to the diet. If not given in the diet or ORS on the first day, an alternative is to give magnesium sulfate 50% solution, 0.3 mL/kg intramuscularly (maximum 2 mL) once, and then include it in the diet. Hypophosphatemia is associated with increased risk of dying and the refeeding syndrome and should be included in the diet. Milk provides an excellent source.

Iron supplements are not recommended during the acute phase because of the risk of producing iron free radicals, as described earlier in the chapter. However, iron supplements are essential once growth and rehabilitation start and muscle and blood are being synthesized. Ferrous

sulfate or fumarate syrup may be given 4 mg elemental iron/kg body weight.[5] As mentioned earlier, electrolyte disturbance is part of the pathophysiology of malnutrition. A triad of interrelated electrolyte deficiencies, potassium, magnesium, and phosphate, occurs. Deficiency of these elements may become especially critical once treatment is instigated and recovery starts.

Regardless of signs of vitamin A deficiency, this should be assumed and retinol palmitate given orally on admission, 50,000 IU for children <6 months age, 100,000 IU for children age 6–12 months, and 200,000 IU for older children. A repeat dose should be given the next day and a third dose after two weeks or on discharge, whichever is sooner.[202] The WHO guidelines recommend a single oral dose of 5 mg folic acid on admission is recommended followed by 1 mg daily.[5] Other vitamins and minerals are best given with the diet or as a daily supplement during recovery.

Children with rickets should be treated with a single intramuscular dose of 600,000 IU of vitamin D as ergocalciferol or cholecalciferol repeated at 12 weeks plus calcium supplements providing 1g calcium as calcium carbonate or lactate daily for 12 weeks. The combination of vitamin D and calcium is more effective than vitamin D alone.[203] Malnourished children, especially those who are predominately breastfed are at risk of vitamin D deficiency. Even children in sunny climates may not get enough exposure to sunlight because of cultural considerations or of the adverse effects of the sun and diets are often low in calcium. Dairy foods to provide calcium should be included in the diet, and vitamin D supplements (400 IU daily) as cholecalciferol or ergocalciferol should be provided, cod liver oil may be a practical alternative.[204]

Anemia

Children with SAM are frequently anemic but this is not usually severe. Blood transfusions should be administered only when the hemoglobin is <5 g/dL as children with less severe anemia have a higher mortality when transfused.[10]

Refeeding Syndrome

The refeeding syndrome is a potentially lethal complication of an increase in calorie intake following starvation. Sudden onset of lethal cardiac failure occurring during refeeding was first reported in the classic studies of experimental starvation in adults in Minnesota and later in starved Japanese prisoners of war, the survivors of concentration camps, and

following the wartime famine in the Netherlands, when, following starvation, there was sudden access to unlimited food.[205–207] This syndrome also occurs in severely malnourished children and may be responsible for considerable mortality, although the precise incidence is difficult to know as, clinically, the syndrome may be difficult to distinguish from pneumonia and sepsis deaths. Sudden death shortly after refeeding has started in a child who seemed to be doing well is suspicious. The syndrome also occurs with parenteral and enteral feeding.

When glucose becomes available during refeeding, especially with carbohydrates, gluconeogenesis is inhibited, and there is a rise in plasma glucose and insulin excretion. Rebound hyperglycemia may occur with polyuria and dehydration. There is a flux of glucose, potassium, magnesium, and phosphate into cells, which may result in rapid lowering of serum levels. The sudden reduction of serum levels of these three electrolytes is thought to explain most of the consequences of the refeeding syndrome.[110,207] Phosphate depletion results in anorexia, adverse neuromuscular effects (e.g., diaphragmatic weakness and respiratory insufficiency), and a reduction in myocardial contractility.[208] The effects of depletion of potassium and magnesium deficiency are similar. Cell membrane electrolyte fluxes are perturbed, and there is an intracellular accumulation of calcium, sodium chloride, and water, which may result in edema and is probably one of the mechanisms that explain recurring edema when children with kwashiorkor are refed too rapidly. Thiamin deficiency may also be important and has been reported in refeeding alcoholics and following starvation treatment of obesity.[207] Its role in the refeeding of malnourished infants is not known. Thiamin is an essential cofactor for various enzymes, and carbohydrate refeeding causes increased thiamin utilization.

The key to prevention of the refeeding syndrome is the avoidance of rapid refeeding, especially with high-carbohydrate diets, whether intravenous or enteral, replenishment with potassium, magnesium, and phosphate, and restricted sodium intake. The safest way to correct phosphate deficiency is with milk diets because oral phosphate salts are laxatives and may well exacerbate diarrhea. Milk is also useful as a good source of magnesium, as well as protein and energy, but lactase intolerance can be a problem. Mixing the milk with a cereal such as rice or sugar reduces the sodium, protein and lactose load, slows gastric emptying and has been used successfully. If intravenous treatment is unavoidable and oral feeding is delayed, phosphate can be given intravenously with care and should be included in parenteral feeding regimens. If children are

dehydrated, magnesium and potassium can be added to oral rehydration therapy as in the ReSoMal recommended by WHO. Vitamins, including thiamin, should be added to the diet from the start. For more details of the management of the refeeding syndrome the reader is referred to several reviews.[207,41]

Skin and Eye Care

The skin lesions of kwashiorkor improve with dietary therapy but, in the initial phase, may become infected and are a potential source of pathogens causing sepsis. Care should be taken to avoid pressure on affected areas and the skin should be kept clean and dry; 1% potassium permanganate solution may be applied to the worst areas. If possible, the perineum and buttocks should be left uncovered and carefully cleaned and dried immediately after defecation. Special care should be taken of the eyes of children with signs of vitamin A deficiency (xerophthalmia). Vitamin A supplements should be started immediately and the eyes should be protected with pads soaked in saline. Atropine and antibiotic eye drops should be applied at least four times daily until there is no sign of inflammation.[209]

Phase 2: Rehabilitation

The initial phase of management lasts while metabolic and electrolyte abnormalities are corrected and infections treated. This phase may last from one to seven days, sometimes longer, if infection is difficult to treat and edema slow to resolve. The recognition of recovery is subjective but usually obvious to staff. There is a general improvement in the child‘s condition. The return of appetite, reduction in apathy, and return of smiling are encouraging signs indicating improvement. Weight gain may not occur at this stage, and in kwashiorkor, there is usually a brisk weight loss as edema disappears. If the child does not seem to be recovering and edema continues without obvious cause, suspect occult infection, for instance, tuberculosis, inadequate potassium repletion, orphosphate deficiency.

Diet

During the rehabilitation phase, the dietary calorie intake should be increased, Figure 14-8. This should be gradual, for instance, an additional 25 kcal/kg/d every other day, to avoid the refeeding syndrome and should be guided by the child‘s appetite and general condition. Diets are calculated based on body weight, so correction will need to be made for

the child‘s changing weight. Most children will reach intakes of between 150 and 250 kcal/kg/d, and rapid weight gain at this stage is desirable as it is associated with the same body composition as lower weight gains.[210] If F75 was used, then F100 should be substituted at this stage if appetite returns, and the child is eating they can be started on RUTF, if available. Whenever possible they can be transferred to rehabilitation care in the community so long as adequate monitoring and support in the community is available. RUTF has facilitated this transition.[110] However, too early discharge in the absence of follow-up rehabilitation leads to very high post discharge mortality.[211]

During this phase, iron supplements should be started at 3–4 mg elemental iron/kg/d and other micronutrient supplements should be continued.

Whatever diet the child eats during this phase as they improve, attention needs to be given to preparing for the home diet. The child may continue to be given RUTF but must ultimately return to eating the family diet. Wherever possible, mothers should be taught about diets based on nutrient-dense local weaning foods and if possible should prepare and feed these preparations in the hospital or community rehabilitation unit. For long-term successful recovery and to prevent relapse, the health personnel needs to work closely with the family during this phase.

An algorithm for the dietary management of the child during this phase is shown in Figure 14-7.

Discharge criteria have been reviewed[6] and increase of 15% weight gain based on % change from admission weight is advised for marasmus and a two weeks after clearance of oedema for children with kwashiorkor.[6] However, recent studies suggest that MUAC is better as this avoids the paradoxical longer stays for children with higher weight on admission which means that when measured as a percentage they have to gain weight in absolute terms.[212]

Physical and Emotional Stimulation

Early physical and emotional stimulation are very important. Even during the initial phase, mothers can be encouraged to talk to and hold their children. They can be taught to massage their children, which has been shown to increase growth recovery. Children should be provided with a visually interesting environment. As soon as possible, play therapy should be instigated. The WHO manual provides examples of easy-to-make toys.[5] Studies in Jamaica and India showed that an early stimulation program

improved growth, complementing micronutrient supplements.[147,149]

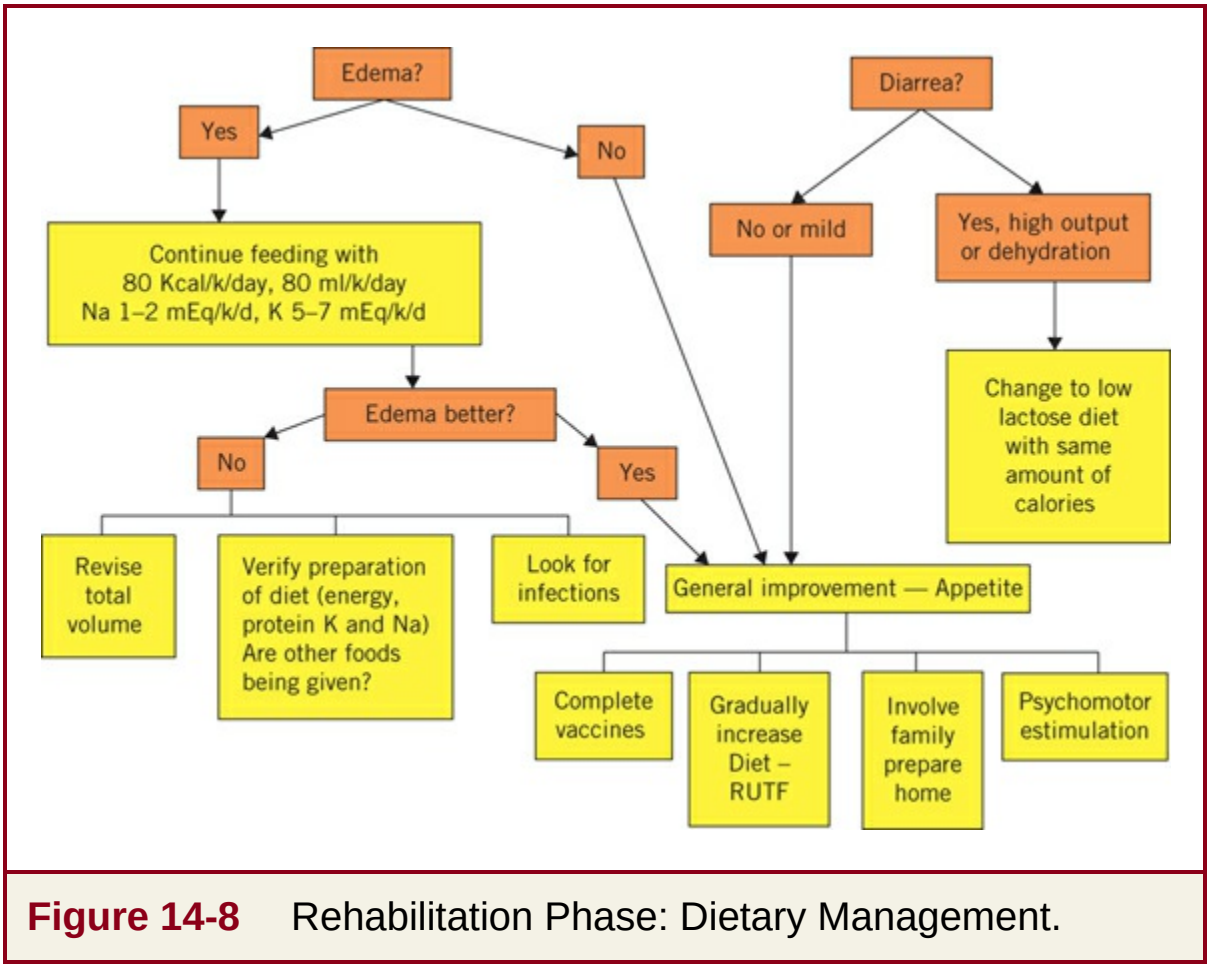


Figure 14-8 Rehabilitation Phase: Dietary Management.

Phase 3

Follow-up of children once they go home is essential as recently recovered malnourished children often relapse, and children may have high mortality rate after discharge.[211] Families should have easy and rapid access to medical care, ideally on the unit where the child is known. The choice of follow-up, like the length of hospital stay, will depend on the availability of services. The availability of a well-designed follow-up program can make all the difference to subsequent risk of dying.[213] Studies following children treated for severe malnutrition have shown variable recovery of nutritional status and intellectual development.[214,215] Sustained recovery of nutritional status is possible, although stunting usually remains.[216] Growth following rehabilitation depends on the favorable home circumstances.[145] When family circumstances remain unchanged, the outlook is poor.[217] A study in Lima comparing children returning home with those who had to be adopted and were integrated into families with

few resource constraints showed both the importance of follow-up care and the potential for recovery of these children.[145] Children should be followed up for at least six months to ensure successful rehabilitation.

CONCLUSIONS

The last few years have brought exciting advances in management of severe malnutrition. Lessons learned in the past are now being applied in hospitals and rehabilitation centers all over the world, although much more need to be done to ensure that this information reaches and changes practice in all of the situations in which severe SAM exists. Greater understanding of the pathophysiology of SAM has contributed to more rational therapy, and some clinical trials of therapies, new diet possibilities, and management strategies are being reported. Nevertheless, there is a great need for more research. The pathophysiology of kwashiorkor is still unclear, but the role of the gut microbiome together with nutrient deficient diets has been a major breakthrough.[15] Even 15 years after the publication of the WHO guidelines there is a lack of evidence in many crucial areas of management.[162]

The long-term goal, of course, is to prevent SAM. Some aspects could be relatively easily achieved. Medical practices, for instance, that deprive children of food while they are ill, especially with diarrhea, have been unequivocally shown to be unnecessary and harmful, but the use of diluted diets or “resting the gut” continues. Attention is now being given to early enteral feeding of critically ill children, which reduces malnutrition and mortality. Great strides have been made in understanding the importance of exclusive breastfeeding and the requirements for adequate complementary feeding practices, but much work is still needed to change complementary feeding practices in many countries and overcome the constraints on food security. Changing child care practices is never easy but a randomized trial showed that nutrition education through the health services was able to prevent stunting in poor periurban population in Peru[218] and an intensive education program in the community was also successful in preventing malnutrition in Bangladesh.[219] Infections, which are very common among the most deprived populations, contribute to malnutrition. Where As children with access to sufficient food of high nutritional value experience only transient effects, those on marginal diets are caught in a cycle of infection, exacerbating malnutrition, and vice versa. Interventions that reduce the burden of disease will reduce malnutrition in these populations. Measles was once a common precursor

to kwashiorkor and severe vitamin A deficiency but, with vaccination, has now disappeared in many countries. Other vaccines against diarrheal diseases could make important contributions. Severe SAM is an entirely preventable disease, but it will continue to killand contribute to the deaths of millions of children while war and natural and man-made disasters continue to affect large parts of the world.

REFERENCES

1. Black RE, Victora CG, Walker SP et al., for the Maternal and Child Nutrition Study Group. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet*. 2013;382:427–51.
2. Caulfield LE, de Onis M, Blossner M, Black RE. Undernutrition as an underlying cause of child deaths associated with diarrhea, pneumonia, malaria, and measles. *Am J Clin Nutr*. 2004;80:193–198.
3. Ahmed T, Hossain M, Sannin KI. Global burden of maternal and child undernutrition and micronutrient deficiencies. *Ann Nutr Metab*. 2012;61(suppl 1):8–17.
4. Bhutta ZA, Salam R. Global nutrition epidemiology and trends. *Ann Nutr Metab*. 2012;61(suppl 1):19–27.
5. World Health Organization. *Management of Severe Malnutrition: A Manual for Physician and Other Senior Health Workers*. Geneva, Switzerland: World Health Organization; 1999.
6. World Health Organization. *Child Growth Standards and the identification of Severe Acute Malnutrition in Infants and Children: A Joint Statement by the World Health Organization and the United Nations Children´s Fund*. New York, NY: World Health Organization and UNICEF; 2009.
7. Ashworth A, Chopra M, McCoy D, et al. WHO guidelines for management of severe malnutrition in rural South African hospitals: effect on case fatality and the influence of operational factors. *Lancet*. 2004;363:1110–1115.
8. Ramirez Prada, Delgado G, Hidalgo CA, et al. Using of WHO guidelines for the management of severe malnutrition to cases of marasmus and kwashiorkor in a Colombia Children´s hospital. *Nutr Hosp*. 2011;26(5):977–983
9. Deen JL, Funck M, Guevara VC, et al. Implementation of WHO guidelines on management of severe malnutrition in hospitals in Africa. *BullWorld Health Organ*. 2003;81(4):237–243.
10. Bachou H, Tumwine JK, Mwadime RKN, Tylleskär T. Risk factors in hospital deaths in severely malnourished children in Kampala, Uganda. *BMC Pediatrics*. 2006;6:7.
11. Schofield C, Ashworth A. Why have mortality rates for severe malnutrition remained so highα *Bull World Health Organ*. 1996;74:223–229.
12. Collins S, Dent N, Binns P, et al. Management of severe acute malnutrition in children. *Lancet*. 2006;308:1992–2000.
13. Venugopalan P, Akinbami FO, Al-Hinai KM, Agarwal AK. Malnutrition in children with congenital heart defects. *SaudiMed J*. 2001;22:964–967.
14. Sharma R, Florea VG, Bolger AP, et al. Wasting as an independent predictor of mortality in patients with cystic fibrosis. *Thorax*. 2001;56:746–750.
15. Smith MI, Yatsunenko T, Manary MJ, etal. Gut microbiomes of Malawian twin pairs discordant for kwashiorkor. *Science*. 2013;339:548–554.

16. Myatt M, Khara T, Collins S. A review of methods to detect cases of severely malnourished children in the community for their admission into community-based therapeutic care programs. *Food Nutr Bull*. 2006;27:S7–S23.
17. Goossens S, Bekele Y, Yun O, et al. Mid-Upper arm circumference based nutrition programming: evidence for a new approach in regions with high burden of acute malnutrition. *Plos One*. 2012;7:1–8. e49320.
18. Briend A, Maire B, Fontaine O, Garenne M. Mid-Upper arm circumference and weight-for-height to identify high-risk malnourished under-five children. *Matern Child Nutr*.2012;8:130–133.
19. WHO/UNICEF/IVACG Task Force. *Vitamin A Supplements: A Guide to Their Use in the Prevention and Treatment of Vitamin A Deficiency and Xerophthalmia*. Geneva, Switzerland: World Health Organization; 1997.
20. Golden MH. Evolution of nutritional management of acute malnutrition. *Indian Pediatr*. 2010;47:667–678.
21. Jahoor F, Badaloo A, Reid M, Forrester T. Protein metabolism in severe childhood malnutrition. *AnnTropPaediatr*. 2008;28:87–101.
22. Frenk S. Metabolic adaptation in protein energy malnutrition. *J Am Coll Nutr*. 1986;5:371–381.
23. Waterlow JC. The nature and significance of nutritional adaptation. *Eur J Clin Nutr*. 1999;53 (suppl 1):S2–S5.
24. Shetty PS. Adaptation to low energy intakes: the response and limits to low intake in infants, children and adults. *Eur J Clin Nutr*. 1999;53 (suppl 1):S14–S33.
25. Gorsky RD, Galloway DH. Activity pattern changes with decrease in food energy intake. *Hum Biol*. 1983;55:577–586.
26. Kerpal-Fronius E, Frank K. Einige besonderheiten de korperzusammensetzung und wasserverteilung bei sauglinsatrophie. *Ann Paediatr*. 1949;173:321–330.
27. Waterlow JS. *Protein-Energy Malnutrition*. Sevenoaks, UK:Edward Arnold; 1992.
28. Garrow JS, Fletcher K, Halliday D. Body composition in severe infantile malnutrition. *J Clin Invest*. 1965;44:417–425.
29. Coward WA, Lunn PG. The biochemistry and physiology of kwashiorkor and marasmus. *Br Med Bull*. 1981;37(1):19–24.
30. Payne-Robinson HM, Coore HG, Golden MH, Simeon DT. Changes in red cell insulin receptors during recovery from severe malnutrition. *Eur J Clin Nutr*. 1990;44:803–812.
31. Brasel J. *Endocrine adaptation to malnutrition*. *Pediatr Res*. 1980;14:1299.
32. Beitins IZ, Graham GG, Kowarski A, Migeon CJ. Adrenal function in normal infants and in marasmus and kwashiorkor:plasma aldosterone concentration and aldosterone secretion rate. *J Pediatr*. 1974;87:444–451.
33. Manary MJ, Broadhead RL, Yarasheski KE. Whole-body protein kinetics in marasmus and kwashiorkor during acute infection. *Am J Clin Nutr*. 1998;67:1205–1209.
34. James WPT. Protein synthesis and breakdown and amino acid catabolism in protein-calorie malnutrition. *Proc Nutr Soc*. 1972;31:225–231.
35. Jahoor F. Effects of decreased availability of sulfur amino acids in severe childhood undernutrition. *NutrRev*. 2012;70:176–187
36. Golden MHN, Ramdath D. Free radicals in the pathogenesis of kwashiorkor. *Proc Nutr Soc*. 1987;46:53–68.
37. Sive AA, Subotzky EF, Malan H, et al. Red blood cell antioxidant enzyme concentrations in kwashiorkor and marasmus. *Ann Trop Paediatr*. 1993;13:33–38
38. Reid M, Badaloo A, Forrester T, et al. In vivo rates of erythrocyte glutathione

synthesis in children with severe protein energy malnutrition. *Am J Physiol Endocrinol Metab*. 2000; 278:E405–E412.
39. Badaloo A, Reid M, Forrester T, et al. Cysteine supplementation improved the erythrocyte synthesis rate in children with severe edematous malnutrition. *Am J Clin Nutr*. 2002;76:646–652.
40. Forrester T, Golden M, Brand S, Swales J. Reduction in vitro of red cell glutathione reproduces defects of cellular sodium transport seen in oedematous malnutrition. *Eur J Clin Nutr*. 1990;44:363–369.
41. Brewster DR. Critical appraisal of the management of severe malnutrition:3. Complications. *J Paediatr Child Health*. 2006;42:583–593
42. Garrow JS. Total body potassium in kwashiorkor and marasmus. *Lancet*. 1965;2:455–458.
43. Alleyne GAO, Millward DJ, Scullard GH. Total body potassium, muscle electrolytes and glycogen in malnourished children. *J Pediatr*. 1970;76:75–81.
44. Alleyne GAO. Studies on total potassium in malnourished infants: factors affecting potassium repletion. *Br J Nutr*. 1970;24:205–212.
45. Mann MD, Bowie MD, Hansen JDL. Total body potassium and serum electrolyte concentrations in protein energy malnutrition. *S Afr Med J*. 1975;49:613–615.
46. Nichols BL, Alvarado J, Rodriguez J. Therapeutic implications of electrolyte, water and nitrogen losses during recovery from protein-energy malnutrition. *J Pediatr*. 1974;84:759–768.
47. Manary MJ, Brewster DR. Potassium supplementation in kwashiorkor. *J Pediatr Gastroenterol Nutr*. 1997;24:194–201.
48. Patrick J, Golden MHB. Leukocyte electrolytes and sodium transport in protein energy malnutrition. *Am J Clin Nutr*. 1977;30:1478–1481.
49. Willis JS, Golden MH. Active and passive transport of sodium and potassium ions in erythrocytes of severely malnourished Jamaican children. *Eur J Clin Nutr*. 1988;42:635–645.
50. Kaplay SS. Erythrocyte membrane Na+ and K+ activated adenosine triphophatase in protein-calorie malnutrition. *Am J Clin Nutr*. 1978;31:579–584.
51. Patrick J. Oedema in protein energy malnutrition: the role of the sodium pump. *Proc Nutr Soc*. 1979;38:61–79.
52. Soliman AT, Alsalmi I, Asfour M. Hypoinsulinaemia has an important role in the development of oedema and hepatomegaly during malnutrition. *J Trop Pediatr*. 1996;42:297–299.
53. Freiman I, Pettifor JM, Gopal MM. Serum phosphorus in protein energy malnutrition. *J Pediatr Gastr Nutr*. 1982;1:547–550.
54. Manary MJ, Hart CA, Whyte MP. Severe hypophosphatemia in children with kwashiorkor is associated with increased mortality. *J Pediatr*. 1998;133:789–791.
55. Waterlow JC, Golden MHN. Serum inorganic phosphate in protein energy malnutrition. *Eur J Clin Nutr*. 1994;48:503–506.
56. Motarjemi Y, Kaferstein F, Moy G, Quevedo F. Contaminated weaning foods: a major risk factor for diarrhoea and associated malnutrition. *Bull World Health Organ*. 1993;71:79–92.
57. World Health Organization. *Complementary Feeding of Young Children in Developing Countries: A Review of Current Scientific Knowledge*. Geneva, Switzerland: World Health Organization; 1998.
58. Bhan MK, Sommerfelt H, Strand T. Micronutrient deficiency in children. *Br J Nutr*. 2001;85(suppl 2):S199–S203.
59. Beisel WR. Herman Award Lecture. Infection-induced malnutrition—from cholera to cytokines. *Am J Clin Nutr*. 1995;62: 813–819.

60. Woolf N. *Cell, Tissue and Disease: The Basis of Pathology*. London, UK:WB Saunders; 2000.
61. Forrester TE, Badaloo AV, Boyne MS, et al. Prenatal factor contribute to the emergence of kwashiorkor or marasmus in severe undernutrition: evidence for the predictive adaptation model. *Plos One*. 2012;7:1–4.
62. Remick DG, Friedland JS. *Cytokines in Health and Disease*. 2nd ed. New York, NY: Marcel Dekker; 1997.
63. Manary MJ, Yarasheski KE, Berger R, et al. Whole body leucine kinetics and the acute phase response during acute infection in marasmic Malawian children. *Pediatr Res* 2004;55:940–946.
64. Muñoz C, Aréval M, López M, Schlesinger L. Impaired interteukin-1 and tumor necrosis factor production in protein-calorie malnutrition. *NutrRes*. 1994;14:347–352.
65. Doherty JF, Golden MHN, Remick DG, Griffin GE. Production of interleukin-6 and tumour necrosis factor-α in vitro is reduced in whole blood of severely malnourished children. *Clin Sci*. 1994;86:347–351
66. Sauerwein RW, Mulder JA, Mulder L, et al. Inflammatory mediators in children with protein-energy malnutrition. *Am J Clin Nutr*. 1997;65:1534–1539.
67. Azavedo ZMA, Luz RA, Victal SH, et al. Increased production of tunmor necrosis factor-α in whole blood cultures from children with primary malnutrition. *Braz J MedBiolRes*. 2005;38:171–183.
68. Schelp FP, Migasena P, Pongpaew P, et al. Serum proteinase inhibitors and other serum proteins in protein-energy malnutrition. *Br J Nutr*. 1977;38:31–38.
69. Hassanein el-S A, Assem HM, Rezk MM, el-Maghraby RM. Study of plasma albumin, transferrin, and fibronectin in children with mild to moderate protein-energy malnutrition. *J Trop Pediatr*. 1998;44:362–365.
70. Rikimaru T, Taniguchi K, Yartey JE, et al. Humoral and cell mediated immunity in malnourished children in Ghana. *Eur J Clin Nutr*. 1998;52:344–350.
71. Ekanem EE, Umotong AB, Raykundalia C, Catty D. Serum Creactive protein and C3 complement protein levels in severely malnourished Nigerian children with and without bacterial infections. *Acta Paediatr*. 1997;86:1317–1320.
72. Doherty JF, Golden MH, Raynes JG, et al. Acute-phase protein response is impaired in severely malnourished children. *Clin Sci (Lond)*. 1993;84:169–175.
73. Morlese JF, Forrester T, Jahoor F. Acute-phase protein response to infection in severe malnutrition. *Am J Physiol* 1998;275(1, pt 1):E112–E117.
74. Manary MJ, Yarasheski KE, Smith S, et al. Protein quantity, not quality, accelerates whole-body leucine kinetics and the acute-phase response during acute infection in marasmic Malawian children. *BrJ Nutr*. 2004;92:589–595.
75. Golden MH. Oedematous malnutrition. *Br Med Bull*. 1998;54:433–444.
76. Golden MH. Protein deficiency, energy deficiency, and the oedema of malnutrition. *Lancet*. 1982;i:1261–1265.
77. Gutteridge JM, Halliwell B. Free radicals and antioxidants in the year 2000. *Ann N Y Acad Sci*. 2000;899:136–147.
78. Evans PS, Halliwell B. Micronutrients: oxidant/antioxidant stress. *Br J Nutr*. 2001;85 (suppl 2):s67–s74.
79. Manary MJ, Leeuwenburgh C, Heinecke JW. Increased oxidative stress in kwashiorkor. *J Pediatr*. 2000;137:421–424.
80. Ashour MN, Salem SI, El-Gadban HM, et al. Antioxidant status in children with protein-energy malnutrition (PEM) living in Cairo, Egypt. *Eur J Clin Nutr*. 1999;53:669–673.
81. Ciliberto H, Ciliberto M, Briend A, et al. Antioxidant supplementation for the

prevention of kwashiorkor in Malawian children: randomized, double-blind, placebo controlled trial. *BMJ*. 2005; 330:1109
82. Decsi T, Koletzko B. Effect of protein energy malnutrition and human immunodeficiency virus-1 infection on essential fatty acid metabolism in children. *Nutrition*. 2000;16:447–453.
83. Franco VH, Hotta JK, Jorge SM, dos Santos JE. Plasma fatty acids in children with grade III protein-energy malnutritionin its different clinical forms: marasmus, marasmic kwashiorkor, and kwashiorkor. *J Trop Pediatr*. 1999;45:71–75.
84. Leichsenring M, Sutterlin N, Less S, et al. Polyunsaturated fatty acids in erythrocyte and plasma lipids of children with severe protein-energy malnutrition. *Acta Paediatr*. 1995;84:516–520.
85. Sammon AM. Dietary linoleic acid, immune inhibition and disease. *Postgrad Med J*. 1999;75:129–132.
86. Houssaini FZ, Foulon T, Iraqi MR, et al. Lipids, lipoproteins, and fatty acids during infantile marasmus in the Fes area of Morocco. *Biomed Pharmacother*. 1999;53:278–283.
87. Holman RT, Johnson SB, Mercuri O, et al. Essential fatty acid deficiency in malnourished children. *Am J Clin Nutr*. 1981;34:1534–1539.
88. Lenhartz H, Ndasi R, Anninos A, et al. The clinical manifestation of the kwashiorkor syndrome is related to increased lipid peroxidation. *J Pediatr*. 1998;132:879–881.
89. Okunade WG, Olorunsogo OO. Effect of reactive oxygen species on the erythrocyte calcium-pump function in protein-energy malnutrition. *Biosci Rep*. 1992;12:433–443.
90. Mayatepek E, Becker K, Gana L, et al. Leukotrienes in the pathophysiology of kwashiorkor. *Lancet*. 1993;342:958–960.
91. Jackson AA. Blood glutathione in severe malnutrition in childhood. *Trans R Soc Trop Med Hyg*. 1986;80:911–913.
92. Brooks SE, Golden MH. The exocrine pancreas in kwashiorkor and marasmus. Light and electron microscopy. *West Indian Med J*. 1992;41:56–60.
93. Spoelstraa MN, Mari A, Mendela M, et al. Kwashiorkor and Marasmus are both associated with impaired glucose clearance related to pancreatic α-cell dysfunction. *Metabolism*. 2012;61:1224–1230.
94. Das BK, Panda BK, Dhingra R, et al. Thyroid hormone studies in protein-energy malnutrition. *J Trop Pediatr*. 1999;45:375–376.
95. Orbak Z, Akin Y, Varoglu E, Tan H. Serum thyroid hormone and thyroid gland weight measurements in protein energy malnutrition. *J Pediatr Endocrinol Metab*. 1998;11:719–724.
96. Onuora C, Maharajan G, Singh A, Etta KM. Thyroid status in various degrees of protein-calorie malnutrition in children. *Clin Endocrinol*. 1983;18:87–93.
97. Kalk WJ, Hofman KJ, Smit AM, et al. Thyroid hormone and carrier protein interrelationships in children recovering from kwashiorkor. *Am J Clin Nutr*. 1986;43:406–413.
98. Chevalier P, Sevilla R, Zalles L, et al. Immuno-nutritional recovery of children with severe malnutrition. *Sante*. 1996;6:201–208.
99. McMurray DN. Cell mediated immunity in nutritional deficiency. *Prog Food Nutr Sci*. 1984;8:193–228
100. Golden MHN, Jackson AA, Golden BE. Effect of zinc on thymus of recently malnourished children. *Lancet*. 1977;ii:1057–1059.
101. Harland PSEG. Tuberculin reactions in malnourished children. *Lancet*. 1965;ii:719–721.

102. El-Gamal Y, Aly RH, Hossny E, et al. Response of Egyptian infants with protein-calorie malnutrition to hepatitis B vaccination. *J Trop Pediatr*. 1996;42:144–145.
103. Teodosio MR, Oliveira EC. Urinary secretory IgA after nutritional rehabilitation. *Braz J Med Biol Res*. 1999;32:421–426.
104. Suskind R, Edelman R, Kukapongs P, et al. Complement activity in children with protein-calorie malnutrition. Am *J Clin Nutr*. 1976;29:1089–1092.
105. Doherty JF, Adam EJ, Griffin GE, Golden MH. Ultrasonographic assessment of the extent of hepatic steatosis in severe malnutrition. *Arch Dis Child*. 1992;67:1348–1352.
106. Cook GC, Hutt MSR. The liver after kwashiorkor. *BMJ*. 1967;3:454–457.
107. Fadden NH, Sayh KI, Shams H, Badrawy H. Myocardial dysfunction in malnourished children. *Ann Pediatr Cardiol*. 2010;3:113–118.
108. Chaithiraphan S. The heart in protein-energy malnutrition. *J Med Assoc Thai*. 1986;69:170–177.
109. Bachou H, Tumwine JK, Mwadime RKN, et al. Reduction of unnecessary transfusion and intravenous fluids in severely malnourished children is not enough to reduce mortality. *AnnTrop Paediatr*. 2008;28:23–33
110. Brewster DR. Inpatient management of severe malnutrition: time for a change in protocol and practice. *AnnTropPaediatr*.2011:31;97–107.
111. Ghignone M, Quintin L. Malnutrition and respiratory function. *Int Anaesthesiol Clin*. 1986;24:65–74.
112. Aubier M, Murciano D, Lecocguic Y, et al. Effect of hypophosphatemia on diaphragmatic contractility in patients with acute respiratory failure. *N Engl J Med*. 1985;313:420–424.
113. Doekel RC, Zwillich CW, Scoggin CH, et al. Clinical semi-starvation:depression of hypoxic ventilatory response. *N Engl J Med*. 1976;295:358–361.
114. Cherian T, Steinhoff MC, Simoes EA, John TJ. Clinical signs of lower respiratory infections in malnourished infants and children. *Pediatr Infect Dis J*. 1997;16:490–494.
115. Fagundez-Neto U, Martins MC, Lima FL, et al. Asymptomatic environmental enteropathy among slum-dwelling infants. *J Am Coll Nutr*. 1994;13:51–56.
116. Black RE, Brown KH, Becker S. Malnutrition is a determining factor in diarrhea duration, but not incidence, among young children in a longitudinal study in rural Bangladesh. *Am J Clin Nutr*. 1984;37:87–94.
117. Bhandari N, Bhan MK, Sazawal S. Importance of persistent diarrhea in childhood mortality. *Acta Paediatr Suppl*. 1992; 381:3–6.
118. Talbert A, Thou N, Karisa J, et al. Diarrhoea complicating severe malnutrition in Kenyan children: A prospective descriptive study of risk factors and outcome. *PLoS One*. 2012;7:1–8.
119. Brunser O. Effect of malnutrition on intestinal structure and function in children. *Clin Gastroenterol*. 1977;2:341–353.
120. Behrens RH, Lunn PG, Northrop CA, et al. Factors affecting the integrity of the intestinal mucosa of Gambian children. *Am J Clin Nutr*. 1987;45:1433–1441.
121. Brewster DR, Manary MJ, Menzies IS, et al. Intestinal permeability in kwashiorkor. *Arch Dis Child*. 1997;76:236–241.
122. James WPT. Sugar absorption and intestinal motility in children when malnourished and after treatment. *Clin Sci*. 1970;39:305–318.
123. Penny ME. The role of the duodenal microflora as a determinant of persistent diarrhoea. *Acta Pediatr Suppl*. 1992;381:114–120.
124. Arora NK, Anand NK, Bhan MK, et al. Nutrient absorption from a fat-enriched diet in young malnourished children: a randomised controlled trial. *Acta Paediatr*.

1998;87:143–148.
125. Viteri FE, Schneider R. Gastrointestinal alterations in protein calorie malnutrition. *Med Clin North Am*. 1974;58:1487–505.
126. Holt E, Tidwell HC, Kirk CM, et al. Studies in fat metabolism.[1]:Fat absorption in normal infants. *J Pediatr*. 1935;6:428–480.
127. Gera T. Efficacy and Safety of Therapeutic nutrition products for home based therapeutic nutrition for severe acute malnutrition: a systematic review. *Indian Pediatr*. 2010:47:709–718.
128. Guerrant RL, Oriá RB, Moore SR, et al. Malnutrition as an enteric infectious disease with long-term effects on child development. *Nutr Rev*. 2008;66:487–505.
129. Chen P, Soares AM, Lima AAM, et al. Association of vitamin A and zinc Status with altered intestinal permeability: analyses of cohort data from northeastern Brazil. *J Health Popul Nutr*. 2003;21(4):309–315.
130. Brown KH, Perez F, Gastañaduy AS. Clinical trial of modified whole milk, lactose-hydrolyzed whole milk, or cereal-milk mixtures for the dietary management of acute childhood diarrhea. *J Pediatr Gastroenterol Nutr*. 1991:12;340–350.
131. Bhan MK, Bhandari N, Bhatnagar S, Bahl R. Epidemiology & management of persistent diarrhoea in children of developing countries. *Indian J Med Res*. 1996;104:103–114.
132. Sive AA, Dempster WS, Rosseau S, et al. Bone marrow and chelatable iron in patients with protein energy malnutrition. *S Afr Med J*. 1996;86:1410–1413.
133. Ramdath DD, Golden MHN. Non-haematological aspects of iron nutrition. *Nutr Res Rev*. 1989;2:29–50.
134. Reddy S, Adcock KJ, Adeshina H, et al. Immunity, transferrin, and survival in kwashiorkor. *BMJ*. 1970;4:268–270.
135. Caballero B, Solomons NW, Batres R, Torún B. Homeostatic mechanisms in the utilization of exogenous iron in children recovering from severe malnutrition. *J Pediatr Gastroenterol Nutr*. 1985;4:97–102.
136. Smith IF, Taiwo O, Golden MH. Plant protein rehabilitation diets and iron supplementation of the protein-energy malnourished child. *Eur J Clin Nutr*. 1989;43:763–768.
137. El Shobaki FA, El Howary MFS, Morcos SR, et al. Iron metabolism in Egyptian infants with protein-calorie deficiency. *Br J Nutr*. 1972;28:81–89.
138. Adams EB, Scragg JN, Naidoo BT, et al. Observations on the aetiology and treatment of anaemia in kwashiorkor. *BMJ*. 1967;3:451–454.
139. Halsted CH, Sourial N, Guindi S, et al. Anemia of kwashiorkor in Cairo: deficiencies of protein, iron and folic acid. *Am J Clin Nutr*. 1969;22:1371–1382.
140. McLaren DS. Skin in protein energy malnutrition. *Arch Dermatol*. 1987;123:1674–1676a.
141. Maverakis E, Fung MA, Lynch PJ et al. Acrodermatitis enteropatica and an overview of zinc metabolism. *J Am Acad Dermatol* 1987;56:116–124
142. Golden MHN, Golden BE, Jackson AA. Skin breakdown in kwashiorkor responds to zinc. *Lancet*. 1980;i:1256.
143. Alvarez JO, Eguren JC, Caceda J, Navia JM. The effect of nutritional status on the age distribution of dental caries in the primary teeth. *J Dent Res*. 1990;69:1564–1566.
144. Alvarez JO, Caceda J, Woolley TW, et al. A longitudinal study of dental caries in the primary teeth of children who suffered from infant malnutrition. *J Dent Res*. 1993;1573–1576
145. Grantham-McGregor SM. A review of studies of the effect of severe malnutrition

on mental development. *J Nutr*. 1995;125:2233S-2238S.
146. Graham GG, Adrianzen TB. Growth, inheritance and environment. *Pediatr Res*. 1971;5:691–697.
147. Grantham-McGregor S, Powell C, Walker S, et al. The long-term follow-up of severely malnourished children who participated in an intervention program. *Child Dev*. 1994;65(2 spec no):428–439.
148. Nahar B, Hossain MI, Hamadani JD, et al. Effects of a community-based approach of food and psychosocial stimulation on growth and development of severely malnourished children in Bangladesh: a randomised trial. *Eur J Clin Nutr*. 2012;66:701–709.
149. Elizabeth KE, Sathy N. The role of developmental stimulation in nutritional rehabilitation. *Indian Pediatr*. 1997;34:681–695.
150. Walker SP, Chang SM, Vera-Hernández M, Grantham-McGregor S. Early childhood stimulation benefits adult competence and reduces violent behavior. *Pediatrics*. 2011;127:849–857.
151. Branca F, Robins SP, Ferro-Luzzi, Golden M. Bone turnover in malnourished children. *Lancet*. 1992;340:1493–1496.
152. Himes JH. Bone growth and development in protein-calorie malnutrition. *World Rev Nutr Diet*. 1978;28:143–187.
153. Walker SP, Golden MHN. Growth in length of children recovering from severe malnutritrion. *Eur J Clin Nutr*. 1988;42:395–404.
154. Ndiaye B, Lemonnier D, Sall MG, et al. Serum osteocalcin regulation in protein-energy malnourished children. *Pediatr Res*. 1995;37:606–610.
155. Holick MF. Resurrection of vitamin D deficiency and rickets. *J Clin Investig* 2006;116:2062–2072.
156. Lemire JM, Adams JS, Kermani-Arab V, et al. 1,25-Dihydroxyvitamin D3 suppresses human T helper/inducer lymphocyte activity in vitro. *J Immunol*. 1985;134:3032–3035.
157. Stroder J, Kasal P. Evaluation of phagocytosis in rickets. *Acta Paediatr Scand*. 1970; 59: 288–292.
158. Muhe L, Lulseged S, Mason KE, Simoes EAF. Case control study of the role of nutritional rickets in the risk of developing pneumonia in Ethiopian children. *Lancet*. 1997;349:1801–1804.
159. Wayse V, Yousafzai A, Mogale K, Filteau S. Association of subclinical vitamin D deficiency with severe acute lower respiratory infection in Indian children under 5y. *Eur J Clin Nutr*. 2004;58:563–567.
160. Wilkinson RJ, Llewelyn M, Toossi Z, et al. Influence of vitamin D deficiency and vitamin D receptor polymorphisms on tuberculosis among Gujarati Asians in west London: a case control study. *Lancet*. 2000;355:618–621.
161. Ashworth A, Jackson A, Khanum S, Schofield C. Ten steps to recovery. *Child Health Dialogue*. 1996;(3–4):10–12.
162. Picot J, Hartwell D, Harris P, et al. The effectiveness of interventions to treat severe acute malnutrition in young children: a systematic review. *Health Technol Assess*. 2012;16(19):1–316.
163. Ahmed T, Ali M, Ullah MM, et al. Mortality in severely malnourished children with diarrhoea and use of a standardized management protocol. *Lancet*. 1999;353:1919–1922.
164. Ashworth A, Chopra M, McCoy D, et al. WHO guidelines for management of severe malnutrition in rural South African hospitals: effect on case fatality and the influence of operational factors. *Lancet*. 2004;363:1110–1115.
165. Deen JL, Funk M, Guevara VC, et al. Implementation of WHO guidelines on

management of severe malnutrition in hospitals in Africa. *Bull WHO*. 2003;81:237–243.
166. Hossain MI, Dodd NS, Ahmed T, et al. Experience in managing severe malnutrition in a government tertiary treatment facility in Bangladesh. *J Health Popul Nutr*. 2009;27:72–80.
167. Ramirez Prada D, Delgado G, Hidalgo Patiño CA, et al. Using of WHO guidelines for the management of severe malnutrition to cases of marasmus and kwashiorkor in a Colombia children`s hospital. *Nutr Hosp*. 2011;26:977–983.
168. Gernaat HBPE, Dechering WHJC, Voorhoeve HWA. Mortality in severe protein energy malnutrition at Nchelenge, Zambia. *J Trop Pediatr*. 1998;44:211–217.
169. Ahmed T, Begum B, Badiuzzaman, et al. Management of severe malnutrition and diarrhea. *Indian J Pediatr*. 2001;68:45–51.
170. Myatt M, Khara T, Collins S. A review of methods to detect cases of severely malnourished children in the community for their admission into community-based therapeutic care programs. *Food Nutr Bull*. 2006;27 (suppl 3):S7–S23.
171. Briend A, Lacsala R, Prudhon C, et al. Ready-to-use therapeutic food for treatment of marasmus. *Lancet*. 1999;1999:1767–1768.
172. Brown KH, Nyirandutiye DH, Jungjohann S. Management of children with acute malnutrition in resource-poor settings. *Nat Rev Endocrinol*. 2009;5:587–603.
173. Linneman Z, Matilsky D, Ndekha M, et al. A large scale operational study of home-based therapy with ready-to-use therapeutic food in childhood malnutrition in Malawi. *MaternChild Nutr*. 2007;3:206–215.
174. Ciliberto MA, Sandige H, Ndekha MJ, et al. Comparison of home-based therapy with ready to use therapeutic food with standard therapy in the treatment of malnourished Malawian children: a controlled randomized effectiveness trial. *Am J clin Nutr*. 2005;81:864–870.
175. Sandige H, Ndekha MJ, Briend A, et al. Home-based treatment of malnourished Malawian children with locally produced or imported ready-to-use food. *J. Pediatr Gastroenterol Nutr*. 2004;39: 141–146
176. Roy SK, Fuchs GJ, Mahmud Z, et al. Intensive nutrition with or without supplementary feeding improves the nutritional status of moderately-malnourished children in Bangladesh. *J Health Popul Nutr*. 2005;23:320–330.
177. Brook OG, Harris M, Salvosa CB. The response of malnourished babies to cold. *J Physiol*. 1973;233:75–91.
178. Dutta P, Mitra U, Manna B, et al. Double blind randomized trial of hypo-osmolar oral rehydration salt solution in dehydrating acute diarrhea in severely malnourished (marasmic) children. *Arch Dis Child*. 2001;84:237–240.
179. Alam NH, Islam S, Sather S, et al. Safety of rapid intravenous rehydration and comparative efficacy of three oral rehydration solutions in the treatment of sevely malnourished children with dehydrating cholera. *J Pediatr Gastroent Nutr*. 2009:48;318–327.
180. Akesh S, Karisa J, Nakamya P, et al. Phase II trial of isotonic fluid resuscitation in Kenyan children with severe malnutrition and hypovolaemia. *BMC Pediatr*. 2010;10:71.
181. Trehan I, Goldbach HS, LaGrone LN, et al. Antibiotics as part of the management of severe acute malnutrition. *N Engl J Med*. 2013;368(5):425–435.
182. De Maayer T, SaloojeeH. Clinical outcomes of severe malnutrition in a high tuberculosis and HIV setting. *Arch Dis Child*. 2011;96;560–564.
183. Fergussen P, Tomkins A. HIV prevalence and mortality among children undergoing treatment for severe acute malnutrition in sub-Saharan Africa: a systematic review and meta-analysis. *Trans Roy Soc Trop Med Hyg*.

2009;103:541–548.
184. Heikens GT, Bunn J, Amadi B, et al; Blantyre Working Group. Case management of HIV-infected severely malnourished children: challenges in the area of highest prevalence. *Lancet*. 2008;371:1305–1307.
185. Lares-Asseff I, Cravioto J, Santiago P, Pérez-Ortíz B. A new dosing regime for metronidazole in malnourished children. *Scand J Infect Dis*. 1993;25:115–121.
186. Penny ME, Paredes P, Brown KH, et al. Lack of a role of the duodenal flora in pathogenesis of persistent diarrhea and diarrhea-related malabsorption in Peruvian children. *Pediatr Infect Dis J*. 1990;97:479–487.
187. Brown KH. Appropriate diets for the rehabilitation of malnourished children in the community setting. *Acta Pediatr Scand Suppl*. 1991;374:151–159.
188. Thomas M, Anand NK, Gupta S. Evaluation of different regimes of management in malnourished children. *Indian Pediatr*. 1977;14:725–730.
189. Kalavi FNM, Muroki NM, Omwega AM, Mwadime RKN. Effect of tempe-yellow maize porridge and milk yellow-maize porridge on growth rate, diarrhoea, and duration of rehabilitation of malnourished children. *East Afr Med J*. 1996;73:427–431.
190. Brewster DR, Manary MJ, Menzies IS, et al. Comparison of milk and maize based diets in kwashiorkor. *Arch Dis Child*. 1997;76:242–248.
191. Hansen JDL, Schendel HE, Wilkins JA, Brock JF. Nitrogen metabolism in children with kwashiorkor receiving milk and vegetable diets. *Pediatrics*. 1960;25:258–282.
192. Hossain MI, Wahed MA, Ahmed S. Increased food intake after the addition of amylase-rich flour to supplementary food for malnourished children in rural communities of Bangladesh. *Food Nutr Bull*. 2005;26:323–329.
193. Briend A. Highly nutrient-dense spreads: a new approach to delivering multiple micronutrients to high-risk groups. *Br J Nutr*. 2001;85 (suppl 2):S175–S179.
194. Manary MJ, Ndkeha MJ, Ashorn P, et al. Home based therapy for severe malnutrition with ready-to-use food. *Arch Dis Child*. 2004;89:557–561.
195. Philip L, Sugana Bai NB, Sathy S, Soman CR. Effect of addition of nicotinic acid to the diet on the rate of weight gain of marasmic children. *Indian Pediatr*. 1982;19:775–777.
196. Golden MH, Harland PS, Golden BE, Jackson AA. Zinc and immunocompetence in protein-energy malnutrition. *Lancet*. 1978;1:1226–1228.
197. Khanum S, Alam AN, Anwar I, et al. Effect of zinc supplementation on the dietary intake and weight gain of Bangladeshi children recovering from protein-energy malnutrition. *Eur J Clin Nutr*. 1988;42:709–714.
198. Penny ME. Zinc supplements in public health. *Ann Nutr Metab*. 2013;62 (suppl1):31–42.
199. Doherty C, Sarkar K, Shakur S, et al. Zinc supplementation and severe malnutrition—higher dosage regimes associated with increased mortality but not through the precipitation of copper deficiency. *J Pediatr Gastroenterol Nutr*. 1998;26:589.
200. Dutta P, Mitra U, Datta A, et al. Impact of zinc supplement in malnourished children with acute watery diarrhea. *J Trop Pediatr*. 2000;46:259–263
201. Cordano A, Baertl JM, Graham GG. Copper deficiency in infancy. *Pediatrics*. 1964;34:324–336.
202. WHO/UNICEF/IVACG Task Force. *Vitamin A Supplements: A Guide to Their Use in the Prevention and Treatment of Vitamin A Deficiency and Xerophthalmia*. Geneva, Switzerland:WHO; 1997.
203. Thacher TD, Fischer PR, Pettiflor JM, et al. A comparison of calcium, vitamin D,

or both for nutritional rickets in Nigerian Children. *NEngl JMed*. 1999;341:563–568.
204. Gartner LM, Greer FR. Prevention of rickets and vitamin D deficiency: new guidelines for vitamin D intake. *Pediatrics*. 2003;111:908–910.
205. Keys A, Brozek J, Henschel A, et al. *The Biology of Human Starvation.* Vols 1 and 2. Minneapolis, MN: University of Minnesota Press; 1950.
206. Solomon SM, Kirby DF. The refeeding syndrome: a review. *JPEN J Parenter Enteral Nutr*. 1990;14:90–97.
207. Fuentebella J, Kerner JA. Refeeding síndrome. *Pediatr Clin N Am*. 2009;56:1201–1210.
208. Kreusser W, Vetter H, Mittmann H, et al. Cardiac function and metabolism in phosphate depleted dogs—effects of hormone stimulation. *Adv Exp Med Biol*. 1982;151:251–258.
209. World Health Organization/UNICEF. *Management of the Child With a Serious Infection or Severe Malnutrition*. Geneva, Switzerland: WHO; 2000.
210. Fjeld CR, Schoeller DA, Brown KH. Body composition of children recovering from severe protein-energy malnutrition at two rates of catch-up growth. *Am J Clin Nutr*. 1989;50:1266–1275.
211. Sadre M, Donozo G, Hedayat H. The fate of the hospitalized malnourished child in Iran. *Environ Child Health*. 1973;29–31.
212. Dale NM, Myatt M, Prudhon C, Briend A. Using mid-upper arm circumference to end treatment of severe acute malnutrition leads to higher weight gains in the most malnourished children. *PLoS ONE*. 2013;8(2): e55404.
213. Pecoul B, Soutif C, Hounkpevi M, Ducos M. Efficacy of a therapeutic feeding centre evaluated during hospitalisation and a follow-up period, Tahoua, Niger, 1987–1988. *Ann Trop Pediatr*. 1992;12:47–54.
214. Stoch MB, Smythe PM. 15 year development study on effects of severe undernutrition during infancy on subsequent physical growth and intellectual functioning. *Arch Dis Child*. 1976;51:327–336.
215. Winick M. Long term effects of kwashiorkor. *J Pediatr Gastroenterol Nutr*. 1987;6:833–840.
216. Banurpurmath CR, Prasad SM, Banurpurmath S, Kesaree N. Follow-up studies of survivors of severe protein-energy malnutrition. *Indian Pediatr*. 1999;36:139–143.
217. Hennart P, Beghin D, Bossuyt M. Long-term follow-up of severe protein-energy malnutrition in Eastern Zaire. *J Trop Pediatr*. 1987;33:10–12.
218. Penny ME, Creed-Kanashiro HM, Robert RC, et al. Effectiveness of an educational intervention delivered through the health services to improve nutrition in young children: a cluster-randomised controlled trial. *Lancet*. 2005;365:1863–1872
219. Roy SK, Fuchs GJ, Mahmud Z, et al. Intensive nutrition education with or without supplementary feeding improves the nutritional status of moderately-malnourished children in Bangladesh. *J Health Popul Nutr*. 2005;23:320–330.

Childhood Malnutrition: Prevention and Control at the National Level

15

Gerardo Weisstaub, MD, MSc, Ana María Aguilar, MD, MSc, Magdalena Araya, MD, PhD, Ann Hill, PhD, and Ricardo Uauy, MD, PhD

NUTRITION A KEY FACTOR FOR HUMAN AND SOCIAL DEVELOPMENT

The nutritional status of children is directly related to their health condition; both in turn are important determinants of the human and social development of communities around the world. Improvements in nutritional and health status increase the chances of child survival and both are preconditions for economic and social development. UNICEF has proposed a conceptual model to understand the causation of malnutrition beyond the obvious immediate determinants; more recently in 2013, the Lancet has updated the classical model focusing on necessary actions to achieve optimum fetal and child nutrition and development.[1–4] Undoubtedly, the basic determinants relate to the social, economic, cultural, and political structure of society. These define what resources are controlled by individual families including access to information and education, household food security, care of women and children, access to

health, and a healthy environment. It is within this context that children have inadequate access to food in terms of quantity and quality; in addition, infection will further compromise nutrition by augmenting nutrient losses and increasing nutrient needs. (Figure 15-1)

In 2011, 6.9 million children under the age of 5 years died worldwide, one third of them related to increased susceptibility to illnesses due to undernutrition. An estimated 178 million children under 5 years are stunted, 55 million are wasted, and 19 million of these are severely affected and are at a higher risk of premature death, the vast majority being from Sub-Saharan Africa and South-Central Asia.[5] The risk factors for undernutrition include low birth weight, inadequate breastfeeding, improper complementary feeding, and recurrent infections. Infectious diseases often coexist with micronutrient deficiencies and exhibit complex interactions leading to the vicious cycle of malnutrition and infections. Diarrhea, often linked with a short duration of exclusive breastfeeding, and the poor selection and intake of complementary food are major contributors to undernutrition.[1]

Nutrition a basic human right and has been expressly recognized as such by international human rights covenants since 1924,[6] yet this "right" is commonly the subject of political demagogy rather than being actively upheld, respected, protected, and promoted. The relationship between poverty and malnutrition has been defined as a vicious cycle, where "poverty conditions nutritional status" and vice versa "poor nutrition leads to poverty" However, the relationship is by no means linear. There are multiple examples that illustrate how nutrition can improve despite economic limitations. While economic development is by no means sufficient to improve nutrition, especially in countries where wealth is concentrated among the few. The UN Human Development Index illustrates how nutrition and health can be dissociated with economic status.[7] Recent analysis of trends reveals significant reductions in malnutrition assessed by underweight or stunted linear growth except in Sub-Saharan Africa and Central America. These regions have experienced protracted civil strife and Africa as a region has been stricken by HIV/AIDS and its consequences. On the contrary, Latin America and the Caribbean combined have reduced malnutrition to about a third of the 1970 figures despite the minimal changes in the proportion of families living in poverty (Figure 15-2).[8] This concept should be stressed since health professionals frequently work under the assumption that nothing can be done in terms of prevention and control of malnutrition unless social conditions change and poverty reduction is achieved. Undoubtedly,

both should proceed in tandem, since improved nutrition will contribute to socio-economic improvement and vice-versa socio-economic progress leads to lower malnutrition rates.

WHO, UNICEF, and other international organizations concerned with malnutrition have recognized for several decades the role of malnutrition as a key determinant of infant mortality? The immediate cause of death may be diarrhea or pneumonia but the risk of dying from these conditions is increased substantially by severe and moderate malnutrition. The work of Puffer and Serrano in the Americas in the 1970s and more recently Pelletier and Habicht working with data from Africa and Asia have documented with precision contribution of malnutrition to infant mortality.[9,10] The conclusion reached by both is that over half of all infant deaths are determined by malnutrition. The recent Lancet series discusses nutrition and infection interactions and the contribution of malnutrition to infant death as crucial factors. Breastfeeding initiation within 24 hours of birth is associated with a 44%–45% reduction in all-cause and infection-related neonatal mortality, and is thought to mainly operate through the effects of exclusive breastfeeding. Vitamin A supplementation also reduced the incidence of diarrhea and measles in children aged 6–59 months, but there was no effect on mortality and morbidity related to respiratory infections. Preventive zinc supplementation in populations at risk of zinc deficiency reduces the risk of morbidity from childhood diarrhea and acute lower respiratory infections.[2]

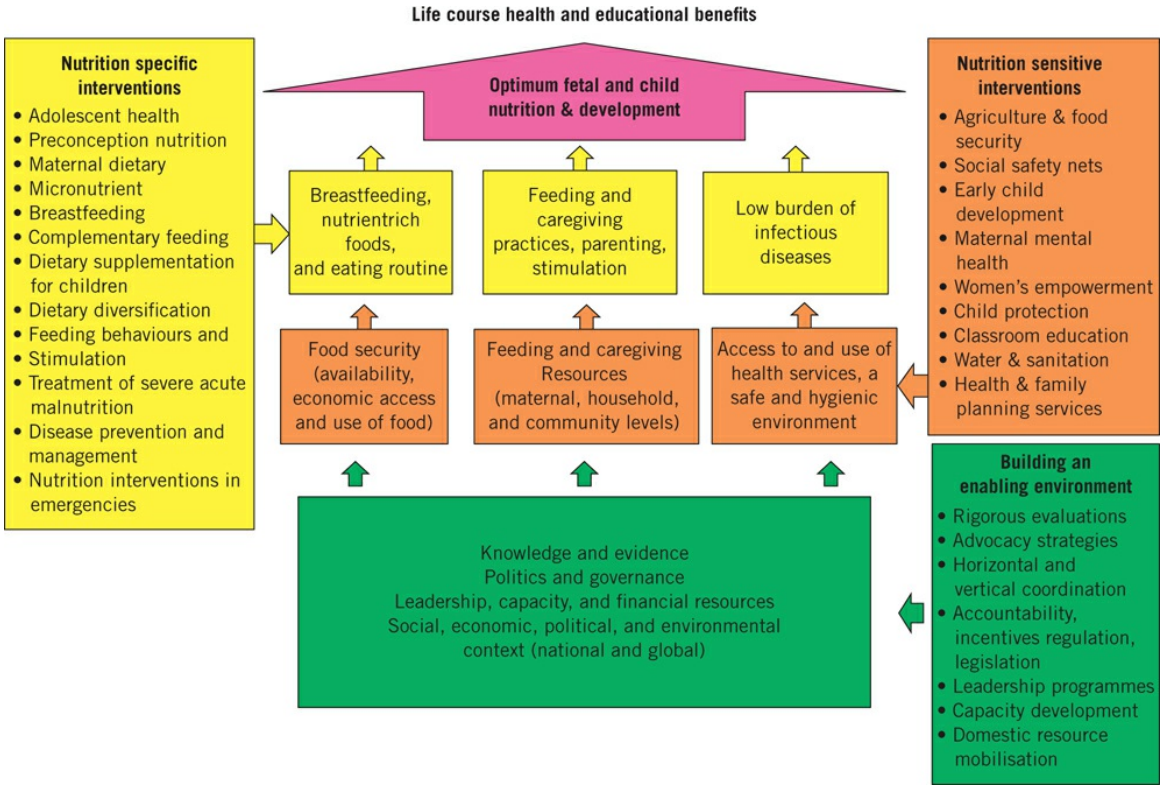


Figure 15-1 Conceptual Model of Child Development useful in the analysis prevention and control of malnutrition. Adapted from Lancet 2013. Modified from Black RE, Victora CG, Walker SP, et al.[1]

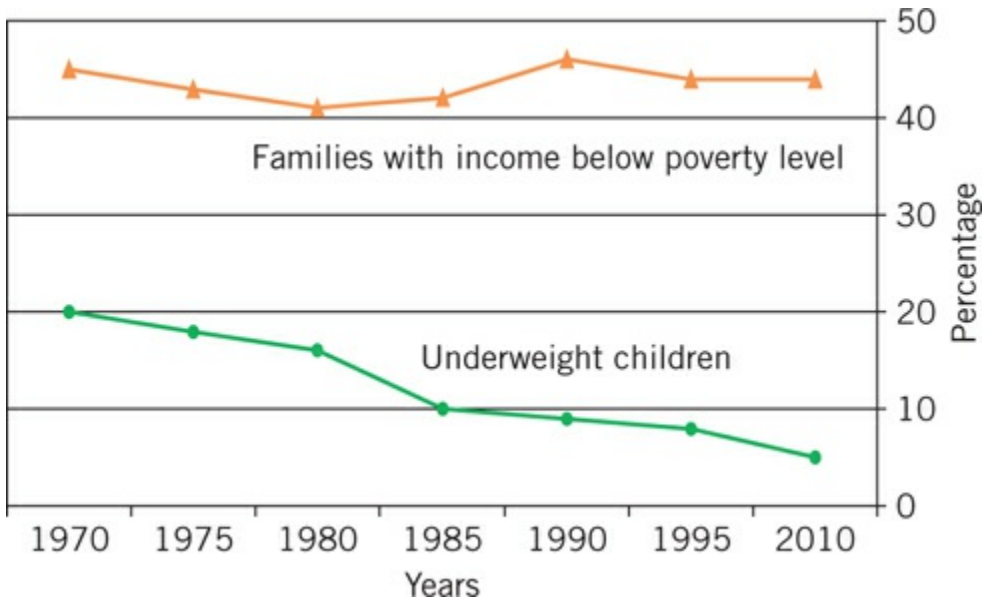


Figure 15-2 Child undernutrition in Latin America and the Caribbean: trends, reasons and lessons. Over the past 30 years, population under poverty line has remained stable, but malnutrition has declined significantly.

MALNUTRITION AS A GLOBAL PUBLIC HEALTH PROBLEM

Weight, height, and mid-upper arm circumference (MUAC) measurements are widely used to assess nutritional adequacy. A low weight-for-age is a useful indicator of malnutrition but has limited clinical significance since it does not distinguish children who are thin (wasted) from those who are short (stunted). Thus, short children may be underweight-for-age but of normal weight-for-height. A low height-for-age represents the cumulative impact of adverse events from conception onward, and the prevalence of stunting in a population is used as a proxy measure for social disadvantage. Additionally, the relationship between child malnutrition (underweight and stunting) and adult adiposity needs to be considered relative to rising prevalence of noncommunicable chronic diseases in developing and transitional countries.[11]

Malnutrition as a public health problem is assessed based on growth indices relative to those derived from a standard population. The child growth standards have been developed by WHO based on the growth of more than 8000 healthy infants and children (from middle income, nonsmoking and well-educated mothers from Brazil, Norway, India, Ghana, USA, and Oman), who were exclusively breast fed for four to six months and then given appropriate complementary foods in addition to breast milk. A finding of the new standard is that child growth during the first years of life is very similar across different ethnic backgrounds. This confirms that existing differences in growth across countries are predominantly environmentally derived and can be narrowed over time. The new standard supports the right of all children to grow equally well and demands that society provides an environment that does not restrict growth. The National Center for Health Statistics (NCHS) growth reference should no longer be used for infants 0 to 36 months, this is derived from children growing in a relatively affluent, predominantly artificially fed, and rural community from Yellow Springs Ohio over 30 years ago.[12]

Most countries have adopted the WHO growth standard which

provides harmonized best practices in child growth assessment and established the breast-fed infant as the norm against which to assess compliance with children's right to achieve their full genetic growth potential.[13]

The sixth report on the world nutrition situation (Administrative Committee on Coordination [ACC]/Sub-Committee on Nutrition [SCN] 2010) provides a broad perspective among regions and a comparison of more than 30 years (ACC/SCN, 1993), this time using the WHO's anthropometric standards.[14] Surveys reporting the prevalence of underweight and stunting in the world measured by serial anthropometry provide a measure of the changes. Based on underweight trends available from 86 countries in Africa, Asia, South-Central America, and Caribbean, the SCN report found that 44% of the countries are improving while 21% deteriorating, although the overall tendency was toward improvement. In Asia, 14/25 were improving and only one deteriorating; in Africa, 18 improved and 14 deteriorated; in South-Central America and Caribbean prevalences are in general lower than other regions and less likely to change accordingly; the report showed no change in 11/19 and 2/19 deteriorating.

There are a number of countries in transition from high to low prevalence (Thailand, Vietnam, Brazil, Bangladesh, India, Sri Lanka, Costa Rica, Mexico, China, and Indonesia) with an average rate of underweight reduction of 0.7–1.0 percentage points per year in the last 30 years or more, this trend is similar for some Asian countries, while the African region has not initiated a consistently downward trend yet. Several factors contribute to this situation among them HIV /AIDS, drought and abrupt climate change, and permanent economic–social distress in selected areas.

Presently, 165 million children < 5 years have a height-for-age *z*-score of –2 or lower (thus are stunted) on the basis of the WHO Child Growth Standards, a 35% decline from an estimated 253 million in 1990. The prevalence decreased from an estimated 40% in 1990, to 26% in 2011—an average annual rate of reduction of 2.1% per year. East and West Africa and South-Central Asia have the highest prevalence [42% for east Africa and 36% for West Africa and South-Central Asia]; the largest number of stunted children, 69 million, live in South-Central Asia. Population-based surveys from 79 countries conducted after 2000 reveal that stunting prevalence in children <5 years was 2.5 times (1.0–7.6) greater in children in the poorest income quintile relative to richest. According to 2011 UN estimates, >100 million children <5 years old, or 16%, were underweight,

and a 36% decrease from an estimated 159 million in 1990. Prevalence of underweight was highest in South-Central Asia 30% and in Western Africa it was 22%. The UN estimate for wasting based on WHO Standards was 8% (52 million) globally in 2011, an 11% decrease from an estimated 58 million in 1990. Seventy percent of the world's wasted children live in Asia, mostly in South-Central Asia, where an estimated 15% (28 million) are affected. Much the same regional pattern occurs for severe wasting in 2011 global prevalence was 3% or 19 million children. Most children with severe wasting can be found in South-Central Asia (5.1%) and Central Africa (5.6%).[1]

In Central America, there is no improvement while in South America progress has been significant. Data on children under 6 years of age show that in Latin America and the Caribbean, the prevalence of low Weight/Age (<−2SD) vary from country to country (from 0.7% to 19.2%). In Haiti and Guatemala, this prevalence is over 15% and in Ecuador, Bolivia, Peru, and El Salvador is close to 6%. As for severe malnutrition (<−3SD Weight/Age) in Haiti, Guatemala, Honduras, Ecuador, and Bolivia, the prevalence vary from 1.2% to 7.3%.[15]

Independent of the improvement of national figures, severe malnutrition including edematous forms can still be detected in hospital settings in some regions of the world.[16,17]

The causes behind the insufficient progress are multiple and complex in their interactions. Political strife, government unresponsive to the needs of the community, stagnant economies, low status of women and unequal rights, poor education and inadequate access to health, poor sanitation and unclean water, high prevalence of infections and low birth weight interact in determining poor postnatal growth and high rates of malnutrition.[18] Since most of these factors are interrelated, it becomes virtually impossible to isolate their effects from retrospective trend data or regression models. Decreases in the prevalence of malnutrition have been described in relation to improvements in household food security, supply of clean water and environmental sanitation, education of women, early treatment of diarrhea with oral rehydration solutions in the community, early treatment of respiratory infections, promotion of breast feeding, appropriate complementary feeding, immunization, growth monitoring, and surveillance for early identification of malnutrition. Smith and Haddad have attempted to explain the key factors responsible for the different rates of progress across regions of the world (see Figure 15-3).[19] They examined the contribution of national food supply, women's status, women's education, and health environment based on proxy indicators

from available country data. Improvements in women's education contributed by far the most, accounting for 43% of the reduction in child malnutrition between 1970 and 1995, while improvements in per capita food availability contributed about 26%. In fact, this information served to define the Millennium Development Goals adopted by heads of state and the UN system to be met by 2015 (http://www.un.org/millenniumgoals/). The overall reduction in the prevalence of child malnutrition in South Asia for the 25-year period was estimated to be 16.5 percentage points.

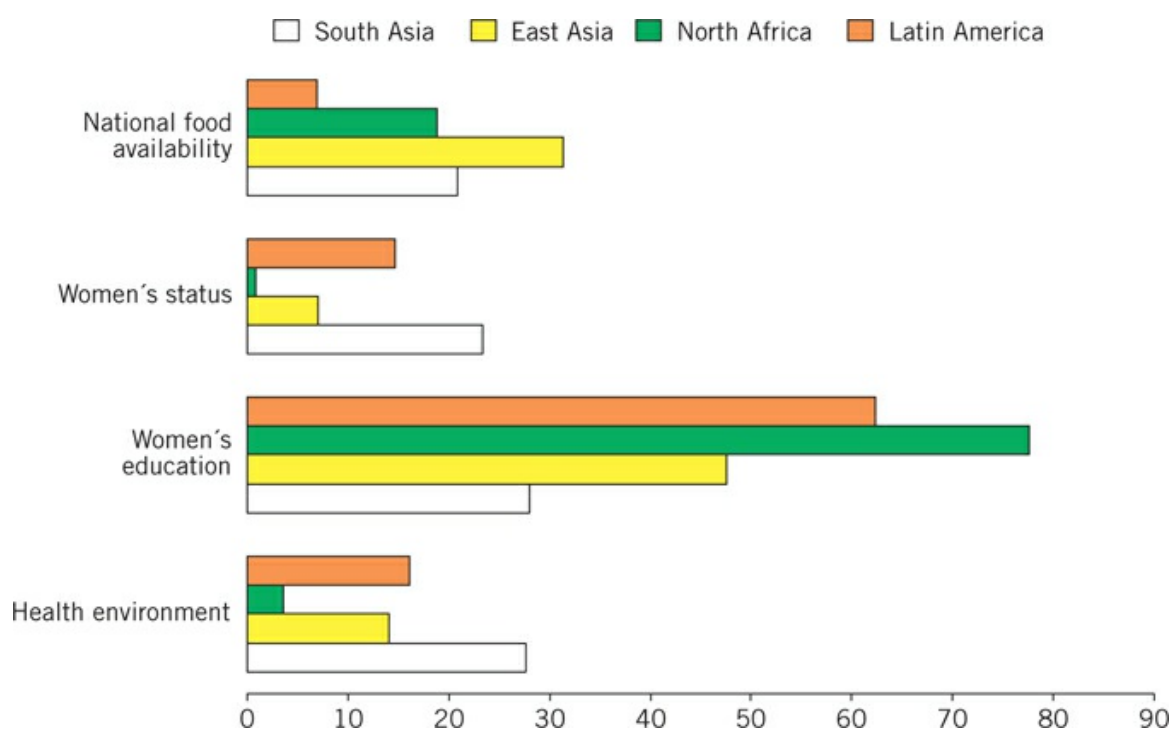


Figure 15-3 Contributions of underlying determinant variables.

The greatest contributions to this reduction came from increased education of women and improvements in health environments; each accounted for about 28%. Gains in the status of women accounted for about 25% of the reduction, and improvements in food availability for about 20%. The total reduction for Sub-Saharan Africa's malnutrition rate over the study period was only 4.2 percentage points. Most of this was explained by increases in women's education, followed by improvements in health environments. Increased education of women made strong contributions in all periods except for the late 1980s, when enrollments actually declined. Improvements in health environments have made their

greatest contribution after 1985. Women's relative status as evidence by gender differences in life expectancy has continually declined in the region since the 1970s, most precipitously after 1985. The consequence has been a recent worsening in prevalence of child malnutrition in the region. Changes in food availabilities have played a large role in both directions. Substantial improvements in the late 1980s and early 1990s were outweighed by deteriorations during the 1970–1985 period.

Latin American and the Caribbean experienced an estimated 11 percentage point reduction in child malnutrition over the study period, most of which took place during 1970–1980. Since then, reductions in child malnutrition have continued at a much slower pace. Like the other regions, the greatest contribution comes from women's education, which explained 62% of the improvement; while the contribution of environmental health to this improvement has steadily declined. Strong improvements in the status of women in the 1970s were followed by very small improvements in the 1980s. Food availability improved in the 1970s, but declined slightly in the early 1980s.

Prevention and control of infections is clearly a critical aspect of malnutrition reduction efforts. Infection has a definite adverse effect on nutritional status, while malnutrition compromises the host defenses and immune responses increasing case fatality and disease susceptibility.[20] Relative risk (RR) of death from infection is directly correlated with severity of malnutrition: RR 8.7 (CI 95%, 5.5–13.7) for severe protein energy malnutrition (PEM), 4.2 (IC 95%, 3.1–5.5) for moderate PEM, and 2.0 (IC 95%, 1.7–2.4) for mild PEM.[21] Yet, since the severely malnourished represent a small proportion of total population most malnutrition related deaths are accounted by moderate malnourished individuals. It has been estimated that close to 80% of all malnutrition infection-related deaths are attributable to moderate and mild malnutrition.

The adequacy of breastfeeding and complementary feeding also plays a key role in malnutrition prevalence. In most countries where malnutrition prevails, the time of onset is >6 months, this is when breast milk may be insufficient to meet the infant energy and micronutrient needs and also a time when mothers have increased competition for their time/dedication to the infant. This is coupled by inadequate, low energy density, and low nutrient density complementary foods. Under these conditions, increased feeding frequency, added oil, and appropriate complementary to increase energy and nutrient density become crucial to prevent malnutrition.[22] A recent study in Bangladesh examined the role of education during the weaning period demonstrating that education on complementary feeding

given to mothers was effective in enhancing weight gain despite the fact that no external sources of foods were provided. The authors suggest the educational efforts on what constitutes adequate complementary feeding and on how to prevent microbial contamination of weaning foods are important as malnutrition prevention strategies.[23]

There are no easy answers or "magic bullets" in combating malnutrition, several options can be suggested based on the success and accomplishments of some countries in malnutrition reduction. In most cases, poverty reduction efforts need to go hand in hand, but should not be considered a pre-condition for success. As presented in Figure 15-1 poverty reduction is a precondition for success, this may be unrealistic since malnutrition is not only a determinant but also a consequence of poverty. However, this does not imply that efforts to improve nutrition should be postponed until major political or economic events improve the economic condition of the poor since reducing malnutrition contributes by itself to poverty reduction. The notion of human capital formation as a major determinant of economic growth has been recognized globally with the award of two Nobel prizes in economics addressing this issue over the past decade.[24]

A strategy for action, based on what has worked in different settings around the world (Costa Rica, Thailand, Cuba, Chile, Kerala-India, among others), is proposed in Figure 15-4. This serves to analyze the necessary interactions among the key social actors required for successful implementation of malnutrition control and prevention programs. The basic and critical component is social mobilization to establish the demands for action from the political actors; this process should lead to community actions as well as responses by government. A responsive government is one that responds to community demands; this requires some degree of participation by society in government, ideally but not necessarily a government by the people, from the people and for the people. Communities that are so empowered and able to participate in the political process will be successful in demanding appropriate actions from governments. Malnutrition needs to become unacceptable to society in order to trigger the level of action required to address the issue frontally. What does it take to trigger this process? In most cases, pro-active academics and/or non-governmental organizations (NGOs) that have a strong voice, independent from government have catalyzed the process and led the way. "Political will" does not come easy nor it is generated from within; professionals, academics, NGOs, community leaders and the press should act in concert to elicit action. The role of the press in generating

public awareness and communicating public concern of malnutrition as a key restriction for national development is crucial. Recruiting and training select journalists will serve to generate widespread recognition of the problem and make people aware that the solutions are credible and feasible by illustrating what has worked elsewhere. Academics and professionals should propose the necessary actions that are required to implement effective programs to control and prevent malnutrition, ideally based on technical consensus rather than political considerations. They should promote a critical review of policy options based on what may work in a given setting; international organizations can assist in reaching a technical consensus by providing evidence and expert advice on successful examples of malnutrition reduction. The private industrial or business sector is usually a latecomer in this scene. Yet, public private partnerships are a must for sustainable programs since industry is a key partner in the implementation of programs; for example in nutrient supplementation, fortification, and other food-based strategies.[25] By the end of the day, unless we activate the political process and include industry, we get no action.

Governments must be committed to generate the necessary actions to solve malnutrition as a public health problem. Governments are usually restricted in meeting social demands by the available monetary resources and are not willing to commit funds unless they see a clear return on investment. Thus, it is crucial to clearly present the cost benefit analysis of various programs for nutrition improvement.

Moreover, if the community is truly aware and the issue has become a political priority, there may be a clear political benefit as well as an economic gain in terms of human capital formation. The involvement of the press in facilitating social communications is important in open societies. For example, the cost of providing half of all critical micronutrient needs per person per day by fortifying a staple food has been estimated to be less than a dollar per year; this is less than a pack of cigarettes in most countries. The World Bank has estimated the cost benefit ratio of micronutrient fortification to be at least 17:1. Not all programs are equal in terms of cost benefit or cost effectiveness, thus the tools of economic assessment should be used in defining policy options. As previously discussed (Figure 15-4), the mix of policies that will work differs in various settings. Working through networks is particularly helpful in countries where human and material resources are limiting, so the potential for complementarity and synergy between groups should be exploited. However, there are multiple preconditions to operate successful

networks. Groups must share clearly stated objectives, common short- and midterm objectives, participants must share decision making, activities must have continuity and be sustained. It is also helpful to geographically map where the problems are and where agencies are located and programs are in place in order to improve resource utilization.

The impact of maternal education has been shown to work across all regions, yet we must bear in mind that access to education by women is in itself an index of multiple social economic and cultural factors. In Peru, Penny et al., working with families living in conditions of peri-urban poverty, where access to food was not a limiting factor demonstrated that nutrition education, delivered through pre-existing health services significantly decreased the prevalence of stunting in young children by 11 points.[26] In our experience in Bolivia, we have come to realize that understanding the cultural representations of the mother, the family and the health team is fundamental to interpret health behaviors that otherwise appear irrational and incomprehensible.[27] Language/cultural barriers can also significantly affect the uptake of services by the community, thus need to be considered and appropriate strategies need to be in place.[28]

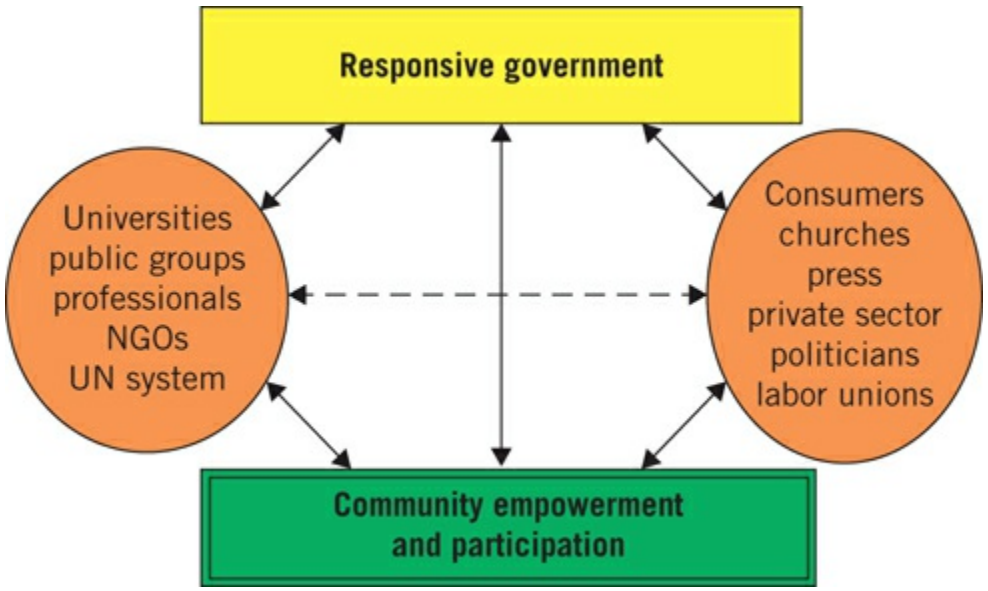


Figure 15-4 Networks and malnutrition. Scheme for strategic analysis of protein energy malnutrition control and prevention. See text for details on dynamic interactions between stakeholders. NGO, Non-governmental organization.

Nutrition and food policies should be integrated in the context of national development strategies. Nutritional problems are determined by economic and labor policies, health and education sector policies as well as welfare policies. The integrated approach may be more difficult to implement in most settings, since government structures are sectorial, yet harmonization of policies is a must to avoid duplication, wasteful use of resources and overburden personnel budgets.[29] Cost effectiveness evaluation of programs should be included at the outset in the present climate of fiscal policies. Today, program sustainability in a world of competing interests will progressively depend on the capacity to demonstrate cost effectiveness over time. Technical considerations should prevail over political interest in defining what should be done, because this is the best way to optimize chances of success. This is true at all levels, national and international programs are not always guided by best technical advice but many times, respond to politically motivated actions.

Nutrition and food policies need to be guided not only by considering the biological and medical aspects but also by the ethical dimension. Human rights concerns including the right to safe and nutritious food can no longer be ignored; in a world of plenty, malnutrition can no longer be justified as a problem of the poor or a problem that affects others but not "us". This includes not only addressing income differences but also ethnic and cultural differences which still represent major barriers for access to health and nutrition in many regions across the world.

Globalization should be not only about business and trade, but mostly about how we as humans face a common future. We cannot afford to have over a third of the people in the planet under conditions of poverty and malnutrition. Pediatricians should be concerned with the status of children worldwide. As we are convinced that our research should not ignore that we are citizens of a common planet, our advocacy and our research should not only serve those around us but also children around the world. Funding for applied research in nutrition and health of children should be as important as the present emphasis given to protection from bio-terrorism and the "war on terror". Providing for a better future for children across the world will do more for peace than the outburst of violence and wars that periodically take place. Controlling and preventing malnutrition should top the list if we are looking for world peace.

Potential Strategies for the Treatment and Control of Severe and Moderate Malnutrition

First, we should clearly establish that prevention and treatment of malnutrition are complementary. Although the adage "prevention is better than cure" is a public health imperative, it cannot be used to withhold treatment from those already affected by malnutrition. Both strategies must work together to avoid malnutrition from occurring and to rehabilitate those who are moderately or severely malnourished and at risk of death and/or severe disability. Although the actions for prevention differ from those necessary for effective treatment, they should be seen as part of a spectrum of efforts needed to combat malnutrition. What can we learn from experience, either success or failure, in the treatment of malnutrition?

Hospital Treatment

Even though severe malnutrition (i.e., severe wasting and/or edema) is commonly seen in pediatric wards in developing countries, few doctors and nurses have been trained in its management. Consequently, case-fatality rates are high. Severely malnourished children die in large numbers if treatment does not take into account the physiological and metabolic changes that arise during reductive adaptation.[30] One way to try to prevent children dying during treatment is to provide case-management guidelines. WHO first produced such guidelines in 1981. These encapsulated the basic principles of care that are still recognized today, namely *stabilization* with small frequent feeds day and night of specially formulated low-protein, low-lactose milk, rehydration with low-sodium fluids, provision of potassium, magnesium and micronutrients, and antibiotics for all children as infections are often silent, followed by *rehabilitation* with a high-energy, high-protein diet to rebuild wasted tissues. With advances in knowledge, notably the importance of withholding iron during the stabilization phase and providing zinc and other trace nutrients and psychosocial stimulation, WHO updated the guidelines in 1999[31,32] and developed a training course.[33] Where the guidelines have been fully implemented, deaths among hospitalized severely malnourished children have fallen dramatically, for example case-fatality rates of 30%-45% have fallen to <10%.[34]

In many hospital facilities around the world, staff have accessed the WHO guidelines and implemented them successfully with minimal or no outside help, and the improvements in the children's rates of recovery and survival have supported the use of the WHO guidelines in becoming hospital policy. In other hospitals, training courses have been the necessary trigger for change. There are also examples, however, where training courses have led only to a transient or partial adoption of improved

practices: the reasons vary; they include high staff turnover and lack of motivation. Factors that favor adoption of improved practices and sustainability are:

- a functioning health system, and basic resources
- leadership, mentoring, and managerial support
- in-service education so that everyone is confident about what to do
- induction of new staff on the treatment of malnutrition
- supervision of junior staff and carers
- regular audit and feedback, and discussion of critical incidents and lessons learned
- good teamwork

A major hindrance to reducing case-fatality rates is that treatment of severe malnutrition is not included in many medical and nursing curricula. The consequence of this omission is illustrated by experiences in a South African hospital where the case fatality rate increased from 18% to 38% with the arrival of newly qualified doctors who had no specific training in the management of severe malnutrition. The sudden rise in case-fatality coincided with lower prescribing of potassium (13% vs 77%), antibiotics with gram-negative cover (13% vs 46%) and vitamin A (76% vs 91%).[35] In many hospitals in developing countries, severely malnourished children comprise the majority of pediatric deaths, so it is imperative to train young doctors and nurses to care for these children correctly. This will require recognition of the importance of malnutrition in clinical practice, establishing a severe malnutrition ward or "corner" where correct treatment is provided day and night, having adequate supplies, and having ward and emergency staff trained in malnutrition case-management.

Community-Based Treatment

Some of the lowest case-fatality rates for severe malnutrition are to be found in humanitarian emergencies where well-trained staffs systematically follow treatment protocols. When thousands of families congregate in crowded camps the risk of disease increases so, as far as possible, case-management should be provided in their communities.[36,37] Likewise, hospital wards expose children to infection so if a child has "uncomplicated" malnutrition and families have access to adequate resources it is best if the child is rehabilitated at home. Community outreach workers screen children for wasting and edema. A MUAC of <115 mm indicates severe wasting in those aged 6 months to 5 years.

Children identified as having mild/moderate edema or are severely wasted but have a good appetite and are alert and appear clinically well, are treated at home with ready-to-use therapeutic food (RUTF). Only those who have poor appetite or are ill, or have severe edema, or edema plus wasting, are referred for inpatient care.[38]

The components of community-based care include:

- community sensitization and active case-finding to identify malnourished children early, before their condition deteriorates
- community mobilization to improve access to care and achieve high coverage
- treatment of moderately malnourished cases
- treatment of severely malnourished children with good appetite and who are clinically well (uncomplicated severe malnutrition).

Carers are instructed how to manage their child's recovery at home. Antibiotics, antihelminthics, and RUTF are provided. Vitamin A is provided for children with eye signs of deficiency, severe diarrhea, or recent measles. The child's progress is monitored every week for programs implemented in an emergency context, or every two weeks in non-emergency settings. In 21 programs in Malawi, Ethiopia, and Sudan, more than 22,000 children have been treated in this way, with a case-fatality rate of 4.8% and coverage rates of more than 70%.[39]

Where an effective system of community-based care is in place, children requiring inpatient management can be discharged after completion of the stabilization phase and their rehabilitation completed at home with supervision by a trained health worker at a health post, or health center, or by home visits where these are feasible.[40] Treatment in the community can:

- benefit children by reducing their exposure to hospital-acquired infections
- benefit families by reducing the time carers spend away from home and the risk of possible neglect of siblings
- benefit families by reducing the opportunity costs associated with hospital treatment
- act as a catalyst for strengthening nutrition activities within clinics for treatment and prevention of malnutrition
- lead to community action to prevent malnutrition

Rehabilitation at home can be achieved by provision of RUTFs or by families using their own resources to make specific mixtures of family

foods. High energy intakes (>150 kcal/kg/d), high protein intakes (4-5 g/kg/d) and provision of micronutrients are essential for rapid weight gain. RUTFs are specially designed for home-feeding in the rehabilitation phase: they contain electrolytes, minerals, and vitamins and have low water content so pathogens cannot grow in them.

Advantages of well-run, effective community-based programs are that:

- timely action should reduce the number of cases requiring inpatient treatment.
- the rehabilitation phase can be completed in the community rather than in hospital, which should reduce length of stay.
- discharged children can be provided with continuity of care.
- treatment is more cost-effective.

Early discharge must always be linked with effective follow-up, but at present there are many countries where this is non-existent. Hospitals with a policy of early discharge and no system of follow-up are usually unaware of high rates of post-discharge mortality. Failure of children to appear at outpatient clinics is easily misinterpreted as parental irresponsibility, rather than to child death.

Nutrition Rehabilitation Centers and Day-Care Centers

There is a long tradition of treatment of moderate and severe cases of malnutrition in residential or daycare nutrition centers, but their low coverage and high opportunity cost make these the least favored option in many settings. Nevertheless, such centers could be a “half-way-house” between hospital and home. For example, attendance for one week could achieve rapid weight gain and provide practical education sessions for mothers and carers and precede rehabilitation at home. Center staff could monitor progress during home rehabilitation through home visits and/or having children return to the center for assessment. Centers could also receive moderately wasted children and treat uncomplicated severe malnutrition. In urban areas with high numbers of severely malnourished children, well-resourced nutrition centers could be alternatives to hospital admission if staffs are sufficiently trained. The centers should be integrated into the child health services and could be attached to a clinic.

Factors for successful rehabilitation. Successful programs share several features:

- all follow the basic principles of treatment of severe malnutrition (i.e., high-energy, high-protein intakes in the rehabilitation phase along with potassium, magnesium, minerals, and vitamins)

- all provide frequent meals (at least 5), comprising specific food mixtures that families can afford, or RUTF
- those not giving RUTF make considerable efforts to teach mothers about child feeding in a memorable way, using a variety of teaching methods and providing opportunities for mothers to practice preparing children's meals
- most address the wider social, economic, and health issues that face poor families: some integrate rehabilitation with poverty alleviation activities
- all have motivated and carefully trained staff.

TABLE 15-1 Characteristics of Different Treatment Modalities for Malnutrition: Advantages and Disadvantages

At Home With RUTF[a]

Advantages

- Independent of home resources
- RUTF contains electrolytes and micronutrients; needs no cooking or refrigeration
- Liked by carers/children: few defaulters
- Can achieve high coverage, rapid scaling-up
- Responsive to fluctuating numbers
- Avoids need for formative research as to which home foods to promote
- Avoids need for intensive teaching of carers on what to give

Disadvantages

- Little opportunity to learn about good child feeding practices and malnutrition prevention
- Need clinic/health post nearby for monitoring progress, and so on
- Need efficient transport and distribution networks
- Need quality control measures if RUTF is locally made.

At Home Using Own Resources

Advantages

- Cost-effective
- Liked by carers: few defaulters
- Teaches mothers about child feeding
- Potential to prevent malnutrition in the long term by learning to make good food mixtures and feed frequently and responsively
- Potential ripple effect

Disadvantages

- Families must have food resources
- Carer must be at home full-time
- Need formative research to develop educational messages
- Need clinic/health post

• Responsive to fluctuating numbers.	nearby to monitor progress and provide timely treatment when ill • Need to provide micronutrient supplements • Need motivated staff; good communicators
Residential or Day-Care Nutrition Centers	
Advantages • Supervised feeding with high chance of success • Opportunity for teaching mothers • Potential for preventing malnutrition in the long-term • Circumvents poor primary health care system	Disadvantages • Need high malnutrition prevalence, or a center attached to a clinic • High institutional cost if "stand-alone" • Burdensome to carers, with risk of defaulting • Low coverage
[a]RUTF ready to use therapeutic feeding..	

The advantages and disadvantages of (1) residential and day-care nutrition centers, (2) home rehabilitation with no free food given, and (3) home rehabilitation with RUTF (are shown in Table 15-1)

Each option has advantages and disadvantages: some options may be better suited to particular contexts, such as urban families, mothers working for a wage, or food insecure communities. Health system infrastructure, accessibility, and staff competencies must also be taken into account. For successful rehabilitation, the system chosen should:

- achieve intakes that will promote catch-up growth and improve immune function
- provide timely treatment of infections and close monitoring of progress.

Ideally, the system should integrate both the treatment and prevention of malnutrition.

Khanum and Ashworth compared the cost-effectiveness of three treatment modalities in 437 severely malnourished Bangladeshi children aged 12 to 60 months. The children were allocated either to (1) inpatient treatment, (2) day-care treatment, or (3) one week of day-care followed by home care. The three groups were comparable at admission: most (98%)

had edema and the mean weight-for-height was <70%. The costs to reach the target of 80% weight-for-height were compared. The days required to reach the target in the three groups were 18, 23, and 35 days, respectively. Although the home-treated group took the longest time, it was the most cost-effective treatment. The respective costs expressed in US$ were $156, $59, and $29 per treated child. Mortality was equally low (<5%) in all three groups (Figure 15-5). Home treatment was the option most preferred by parents: day-care was the least preferred option.[41,42]

The next section includes a case study of how Chile succeeded in reducing malnutrition and shows the need to examine not only the specific actions to treat severely and moderately malnourished children but also the multiple programs that acted in synergy to achieve this goal. A key component was the primary health infrastructure with wide coverage and the human/material resources necessary for effective action. The capacity to identify and refer the severely malnourished to nutrition recovery centers when appropriate was vital for success. A nutrition recovery program focused on treatment of the severely malnourished working in isolation will have little or no impact unless it is coordinated with an effective prevention program working through the primary health system.[43]

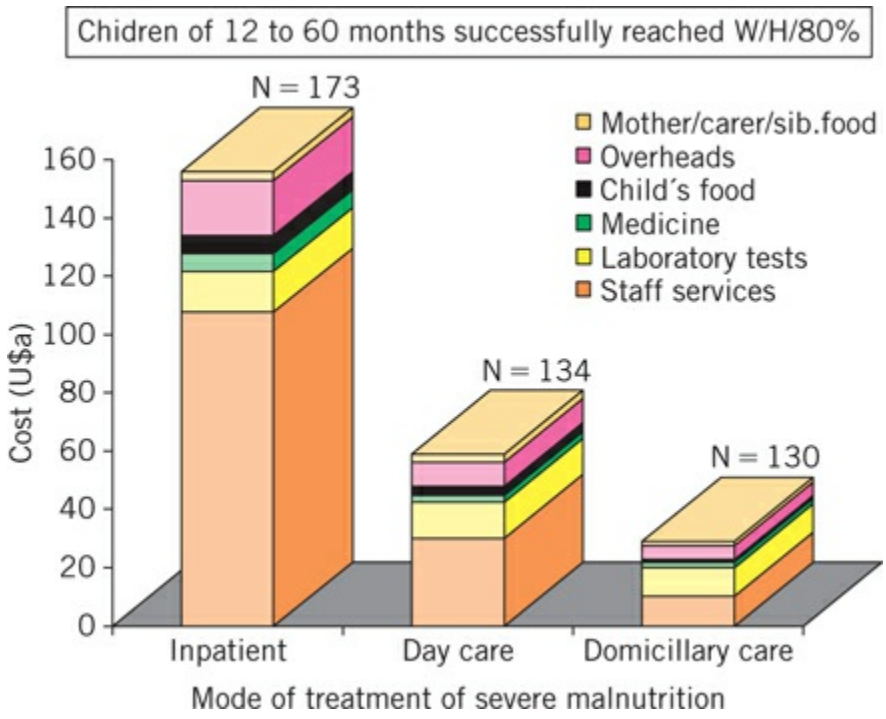


Figure 15-5 Cost effective treatment in severe malnutrition. Cost

effectiveness of various treatment for severe malnutrition. Children were allocated to hospitalization (*n* = 173), day care nutrition recovery center (*n* = 134), and ambulatory treatment (*n* = 130). All achieved a similar degree of recovery (weight for height 80% of Word Health Organization Standard). Cost is expressed in US dollars per treated child for treatment to reach the goal.

IMPLEMENTATION OF PROGRAMS TO PREVENT AND REDUCE MALNUTRITION AT THE NATIONAL LEVEL: THE EXPERIENCE OF CHILE AND BOLIVIA

Chile stands out among countries of Latin America and the Caribbean for the notable improvement of its health and nutrition situation, particularly in pregnant women and young children. This improvement has been achieved progressively and continuously through comprehensive health, nutrition, sanitation, education, and reproductive health strategies initiated in the 1940s. Results came 30-40 years later but much earlier than the considerable economic progress experienced by the country during recent decades. A key element in the success of the Chilean experience has been the uninterrupted application of programs defined based on technically consensus independent of major political, social, and economic changes. The programs remained in place and were even expanded in periods of social instability, radical political changes, and serious economic crisis. Only recently, poverty reduction has contributed to improved food security and to the improvement of the quality of life of low income groups.

The achievements obtained in children's health can be appreciated in the declining rates of infant mortality, mortality due to acute diarrhea and to bronchopneumonia. Infant mortality dropped by approximately 90%, and mortality due to respiratory infections and acute diarrhea by over 95%. Between 1940 and 1960, total infant mortality was reduced in Chile from 193 to 120 per thousand live births. From 1960 to 1990, it fell to 17 per thousand, and over the past decade, it dropped to around 10 per thousand. See Table 15-2 for degree of achievements of 2000 goals for nutrition of children and women, note the virtual eradication of malnutrition and the rise of obesity as the major diet and nutrition related public health problem.[44,45]

A key strategic factor in the development of Nutritional Programs in Chile has been the emphasis placed on the political dimension of

nutritional problems. Nutrition of the population, and especially its more vulnerable sectors, has been at the center of the political debate for the past 40 or 50 years. It has been transformed into a key indicator to assess social justice and equal opportunity for all. The 1970 Presidential election campaign is a good example of this. All three main candidates to the Presidency (one representing the conservative right, a second representing the center left Christian Democratic, and the third representing the Socialist left) proposed the eradication of malnutrition as a key target of their respective future governments. Salvador Allende, the socialist candidate was elected on that occasion with the promise of providing half a liter of milk per day to every child up to 15 years of age in the country.

The community demanded access to health and food, not only to meet this basic need but as a fundamental human right. The process initiated by academic researchers led to policy development and implementation of nutritional programs. The expansion of these programs became part of the social demands made by the population to the governments, and those politicians that satisfied these demands of providing more food to the economically vulnerable groups were elected and maintained in office. Community organizations, political parties, the Catholic Church, concerned academics, labor unions as well as multiple social organizations played key roles in demanding action. Most of the nutrition programs were eventually built into the law specifying universal coverage and the respective yearly budget allocations. Any changes to the supplementary feeding programs have always been undertaken with extreme caution by governments, since they are reluctant to take risks on a matter of such high political sensitivity.[40,41]

TABLE 15-2 Degree of Achievement of Nutritional Goals Chile 2000–2005

Indicator (%)	Base 87–89 (%)	Goal 2000 (%)	2005[a] (%)
Pregnant women with low W/H	25	15	12
Anemia in infants	30	10	3
Anemia in pregnant women	25	10	25
Newborn with low birth weight	6.9	6.0	5.0
Newborn with insufficient weight	21	15	12.5
Breastfeeding exclusively to 4 mo	44	80	60
Breastfeeding to 12 mo	20	35	34
Student gr. 1 elem. with height <	33	20	10.4

−1 SD			
Under 6 yr old with W/A <−2 SD	2.2	2.0	0.8
Food insecure households	35	20	14
Indigent population	18	15	5
Obesity in preschool children	4.6	3	14
Obesity in children entering school	6.5	4.5	19.5
Obesity in pregnant women	12	8	36

[a]Latest figures Ministry of Health Chile 2001.

Source: Plan Nacional de la Infancia, UNICEF 1994.

The Bolivian Experience

In 2007, the Government of Bolivia launched its first national program which was designed to address wasting, stunting, and childhood anemia. The program's design was based on "evidence-based interventions" as described in The Lancet Series of mother and child malnutrition 2008.[46] The program was able to raise awareness of the effects of early malnutrition in later years and was able to build a wide social and political commitment to fight this condition as a national priority.

The main characteristics of the program were: taking a multi-sectorial approach, emphasizing social mobilization across all ethnic/cultural groups, focusing on geographic/regional vulnerability to food insecurity, and addressing the differential age vulnerability of children, prioritizing children under two years of age. Technical features include the use of the 2006 WHO Multicenter Growth Study, use of height for age and weight for height indicators to diagnose acute and chronic malnutrition in the entire health system and the Integrated Management of Common Illness (IMCI) approach which was modified to reinforce prevention and early interventions for malnutrition.

Nutrition-focused IMCI was delivered through primary healthcare clinics. Health workers were trained to measure height/length, identify severely malnourished and treat those with mild and moderate forms. Nutritional care in the community clinics was upgraded. In summary, the implementation meant developing support mechanisms within established community organization to prevent disease, helping families to care for sick children, and getting children to clinics or hospitals when needed.

After five years, the program was able to reduce mortality of severe malnourished children in the country from 25% to less than 5% based on widespread implementation of the WHO's 10 steps protocol for hospitalized care and the application of community malnutrition

management. Stunting (chronic malnutrition) fell from 27% to 20% in children from 3 to 23 months (DHSS 2003–2008)

SELECTING THE BEST APPROACH TO TREAT SEVERELY MALNOURISHED CHILDREN

The selection of treatment modality for malnourished children starts by defining the severity of the malnutrition. Gomez in the late 1940s proposed the first classification of PEM based on weight expressed as a % of normal value for age, despite the limitations of this expression it is still used by many because of its simplicity. First degree or mild PEM is defined as between 75% and 90% of norm, second degree or moderate 60%–75%, and third degree or severe as <60% of expected for age.[47] Bengoa with his functional classification in the mid-1950s established treatment modalities based on the Gomez criteria, indicating that those with first degree should be treated in the primary health or outpatient health clinic by providing education and dietary advice; those with second degree should be treated in the day care nutrition recovery center using complementary foods in addition to education. Severe PEM should be treated in the hospital. Hospitalization should last for strictly the minimal time required "to treat complications and stabilize the patient", then provide nutrient and energy dense foods as tolerated. Closed nutritional recovery centers should be reserved to situations where PEM is highly prevalent, particularly in young infants and family conditions do not permit recovery at the home. The choice of treatment modality should always consider parental choice, cost effectiveness of treatment under real life situations, access to primary healthcare system, availability of health infrastructure, social support system available in the community, maternal education and overall condition, and psychosocial interactions within the family.

The main barriers or impediments in control and treatment of malnutrition relate to the multiple conditioning factors presented earlier. Translating theoretical concepts into practices that lead to improved nutrition and health of children is also a challenge. Treatment norms and preventive strategies may be well known but they are harder to establish as standards of practice. In addition, physicians commonly depart from rules to demonstrate their autonomy and status, or challenge specific aspects of the norm based on anecdotal personal experience rather than on firm evidence. Health professionals often consider malnutrition as an unavoidable consequence of poverty, thus may accept it as a social problem outside the health realm. The common acceptance of malnutrition

by communities and the political leadership as a chronic unsolvable problem inherent to social structure for ages leads to passive acceptance and inaction. Physicians often define health expenditure priorities, particularly those with greater status such as surgeons and internists; unfortunately, the choice is often based on what is more attractive for medicine per se, rather than basing decisions on objective measures of impact on burden of death and disability. Physicians often have greater incentives and are more interested in providing individual health care rather than addressing public health problems.

The organizational environment is also an important barrier, physician lead administration usually centers on medical interests rather than on patient's needs. Moreover, other health professionals are seen as subservient to the physician rather than team members with equal participation. Physicians in general have limited skills for networking and to develop partnerships, they are poorly trained for team work; they associate success with individual recognition rather than group effort. These traits prevent physicians from leading multi-professional multidisciplinary teams such as those needed to address malnutrition effectively. Promoting new attitudes and skills, training for team work, active listening, and defining funding priorities based on public health needs of the community are mandatory to establish effective malnutrition prevention and treatment programs.

Factors That May Explain the Effect of Nutrition Interventions in Rising Obesity Trends

The transit from undernutrition to good nutrition often leads to increasing rates of overweight and obesity, countries may reach the goals linked to reduced undernutrition but at the same time have rising prevalence of obesity and related chronic disease.[48] In order to prevent this from occurring PEM prevention programs should include from the outset the need to consider obesity prevention strategies. For example, there is a need to monitor not only of weight-for-age, but also of length-for-age as well as weight-for-length. The use of combined indicators should permit identification of the stunted overweight or obese child. Beneficiaries should not be selected based solely on socioeconomic conditions, but on true assessment of nutritional status. Stunted children of low weight-for-age but normal weight-for-length should not be given therapy that provides excess energy, since this will lead to obesity. The quality of the foods provided is crucial in pre-school and school feeding programs; fruits and

vegetables should be included in the diet of children to provide micronutrients and prevent energy excesses. Obesity prevention requires optimizing linear growth and lean tissue mass gain; thus, micronutrients and quality of the protein source are important.

PEM prevention programs need a periodic evaluation, including targeting of beneficiaries, definition of real needs and impact on obesity. Food products may need to be modified in urban settings. For example, in situations where stunting and obesity coexist, providing Fe- and Zn fortified skimmed milk in pre-school feeding programs, rather than full-fat products, is highly recommended.

Programs should be redesigned on the basis of actual need, considering the rapid transition of most urban communities in developing countries. Planners should consider the assessment of physical activity; if low, increased activity should be promoted. Food programs designed to combat PEM may become outdated as rapid improvements in socioeconomic and environmental conditions occur. Slashing social benefits without redirecting them is always unpopular. Policy makers can prevent this reaction by reallocating these funds to alternative social programs that enhance the quality of life of the poor. Thus, the changes will not be seen as taking away benefits from the poor, but rather as responding to a new situation where other needs become more relevant. Ideally, the right to food should be upheld by providing access to safe and nutritious foods, based on what people can buy or grow for themselves.[49]

Cost of Hunger and Malnutrition (The Case of Latin America)

Child malnutrition is a recognized cause of mortality and morbidity. The economic costs of child malnutrition are extremely high. The total cost is a result of higher expenses for health treatment, educational process inefficiencies and lower productivity. The increased chance of having malnourished children proportionally increases costs in the health sector which in the aggregate, equal to the sum of the interactions between the probability of malnutrition in each population group, the likelihood that this group will suffer each of the diseases because of that malnutrition and costs of treating the pathology (diagnosis, treatment, and control). To this is added the costs paid by individuals and their families product of time and quality of life lost.

What less recognized is that it also generates permanent mental and physical consequences that have serious consequences, not only economic

but also human. Malnourished children are more vulnerable to infections, which in turn increase the riskier malnutrition and premature death. Besides the above, malnutrition interferes with proper physical and brain, the latter is expressed not only in lower cognitive learning and lower capacity but in multiple other brain functions necessary for a healthy and productive life.

In Costa Rica, El Salvador, Guatemala, Honduras, Nicaragua, Panama, and Dominican Republic, it was estimated that in 2004, the cost of malnutrition $6.658 million. In Bolivia, Ecuador, Paraguay, and Peru, the cost of undernutrition in these countries totaled US $4.331 million in 2005, equivalent to 3.3% of the combined GDP of these countries. The results show that the economic impact in these countries ranges between 2.0% and 5.9% of GDP. The productivity losses for higher incidence of death and lower levels of education account for 95% of the price paid by society over a life course.[50,51]

Finally, international experience indicates that it is more cost-effective to invest in eradicating child malnutrition than to suffer the social and economic consequences. To achieve this we need not only resources, but well-defined evidence based policies and cost-effective management models that maximize impact and efficiency. Thus, it is necessary to achieve the commitment and active participation of all sectors of society. It is estimated that approximately one million Central Americans and Dominicans (2004) have dropped out of school as a result of malnutrition; this results in a two year lower school attendance in the malnourished which translate in social and economic losses not only to those affected but to society at large.

CONCLUSIONS

Undernutrition affects close to a billion people, stunting and underweight affects close to 180 million children under 5 years of age mostly found in South Asia and Sub-Saharan Africa. The overall trend is toward a small decrease in the proportion of undernourished children with a minute decrease in the absolute number. Unfortunately, the goal established by the UN for year 2015 two decades ago to reduce malnutrition by 50% was not met. The present level of progress is less than half of the decrease required to reach this goal by the next 20 years. Moreover, some regions like Sub-Saharan Africa show a rising prevalence of malnutrition. This situation is worse in countries affected by civil strife and draught, were famine condition the death of children at rates 20-40 times greater than those

found in the general population. We can firmly state that malnutrition and growth failure of children remains “an issue” which affects not only survival but also the quality of life of children in many parts of the world. Prevention is the best form of treatment. There is no doubt that sufficient knowledge exists to treat and prevent primary malnutrition in virtually all children. The condition should be considered a preventable disease, but its eradication requires full commitment by society to assign a high priority to the well-being of children. Changing the social, economic, and political forces that condition abnormal growth and development in PEM goes beyond the realm of specific medical action but concerted participation of physicians as advocates for children in the political process. Placing demands on political leaders is essential to assure that children from the time of conception throughout their various development stages are provided adequate nutrition to allow for the full expression of their genetic potential. Acceptance of this basic premise by the community is essential before appropriate prevention strategies can be successfully undertaken.[8]

Treatment of childhood malnutrition initiated as a response to a basic humanitarian duty to prevent death during the first half of the past century remains a problem in many regions of the developing world. Presently, the mandate is based on the right to adequate food and nutrition for all children as part of the global human and social development agenda. National and International Nutrition and Food Programs developed over the past 50 years have been implemented as integral components of broader strategies of primary healthcare and education, oriented toward preventing deaths, and improving quality of life of low socioeconomic groups. Pediatricians worldwide also should be aware of malnutrition as a conditioning factor delaying recovery from illness and as a major determinant of the quality of life of children with chronic disease.[8]

The key for effective malnutrition prevention is integrating health care, household food security and care as proposed by the updated conceptual model presented at the outset. The following are some of the components of malnutrition primary prevention programs that have been developed and tested over past decades throughout the world in a variety of political and social settings:

1. Promote early contact between mother and infant to improve chance for successful breastfeeding. Establish hospital and other health routines that support breastfeeding while in the maternity ward and later, successful early bonding and documented breast feeding is vital. Support breastfeeding by health team and by

legal framework that promotes, protects, and sustains the right of working women to practice it.

2. Monitor growth and development with adequate standards (present standards are being revised based on present recommended feeding modes). Intervene only when appropriate to prevent malnutrition and specific micronutrient deficits. Mothers should be familiar with growth monitoring cards and be ready to take appropriate actions when growth faltering occurs. The degree of actions taken by the community will depend on the strength of the primary health care.
3. Introduce appropriate micronutrient rich complementary foods and supplements at 6 months of age. If complementary foods are needed earlier consider the risks associated with interference of breastfeeding. Ideally, these should be based on local foods that are accessible to the population. Micronutrient will be required in most cases, new developments include fortification at the household level either with tablets, sauces or sprinkles. Food donations should be incorporated into existing programs and not be dependent on whether surplus exists in industrialized countries. Occasional food donations no matter how well meaning can have adverse consequences in the long term when they disappear.
4. Identify infants at risk for malnutrition and growth failure based on biological and social risk factors. Provide adequate social and medical support for families with children at risk. Early identification should be based on community surveillance not only of growth but also of caring practices and of critical food insecurity. Early interventions at this level are significantly more cost effective. This area is presently receiving insufficient attention despite being at the core of the problem.
5. Educate parents and adolescent girls (who will be mothers) on how to promote growth and development through appropriate home environment, care, and stimulation. Verbal and cognitive stimulation for malnourished children results in higher growth rates than for children without such stimulation. Interactions with parents, caregivers and other children are essential for the young child and these interactions can be improved by education of parents and other caregivers. Care initiatives should go beyond focusing on individual practices and behaviors to bring in dimensions of care for the family and the

community.

6. Provide universal coverage of children to basic healthcare services, full coverage for all children with immunizations to prevent infectious disease and avoid their adverse effects on nutritional status and provide early diagnosis and treatment of diarrheal disease at the community level using oral rehydration. Linking this effort to the community-based surveillance for effective prevention and control of mild and moderate PEM. Depending on what is available in the country the approach may not require expensive infrastructure but rather be community based and sustained.

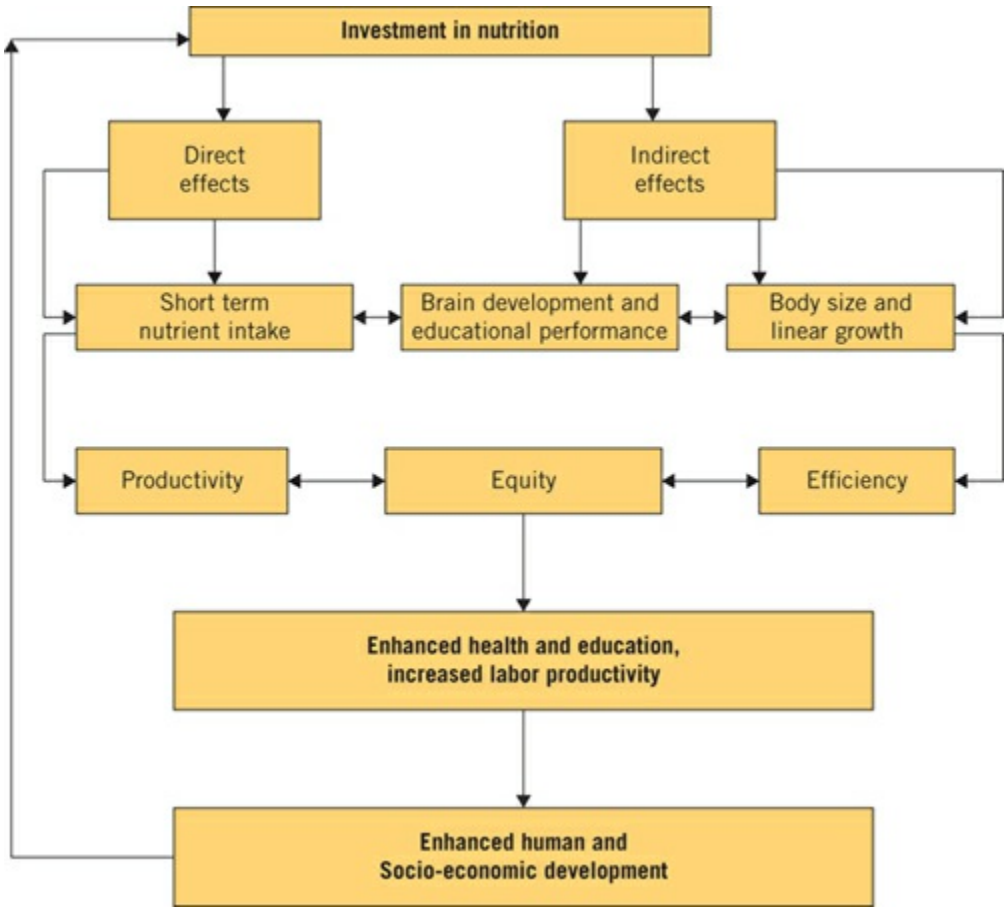


Figure 15-6 Investment in nutrition and socio-economic development.

Treating hundreds or thousands of affected children will not solve the problem of malnutrition as a global public health problem. Unless society at large confronts this issue in its full dimension the problem will continue.

Access to adequate amount and quality of food represents a basic human right and is a necessary precondition for health. In turn, good nutrition and health are prerequisites for human, social, and economic development (Figure 15-6). Physicians and specially pediatricians should not be passive bystanders but rather be activists in this process. Reducing malnutrition of infants and young children means a brighter future for humankind and will contribute in giving world peace a better chance.

REFERENCES

1. Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet*. 2013; 382(9890):427-451. 10.1016/S0140-6736(13)60937-X.
2. Bhutta ZQ, Das JK, Rizvi A, et al. The Lancet Nutrition Interventions Review Group, and the Maternal and Child Nutrition Study Group. Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *Lancet*. 2013;382(9890):452–477. doi: 10.1016/S0140–6736(13)60996–4.
3. Ruel MT, Alderman H and the Maternal and Child Nutrition Study Group. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *Lancet.* 2013; 382(9891):536–551. doi: 10.1016/S0140–6736(13)60843–0.
4. Gillespie S, Haddad L, Mannar V, et al. The politics of reducing malnutrition: building commitment and accelerating progress. *Lancet*. 2013; 382(9891):552–569. doi: 10.1016/S0140–6736(13)60842–9
5. Bhutta ZA, Salam RA. Global nutrition epidemiology and trends. *Ann Nutr Metab*. 2012;61(suppl 1):19–27
6. UNICEF. *The State of the World's Children 1998*. New York: Oxford University Press; 1998
7. UNDP. *Human Development Report UN Nations Development Program*. New York: UNDP; 2013
8. ACC/SCN Commission on the Nutrition Challenges of the XXI Century. *Food Nutr Bull*. 2000;21(3):1–88
9. Puffer RC, Serrano CV. *Patterns of Mortality in Childhood*. Scientific Publication N° 262. Washington DC: PAHO; 1973.
10. Pelletier D, Frongillo E, Schroeder DG, Habitcht JP. A methodology for estimating the contribution of malnutrition to child mortality in developing countries. *J Nutr*. 1994;124:2106S-2122S.
11. Adair LS, Fall CH, Osmond C, et al. Associations of linear growth and relative weight gain during early life with adult health and human capital in countries of low and middle income: findings from five birth cohort studies. *Lancet*. 2013; 382(9891):525–534. doi:10.1016/S0140–6736(13)60103–8.
12. Johnson W, Soloway LE, Erickson D, et al. A changing pattern of childhood BMI growth during the 20th century: 70 y of data from the Fels Longitudinal Study. *Am J Clin Nutr*. 2012;95:1136–1143.
13. de Onis M, Onyango A, Borghi1 E, et al. Worldwide implementation of the WHO Child Growth Standards. *Public Health Nutr*. 2012;15:1603–1610.
14. ACC/SCN. *Sixth Report on The World Nutrition Situation*. Geneva, Switzerland,

UN Sub-Committee on Nutrition; 2010
15. Lutter CK, Chaparro CM. *Malnutrition in infants and young children in Latin America and the Caribbean: Achieving the Millennium Development Goals*. Washington DC: Pan American Health Organization; 2008.
16. Weisstaub G, Soria R, Araya M. Improving quality of care for severe malnutrition. *Lancet*. 2004;363:1110 (letter).
17. Forrester TE, Badaloo AV, Boyne MS, et al. Prenatal factors contribute to the emergence of kwashiorkor or marasmus in severe undernutrition: evidence for the predictive adaptation model. *PLoS One*. 2012;7(4):e35907. doi: 10.1371/journal.pone.0035907.
18. de Onis M, Frongillo EA, Blössner M. Is malnutrition declining? An analysis of changes in levels of child malnutrition since 1980. *Bull World Health Organ*. 2000;78:1222–1233.
19. IFPRI. Explaining child malnutrition in developing countries: a cross- country analysis. In: Smith CL and Haddad L, eds. *IFPRI*. Washington DC; 2000;1–107.
20. Rice AL, Sacco L, Hyder A, Black R. Malnutrition as an underlying cause of childhood deaths associated with infectious diseases in developing countries. *Bull World Health Organ*. 2000;78:1207–1221.
21. Caulfield LE, de Onis M, Blössner M, et al. Undernutrition as an underlying cause of child deaths associated with diarrhea, pneumonia, malaria, and measles. *Am J Clin Nutr*. 2004;80:193–198.
22. Brown KH, Lutter CK. Potential role of processed complementary foods in the improvement of early childhood nutrition in Latin America. *Food Nutr Bull* 2000;21:5–11.
23. Brown LV, Zeitlin MF, Peterson KE, et al. Evaluation of the impact of weaning food messages on infant feeding practices and child growth in rural Bangladesh. *Am J Clin Nutr*. 1992;56:994–1003.
24. Fogel RW. Economic growth, population theory and physiology: the bearing of long term processes on the making of economic policy. *Am Econ Rev*. 1984;84:369–394.
25. Uauy R, Hertrampf E, Reddy M. Iron fortification of foods: overcoming technical and practical barriers. *J Nutr*. 2002;132:849S-852S.
26. Penny ME, Creed-Kanashiro HM, Robert RC, et al. Effectiveness of an educational intervention delivered through the health services to improve nutrition in young children: a cluster-randomised controlled trial. *Lancet*. 2005;365:1863–1872.
27. Castillo Carnigia A, Weisstaub S, Aguirre P, et al. Identifying cultural representations of families and the health team to improve the management of severe malnutrition in childhood. *Qual Health Res*. 2010;20:524–530.
28. Morales R, Aguilar AM, Calzadilla A. Geography and culture matter for malnutrition in Bolivia. *Econ Hum Biol*. 2004;2:373–389.
29. Philip J, Kaare N, Suttilak S, Swaminathan M.S., Tagwireyi J, Uauy R, Mahbub ul Haq. Ending malnutrition by 2020: an agenda for change in the millennium. *Food Nutr Bull*. 2000;21(13):88.
30. Schofield C, Ashworth A. Why have mortality rates for severe malnutrition remained so high? *Bull World Health Organ*. 1996;74:223–239.
31. World Health Organization. *Management of Severe Malnutrition: A Manual for Physicians and Other Senior Health Workers*. Geneva, Switzerland: WHO; 1999. http://whqlibdoc.who.int/hq/1999/a57361.pdf.
32. Ashworth A, Khanum S, Jackson A, et al. *Guidelines for the Inpatient Treatment of Severely Malnourished Children*. Geneva, Switzerland: WHO; 2003.

http://www.who.int/nutrition/publications/guide_inpatient_text.pdf
33. World Health Organization. *Training Course on the Management of Severe Malnutrition*. WHO/NHD/02.4(P)1. Geneva, Switzerland: WHO; 2002.
34. Jackson AA, Ashworth A, Khanum S. Improving child survival: Malnutrition Task Force and the paediatrician's responsibility. *Arch Dis Child.* 2006; 91:706–710.
35. Ashworth A, Chopra M, McCoy D, et al. WHO guidelines for management of severe malnutrition in rural South African hospitals: effect on case fatality and the influence of operational factors. *Lancet*. 2004;363:1110–1115.
36. Collins S. Changing the way we address severe malnutrition during famine. *Lancet*. 2001;358:498–501.
37. Collins S. *Community-Based Therapeutic Care. A New Paradigm for Selective Feeding in Nutritional Crises. Humanitarian Practice Network.* London: Overseas Development Institute; 2004.
38. Collins S, Sadler K, Dent N, et al. Key issues in the success of community-based management of severe malnutrition. *Food Nutr Bull*. 2006;27:S49-S82.
39. Collins S, Dent N, Binns P, et al. Management of severe acute malnutrition in children. *Lancet*. 2006;368:1992–2000.
40. Ashworth A. Efficacy and effectiveness of community-based treatment of severe malnutrition. *Food Nutr Bull*. 2006;27:S24-S48.
41. Khanum S, Ashworth A, Huttly SRA. Controlled trial of three approaches to the treatment of severe malnutrition. *Lancet*. 1994;344:1728–1732.
42. Ashworth A, Khanum S. Cost-effective treatment for severely malnourished children: what is the best approach? *Health Policy Planning*. 1997;12:115–121.
43. Monckeberg F, Riumallo J. Nutrition recovery centers: the Chilean experience. In: Underwood B, ed. *Nutrition Intervention Strategies in National Development*. New York: Academic Press; 1983;189–199.
44. Uauy-Dagach R. The practice of clinical nutrition in a developing nation. *J Nutr*. 1994;124:1449S-1454S.
45. Albala C, Vio F, Kain J, Uauy R. Nutrition Transition in Latin America: The case of Chile. *Nutr Rev*. 2001;59:170–176.
46. Bhutta ZA, Ahmed T, Black RE, et al. Maternal and Child Undernutrition Study Group. What works? Interventions for maternal and child undernutrition and survival. *Lancet*. 2008;371:417–440.
47. Gómez F, Ramos Galvan R, Frenk S, et al. Mortalidad asociada a la malnutrición de segundo y tercer grado (Mortality in second and third degree malnutrition). *Bull World Health Organ*. 2000; 78:1275–1280.
48. Uauy R, Albala C, Kain J. Obesity trends in Latin America: transiting from under- to overweight. *J Nutr.* 2001;131:893S-899S.
49. Uauy R, Kain J. The epidemiological transition: need to incorporate obesity prevention into nutrition programmes. *Public Health Nutr*. 2002;5:223–229.
50. Martínez R, Fernández A. *The Cost of Hunger: Social and Economic Impact of Child Undernutrition in Central America and Dominican Republic*. Santiago, Chile: CEPAL; 2007.
51. Martínez R, Fernández A. *The Cost of Hunger: Social and Economic Impact of Child Undernutrition in the Plurinational State of Bolivia, Ecuador, Paraguay and Peru*. Santiago, Chile: CEPAL; 2009.

16 Community Nutrition and Its Impact on Children: Highly Industrialized Countries

Johanna Dwyer, DSc, RD, Kristy Hendricks, DSc, RD, Janice H.Y. Chow, MS, RD, and Melanie Reyes, MS, RD, LD

If the future of a society is to be shaped by its children, then a strong national commitment is needed to develop their bodies, minds, and spirits. Good nutrition plays a role in the growth, development and later health, prevention, control and treatment of disease, and improvement of quality of life. Community efforts are the key to making this happen. This chapter focuses on issues of public health importance in community nutrition using examples from English-speaking North America.

REASONS FOR CONCERN

Nutrition is an important component of health care for infants, children, and adolescents. Infants grow rapidly and have requirements for protein, energy, and other essential nutrients that are higher per unit of body weight than at any other time in life, making them vulnerable to dietary

deficiencies and growth failure. Dietary habits of toddlers and preschoolers are important in shaping their later health and may play a role in increasing or decreasing obesity, high blood pressure, other cardiovascular diseases such as high serum cholesterol, and dental caries later in life. Adolescence, a time of emotional, physical, social, biologic, and educational transitions, is a period of particular susceptibility to weight concerns, eating disorders, alcohol abuse, and other poor dietary habits. Children with special health care needs at all ages, because of a variety of disabilities, handicaps, and chronic illnesses and conditions that afflict them, often need special complex and involved care and require particular attention to enable them to achieve their maximum potential.

Eating patterns begin to develop early in childhood and can profoundly affect health throughout life. In childhood, the immediate effects of inappropriate diet are most apparent in such conditions as failure to thrive, obesity, dietary deficiency diseases, and fetal alcohol effects. Later, the risk of chronic degenerative disease increases. Today, the leading causes of death and disability for adults include chronic degenerative disease such as cancers, cardiovascular diseases, osteoporosis, and alcoholism. Each involves risks associated with dietary patterns, as well as other preventable factors. Programs for the family and community must be designed to accommodate and deal with all these diet-related problems.

COMMUNITY NUTRITION AND CHILDREN TODAY: THE UNITED STATES

In the year 2011, there were 74 million children younger than 18 years in the United States (about 24% of the population) down from the peak of 36% in 1964 at the end of the baby boom.[1] American children are racially and ethnically diverse. In 2011, 53% of US children were white non-Hispanic, 15% were black non-Hispanic, 5% were Asian or Pacific Islanders, 1% were American Indians and Native Americans, and 2% were children identified with two or more racial groups. In the same year, 65% of children under 18 lived in a family with two married parents, 27% with one parent, 4% without parents, and 4% unknown. Among children living with neither parent, more than 50% of them lived with a grandparent.

For many socioeconomic and health indicators, gains have been impressive over the past decade. For example, with respect to poverty, now the rate of children living with their families is 10%, which is lower than it has been for several decades. In 2010, 22% of children lived in households reporting any level of food insecurity, slightly down from 23%

in 2009. But problems still exist. For example, children in families below the poverty line are much more likely to experience food insecurity and hunger than their peers in families above the poverty line. Younger preschool children have better diets than older children, but all could use improvement. According to a study using the Healthy Eating Index in 2005, children aged 2–5 years had significantly better diets than children aged 6–11 years, in the categories of "total fruits," "whole fruits," "milk," and "extra calories." Children in families living in poverty were less likely than those from higher income families to have a dietary pattern rated as "good." They also tended to be in poorer health than their more affluent counterparts. Therefore, many health and nutritional problems remain. This chapter focuses on the ways of ameliorating them by community-wide interventions.

As a nation, the United States has made great strides in nutrition over the past century. The situation today differs dramatically from that in 1900, when infectious diseases, dietary deficiency diseases, undernutrition, and food insecurity were major societal problems.[2] Although there are still some individuals who suffer from these problems, there are new faces to malnutrition today. Overnutrition and obesity are increasing; for example, in 2003–2004, 18% of children aged 6–17 years were overweight. Sometimes overweight even coexists in the same individuals who suffer from other dietary deficiencies. Although malnutrition, undernutrition, and other dietary deficiency diseases will probably continue to be problems in some small subpopulation groups in the United States in the future, more prevalent nutrition problems involving overconsumption and imbalances in the types and amounts of food consumed and excesses of certain nutrients are more frequent challenges, particularly in an environment of physical inactivity.[2] Of all the many nutrition problems affecting young children, obesity is the most prevalent and a growing problem.[3] Its causes are many. Social and economic changes, such as altered family structures with more single parents, increasing ethnic diversity, a predominantly urban maternal and child population, high rates of poverty among children and their families, and more eating outside of the home, are some key factors in explaining the new forms of malnutrition we see today.[4]

COMMUNITY NUTRITION AND CHILDREN TODAY: CANADA

In 2011, the population of Canada was 34.5 million, and the number of

Canadian children and youth younger than 14 years was 5.6 million (approximately 16% of its total population).[5–7] The increase in the five years between 2006 and 2011 was 11.0% among children aged 4 and younger; this was the highest growth rate in this age group since the baby boom of 1956–1961.[8]

Both the United States and Canada are culturally and ethnically diverse countries. There were more than 1 million Aboriginal people in Canada in 2006, which included the First Nations (original native peoples), Métis (mixed First Nations and European ancestry), and Inuit (aboriginal people in the Arctic region).[9] Around 5 million Canadians (16% of the population) belong to a "visible minority" (e.g., non-Caucasian, non-Aboriginal ancestry). Note that the terms used to identify minority status in Canada are different from those used in the United States. Visible minority is further defined by ethnicity or native region (i.e., instead of Asian, the percentages of Chinese, South Asians, Japanese, and Korean are reported separately).[10] Community programs targeting ethnic diversity have been created, and these will be summarized later in the section Existing Programs: Canada.

THE FUTURE

There will be more children and a greater percentage of children in the population by the year 2050 than there are today in both the United States and Canada.[1] For this reason alone, continued efforts to improve child health at the community level are necessary. Whether North American children will be healthier in the future will depend on sustained efforts on the part of parents and caretakers, as well as the larger community. Systems for tracking health promotion and disease prevention objectives that include nutrition and other aspects of child and family health are now available that permit us to monitor our progress and to make corrections to improve health when they are necessary.

Table 16-1 summarizes future directions that have been suggested for community nutrition programs in the United States and Canada.

TABLE 16-1 Future Directions for Community Nutrition Programs in the United States and Canada

The United States	Canada

Work with the Canada on universal health care and update nutrition guidelines.

- With Affordable Care Act, the United States is working on providing universal health care for citizens. Collaboration with Canada will be helpful.

Revise current policies to improve food security in certain regions and within certain ethnic communities.

- Food insecurity remains high in the United States, particularly in ethnic communities and "food desserts." Revised policies and community programs are needed to improve the current situation.

Develop a national database for community nutrition resources in each province.

- Due to the absence of a national database for community nutrition resources, it is hard to keep track of program changes and sources of funding. Also, it is difficult to obtain information from different departments within each provincial government.

Develop a national School Nutrition Program.

- There is no federal program that supports school meals. To make sure the continuation of funding sources and provision of standard nutrition, there is a need to implement a national school nutrition program.

Work with the United States on affordable health care and update nutrition guidelines.

- Through the Canada Health Act, Canadians are able to gain access to health services through public health care insurance funded by the government.[69] As the United States is starting to look upon the development of affordable health care system, the collaboration between the United States and Canada is crucial.
- There is a continuous need to update nutrition guidelines under the partnership of Institute of Medicine and Health Canada.

DEFINITIONS

Public Health

Public health is "the science and art of preventing disease, prolonging life, and promoting health and efficiency through organized community effort, so organizing these benefits as to enable every citizen to realize his birthright of health and longevity."[11] The goal of public health is to attain the highest level of physical, mental, and social well-being and longevity consistent with available knowledge and resources at a given time and place.[12] It accomplishes these objectives by focusing on the community or

on large population groups rather than on individuals, and on planning and monitoring rather than on delivering services.

Community Nutrition

Communities are groups of individuals or families living in a defined geographic area that live and work together, hold similar values, are influenced by the same environment and share a number of common concerns. An understanding of the influences of society and the community is necessary for planning effective nutrition interventions. Community nutrition applies scientific knowledge in food, nutrition, and health to a specific group of people to improve their health and welfare. To be successful, it must take into consideration the wide range of societal influences and constraints.

The community nutritionist must have a public health perspective and take into account an individual's cultural eating habits, ethnic background, income, health history, clinical assessment, dietary intake, and use of community nutrition programs to screen, assess needs, and intervene effectively.

The objectives of community nutrition are to promote health; prevent nutrient deficiencies, excesses, and imbalances; prevent diet-related diseases or conditions; and rehabilitate those suffering from nutrition-related problems. This is done by promoting healthful eating habits, ensuring access to food, and integrating nutrition into primary, secondary, and tertiary health care services.[2] Thus, it represents the traditional public health concerns (such as mass prevention and intervention measures) as well as a personal health services focus to ensure that high-risk, high-priority children especially receive health and other services they need. In this respect, community nutrition represents a fusion of both the personal health service's focus on individuals and the public health focus on the entire population.

GOALS OF COMMUNITY NUTRITION

Every decade, the US Department of Health and Human Service (DHHS) produces a document entitled "Healthy People" that summarizes goals and objectives for health promotion and disease prevention for the country. These objectives include nutrition. "Healthy People 2020", the latest edition of the plan, singles out nutrition as essential to promote optimal health and prevent chronic degenerative disease and provides nutrition

goals for children and adolescents, which are listed in Table 16-2.[13] This chapter focuses on needs and meeting these Healthy People nutrition goals at the community level.

TABLE 16-2 Healthy People 2020 Nutrition-Related Goals for Children and Adolescents

- Increase rate of having medical insurance for children younger than 18 years from 94% to 100%.
- Increase the proportion of schools with a school breakfast program to 75% of schools overall.
- Reduce the proportion of people with physical disabilities reporting barriers to local health and wellness programs.
- Target 10% improvement in children and youth younger than 21 years with disabilities living in congregate care housing.
- Increase the proportion of children with disabilities (0–2 years) receiving early intervention services at home or community-based setting from 91% to 95%.
- Target 10% improvement in the proportion of elementary, middle, and senior high schools that require school health education to prevent unhealthy dietary patterns and inadequate physical activity.
- Increase the proportion of preschool Early Head Start and Head Start programs on topics like dietary patterns, physical activity, and dental health.
- Target 10% improvement in the proportion of having official school policies and practices that promote a healthy and safe physical school environment in elementary, middle, and high schools.
- Target 10% reduction in pregnancies among adolescent females from 15 to 19 years old.
- Reduce infections caused by pathogens transmitted through food and number of outbreak-associated infections.
- Target 10% improvement in reducing the proportion of children and adolescents with hypertension.
- Reduce the rate of all infant deaths within 1 year to 6 infant deaths per 1000 live births.
- Reduce low birth weight and very low birth weight and preterm births.
- Increase abstinence from alcohol, cigarettes, and drugs among pregnant women close to 100%.
- Increase proportion of women with childbearing potential with at least 400 µg folic acid daily intake.
- Increase the proportion of infants who are breastfed.
- Target 10% improvement in the proportion of children with special care

having access to medical homes or family-centered and comprehensive programs.

- Target 10% improvement in reducing the proportion of children and adolescents of 2–19 years old who are considered obese.
- Reduce food insecurity among children from 1.3% to 0.2%.
- Increase the consumption of fruits and vegetables in children of 2–19 years old to 2 cups per 1000 calories daily.
- Reduce consumption of sodium, saturated fat, solid fats, and added sugars in children of 2–19 years old.
- Target 10% improvement in reducing iron deficiency among young children and female adolescents.
- Target 10% improvement in the proportion of required daily physical education for students in public or private schools.
- Increase the proportion of children and adolescents who do not exceed recommended limits for screen time.

Source: Adapted from US Department of Health and Human Services.

The goals of child nutrition programs are to optimize growth and to decrease the risk of all diet-related diseases. Strategies to accomplish the goals in the community must include consideration of the child's family; the efforts of the child, the family, and many members of the health care team; and contributions by institutions as well as economic resources in the health, food, education, and welfare sectors of the community. A supportive physical environment is also important.

In the community setting, strategies should focus on health promotion, early intervention (EI), and prevention of diet-related diseases in groups of children and their families. In addition, community-level efforts should focus on ensuring that preventive and curative health services are in place.

Federal and state governments have been committed to promoting child health since early in this century.[14] However, some programs that have been instituted are ineffective unless communities make them available and families take advantage of them. Both states and communities exhibit considerable variation in the extent to which they provide supportive services and extra funds for reaching nutrition objectives. Detailed advice is available for discovering and mobilizing community nutrition resources.[15]

In the clinical setting, community nutrition–related strategies should focus on providing anticipatory guidance, preventive activities, and control of clinical problems and diseases involving both primary and secondary malnutrition in all children but particularly in high-risk children, such as

those who are developmentally disabled.

All health care team members need to understand the importance of community nutrition, know what programs are (or should be) available, know how to obtain them for patients and clients, and know how to advocate their extension. Otherwise, those who provide direct care to children may fail to meet all of their children's nutrition needs.

HISTORY OF COMMUNITY NUTRITION AND HEALTH: THE UNITED STATES

Only in the twentieth century did the federal government begin to play a role in ensuring and safeguarding the health of American children. The Children's Bureau was established in the Department of Labor in 1912 to "investigate and report…upon all matters pertaining to the welfare of children and child life among all classes of our people."[14] It was the forerunner of efforts now centered in the Maternal and Child Health Bureau of the DHHS. The first federal grants-in-aid to the states for planning to improve infant and maternal health were authorized by the Sheppard-Towner Act of 1921. This legislation was first administered by the Children's Bureau in the Labor Department, but since the 1960s, the US Public Health Service in the DHHS has administered such programs.

Title V of the Social Security Act of 1935 initiated the federal and state partnership for maternal and child health that served as the major impetus for the development of public health nutrition services for mothers and children. The states used these federal funds for planning and developing a wide range of programs to improve the health of mothers and children. In 1981, Title V changed with the creation of the Maternal and Child Health Service Block Grant, which provided monies with fewer specific requirements and more state discretion within the general realm of maternal and child health needs. Fifty-nine states and jurisdictions now receive funding. The federal government also provides small amounts of federal set-aside funds to support special projects of regional or national significance. Other categorical programs for specific purposes are funded from the DHHS, the US Department of Agriculture (USDA), and other branches of government. With these funds and their own matching resources, the states provide a variety of services. However, access to health care remains an unresolved issue today for many Americans and their children.[16,17] Lack of access to medical nutrition therapy and other health services is a major problem for children who have health problems

that cannot be solved by community nutrition or public health efforts. Community efforts supplement, but are not substitutes for, individual health services.[18]

Today more than 2000 public health nutritionists are employed in federal, state, and local public health agencies. They also serve as members of health care teams, where they have the responsibility of assessing community nutrition needs as well as planning and directing programs of nutrition services for individuals. Many other health professionals, including around 70,000 registered dietitians, devote some of their time and effort beyond the scope of individual counseling to community programs and also play a role in ensuring that community nutrition needs are met.

HISTORY OF COMMUNITY NUTRITION AND HEALTH: CANADA

Canada is renowned in North America for its health care system, which includes universal health insurance. “Medicare Canada”, which is Canada’s national health insurance program, was established by legislation under Canada Health Act (CHA) in 1957.[19] It was developed to ensure universal coverage and provide reasonable access to medical services for *all* citizens and permanent residents. Medicare Canada is a national program consisting of 13 provincial and territorial health insurance plans; each shares basic coverage and common features. Under the CHA, provincial and territorial governments are responsible for management and delivery of services for their residents, and for setting criteria for insurance plans to obtain federal funding.[20]

NUTRITION SCREENING, ASSESSMENT, AND INTERVENTION

Nutrition screening, assessment, and intervention are important at both the level of the individual and that of population groups. A systematic approach and an integrated structure for screening, assessing, and intervening are needed to deal with nutrition problems in both health and social service settings.[21] The approach is summarized in Table 16-3. Details on how nutrition screening and assessment apply to individuals are provided elsewhere in this book.

The first step in developing an appropriate care plan for a community

is nutrition screening of the entire maternal and child population to identify major problems. This may involve examination of relevant statistics and resources of actual surveys. Next, the specific subpopulation of interest must be considered. Screening aims both to increase nutrition awareness among communities, families, and health professionals and to identify factors increasing nutritional risks in target groups. In the community nutrition context, the screening process might involve review of health indicators, such as the prevalence of low birth weight, with further delineation of high-risk groups using income, ethnicity, age, and other characteristics of parents or infants. Those groups who are found to be at risk are then further assessed.

The second step in care planning is a comprehensive nutritional assessment of the community in which the children live. Nutrition screening and assessment should be used in an effort to find groups at especially high nutritional risk. Some nutritional issues can be easily assessed and remedied by the health and social service systems. Others require further medical or social assessment before the causes of the malnutrition can be assessed. The process of setting priorities for intervention among the groups found to be in need of services and allocating resources appropriately to these efforts depends on the individual initiative and political will of the group's providers.

The final step in care planning is intervention, which may be at either the individual or the community level. This may involve nutritional measures such as food or counseling or other interventions that improve nutritional status more indirectly, such as oral health, pharmacy, and social and mental health services. The ultimate goal is to prevent, control, or ameliorate nutrition-related problems. The end result should always be to improve or ameliorate children's health.

TABLE 16-3 Pediatric Nutrition Screening, Assessment, and Intervention in the Community

Screening	To look for possible risk factors and identify the needs for further assessment for potential health problems and nutritional implications. • Anthropometrics • Medical history • Diet and feeding ability • Laboratory values if available (e.g., serum proteins, iron)

	• Socioeconomic status and related information • Oral and mental health
Assessment	To determine any diet-related problems, inadequacy of nutrition for growth with dietary deficiency or excess in nutrition and imbalance secondary to diseases. • CDC Clinical Growth Charts (2000) - Length of height - Weight - Head circumference • BMI for weight status and risk of overweight and obesity • Clinical signs and symptoms for malnutrition or nutritional deficiencies
Intervention	To use approaches that are specific, family-centered, community-based, comprehensive, and culturally competent. • Social services • Behavioral and mental health • Nutrition counseling, education, and support • Medical care

Source: Adapted from US Department of Agriculture, Early Childhood Learning & Knowledge Center (ECLKC), WIC and Head Start.

NUTRITIONAL MONITORING AND SURVEILLANCE AT THE STATE AND NATIONAL LEVELS: THE UNITED STATES

Monitoring and surveillance are analogous to repeated screening and evaluation efforts at the individual level. Monitoring is the periodic measurement of factors that indicate that changes in nutritional status have occurred. It applies to specific individuals, communities, and the general population. Surveillance is the continuous and regular collection of data that are compiled to detect warning signs of problems at the community level early enough to provide feedback for health care team members.

The purpose of monitoring and surveillance is to provide continuous, reliable information on a community's nutritional status and the factors that influence it to find ways to improve it.

National nutrition monitoring involves measurement of health status, food supply, food consumption and composition, and dietary knowledge and attitudes.[21] At the federal level today in the United States, nutrition

monitoring and surveillance are conducted by several different government agencies. Within the DHHS, the National Center for Health Statistics of the Centers for Disease Control and Prevention (CDC) conducts the National Health and Nutrition Examination Survey (NHANES). NHANES is a periodic, population-based survey providing national data on specific health status measures, including dietary intake, eating habits, and biochemical and anthropometric indices related to nutrition. NHANES is now continuously in the field. The NHANES data provide useful benchmarks for the health of these groups. However, the number of children and adolescents surveyed is small, and not all relevant problems can be examined in each survey. Therefore, occasionally, special surveys have been conducted that oversample these groups.

In 1999, the USDA's Continuous Survey of Food Intake of Individuals was merged with the NHANES to produce an ongoing survey with a greater sample size. Both the USDA and the CDC's National Center for Health Statistics now partner on the dietary portion of the survey. The combined survey has been conducted each year since then, and recent results are available at http://www.cdc.gov/nchs/nhanes.htm. Documentation about the dietary screener questionnaire, the interview on individual foods and total nutrient intake, and dietary supplement usage can be found on the website.

The USDA monitors food security in the United States through an annual survey of approximately 40,000 US households conducted as a supplement to the US Census Bureau's nationally representative Current Population Survey. The most recent food security survey revealed that 85% of US households were food secure throughout the entire year. The remaining 15% were food insecure at least some time during the year; that is, these households were uncertain about having or were unable to acquire enough food to meet the basic needs of all household members because they had insufficient money or other resources. About 6% reported that they had very low food security, which signified that there was a reduction in intake or disruption in eating patterns for at least one household member due to lack of resources for food.[22] Minority groups of Black and Hispanic households and single female- or male-headed households had rates of food insecurity and hunger that were substantially higher than the national average.

TABLE 16-4 Health Surveillance Tools Used in Canada

Survey	Comments
Canadian Health Measures Survey	Canadian Health Measures Survey collects important health information through household interview and direct physical measures at a mobile clinic, and it helps to create national baseline data for major health concerns such as obesity, sexual behavior, lifestyle and physical activity, environmental and housing characteristics, as well as demographic and socioeconomic variables. The website is available at: http://www23.statcan.gc.ca/imdb/p2SV.pl?Function=getSurvey&SDDS=5071&lang=en&db=imdb&adm=8&dis=2
Census of Canada	Census of Canada is conducted every five years. It provides demographic information about the Canadian population, including age, gender, ethnicity, education, household salaries, social benefits, etc. It is crucial for determining public health policies and recommendations on a population level. The website is available at: http://www12.statcan.gc.ca/census-recensement/index-eng.cfm.

The USDA also provides funds for grants and contracts to universities and nonprofit organizations to perform analysis and research in support of improving food consumption practices. These include rating food intakes of various portions of the population from national survey data using the Healthy Eating Index, which is currently available at http://www.cnpp.usda.gov/Publications/HEI/healthyeatingindex2005factsh Also program-specific analyses such as studies of the National School Lunch and Breakfast Programs are available.[24] Many other publications on these and other programs are available at http://www.ers.usda.gov.

The CDC of the DHHS conducts ongoing state-based nutritional monitoring activities. The database consists of data collected on children and pregnant women enrolled in health department clinics, the Special Supplemental Nutrition Program for Women, Infants, and Children (WIC), and the Head Start programs at the state and local levels. Programs include the Pediatric Nutrition Surveillance System, the Pregnancy Surveillance System, and the Severe Pediatric Undernutrition Surveillance Systems. These surveys are not necessarily representative of the entire population but provide much useful data on groups at special risk.[25]

NUTRITIONAL MONITORING AND SURVEILLANCE AT THE PROVINCIAL AND NATIONAL LEVELS: CANADA

Table 16-4 provides a summary of two surveys that are used for health

surveillance in Canada.

DIVISION OF LABOR: MANAGEMENT OF COMMUNITY NUTRITION SERVICES

The management process consists of planning, organizing, implementing, directing, controlling, and monitoring. The dynamic aspects of the process of community nutrition service management occur at two levels. First, there is program management at the indirect service level, which includes macro-level planning, administration, supervision, consultation, and training, as well as screening and assessment of problems at the community level. Program management and evaluation efforts generally occur at this level. Second, direct service delivery may be provided to both groups and individuals, and this includes screening, assessment, intervention, and follow-up. Both levels must focus on the child and his/her family if the programs are ultimately to be effective.

Planning

Planning is a process of devising a course of action to reduce uncertainty and to anticipate and initiate change. Community nutrition planning involves analyzing subjective and objective data, writing mission statements, setting goals and objectives, and activating means of achieving objectives. Nutrition planning is influenced by the internal culture and capabilities of each organization involved, as well as by the public agenda, state of scientific knowledge, wants and needs of the families, and advocacy of experts, parents, and professional groups.

Community nutritionists should participate in the planning processes of the agency, study the agency or community health plans, know those who participate in the planning group, get involved, and involve others. Planning goes beyond administrators, leaders, and program managers and should include those who will be responsible for implementation and those who will be recipients of services.[26]

Planning for high-priority nutrition problems can be aided by forming alliances with related constituencies in the service network. Some examples are collaborative planning by nutrition service providers, advocates, and patients. This sort of collaboration may build political support and bridge organizational and ideological boundaries, identify mutual goals, and build cohesion and capacity to act together if potential adversaries are approached, co-opted, and included in the process. In some

instances, it may assist in making difficult decisions about using limited resources.

Implementing

Planning identifies what to do. Implementation begins the process of doing. Effective community nutrition program implementation requires administrative support (budget, staff, space, and equipment); commitment of staff to program goals, objectives, and interventions; and support and respect of a target population who is also convinced of the benefits of the program and willing to commit time and effort to participate.

Screening and assessment at the community level are the responsibilities of the community nutritionist, who ensures that systems are in place in community programs and encourages their institution in personal health services. Nutrition screening and assessment also occur at the direct service delivery level, where they are usually administered by health or social welfare care team members such as dietitians, physicians, nurses, social workers, and physical therapists.

The role of the community nutritionist in implementing these programs varies. Depending on the health and nutritional problems found, children and/or families are referred to different community and personal health services for interventions, including nutritional counseling, food, welfare education, and health programs, or to private physicians.

Monitoring and Evaluation

Community nutrition program implementation must be systematically monitored by the collection and analysis of appropriate data to provide the continuous feedback needed to assess performance. Evaluation involves the systematic measurement of results by comparing the data collected with pre-established standards or controls to decide whether to continue, expand, redesign, or terminate the program.

Monitoring must also be accomplished at both direct and indirect service levels. At the direct personal health service delivery level, the health care team evaluates program impact on a population or community by observing improvements in health. At the indirect level, the management of the community nutrition program may also evaluate effectiveness, efficiency, and cost through cost-effectiveness analysis to determine whether the program's benefits are sufficient to offset its cost.

One current effort of this type in the United States is the Preventive Health Services Task Force. Its publications on health services that are

viewed as effective are increasingly being used as a guide for services.[27] Another key document for monitoring progress at the community level is the tracking system for the Healthy People 2020 objectives.[28,29] In Canada, Canadian Health Measures Survey and Census of Canada are health surveillance tools that are in place.

CURRENT NUTRITIONAL STATUS OF INFANTS, CHILDREN, AND ADOLESCENTS: THE UNITED STATES

This section summarizes the major problems of different age groups today in the United States and some diet-related risk factors. It provides recommendations needing incorporation in both community nutrition programs and personal health and social services for children. Some nutrition problems such as obesity, food insecurity, excessive consumption of calories from solid fats, saturated fat, sodium and added sugars, and iron deficiency affect some children at all ages and are matters of concern across the entire developmental period.[29] Others, as described in the following sections, affect specific age or life stage groups.

Pregnant Women and Infants

Community concern about good nutrition should start before birth of the child. Nutritional needs in pregnancy and lactation at both the individual and community levels have been well summarized elsewhere.[30,31] High-risk women who are poor need to be enrolled in the WIC and provided with nutrition counseling, food education, and other specialized health services.

Adequate nutrition is vital for premature, small-for-date, and other low-birth-weight infants who are at risk of developmental disadvantage. (See other chapters in this volume for more details.) EI programs to encourage and promote good nutrition among those infants most in need are of particular concern.[32]

Nutrition is especially critical in the first year of life.[33] Nutrient needs are high because growth and development are more rapid in infancy than at any other time after birth. Weight usually triples and height doubles in the first year. Approximately one-third of calories consumed is used for growth in very early infancy, and it remains high during the first four to six months. This rapid growth in the healthy term infant during the first

four to six months of life requires more energy, protein, and other essential nutrients per unit of body weight than at any other time in infancy or childhood. The feed of choice for young infants is human milk, or if this cannot be provided, iron-fortified infant formula, in which its nutrient content is specified by law. Feeding additional nourishing foods to infants thereafter promotes adequate growth and good health.

The gastrointestinal tract of the newborn infant is well developed but immature. All of the secretions of the digestive tract contain enzymes especially suited to the digestion of human milk. The ability to handle foods other than human milk or formula depends on the physiologic development of the infant. Thus, it is important to encourage breastfeeding and to feed infants age-appropriate foods selected on the basis of their developmental readiness.[34] This will not only help to prevent complications but also help to build positive attitudes about food and eating.

Attention to feeding the appropriate type and amount of food is essential. All parents and caretakers need appropriate advice and guidance. For the poor with high nutritional risk, WIC, which provides food, health services, and education, may be in order. For the ill, Title V and Medicaid-supported health services, as well as special services for those with especially severe handicapping conditions, may be in order.

Some of the common nutritional and health problems and related risk factors in infants that need attention by community nutritionists are summarized in Table 16-5. Table 16-6 provides recommendations for promoting good nutritional status during this period.

Preschool and School-Aged Children

Table 16-7 outlines some of the current nutritional concerns, and Table 16-8 provides recommendations on promoting good nutrition after infancy in the preschool and school years. New findings in the Feeding Infants and Toddlers Survey 2008 can be assessed at http://download.journals.elsevierhealth.com/pdfs/journals/0002-8223/PIIS0002822310014793.pdf. This population-based survey is one of the latest and most recent studies of very young children in the United States. These data and the NHANES survey provide a glimpse at the dietary status of American infants and young children at present. The US DHHS is now considering to develop dietary guidelines for infants and children younger than 2 years.

In late infancy, children begin to learn to feed themselves independently. They progress from eating with their hands to using

utensils. Children need to be independent and able to try new skills to help them develop positive attitudes about food and eating.

Children's growth rates slow considerably after the first year of age, resulting in decreased appetite and food intake. This normal phenomenon is often mistaken for poor appetite, although it is not. In spite of the slower growth rates, appropriate nutrition is vital to provide needed nutrients and energy to maintain resistance to infections, materials needed to build new tissues as growth proceeds, and adequate nutrient stores.

Good nutrition is important to sustain growth and permit development. It is especially important in the weaning period to prevent the risks of iron deficiency anemia, dental caries, and obesity and diet-related risks that may contribute to later hyperlipidemia and hypertension.

Obesity is increasing in Americans, with estimates of obesity about 20%–30% in children aged 6–19 years.[35,36] Therefore, attention to nutrition must not be neglected in the preschool, primary, and middle school years since problems can develop at any time. In 2008, more than one-third of children and adolescents were overweight, and these figures have tripled since 1980. Also, associated conditions such as cardiovascular diseases and type 2 diabetes are increasing among children, and diet-related chronic disease risks linked to obesity are also on the rise. Minorities and low-income families are often particularly afflicted. In 2010, the White House Task Force on Childhood Obesity published a report "Solving the Problem of Childhood Obesity Within a Generation" to President Obama. Along with the First Lady Michelle Obama's Let's Move Campaign, this report emphasized that childhood obesity was becoming a "national health crisis."[36] Therefore, there is a need for immediate action. The National Institutes of Health have produced many useful materials that are online to help that can be accessed at http://www.nhlbi.nih.gov/health/public/heart/obesity/wecan/, and additional publications are available at the CDC's Division of Nutrition and Physical Activity (access at http://www.cdc.gov/nccdphp/dnpao/publications/index.html).

TABLE 16-5 Current Nutritional Issues and Recommendations for Infants in the United States

Problem	Comments
Growth retardation and underweight	Dietary inadequacies Low birth weight

	Inappropriate introduction of foods
Dental caries	Excessive or frequent consumption of sweets that are retained in the mouth Baby bottle tooth decay Lack of fluoride
Iron deficiency anemia	Early introduction of cow's milk Malnutrition Inadequate dietary intake of iron
Lack of breastfeeding	Inadequate nutrients Increased risk of maldigestion Increased risk of infections, obesity, and type 2 diabetes
Other nutrient deficiencies	Inadequate intake of certain nutrients Poor dietary quality Lack of appropriate supplements
Infections	Malnutrition Lack of breastfeeding

Recommendations:

- Improve nutrition training of health professionals
- Increase nutrition counseling and education of parents and caretakers
- Strengthen breastfeeding support
- Develop nutrition and feeding guidelines for agencies responsible for the administration of health social welfare and related program services
- Enhance research base for current issues in infant nutrition
- Prevent or treat inappropriate weight gain

TABLE 16-6 Recommendations for Enhancing Nutritional Status of Infants in the United States

Early infancy (0–6 months)

- Exclusive breastfeeding until 6 months of age
- If breast milk is contraindicated, feed with a nutritionally adequate and heat-treated infant formula until 6 months of age
- Supplement with vitamin D (400 IU per day) if infant is solely breastfed for the first 6 months of life or if these are not provided in formula
- Avoid providing regular cow's milk and skim milk to infants younger than 1 year and encourage breastfeeding and adequate supplementation
- Ensure that the infant receives enough food by monitoring growth
- Use proper feeding techniques to avoid gas, aspiration, and other problems

- Delay introduction of complementary foods until 4–6 months when the infant is developmentally ready to deal with them
- Avoid foods commonly causing allergies, intolerances, or hypersensitivities
- Ensure that water intakes are adequate
- If family resources are inadequate, obtain assistance from federally sponsored food programs for infants and children
- Obtain relevant anticipatory guidance on feeding and health

Later infancy (6–12 months) Introduce complementary food rich in iron and zinc

- Supplement with fluoride after 6 months of age (limited in communities where fluoride concentration in water is <0.3 ppm)
- Introduce complementary solid foods after 6 months of age, along with continued breastfeeding for 1 year or more. Introduce regular cow's milk or skim milk after 1 year of age.
- Ensure a gradual transition to family diets rather than weaning off suddenly and encourage self-feeding
- Take common problems in stride—temporary refusals of solid foods at weaning and failures to switch over from pureed or baby foods to foods that must be chewed are normal setbacks
- Consult health providers if feeding problems persist
- Realize that some decrease in appetite in late infancy is to be expected as growth slows and that is no cause for alarm
- Avoid struggles and battles of will with the child about food
- Keep environments safe (especially with respect to lead and common household chemicals) and begin to teach child to shun mouthing and eating nonfood objects
- Begin oral hygiene measures

Source: Adapted from (1) American Academy of Pediatrics, Policy Statement on Breastfeeding and Use of Human Milk, 2012; (2) American Academy of Pediatrics, *Pediatric Nutrition Handbook*, 5th Ed.

Strategies that communities can use in helping to address overweight and obesity especially in children and adolescents involve "CARE" (**C**ommunication of the problem and steps to correct it, **A**ctions, **R**esearch, and **E**valuation). These action options include the following:

1. *Physical activity.* Require daily, quality physical education at all school grades. In 2011, about 30% high school students participated in at least 60 minutes of physical activity per day. It is also important to provide safe and accessible facilities for the recreation of people of all ages. Community facilities should be available for physical activity for all people, including on the weekends, and there should be more opportunities for physical

activity at work sites. At the same time, it is important to reduce the time people spend in watching television and other sedentary behaviors. According to a recent survey by the Kaiser Family Foundation, children and adolescents aged 8–18 spend around 7.5 hours per day in front of screen for entertainment; half of time is spent in watching television.[37] The screen time spent can cause further physical inactivity.

2. *Food options*. Ensure the availability of more food options that are low in saturated fat and calories, as well as fruits, vegetables, whole grains, and low-fat or nonfat dairy products, on school campuses and at school events. It is important to enforce existing USDA regulations that prohibit serving foods of minimal nutritional value during mealtimes in school food-service areas, including vending machines. The ChooseMyPlate.gov website provides health and nutrition information for pregnant women, preschoolers and children older than 5 years.
3. *Communications*. Change the perception of obesity so that health, and not simply personal appearance, becomes the chief concern. This means providing appropriate education in schools and communities and training of health professionals about healthful eating habits and regular physical activity, based on the Dietary Guidelines for Americans, for people of all ages.
4. *Growth monitoring*. New growth charts are now available for children and adolescents to evaluate their size and growth.[38] They need to be used on a continuing basis and can be accessed at http://www.cdc.gov/nchs/about/major/nhanes/rowthcharts/htm.

TABLE 16-7 Current Nutritional Issues and Recommendations for Children in the United States

Problem	Comments
Obesity	Excessive energy intake Lack of physical activity
Failure to thrive	Dietary adequacies
Iron deficiency anemia	Malnutrition Inadequate dietary iron intake
Dental caries	Excessive or frequent consumption of sugary foods

	Poor oral hygiene
High blood pressure	Overweight High sodium intake
High blood cholesterol	Excessive intakes of saturated fat, trans fat, and cholesterol
Food insecurity	Lack of access to social service programs Inadequate nutrients

Recommendations:

- Achieve adequate nutrition services in daycare programs and early childhood education
- Increase participation and coverage of child nutrition programs
- Increase parent and community nutrition education
- Improve nutrition training of health professionals
- Enhance the research base to answer existing questions in child nutrition
- Change policy to integrate nutrition services into all aspects of child health care
- Improve policies on increasing access of food and venues for physical activity for children

TABLE 16-8 Recommendations for Enhancing Nutritional Status of Children in the United States

Preschool years

- Ensure that levels of physical activity and rest suffice to maintain normal growth, development, and appetite
- Provide nutritious food at home and school where child eats and encourage healthy food choices
- Teach child to choose and eat nutritious foods for meals and snacks
- Avoid sugary and starchy snacks that can retain in the mouth and encourage oral hygiene and good dental practices
- Foster appropriate eating behaviors by providing suitable role models, by permitting the child to choose food portions, by giving some latitude in food preferences, by exposing him/her to new foods, by fostering table manners appropriate to his/her developmental level, and by keeping meal as times for social exchange
- Recognize and deal with feeding problems early before they become well established
- Handle struggles over food reasonably to avoid the development of feeding problems

- Feed and hydrate children appropriately during illness
- Ensure that iron needs are met by food choices
- Ensure that the child learns to distinguish food and nonfood objects
- Limit screen time to two hours maximum for children older than 2 years.

School years

- Monitor growth in height and weight to monitor nutritional status
- Provide good examples and guidance to instill healthful habits and attitudes about food and eating
- Establish consistent guidelines and follow them to ensure adequate nutrition for the child's diet
- Promote healthy eating and physical activity if child's body weight is concerning
- Ensure that diet-related risks for dental caries are minimized
- Help the child to distinguish reliable sources of information on food choices and promotional messages
- Encourage moderation consumption of sodium, solid fats, saturated fat, and added sugar; encourage diet adequate in fiber
- Limit screen time to two hours maximum for children older than 2 years.

Adolescents

Puberty is a unique and complex developmental period marking the transition from childhood to adulthood. It is accompanied by a series of physical, physiologic, biochemical, hormonal, social, and psychological changes. Adolescence is a nutritionally vulnerable period for several reasons. A dramatic increase in physical growth occurs during puberty. The growth spurt is highly sensitive to nutrient deprivation. Inadequate nutrition during this time may delay or stunt linear growth. Alterations in body composition in puberty and during the consolidation that occurs in later adolescence, as maturity is reached, increase nutrient and energy needs. During the second decade of life, children gain about 20% of their adult height and 50% of their adult weight.[39] After puberty, although linear growth ceases, changes continue until maturity is reached. The larger body requires increased amounts of nutrients. Calcium is a nutrient of particular concern because bone growth and bone mass accumulate rapidly during this time of life. Osteoporosis is a disease that afflicts mostly the elderly, but just as is the case with atherosclerosis; its seeds are sown in childhood. One of the most effective ways to prevent osteoporosis is to build a strong, dense skeleton during the growing years to act as a reserve later in life.[40]

Moreover, the changes in individual behaviors and lifestyle preferences in younger children may last through adolescence and into adulthood, again contributing a lifetime of optimal bone health. An increasing attention is being paid to school programs that will increase bone accretion and muscular strength in children starting in early elementary school, aged 5–7 years, and continuing into adolescence. The interventions now being tested include weight-loading physical activity and calcium-rich snacks for children as well as age-appropriate, behaviorally focused active learning to promote skill building and self-competence while building bone quality and bone strength.

Growing independence, the need for peer acceptability, concern with appearance, and active lifestyles of adolescents often affect eating habits, food choices, nutrient intake, and thus nutrition status. Adolescents are seeking to establish their identity and are susceptible to peer pressure and mass media. They are also preoccupied with physical appearance and body shape and size. Desires to be lean and to have an attractive body, societal pressures, and an abundance of food in the environment can lead to inappropriate weight reduction, dietary aberrations, and nutrient deficiencies.[41] Chronic dieters are at high risk for inadequate diets. In adolescent females, such behavior may lead to eating disorders, such as anorexia nervosa and bulimia nervosa.[42,43]

Common nutritional problems in adolescents are provided in Table 16-9. They include obesity, hyperlipidemia, eating-related disorders, and hypertension.[41–43] These problems of dietary imbalance and excesses may coexist with undernutrition with respect to iron, energy, and other nutrients in some individuals. Dietary factors increasing the later risk for chronic diseases such as osteoporosis (inadequate intakes of calcium and vitamin D), heart disease (excessive intake of saturated fat and cholesterol and low intake of fiber), and some types of cancer may be present.[39,41,44] Table 16-10 provides recommendations to enhance nutritional status during adolescence.

TABLE 16-9 Current Nutritional Issues and Recommendations for Adolescents in the United States

Problem	Comments
Hypertension	Overweight Excessive sodium consumption Low potassium intake

Underweight	Undernutrition Anorexia nervosa Bulimia nervosa
Obesity	Excessive energy intake Sedentary lifestyle
Iron deficiency anemia	Malnutrition Inadequate dietary iron intake Pregnancy without iron supplementation
Nutrient deficiencies	Inadequate nutrient intakes Extended periods of dieting
Eating disorders	Societal pressure to be thin Peer pressure Diet fads
Alcohol and substance abuse	Peer pressure

Recommendations:

- Include nutrition guidelines for school-based nutrition education programs
- Increase nutrition education to parents and communities
- Improve training and continuing education for all health care providers in the area of nutrition
- Integrate nutrition screening, education, and intervention services into health services aimed at teenagers
- Expand school-based health clinics especially for teenagers who have no primary care
- Expand research on nutritional needs of adolescents
- Provide and promote social services for adolescents for behavioral and mental health

Pregnancy and lactation in adolescence also increase nutrition risk because the young female needs a supply of nutrients to support her own growth as well as the growth of the fetus and nutrition of the suckling infant.[45,46] Nutritional needs of pregnancy and lactation are discussed elsewhere in this book.

CURRENT NUTRITIONAL STATUS OF INFANTS, CHILDREN, AND ADOLESCENTS: CANADA

Table 16-11 summarizes nutritional issues of concern for children and youth in Canada. Factors that affect the nutritional status of children in Canada are similar to those in the United States. They include obesity, excess sodium intake, limited physical activity, and lack of fruit and

vegetable intake. Undernutrition, malnutrition, and food insecurity still occur in some Canadian children and youth, particularly among the poor. However, mean overall intakes of Canadian children between 1 and 8 years of age in recent surveys provided adequate amounts of most vitamins and minerals and appropriate proportions of energy from protein and carbohydrate.[47]

TABLE 16-10 Recommendations for Enhancing Nutritional Status of Adolescents in the United States

- Maintain normal healthy weights recommended. Avoid obesity and eating disorders by developing healthful attitudes toward food, appropriate energy intakes, and a physically active lifestyle, including both aerobic and strength-building exercise
- Abstain from self-induced vomiting, bulimia, and laxative abuse
- Eat a variety of foods to ensure dietary sufficiency of protective nutrients, including protein, vitamin, minerals, and fiber
- Identify dietary imbalances and avoid them by educating adolescents on moderate intake of fat, saturated fat, sugars, and salt
- Avoid alcohol and substance use
- Provide assistance on teenage pregnancy, diets for competitive athletes, diets for those with allergies and disease
- Ensure adequate intakes of calcium and vitamin D to maximize bone density

TABLE 16-11 Current Nutritional Issues of Children and Youth Canada

Problem	Comments
Obesity	BMI: 32% of Canadian children and youth aged 5–17 years were overweight or obese in 2009–2011 data.[70,71] Excess energy intake: One in five Canadian children have energy intakes exceeding their needs.[47]
Lack of fruit and vegetable intake	Only 49% of children and youth eat fruit and vegetables more than five times daily.[72] In 2011, 40% of Canadians aged 12 and older reported that they consumed fruit and vegetables five or more times per day.[73]
Excess sodium intake	More than 75% of 1–8 year olds had usual intake of sodium exceeding the upper limit set for their age group (1500–1900 mg/d).[47]

Limited physical activity	Increased screen time: Canadian children see more than 50 fast food commercials on television each month.[72] Sedentary time: Nearly 9 hours or 62% of waking hours is the average of the total daily sedentary time for Canadian children and youth. Sedentary time rises with increasing age.[74] Amount of physical activity: 7% of Canadian children and youth accumulate at least 60 minutes of moderate-to-vigorous activity at least 6 days a week.[74]
Undernutrition and food insecurity	10% of households with children are food insecure as a result of financial challenges faced in having access to adequate food.[70] Fewer servings of fruits, vegetables, and milk products were consumed by children and youth younger than 18 years, demonstrating constraints in food intake.[75]

NUTRITION FOR CHILDREN WITH SPECIAL NEEDS: THE UNITED STATES

Some of the most common nutritional issues of children with special needs are outlined in Table 16-12. Children with special health care needs make up approximately 10%–20% of the pediatric population, depending on the definition used.[48] The term special needs describes a group of infants, children, and youth with or at risk for physical or developmental disability or with a chronic medical condition caused by or associated with genetic/metabolic disorders, birth defects, prematurity, trauma, or infection (including HIV infection). They present a particular challenge to community nutritionists today because many children with special needs who were formerly institutionalized now dwell with their families in the community.

Table 16-13 provides some recommendations for enhancing the nutritional status of children with special needs. Some of the nutrition needs of children with special health care needs are unique (e.g., the child with a metabolic disorder controllable only by diet), but in most aspects, their nutritional needs are similar to those of other children. The child with "special needs" in the family is first of all a child who has the same nutrient needs for optimal health, growth, and development as any other child. Superimposed on these needs, however, may be multiple risks, ranging from social/environmental factors, such as decreased access to food because of limited finances, to factors involving disorders, such as feeding problems or the need for chronic medication.[49] These factors may place a child at risk for inadequate nutrient intake, impaired nutrient absorption or use, or increased nutrient excretion. Such insults, if they are

severe and/or prolonged, and especially if they occur early in life, can have lasting effects on the child's growth and cognitive development. Various handicapping conditions have different nutrition-related problems and needs.[50] These will be discussed in detail in Chapter 47.

TABLE 16-12 Current Nutritional Issues and Recommendations for Children With Special Needs in the United States

Problem	Comments
Growth retardation or underweight	Increased needs Inadequate intake caused by mechanical difficulties with chewing, swallowing, or feeding oneself and malabsorption
Obesity	Reduced resting metabolism Decreased energy expenditure Excessive energy intake
Gastrointestinal complications	Diet inadequate of fiber and water
Nutrient deficiencies	Inadequate intake of certain nutrients Lack of supplementation Increased nutrient needs Drug–nutrient interaction
Infections	Malnutrition
Lack of physical activity	Muscle wasting Overweight Suboptimal bone health and strength

Recommendations:

- Ensure that community programs for children with special needs are in place to screen and assess nutritional problems
- Implement use of standardized screening and referral tools for service providers
- Improve nutrition training of health professionals
- Increase nutrition counseling and education for families
- Develop nutrition and feeding guidelines for agencies responsible for the administration of health, social welfare, and related program services
- Enhance the research base for current issues in nutrition for children with special needs

An amendment (PL 99-457) to the Federal Education for the Handicapped Act gives nutrition professionals the opportunity to have a voice in establishing nutrition policy and standards of care for young

handicapped and high-risk children. Preventive services now extend to children as young as 3 years of age, and Part H of the law provides financial incentives for states to provide services to children with special health care needs from birth to 2 years of age. Nutrition should be included in these services.

TABLE 16-13 Recommendations for Enhancing Nutritional Status of Children With Special Needs in the United States

- Recognize that expectations for growth may be different; develop disease or condition-specific growth charts and guidelines
- Provide adequate nutrition to achieve optimal growth potential
- Consider the extra or lessened energy needs owing to the child's condition and adjust intake accordingly
- Use vitamin and mineral supplements if directed by physician
- Ensure adequate intake of fiber and water
- Consult an interdisciplinary team and develop specific care plan that includes feeding skill progression, appropriate physical activity, recognition of developmental readiness, and enrollment in early intervention programs
- Adjust feeding positioning to allow for adequate food intake, use appropriate nipples and feeding devices for children unable to use standard equipment, and adjust pace of feeding
- Determine family's need for food assistance, financial support, diet information, and emotional support

NUTRITION FOR CHILDREN WITH SPECIAL NEEDS: CANADA

The Canadian Paediatric Society's 2009 position paper on the nutritional status in children with neurological impairment, a group with high risks of problems in nutritional status, is summarized in Table 16-14. Approximately 3.7% of children and youth of ages of 0–14 years have disability in 2006 with a greater proportion of boys (4.6%) than girls (2.7%).[51] The nutritional status of children with special needs is difficult to assess because it varies from condition to condition, and few studies have been done.

EXISTING COMMUNITY NUTRITION PROGRAMS: THE UNITED STATES

Federal Programs

Either the USDA or the DHHS administers most federally funded food and/or nutrition programs for mothers and children. The programs are administered on the state level by the Department of Welfare, Department of Education, or Department of Health depending on their particular focus. One type of federal support is through benefits or cash assistance that goes directly to eligible recipients (such as the Supplemental Nutrition Assistance Program [SNAP], formerly called the Food Stamp program), which provides food vouchers or the School Lunch program that provides inexpensive or free meals). Another type is in-kind services (such as the USDA's Expanded Food and Nutrition Education Program [EFNEP], which provides nutrition education to low-income families).

TABLE 16-14 Key Points From the Canadian Pediatric Society's Paper: "Nutrition in Children With Neurological Impairment"

- A significant number of children with neurodevelopmental disabilities are undernourished. In particular, most children with cerebral palsy are undernourished. However, this is not the only problem; about 8%–14% of them are overweight.
- The causes of malnutrition include inadequate intake, increased losses, altered metabolism, oromotor dysfunction, specific syndromes, endocrine dysfunction, ethnicity, genetic potential, and associated factors such as pubertal status in these children.
- Frequent medical problems of these children include gastroesophageal reflux, pulmonary aspiration, and osteoporosis.
- Improvement of oral intake is crucial if it can be done safely. Oromotor therapy, adjustment of food consistency, and increase in food caloric density can be helpful.
- Multidisciplinary management is important in the nutritional care in children with neurological impairment.
- Careful monitoring of nutrition interventions is needed to avoid excess weight gain or any nutrient deficiencies, and of the need for enteral or parenteral nutrition.

Source: Marchand; Canadian Paediatric Society, Nutrition and Gastroenterology Committee.[76]

Means-tested programs require assessment of economic means or

income before one qualifies for them. They are special cash or in-kind services targeted to provide a safety net to protect children in low-income families. Eligibility for such programs, such as SNAP, Medicaid, general relief payments, Title V funded child health services, or free school lunch and breakfast under the National School Lunch and Breakfast Program legislation, is assessed by review of family incomes. These programs are valuable contributions for maintaining or improving the nutritional status of low-income families. Such families may have limited access or resources available to buy food, and those who are particularly vulnerable from the physiologic standpoint include infants, young children, and pregnant and lactating women.

Community nutrition services for families and children are also available as part of other health and social services, such as intensive infant care programs, services for handicapped children, dental health programs, family planning service, school health, and teenage pregnancy/parenting programs.

The federal government also provides grants for planning and service provisions to states for program development to provide food assistance, nutrition education, and counseling to eligible families at the local level through health centers, school systems, and community action agencies.

Health care providers need to know about the many food and nutrition programs available in their local areas and how and when to refer their patients to them.

USDA Programs

The USDA provides various public health programs at the community level. These include SNAP, School Lunch and Breakfast, Special Milk Program for Children, Child Care Food Program, Summer Food Service Program for Children, WIC, EFNEP, and Commodity Supplemental Food Program (CSFP). About one in every four Americans participates in at least one of these programs in 2011. These programs account for over two-third of USDA's budget in the fiscal year of 2011, totaling $103.3 billion.[52] Food pantries and soup kitchens are also available for some families that lack food. Surplus foods, both dry and perishable, are distributed to food pantries and soup kitchens. Table 16-15 presents a summary of current expenditures on these programs.

Supplemental Nutrition Assistance Program

What It Is

This is the largest and most far-reaching food assistance program for low-income families in the United States to help them buy the food they need for a nutritionally adequate diet. The amount provided is based on the USDA's Thrifty Food Plan, a market basket of suggested amounts of food that make up a nutritious diet and that can be purchased at a relatively low cost. The federal government pays for all benefits issued through the program and shares administrative costs with the states. Puerto Rico, American Samoa, and the Northern Mariana Islands receive cash grants instead under the Nutrition Assistance Program that permits them to provide food assistance programs for their low-income residents. SNAP is a means-tested entitlement program; any house-hold that meets program eligibility requirements may receive them. Households participating in the program actually receive their benefits by an electronic benefits transfer (EBT) card system rather than by stamps or coupons, as in days gone by. Benefits can be used to buy food but not alcohol, tobacco, medicines, or nonfood items such as soap or paper supplies. Nationally, the majority of all SNAP recipients are children, and the preponderance of SNAP benefits is paid to families with children.

Who Qualifies

For families with children, the principal eligibility requirement is that gross household income is below 130% of the federal poverty level and that net income, after various deductions, is at or below 100% of the poverty level. The value of a household's assets (e.g., cash on hand, checking or savings accounts, stocks and bonds, recreational boats and vehicles, some cars, and land that is owned but not inhabited by the household) is also accounted for in determining eligibility.

TABLE 16-15 Food and Nutrition Assistance Program Costs, USDA (Fiscal 2010)

Assistance Program	Program Cost, 2010 ($ Millions)
Food-assistance related programs	
Supplemental Nutrition Assistance Program	68,180
Nutrition Assistance Program for Puerto Rico, American Samoa and the Northern Marianas	2049
Child nutrition programs	
National School Lunch Program	10,458

School Breakfast Program	2843
Child and Adult Care Food Program	2634
Summer Food Service Program	357
Special Milk Program	12
Supplemental food programs	
WIC	6757
Commodity Supplement Food Program	203
Food donation programs	
Food Distribution on Indian Reservations Disaster Feeding Program	93
The Emergency Food Assistance Program	468

Abbreviation: WIC, Special Supplemental Nutrition Program for Women, Infants, and Children.

Source: Adapted from USDA Food and Nutrition Assistance Research 2010. http://www.ers.usda.gov/topics/food-nutrition-assistance/food-nutrition-assistance-research/charts/costs-and-participation.aspx

How to Enroll

Contact the state or local welfare office and obtain an application.

Child and Adult Care Food Program

What It Is

This program provides cash reimbursement and/or commodities for the provision of meals and snacks to institutions such as child care centers and family and group daycare homes providing nonresidential child care to children from low-income families. This program funds the meals provided in Head Start. This is not an entitlement program. Subsidies are approved to provide two meals and one snack per day for children younger than 12 years in independent centers and sponsoring organizations. Children and youth (18 years or younger) are eligible for free meals and snacks if they are in the "at-risk" afterschool care programs. Meals must meet USDA-specified requirements. Family or group daycare homes are paid a fixed reimbursement, depending on the type of meals served. Daycare centers are reimbursed for meals at rates depending on the type of meal and eligibility of children for free, reduced-price, or paid meals.

Who Qualifies

Institutions eligible to participate include child care centers, settlement houses, neighborhood and Head Start centers, and institutions providing day care for the handicapped and their families or group daycare homes. Private daycare centers can participate only when at least 25% of their

enrolled children are from low-income families receiving benefits through Title XX of the Social Services Act.

How to Enroll

Contact your local state agency, the local school authority, or child care center.

School Lunch and School Breakfast Programs

What They Are

Over 90% of all schools in the United States participate in the National School Lunch Program. They receive cash assistance to help purchase food and pay labor costs as well as direct donations of agricultural food commodities. Participating schools must agree to serve meals at a reduced price or free to children who are unable to pay the locally established full price. The federal reimbursement level is such that all meals fed to children receive some subsidies, but they are largest for the poorest children, who receive meals free. To receive a free or reduced-price meal, means tests are required. Each year, the Secretary of Agriculture issues uniform national standards for free and reduced-price meal eligibility based on national poverty guidelines.

To ensure that the nutrition goals of the National School Lunch and Breakfast programs are met, minimum meal pattern requirements are specified, and the USDA periodically updates these. USDA updated the program's meal pattern and nutritional standards based on the latest *Dietary Guidelines for Americans*, which was passed by Congress in the Healthy, Hunger-Free Kids Act of 2010. Table 16-16 outlines the updated breakfast and lunch meal patterns. The new meal pattern goes into effect starting in school years of 2012–2014. There is an increased amount of vegetables, fruits, and whole grains in the menu. Age-appropriate calorie limits are set for grades K-5, 6-8, and 9-12. In addition, sodium content in meals will be gradually decreased, with target goals to be reached by 2023.

Who Qualifies

Any public or nonprofit private school of high school grade or under is eligible. Public and licensed nonprofit private residential child care institutions such as orphanages, homes for retarded children, juvenile detention centers, and temporary shelters for run-away children are also eligible.

TABLE 16-16 USDA Updated Meal Patterns and Dietary Specifications for School Meals in the United States

Meal Pattern	Breakfast Meal Pattern			Lunch Meal Pattern		
	Grades K–5	Grades 6–8	Grades 9–12	Grades K–5	Grades 6–8	Grades 9–12
Amount of food per week (minimum per day)						
Fruits (cups)	5 (1)	5 (1)	5 (1)	2 1/2 (1/2)	2 1/2 (1/2)	5 (1)
Vegetables (cups)	0	0	0	3 3/4 (3/4)	3 3/4 (3/4)	5 (1)
• Dark green	0	0	0	1/2	1/2	1/2
• Red/orange	0	0	0	3/4	3/4	1 1/4
• Beans/peas (legumes)	0	0	0	1/2	1/2	1/2
• Starchy	0	0	0	1/2	1/2	1/2
• Other	0	0	0	1/2	1/2	3/4
Additional vegetables to reach total	0	0	0	1	1	1 1/2
Grains (oz)	7–10 (1)	8–10 (1)	9–10 (1)	8–9 (1)	8–10 (1)	10–12 (1)
Meats/meats alternates (oz)	0	0	0	8–10 (1)	9–10 (1)	10–12 (1)
Fluid milk (cups)	5 (1)	5 (1)	5 (1)	5 (1)	5 (1)	5 (1)
Other specifications: Daily amount based on the average for a five-day week						
Minimum to maximum calories (kcal)	350–500	400–550	450–600	550–650	600–700	750–850
Saturated fat (% of total calories)	<10	<10	<10	<10	<10	<10
Sodium (mg)	≤430	≤470	≤500	≤640	≤710	≤740
Trans fat	Nutrition label or manufacturer specifications must indicate zero grams of trans fat per serving.					

Source: Adapted from USDA, Food and Nutrition Service, 2012. http://www.fns.usda.gov/cnd/Governance/Legislation/dietaryspecs.pdf

How to Enroll

Contact the local school authority.

Special Milk Program for Children

What It Is

This program reduces the cost of each half-pint of milk served to children by providing for cash reimbursement at an annually adjusted rate to public and nonprofit schools, child care centers, summer camps, and similar institutions that do not participate in any other federally assisted nutrition program. Participants can choose to provide milk free or at low cost to all children or may elect to serve free milk to children whose families are below 130% of poverty. Schools and institutions can offer only pasteurized fat-free or low-fat (1%) fluid milk; all milk should contain standard levels of fortification of vitamin A and D (vitamin A: 6000 IU per quart; vitamin D: 800 IU per quart) established by the Food and Drug

Administration.

Who Qualifies

This program is available only to institutions that do not participate in the National School Lunch, School Breakfast, or Commodity Programs.

How to Enroll

Contact the local school authority.

Summer Food Service Program for Children

What It Is

This program provides nutritious meals for preschool and school-aged children younger than 18 years in recreation centers or summer camps, during vacations in areas operating under a continuous school calendar, or in areas with poor economic conditions. Meals are served free and must meet the minimum standards established by the USDA.

Who Qualifies

Sponsors of the program must be public or private–nonprofit school food authorities; state, local municipal, or county governments; or public, private–nonprofit residential summer camps. The programs must be in areas where at least half of the children are from households with incomes at or below 185% of federal poverty guidelines; there is no income test for eligibility in these low-income areas, and any child in the program may participate. Other programs that contain at least half of children from families at or below 185% of the federal poverty guidelines may also provide food to all children in the program. Individual children in these programs may participate without regard to their family's income, and they may receive as many as two meals and two snacks per day. The USDA reimburses sponsors of the program.

How to Enroll

Contact the local school authority.

Women, Infants, and Children

What It Is

Since its inception in 1972, the WIC program grew from $20 million to more than $7 billion yearly in authorized funds. The USDA provides annual cash grants to state health departments or comparable agencies on

the basis of a formula that considers food costs and administrative costs. The program is very large, but it is not an entitlement program; rather, it is a grant program with funding limits that are set annually. Participation is an option for states and localities, but today it is available in virtually all parts of the country.

The WIC program differs from all other federal food assistance programs in its close association with health care services and inclusion of food, health, and nutrition education services together.[53,54] These services are defined as ongoing, routine pediatric and obstetric care. Health care itself, other than screening and limited assessment and care provided by WIC, must be supported by state, local public, and private–nonprofit health or human service agencies as well as the private sector. The amount and kind of health services provided to WIC recipients vary from state to state and locally.

The benefits of the program include checks or vouchers or EBT (all WIC state agencies are required to implement the EBT by October 2020) for the purchase of specific foods tailored to the dietary needs of infants, children, and pregnant or postpartum women. Physicians or health professionals may also prescribe special infant formulas and certain medical foods for WIC participants with special medical conditions. In addition to food, WIC also provides referrals to health and social services, including prenatal care, and nutrition counseling and education sessions. In addition, many pregnant women gain initial access to prenatal care via their WIC screening. Efforts are made to adapt the education activities to the individual participant's nutritional needs, cultural preferences, and education levels. Approximately 76% of the current participants are children ranging in age from newborn to 5 years.

Who Qualifies

Low-income pregnant and postpartum women, lactating mothers, infants, and children up to 5 years who are determined to be at nutritional risk by a health professional are eligible if they fall below 185% of federal poverty guidelines. Automatic eligibility is offered for anyone who participates or has family members participating in other benefit programs, such as SNAP and Medicaid. The criteria for nutritional risk include the following: dietary risk, anthropometric risk (e.g., overweight, underweight), biochemical risk (e.g., low hematocrit), and medical risk (e.g., diabetes mellitus) and other factors such as homelessness. A priority system is in place that categorizes participants on the basis of severity of potential effects and outcomes so that those at highest risk are served first.

Participants must be recertified every 6 months to continue receiving benefits.

How to Enroll

To obtain WIC benefits, an individual must apply at a WIC clinic.

WIC's Farmers' Market Nutrition Program

What It Is

This program is associated with WIC. It aims to provide fresh, nutritious and locally grown fruits and vegetables through farmers' markets and roadside stands to WIC participants. In addition to their regular WIC benefits, eligible WIC participants are given Farmers' Market Nutrition Program (FMNP) checks or coupons. These checks or coupons are used to buy eligible foods from farmers at farmers' markets and/or roadside stands that are approved by state agency to accept FMNP coupons. Nutrition education about how to select, store, and prepare fresh fruits and vegetables is provided to FMNP recipients by state agency through local WIC agency.

Who Qualifies

Women, infants over 4 months old, and children who are certified to receive WIC program benefits or who are on a waiting list for WIC certification are eligible.

How to Enroll

Contact your local WIC agency or your FMNP contact person for each state agency. A list of state contacts can be found at http://www.fns.usda.gov/wic/Contacts/farm.HTM.

Commodity Supplemental Food Program

What It Is

This in-kind program provides food and is available at local option in several states. Providing commodity supplemental foods and nutrition education to participants, the CSFP is designed to supplement their diets with nutritious foods. The foods supplied by CSFP are different from WIC foods in their variety (i.e., they consist of canned meat or poultry and canned vegetables). CSFP distributes food rather than vouchers for redemption at the grocery store. Also, the provision of health care to recipients is encouraged but not mandated, unless it was already in

existence prior to 1978. Simultaneous participation in WIC and CSFP is prohibited. The supplemental foods (usually surplus agricultural commodities) are distributed to state agencies, Indian tribes, or groups responsible for administering the program. Each state selects local, private, nonprofit agencies to administer the program at the local level. Local agencies determine the eligibility of applicants, distribute the supplemental foods, and provide nutrition education. Federal funds provide for some of the administrative costs such as outreach, warehousing of food, and transportation to obtain food if necessary.

Who Qualifies

Low-income pregnant, lactating, and postpartum women, infants, and children up to 6 years old are eligible. Women, infants, and children who are below 185% of federal poverty levels and *not* participating WIC are qualified.

How to Enroll

Contact your CSFP State Distributing Agency. A list of state contacts can be found at www.fns.usda.gov/fdd/contacts/sdacontacts.htm.

Expanded Food and Nutrition Education Program

What It Is

This program provides instruction in basic food and nutrition-related topics to low-income families and individuals and is operated through the state cooperative extension service. It is not an entitlement program. Its aim is to develop understanding and awareness through a variety of methods, including direct teaching by nutrition assistants in existing groups (such as employment and training programs and shelters for homeless families), cluster groups consisting of two to five participants, one-on-one home visits, mailings, telephone, and mass media efforts.

EFNEP teaches participants how to plan for daily food needs; prepare nutritious, low-cost meals that meet the daily recommended dietary allowances (RDAs) of nutrients; select and buy food economically; and effectively use other supplemental programs available, such as SNAP, WIC, School Meal Programs, and CSFP.

Who Qualifies

Families with children younger than 19 years, pregnant women, and individuals who are responsible for purchasing and preparing foods (such

as special needs adults, teens living independently) are eligible. To qualify, applicants must have income at or below 185% of federal poverty guidelines.

How to Enroll

Look for state or regional EFNEP contacts on http://www.extension.purdue.edu/cfs/topics/EFNEP/national_efnep/directo

Food Distribution Program on Indian Reservations

What It Is

This program provides commodities to low-income households on participating reservations and to Native American families living in designated areas near reservations. This is an alternative to the SNAP program for American Indians who do not have easy access to food stores. Participants receive a monthly food package containing a variety of foods to meet their health needs and preferences.

Who Qualifies

Program eligibility is based on a person's household income, assets, and residence on or near a reservation, or in Oklahoma (since most native Americans live in that state) that contain at least one person who is a member of a federally recognized tribe. Households may not participate in Food Distribution Program on Indian Reservations (FDPIR) and SNAP in the same month.

How to Enroll

Contact your local Indian Tribal Organization or the Food and Nutrition Service (FNS) Regional Office in your area for further assistance. The FDPIR contacts that administer this program can be found at http://www.fns.usda.gov/fdd/contacts/fdpir-contacts.htm.

The Emergency Food Assistance Program

What It Is

First started as a way to reduce inventories and storage costs of surplus commodities by distributing them to needy households, it now also includes funds appropriated by Congress to purchase additional commodities. The USDA buys the food, processes and packages it, and ships it to the states based on a formula that takes into account the number of people below the poverty level and the number unemployed.

Who Qualifies

States set their own eligibility criteria and select emergency feeding organizations such as soup kitchens, food recovery organizations, and food banks to distribute the food.

Fresh Fruit and Vegetable Program

What It Is

Fresh Fruit and Vegetable Program (FFVP) provides all students in selected elementary schools with healthy fresh fruits and vegetables of reduced price or free. The aim of this program is to introduce school children to a variety of produce and educate them with healthy eating habits. Established in 2002 as a pilot program, FFVP is now available in selected schools nationwide.

Who Qualifies

To be selected for the FFVP, a school must be an elementary school, represent the highest percentages of students certified for free and reduced price benefits, participate in the National School Lunch Program, and complete an annual application.

How to Enroll

Contact your state agency to obtain an application package.

DHHS Programs

The US DHHS provides a wide variety of public health programs at the community level, as well as payments for health services. The public health programs include the Head Start and EI programs. In addition, limited monies are provided for nutrition services such as assessment and counseling. Title V of the Social Security Act of 1935 authorized grants to states to plan for provision of comprehensive health services to mothers and children. In subsequent years, funding of categorical programs provided monies for planning and provision of special services, such as lead poisoning prevention, services for handicapped children, and family planning. Monies for planning such programs are provided from federal funds by the DHHS. Demonstration grant funds have also been available on an increasingly limited basis over the past few decades for high-risk communities and groups. The comprehensive services include medical and dental care, nursing, nutrition, and social work. The American Recovery and Reinvestment Act of 2009 (Recovery Act) provides Recovery Act

funds that aim to improve health and human services.[55]

Head Start

What It Is

This program is particularly well known for its multifaceted approach to child health and development, including social and educational development. Administered by DHHS and funded directly to local agencies, Head Start provides low-income preschoolers and their families with day care, access to medical care and social services, and nutrition services, including meals, nutrition education, and staff training.

Head Start programs are now funded through grants from the Recovery Act and Consolidated Appropriations Act of 2012. These grants are awarded to a variety of local public agencies, private–nonprofit organizations, and public school systems. The program is very large but not an entitlement. It is at the discretion of states and localities and serves primarily children from poor families.

Head Start programs serve a multiracial, multiethnic group of low-income children and families. Each program must offer four major components: education, social services, parent involvement, and health services. With the goal of meeting each child's individual needs, the program offers a wide variety of learning experiences aimed at fostering intellectual, social, and emotional growth. Head Start also aims to meet the ethnic and cultural characteristics of the communities served.

Head Start's nutrition program is part of the health component offered and provides nutritious meals from the Child Care Food Program, as well as nutrition education for children, staff, and parents.

Who Qualifies

To be eligible for Head Start, a child must be between 3 and 5 years of age and come from a low-income family whose total annual income does not exceed 100% of the federal poverty guideline or who is receiving public assistance from programs such as Temporary Assistance for Needy Families (TANF) or Social Security's Supplemental Security Income Program.

How to Enroll

Contact the office of Head Start is located in the Administration for Children and Families, DHHS.

Head Start for Children With Special Needs

What It Is

Head Start is the largest provider in this country of services for preschool children with handicapping conditions.

Who Qualifies

It is mandated to serve preschool children who have a broad range of handicaps. Children with severe or multiple handicapping conditions present additional challenges to Head Start staff in the planning and provision of individual services. Program requirements call for individualized plans for special education, treatment, and related services for all children with handicapping conditions served by Head Start. These plans must be based on the child's specific handicapping conditions, as well as the unique needs arising from those conditions.

How to Enroll

Contact the Office of Head Start, which is located in the Administration for Children, Youth, and Families, DHHS.

Early Intervention Programs and Services

What It Is and Who Qualifies

EI programs and services for infants, toddlers, and their families have existed in many areas of the United States for more than 25 years. These services are organized to address the developmental needs of young children between birth and 3 years of age who may be at risk for poor developmental outcome because of an identified disability or because of biologic or environmental risk factors.[56,57] Although all these programs share the common goal of optimal infant development, they approach it in different ways. That is why collaboration is essential in the development of an Individual Family Service Plan, which addresses all of the child's needs that must be met for the child's potential to be reached. Some EI programs are home-based models, others are center-based models, and others are a combination of those two approaches. Intervention programs also differ with respect to their program philosophy and goals, the population that is served, team composition, program content, and program evaluation. Services are generally multidisciplinary, in recognition of the interrelationship of development in various domains, and may include input by developmental educators, occupational and physical therapists, speech pathologists, psychologists, nurses, dietitians, and social workers.

Federal law significantly influences the development of EI programs. The most applicable law, enacted in 1986, is Title I, Part H, Handicapped Infants and Toddlers, of PL 99-457, Education of the Handicapped Amendments of 1986. That law extends public education to children aged 3 years and older and provides incentives to states to serve children with special needs from birth onward. It includes nutrition as one of eight disciplines to be included on the team providing services to these handicapped infants and their families.

Part H of PL 99-457 requires that services designed for children from birth through 36 months meet their developmental needs. It mandates psychological services, parent and family training and counseling, transition services, diagnostic medical services, case management, and health services that will enable the child to benefit from other EIs. The eight disciplines specified by PL 99-457 include special education, nursing, nutrition, psychology, physical therapy, occupational therapy, speech-language pathology, and social work. This law provides great opportunities for nutrition intervention in preschool children with handicapping conditions and their families. To date, however, most states have done little to develop community-wide networks to provide EI services that include nutrition.

How to Enroll

Visit the National Early Childhood Technical Assistance Center for more information about EI in your state: http://www.nectac.org/contact/ptccoord.asp.

"Let's Move" Child Care

What It Is

"Let's Move!" is an initiative launched by the First Lady, Michelle Obama in 2010. Its aim is to tackle the problems associated with childhood obesity through comprehensive approach. Let's Move Child Care is one of the programs under this initiative. There are five main goals in this program: (1) at least one hour of physical activity throughout the day; (2) no screen time for children younger than 2 years; (3) serve fruits and vegetables at every meal and provide no fried foods; (4) provide access to water during meals and no sugar-sweetened beverages; and (5) promote breastfeeding. Participants will receive certificates, updated resources on promoting children's health and be able to create an Action Plan to reach specific goals.

Who Qualifies

Anyone who is a parent or early care and education provider is eligible.

How to Enroll

To sign up, go to http://www.healthykidshealthyfuture.org/content/hkhf/home/startearly/sign

Maternity Group Homes for Pregnant and Parenting Youth Program

What It is

This program provides support for homeless pregnant and/or parenting young people, as well as their dependent children. Services provided include accommodations, life-building skills education, job assistance, education opportunities, health care and any related resources. The Family and Youth Services Bureau funds Maternity Group Homes for Pregnant and Parenting Youth Program as part of the Transitional Living Program, under the provisions of the Reconnecting Homeless Youth Act of 2008 (Public Law 110-378).

Who Qualifies

Homeless pregnant and/or young people aged between 16 and 22 years and their dependent children are eligible. Services are provided for dependent children up to 21 months or until a young person turns 18 years.

How to Enroll

Contact your regional grantee programs under Family and Youth Services Bureau at http://www.acf.hhs.gov/programs/fysb/resource/fysb-grantee-map.

STATE AND LOCAL PROGRAMS: THE UNITED STATES

There is a great deal of variability between states and localities in the extent to which they supplement federal nutrition funds or provide additional in-kind resources, such as health services. These variations make it essential for community nutritionists to know the specifics of state and local programs in their areas.

EXISTING PROGRAMS: CANADA

Federal Programs

Health Canada

Health Canada is the federal department responsible for the health of the Canadians. It offers evidence-based guidelines, research funding, and health promotion and education for citizens and residents in Canada. Table 16-17 summarizes the existing community nutrition programs for Canadian children under the regulation and monitoring of Health Canada.

Division of Childhood and Adolescence Under Public Health Agency of Canada

The Public Health Agency of Canada is another important institution in Canadian nutrition. It is the major federal public health agency which promotes and protects the health of Canadians through various public health measures. It provides relevant statistics on the incidence of child abuse and general trends related to child and youth health. Table 16-18 summarizes the community nutrition programs for Canadian children under the Public Health Agency of Canada.

Provincial and Community Programs

There are 10 provinces in Canada: Ontario, British Columbia, Manitoba, Saskatchewan, Alberta, Quebec, New Brunswick, Nova Scotia, Prince Edward Island, and Newfoundland and Labrador. Each province has jurisdiction power over many public goods, such as health care, education, welfare, and intraprovincial transportation. Canada also has three territories in addition to provinces: Northwest Territories, Yukon, and Nunavut. The territories have delegated powers directly under the Parliament of Canada (federal government). With different characteristics among the provinces and local communities, each has developed a different version of a similar program. In addition, each province may develop its own policies and guidelines. One example is the school nutrition program. Currently, there is no national school nutrition program in Canada.[58] Provincial government and organizations provide meals or funding to support giving nutritious food to Canadian children. Table 16-19 summarizes the provincial and community efforts on school foods.

WEB-BASED RESOURCES FOR COMMUNITY

NUTRITION: THE UNITED STATES

The US DHHS has developed a number of web-based resources that may be helpful in planning community nutrition efforts. Many nutrition resources not specified in this chapter are listed at http://www.nutrition.gov. The gateway to reliable consumer health information on the Internet is provided at http://www.healthfinder.gov. Healthy People 2020–related materials include a general description of the initiative, which is available at http://www.healthypeople.gov/2020/default.aspx. Information on TANF is available at http://www.neighborhoodlaw.org/aprimer.htm. The Healthy People 2020 Consortium Toolkit, which helps Consortium members in understanding, implementing, and marketing Healthy People, is available at http://healthypeople.gov/2020/consortium/hpConsortium.aspx.

TABLE 16-17 Community Nutrition Programs for Canadian Children Sponsored by Health Canada

Canada Prenatal Nutrition Program (in collaboration with Public Health Agency of Canada)	It is a community-based program that offers support to improve health and well-being of pregnant women, new mothers, and their babies. This program is run on provincial and territorial levels, which allows more flexibility and specificity for regional needs. Main objectives of the program include increasing the rate of breastfeeding initiation and decreasing the rate of unhealthy birth weights. Program details can be accessed at http://www.phac-aspc.gc.ca/hp-ps/dca-dea/prog-ini/cpnp-pcnp/index-eng.php.
Nutrition North Canada	This is a food subsidy program that aims to improve access of perishable healthy food to people living in isolated northern communities in the provinces. Country foods (traditional foods for the First Nations, Métis and Inuits) are also included in the subsidies. This program also funds retail and community-based nutrition education initiatives to increase knowledge and skills on selecting and preparing foods to promote healthy eating. For more information, go to their website at http://www.nutritionnorthcanada.ca/mr/fs_trns-eng.asp.
Canadian Hypertension Education Program	This program is run by Hypertension Canada. It is not a direct community nutrition program, but it targets educating health professionals in both clinical and community settings for treating hypertension, which in turn can educate the public. Education materials and evidenced-based guidelines are developed and can be accessed at http://www.hypertension.ca/education.

TABLE 16-18 Community Nutrition Programs for Canadian Children Directed by the Public Health Agency of Canada

Institution	Comments
Aboriginal Head Start in Urban and Northern Communities	This program focuses on the early development of children in the First Nations, Inuit and Métis. In terms of nutrition, the program ensures that children are provided with essential nutrients, fed at appropriate times, met the nutritional needs by the Aboriginal Food Guide, and educated about the nutritious and healthy eating habits. For more information, visit their website at http://www.phac-aspc.gc.ca/hp-ps/dca-dea/publications/ahsuni-papairun/index-eng.php#a17.
Community Action Program for Children	This program provides long-term funding to coalitions and groups that create programs that promote the healthy development of young children (<6 years old). It is managed at provincial and territorial levels to allow more flexibility. Local CAPC sites will provide locations for families to connect with their local communities and obtain information about access to public health and social services. These include family resources, parenting education, parent/child support groups, home visiting, nutrition education, and so forth. The website is at http://www.phac-aspc.gc.ca/hp-ps/dca-dea/prog-ini/capc-pace/index-eng.php.
Centre of Excellence for Children's Well Being	It provides resources, information, and tools to improve the health of children and youth in Canada. In addition, it collects and analyzes health information and data. It also conducts focus-oriented researches and generates education resources to the public. Moreover, it helps to build local, national, and international networks that involved in children's well-being. More resources can be found at http://www.phac-aspc.gc.ca/hp-ps/dca-dea/publications/centres-program/index-eng.php.
Fetal Alcohol Spectrum Disorder Initiative	This initiative aims to prevent fetal alcohol spectrum disorder and improve the outcomes for individuals with the condition and their families. It also contributes to the development of the Low-Risk Alcohol Drinking Guidelines, which can be found at http://www.ccsa.ca/eng/priorities/alcohol/canada-low-risk-alcohol-drinking-guidelines/Pages/default.aspx. More resources can be found at http://www.phac-aspc.gc.ca/hp-ps/dca-dea/prog-ini/fasd-etcaf/index-eng.php.
Pan-Canadian Joint Consortium School Health	It facilitates the development of knowledge, leadership, and capacity building that supports comprehensive school health. The website is at http://www.jcsh-cces.ca/.

TABLE 16-19 Provincial and Community Efforts on Canadian School Nutrition Program

Province or Community	Comments
Ontario	Ontario Student Nutrition Program has 14 regional and multiple local offices located throughout the province. The funding comes from the Ontario Ministry of Children and Youth Services and aims to provide students with breakfast, lunch, and snacks. Also, its guidelines are in alignment with the Canada's Food Guide.
British Columbia	School Meal and School Nutrition Programs include breakfast, lunch, and snack programs, which are funded under the British Columbia Ministry of Education.
Manitoba	Child Nutrition Council of Manitoba provides funding to schools for nourishment programs and education resources.
Saskatchewan	Multiple efforts are providing funding to schools in Saskatchewan. • Government of Saskatchewan offers school nutrition programs for children enrolled in Community schools. • Child Hunger and Education Program supports the provision of meals for children in community centers and schools.
Alberta	E4C provides School Lunch Program and School Snack Program to schools with demanding needs in the Edmonton area.
Quebec	Club des petit dejeuners du Quebec under Government of Quebec provides school breakfast to those who are enrolled in the Club.
New Brunswick	The Department of Education has the Healthy Minds School Nutrition Program which sets standards for school nutrition policies and guidelines for school lunch programs.
Nova Scotia	Nova Scotia Departments of Education, Health Promotion and Protection develops provincial breakfast program standards with Breakfast for Learning Canada and funds the school breakfast programs.
Prince Edward Island	Department of Health provided Prince Edward Island Eating Alliance with funding for school breakfast and snack programs for three years. PEI Eating Alliance has also developed school nutrition policies that are adopted province-wide.
Newfoundland and Labrador	Kids Eat Smart Foundation supports children with nutrition through Kids Eat Smart Clubs, which are nutrition programs run by volunteers at schools and community centers. Around 20,000 children participate in this program.
Breakfast for	It is a largest national charity which operates in all 10 provinces

Learning Canada	to support school breakfast, lunch, and snacks program. In 2011, it provided meals and snacks to over 400,000 children in Canada.
Breakfast Clubs of Canada	It is a nonprofit organization that offers services and funding to school breakfast programs in Canada. They also provide breakfast programs in the Aboriginal communities.

Several websites focus on physical activity and health. The President's Council on Fitness, Sports, and Nutrition is available at http://www.fitness.gov. Tips for promoting better health for young people through physical activity and sports are provided at http://www.cdc.gov/healthyyouth/npao/publications.htm. For obesity prevention, see http://www.cdc.gov/youthcampaign/. For Let's Move campaign, see http://www.letsmove.gov/.

The USDA provides several websites to improve access to food stamp and food program–related nutrition resources for assisting community nutrition effort.

All USDA websites are available at the National Agricultural Library's homepage: http:/www.nal.usda.gov. Some specific websites include the following:

- *The Food and Nutrition Information Center* is a large, web-based information center backed up with trained nutritionists who can help in locating information and borrowing additional resources from the collection of the National Agricultural Library. Access it at http://fnic.nal.usda.gov/.
- *The SNAP-Ed Connection* is an online resource center for state and local SNAP-Ed providers. It provides professional resources for training and continuing education for educators and offers educational materials developed for SNAP participants. These materials are available at http://snap.nal.usda.gov/.
- The *WIC* program has a website that provides publications on recent studies and educational materials. It is located at http://www.fns.usda.gov/wic/publications.htm.
- *The Healthy School Meals Resource System* is an online resource center for Team Nutrition, which is an initiative of the USDA Food and Nutrition Service to support child nutrition programs through training and technical assistance for personnel working in food service, nutrition education, and school and community health areas. It offers training and educational materials for these personnel. Access it at http://healthymeals.nal.usda.gov/.

- *The Dietary Guidelines for Americans* are available at http://health.gov/dietaryguidelines/. Possible implications for US agriculture from adopting the Dietary Guidelines can be assessed at http://www.ers.usda.gov/media/870329/err31_reportsummary.pdf.
- *CDC's Communities Putting Prevention to Work* has an online resource center for different areas of preventative health work, including tobacco, obesity, nutrition, physical activity, schools, and foundational skills: http://www.cdc.gov/CommunitiesPuttingPreventiontoWork/resources/ir
- *Public Health Workforce* has an online database for nutrition-related resources. It can be assessed at http://phpartners.org/nutrition.html.

RESOURCES FOR COMMUNITY NUTRITION: CANADA

Table 16-20 summarizes the current food and nutrition guidelines in Canada. They include food guides based on nutrient standards and are available on the Internet. Table 16-21 provides some community nutrition resources for Canadian children, including the websites for the major agencies and journals.

LESSONS LEARNED FROM COMMUNITY NUTRITION EFFORTS

Community Investments in Child Health and Nutrition Make Sense

As early as the fourth century BC, the philosopher Plato stressed the importance of investing in children from an early age. Several millennia later, numerous studies confirm Plato's suppositions about the importance of investing in our children.[59] For example, supplementing the diets of pregnant women and infants and immunizing children against early childhood diseases save lives and improve health. It is to be hoped that, in the future, programs such as Head Start, EI, and WIC, with demonstrated success in strengthening the bodies and minds of children, will reach more of their target populations. Also, special attention must be paid to nutrition for children with special needs and other chronic health problems. Current programs do not cover them adequately.

TABLE 16-20 Current Food and Nutrition Guidelines in Canada	
Publication	**Comments**
Canada's Food Guide (Health Canada)	Canada's Food Guide is the most recent guidelines for food and nutrition intake, which can be accessible at http://www.hc-sc.gc.ca/fn-an/food-guide-aliment/context/evid-fond-eng.php. The food guide's goal is to assist the people of Canada in making food choices that promote health and reduce the risk of nutrition-related chronic disease. Development of the food intake pattern was built on findings from reviewing previous Food Guides. The nutrient standards and assessment methods are based on Health Canada/US IOM DRI reports.
Eating Well with Canada's Food Guide—First Nations, Inuit and Métis (Health Canada)	Based on Canada's Food Guide, this tailored food guide is for the First Nations, Inuit and Métis and includes both traditional and store-bought foods. It shows pictures of store-bought foods that are generally available in rural and remote locations and provides unique images and content developed for Aboriginal populations. The Eating Well with Canada's Food Guide—First Nations, Inuit and Métis can be accessed at http://www.hc-sc.gc.ca/fn-an/alt_formats/fnihb-dgspni/pdf/pubs/fnim-pnim/2007_fnim-pnim_food-guide-aliment-eng.pdf.
DRI (Health Canada in collaboration with IOM)	The DRI contains reference nutrient values that are the basis of food and nutrition guidelines in both the United States and Canada. These reference values are specific for age, gender and life stages. The DRI include adequate intake amounts (RDA) and UL for different nutrients and can be accessed at http://www.iom.edu/Activities/Nutrition/SummaryDRIs/DRI-Tables.aspx.
Nutrition for Healthy Term Infants: Recommendations from Birth to 6 Months	The guidelines offer evidence-based recommendations for health care professionals on infant nutrition from birth to 6 months. Breastfeeding and having adequate amounts of specific nutrients are emphasized. The recommendations can be accessed at http://www.hc-sc.gc.ca/fn-an/nutrition/infant-nourisson/recom/index-eng.php.

Abbreviation: DRI, Dietary Reference Intakes; UL, upper levels

Table 16-15 shows current federal spending for a number of key health and food programs involving children.

Community Health and Children's Health Care Must Be Equal Partners

Community health and nutrition efforts expand the traditional focus of direct health services. Both are necessary to ensure child and family health. Also, broader economic factors that affect food insecurity must be

dealt with.[60] The distinctions between traditional public health and personal health services are slowly blurring in this country. In the future, community nutrition concerns will need to be incorporated into both systems. There is the danger that nutrition will "fall through the cracks" because there are other pressing needs. Thus, it behooves all of those who are interested in the health of children to pay attention to ensure that federal, state, and local governments employ nutritionists to look after community needs in nutrition. These professionals must work with others in health, welfare, education, and food agencies in the public and private sectors to ensure that all children are well nourished. At the same time, dietary concerns must be increasingly incorporated into personal health services. Only when these steps are taken will community nutrition realize its potential to have a positive impact on children.

TABLE 16-21 Community Nutrition Resources for Canadian Children

Public Health Agency of Canada	It promotes and protects the health of Canadians through various publi and provides relevant statistics on the incidence of child abuse and ger to child and youth health. More specific information can be found, unde Health" at http://www.phac-aspc.gc.ca/hp-ps/index-eng.php#ch.
Health Canada	Health Canada's website provides specific links to Canada's Food Guic information about Canadians' food consumption and nutrient intake, gu nutrition labeling, and promotional information on infant feeding. Some follows: • Canada's Food Guide: http://www.hc-sc.gc.ca/fn-an/food-gu eng.php • Food and Nutrition Surveillance: http://www.hc-sc.gc.ca/fr eng.php • Nutrition Labeling: http://www.hc-sc.gc.ca/fn-an/label-etiqu eng.php • Infant feeding: http://www.hc-sc.gc.ca/fn-an/nutrition/infan eng.php
Canadian Journal of Public Health	Published by the Canadian Public Health Association, this is a professi reviewed journal carrying articles related to various components of pub epidemiology, nutrition, family health, public policy, etc. This can be ac http://journal.cpha.ca/index.php/cjph.
Dietitians of Canada (DC)	The national professional association for dietitians in Canada. Nutrition educational materials for the public are available on its website. There i on the website for Registered Dietitians in Canada which provides reso continued education, literature reviews, practice guidelines, and career This is accessible at http://www.dietitians.ca/.

Provincial Ministries of Health	Each province in Canada has its own Ministry of Health (the title of the differ) with the aim of establishing a sustainable provincial health syste legislation and policies and monitoring the health of provincial residents summarizing the programs and resources in the province. Websites for Health for each province are as follows: • Ontario: http://www.health.gov.on.ca/en/ • British Columbia: http://www.gov.bc.ca/health/ • Manitoba: http://www.gov.mb.ca/health/ • Saskatchewan: http://www.health.gov.sk.ca/ • Alberta: http://www.health.alberta.ca/ • Quebec: http://www.msss.gouv.qc.ca/en/ • New Brunswick: http://www.gnb.ca/0051/index-e.asp • Nova Scotia: http://novascotia.ca/dhw/ • Prince Edward Island: http://www.gov.pe.ca/health/ • Newfoundland and Labrador: http://www.health.gov.nl.ca/health
Canadian Paediatric Society	A national association of pediatricians in Canada with the mission of pr promoting the health and well-being of children and youth. It issues pra and clinical tools and runs several programs—some along with the fede which include Neonatal Resuscitation Program, Canadian Paediatric Su Program, and IMPACT (a vaccine surveillance program). For more info website http://www.cps.ca/en/.
Canadian Diabetes Association	This association helps patients with diabetes living healthy lives and res related issues. It provides basic information about diabetes, manageme nutrition information for diabetes patients. It also updates clinical guidel years to provide current evidence-based practice data for health care p recently the 2008 Clinical Practice Guidelines, http://www.diabetes.ca/f professionals/resources/2008-cpg/). More information is on the followin http://www.diabetes.ca/.
Heart and Stroke Foundation of Canada	The Foundation is a federation of 10 provincial Foundations, which aim reduce impacts from heart disease and stroke through research, promo living and advocacy. Its “Healthy Kids” program within the organization healthy eating and activity for families with children and youth. More inf found at http://www.heartandstroke.com/site/c.ikIQLcMWJtE/b.3479025/k.802B/ It provides general information for heart disease and stroke, and curren related issues. The website is at http://www.heartandstroke.com/site/c.ikIQLcMWJtE/b.2796497/k.BF8B

Child Nutrition and Physical Activity Are Inextricably Linked, Like Siamese Twins

According to the World Health Organization, both physical activity and sound nutrition in the community help to keep children healthy and ready to learn and also prevent later chronic disease risk.[61] Long-term

community efforts at the school, family, and individual levels are necessary to yield the benefits of lower health risks over the long term. Community leaders, experts in health and education, and parents themselves must be involved in school-based and other prevention-oriented programs.

School-Based Programs Offer Much Potential for Health Promotion and Primary Prevention of Diet- and Physical Activity–Related Risk Factors if Supplemented by Parent, Family, and Community-Wide Efforts

School-based programs are excellent sites for these efforts. However, changes in the curriculum alone are not sufficient to bring about alterations in risk factors for cardiovascular disease or obesity; changes in school meals and alterations in physical education programs are also needed. More attention to physical training components that increase strength may be important in preventing later osteoporosis risk. Thus, multiple risk factors need to be targeted. Also, community efforts to reinforce these efforts are needed.[62]

In addition to school-based efforts, family and community efforts are also required. Children do not spend all of their waking hours in school. The microenvironment of the family is an important milieu for enhancing and carrying forward school-based health promotion and physical education efforts. The macroenvironment of schools and the community must also be conducive to healthy lifestyles.[63,64]

Healthful Eating Patterns for the Whole Family Are Critical

Foods must be eaten to be nutritious, and overall diets are the most important. Children and their families eat foods, not individual nutrients. There is often a gap in teaching about the merits and demerits of individual foods, and broader guidance on healthful diets or eating patterns is needed. Existing scientific evidence shows that overall eating patterns and diets and not consumption of individual foods or nutrients are most closely linked to positive effects on health. Therefore, families need information about healthful, enjoyable eating patterns that provide a broad, food-based context for healthful eating and about healthful physical activity and exercise patterns. When needs cannot be met by food-based solutions alone, supplementation with vitamins and minerals may be considered.

Information on Healthful Eating and Physical Activity Must Be Communicated More Effectively at the Community Level

Consumers and policy makers must view the Dietary Guidelines for Americans and other nutrition education guidance and programs as acceptable and actionable as well as authoritative if they are to be used. The Dietary Guidelines, the USDA's MyPlate (which is based on the Dietary Reference Intakes and the Dietary Guidelines), food labels, and health claims provide such guidance to the community.[65–67] Information about them needs to be communicated, coordinated, and harmonized to facilitate food purchases and consumption decisions, and diet-related efforts must be accompanied and reinforced by physical activity programs. This challenge is one that food producers, marketers, public policy makers, and educators face together. Intervention strategies from the social and behavioral sciences may be helpful in tailoring messages to our diverse population.[68]

CONCLUSION

Community efforts in the United States and Canada are continuing to increase healthy life expectancy, ensure universal access to quality health care, achieve greater equality in health status, and prevent acute and chronic diseases through public health measures. With the joint efforts of federal and state or provincial governments and with the involvement of nongovernmental organizations, private sectors, and global organizations, these goals can be achieved gradually.

REFERENCES

1. Federal Interagency Forum on Child and Family Statistics. *America's Children in Brief: Key National Indicators of Well-Being, 2012–Demographic Background*. http://www.childstats.gov/americaschildren/demo.asp. Updated 2012. Accessed November 12, 2012.
2. Frazao E. *America's Eating Habits: Changes and Consequences*. Washington, DC: Economic Research Service, US Department of Agriculture; 1999.
3. Styne, DM. Clinical Management of Pediatric Obesity: An Overview. In: Sothern MS, Gordon ST, Kristian von Almen T, eds. *Handbook of Pediatric Obesity*. 6th ed. Boca Raton, FL: CRC Press; 2006:1–5.
4. Rogers CC. A look at America's children and their families. *Food Rev.* 2001;4(2):2–7.
5. Statistics Canada. *Canada at a Glance 2012–Populaton*. http://www.statcan.gc.ca/pub/12–581-x/2012000/pop-eng.htm#t01. Updated April

2012. Assessed November 12, 2012.
6. Statistics Canada. *The Canadian Population in 2011: Age and Sex–Figure 1, Number of Children Aged 14 and Under and of People Aged 65 and Over, Canada, 1921 to 2011*. http://www12.statcan.gc.ca/census-recensement/2011/as-sa/98–311-x/2011001/fig/fig1-eng.cfm. Updated May 2012. Accessed November 12, 2012.
7. The World Bank. *Population Ages 0–14 (% of total)–Canada*. http://data.worldbank.org/indicator/SP.POP.0014.TO.ZS. Updated 2012. Accessed November 12, 2012.
8. Statistics Canada. *The Canadian Population in 2011: Age and Sex*. http://www12.statcan.gc.ca/census-recensement/2011/as-sa/98–311-x/98–311-x2011001-eng.cfm. Updated May 2012. Accessed November 12, 2012.
9. Statistics Canada. *2006 Census: Aboriginal Peoples*. http://www12.statcan.gc.ca/census-recensement/2006/rt-td/ap-pa-eng.cfm. Updated April 2012. Accessed November 25, 2012.
10. Statistics Canada. *2006 Census: Ethnic Origin and Visible Minorities*. http://www12.statcan.gc.ca/census-recensement/2006/rt-td/eth-eng.cfm. Updated May 2012. Accessed November 25, 2012.
11. Winslow CEA. The untilled field of public health. *Mod Med.* 1920;2:183–191.
12. Kaufman M. Understanding public health. In: Kaufman M, ed. *Nutrition in Public Health.* Rockville, MD: Aspen; 1990:3–13.
13. US Department of Health and Human Service. *Topics & Objectives Index–Healthy People*. http://www.healthypeople.gov/2020/topicsobjectives2020/default.aspx. Updated August 2012. Accessed December 31, 2012.
14. US Department of Health and Human Services. *Maternal and Child Health Bureau*. http://www.mchb.hrsa.gov/timeline/. Updated 2012. Accessed December 26, 2012.
15. CORE Group, Nutrition Working Group. *Nutrition Program Design Assistant: A Tool for Program Planners*. Washington, DC; 2010. http://www.coregroup.org/storage/documents/Workingpapers/NPDA_RefGuide_w Accessed December 26, 2012.
16. US Department of Health and Human Services. *Maternal and Child Health Bureau. The National Survey of Children's Health 2007*. Rockville, MD: US Department of Health and Human Services; 2009. http://mchb.hrsa.gov/nsch/07main/index.html. Accessed December 26, 2012.
17. Institute of Medicine, Board of Health Care Services. *America's Uninsured Crisis: Consequences for Health and Health Care*. Washington, DC: The National Academies Press; 2009. http://books.nap.edu/openbook.php?record_id=12511. Accessed December 26, 2012.
18. US Department of Health and Human Services, Agency for Healthcare Research and Quality. *Improving Health Care Quality.* http://www.ahrq.gov/news/qualfact.htm. Updated September 2002. Accessed December 26, 2012.
19. Canadian Museum of Civilization. *Making Medicare: The History of Health Care in Canada, 1914–2007*. http://www.civilization.ca/cmc/exhibitions/hist/medicare/medic01e.shtml. Updated April 2010. Accessed on November 27, 2012.
20. Health Canada. *Canada's Health Care System (Medicare)*. http://www.hc-sc.gc.ca/hcs-sss/medi-assur/index-eng.php. Updated December 2010. Accessed December 27, 2012.

21. Gallagher-Allred C. *Implementing Nutrition Screening and Intervention Strategies*. Washington, DC: The Nutrition Screening Initiative; 1993.
22. Coleman-Jensen A, Nord M, Andrews M, Carlson S. *Household Food Security in the United States in 2011*. Washington, DC: US Department of Agriculture, Economic Research Service; 2012. http://www.ers.usda.gov/media/884525/err141.pdf. Accessed December 26, 2012.
23. Guenther PM, Reedy J, Krebs-Smith SM, et al. *Development and Evaluation of the Healthy Eating Index-2005: Technical Report*. Washington, DC: Center for Nutrition Policy and Promotion, U.S. Department of Agriculture; 2007. http://www.cnpp.usda.gov/HealthyEatingIndex.htm. Accessed December 26, 2012.
24. U.S. Department of Agriculture, Office of Research, Nutrition, and Analysis. *School Nutrition Dietary Assessment Study-III: Summary of Findings*. Washington, DC: Food and Nutrition Service, U.S. Department of Agriculture; 2007. http://www.fns.usda.gov/ora/menu/Published/CNP/FILES/SNDAIII-SummaryofFindings.pdf. Accessed December 26, 2012.
25. Briefel RR, McDowell MA. Nutrition monitoring in the United States. In: Erdman JW, Macdonald IA, Zeisel SH, eds. *Present Knowledge in Nutrition.* 10th ed. Washington, DC: ILSI Press and Wiley-Blackwell; 2012:1082–1109.
26. Nnakwe N. *Chapter 12: Principles of Planning Effective Community Nutrition Programs*. *Community Nutrition: Planning Health Promotion and Disease Prevention.* 2nd ed. Sudbury, MA: Jones & Bartlett Publishing; 2012.
27. US Department of Health and Human Services, Agency for Healthcare Research and Quality. *Guide to Clinical Preventive Services, 2012*. http://www.ahrq.gov/clinic/pocketgd.htm. Updated October 2012. Accessed on December 26, 2012.
28. US Department of Health and Human Services. *Implementing Healthy People 2020–Track.* http://www.healthypeople.gov/2020/implementing/track.aspx. Updated November 2011. Accessed on December 27, 2012.
29. US Department of Health and Human Services. *What's New For 2020*. http://www.healthypeople.gov/2020/about/new2020.aspx. Updated December 2012. Accessed October 8, 2012.
30. Allen LH. Maternal nutrient metabolism and requirements in pregnancy and lactation. In: Erdman JW, MacDonald IA, Zeidel SH, eds. *Present Knowledge in Nutrition.* 10th ed. Washington, DC: ILSI Press and Wiley-Blackwell; 2012:608–623.
31. US Department of Health and Human Services. *2020 Topics and Objectives*. http://www.healthypeople.gov/2020/topicsobjectives2020/overview.aspx?topicid=26. Updated August 2012. Accessed September 9, 2012.
32. The National Early Childhood Technical Assistance Center. *The Importance of Early Intervention for Infants and Toddlers With Disabilities and Their Families.* http://www.nectac.org/~pdfs/pubs/importanceofearlyintervention.pdf. Updated July 2011. Accessed December 27, 2012.
33. Heird WC. Infant nutrition. In: Erdman JW, MacDonald IA, Zeidel SH, eds. *Present Knowledge in Nutrition*. 10th ed. Washington, DC: ILSI Press and Wiley-Blackwell; 2012:624–636.
34. Centers for Disease Control and Prevention. *School Health Guidelines to Promote Healthy Eating and Physical Activity*. Morbidity and Mortality Weekly Report, Vol. 60. No. 5. Atlanta, GA: Centers for Disease Control and Prevention; 2011. http://www.cdc.gov/mmwr/pdf/rr/rr6005.pdf. Accessed October 15, 2012.
35. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in

body mass index among U.S. children and adolescents, 1999–2010. *JAMA.* 2012;307(5):483–490.
36. Executive Office of the President of the United States, White House Task Force on Childhood Obesity. *Solving the Problem of Childhood Obesity Within a Generation.* http://www.letsmove.gov/sites/letsmove.gov/files/TaskForce_on_Childhood_Obes Updated May 2010. Accessed December 27, 2012.
37. Kaiser Family Foundation. *Generation M2: Media in the Lives of 8- to 18-Years-Olds*. http://www.kff.org/entmedia/upload/8010.pdf. Updated January 2010. Accessed December 30, 2012.
38. Kuczmarski RJ, Ogden CL, Guo SS, et al. *CDC Growth Charts for the United States: Methods and Development*. Vital Health Statistics Series 11, No. 246. Atlanta, GA: National Center for Health Statistics; 2002.
39. Maqbool A, Dougherty KA, Parks EP, Stallings VA. Adolescence. In: Erdman JW, MacDonald IA, Zeidel SH, eds. *Present Knowledge in Nutrition.* 10th ed. Washington, DC: ILSI Press and Wiley-Blackwell; 2012:637–653.
40. American Academy of Orthopaedic Surgeons. *Early Prevention of Osteoporosis*. http://orthoinfo.aaos.org/topic.cfm?topic=a00330. Updated July 2007. Accessed December 27, 2012.
41. Lytle LA, Kubik MY. Nutritional issues for adolescents. *Best Pract Res Clin Endocrinol Metab*. 2003;17(2):177–189.
42. Coughlin JW, Guarda AS. Behavioral disorders affecting food intake: eating disorders and other psychiatric conditions. In: Shils ME, Olson JA, Shike M, et al. eds. *Modern Nutrition in Health and Disease.* 10th ed. Baltimore, MD: Williams & Wilkins; 2005:1353–1361.
43. Wilson GT. The Controversy over Dieting. In: Fairburn CG, Brownell KD, eds. *Eating Disorders and Obesity: A Comprehensive Handbook.* 2nd ed. New York, NY: Guilford Press; 2005:93–97.
44. Deckelbaum RJ, Williams CL. Childhood obesity: the health issue. *Obesity.* 2001;9(S11):239S-243S.
45. Montgomery KS. Improving nutrition in pregnant adolescents: recommendations for clinical practitioners. *J Perinat Educ.* 2003;12(2):22–30.
46. KidsHealth. *Having a Healthy Pregnancy*. http://kidshealth.org/teen/your_mind/emotions/pregnancy.html. Updated February 2012. Accessed December 27, 2012.
47. Health Canada. *Do Canadian Children Meet Their Nutrient Requirements Through Food Intake Alone?* http://www.hc-sc.gc.ca/fn-an/alt_formats/pdf/surveill/nutrition/commun/art-nutr-child-enf-eng.pdf. Updated January 2012. Accessed November 16, 2012.
48. Academy of Nutrition and Dietetics (Formerly Known as American Dietetic Association). Position of the American Dietetic Association: providing nutrition services for people with developmental disabilities and special health care needs. *J Am Diet Assoc.* 2010;110:296–307
49. Washington State Department of Health. Yang Y, Lucas B, Feucht S, eds. *Nutrition Interventions for Children With Special Health Care Needs.* 3rd ed. Olympia, WA: Washington State Department of Health; 2010. http://here.doh.wa.gov/materials/nutrition-interventions/15_CSHCN-NI_E10L.pdf. Accessed December 26, 2012.
50. Academy of Nutrition and Dietetics (Formerly Known as American Dietetic Association). Lucas B, ed. *Children With Special Health Care Needs: Nutrition Care Handbook.* Chicago, IL: American Dietetic Association; 2004.

51. Statistics Canada. *Participation and Activity Limitation Survey 2006: Families of Children With Disabilities in Canada*. http://www.statcan.gc.ca/pub/89–628-x/89–628-x2008009-eng.htm. Updated November 2008. Accessed on November 11, 2012.
52. Oliveira V. *The Food Assistance Landscape: FY 2011 Annual Report. Economic Information Bulletin.* No. 93. Washington, DC: US Department of Agriculture, Economic Research Service; 2012.
53. Institute of Medicine. *WIC Nutrition Risk Criteria: A Scientific Assessment.* Washington, DC: The National Academies Press; 1996.
54. Institute of Medicine. *Dietary Risk Assessment in the WIC Program.* Washington, DC: The National Academies Press; 2002.
55. US Department of Health and Human Services. *Head Start Recovery Act Implementation Plans.* http://www.hhs.gov/recovery/reports/plans/pdf20100610/ACF%20Head%20Start% Updated June 2010. Accessed September 30, 2012.
56. Schuman A, Benham R. Early childhood intervention: a nation-wide resource. *Semin Pediatr Gastroenterol Nutr*. 1993;4(1):8–11.
57. Taft Bayerl C, Ries JD, Bettencourt MF, Fisher P. Nutritional issues of children in early intervention programs: primary care team approach. *Semin Pediatr Gastroenterol Nutr*. 1993;4(1):11–15.
58. Breakfast for Learning Canada. *Canada's Children Need a National Nutrition Program*. http://www.breakfastforlearning.ca/en/how-to-help/canadas-children-need-a-national-nutrition-program. Updated 2012. Accessed November 12, 2012.
59. Healy T, Cote S, Helliwell JF, Held S. *The Well-Being of Nations: The Role of Human and Social Capital.* Paris, France: Organization for Economic Cooperation and Development; 2001.
60. Pelletier DL, Olson CM, Frongillo EA. Food insecurity, hunger and undernutrition. In: Erdman JW, MacDonald IA, Zeidel SH, eds. *Present Knowledge in Nutrition.* 10th ed. Washington, DC: ILSI Press and Wiley-Blackwell; 2012:1165–1181.
61. World Health Organization. *Global Strategy on Diet, Physical Activity and Health: Physical Activity and Young People*. http://www.who.int/dietphysicalactivity/factsheet_young_people/en/index.html. Updated 2012. Accessed December 27, 2012.
62. Institute of Medicine, Board on Children, Youth and Families. *Children's Health, The Nation's Wealth: A ssessing and Improving Child Health.* Washington, DC: The National Academies Press; 2004.
63. Patrick H, Nicklas TA. A review of family and social determinants of children's eating patterns and diet quality. *J Am Coll Nutr*. 2005;24(2):83–92.
64. Bauer KW, Berge JM, Neumark-Sztainer D. The importance of families to adolescents' physical activity and dietary intake. *Adolesc Med State Art Rev.* 2011;22(3):601–613, xiii.
65. US Department of Agriculture, US Department of Health and Human Services. *Dietary Guidelines for Americans, 2010*. 7th ed. Washington, DC: U.S. Government Printing Office; 2010.
66. US Department of Agriculture. *MyPlate*. Washington, DC: US Department of Agriculture, Center for Nutrition Policy and Promotion; 2011.
67. Murphy SP. Nutrition guidelines to maintain health. In: Coulston AM, Rock CL, Monsen ER, eds. *Nutrition in the Prevention and Treatment of Disease.* 2nd ed. San Diego, CA: The Academic Press; 2008:205–221.
68. Center for the Study of Social Policy. *Results-Based Public Policy Strategies for*

Promoting Children's Social, Emotional and Behavioral Health. http://www.cssp.org/policy/papers/Promote-Childrens-Social-Emotional-and-Behavioral-Health.pdf. Updated March 2012. Accessed December 27, 2012.
69. Health Canada. *Canada Health Act*. http://www.hc-sc.gc.ca/hcs-sss/medi-assur/cha-lcs/index-eng.php. Updated April 2012. Accessed November 12, 2012.
70. Statistics Canada. *Body Mass Index of Canadian Children and Youth, 2009 to 2011*. Catalogue No. 82–625-X. http://www.statcan.gc.ca/pub/82–625-x/2012001/article/11712-eng.pdf. Updated September 2012. Accessed November 12, 2012.
71. Shields M. Overweight and obesity among children and youth. Statistics Canada. *Health Rep*. 2006;17(3). http://www.statcan.gc.ca/pub/82–003-x/2005003/article/9277-eng.pdf. Accessed November 12, 2012.
72. Public Health Agency of Canada. *Curbing Childhood Obesity: A Federal, Provincial and Territorial Framework for Action to Promote Healthy Weights*. http://www.phac-aspc.gc.ca/hp-ps/hl-mvs/framework-cadre/2011/overview-resume-eng.php. Updated November 2011. Accessed November 25, 2012.
73. Statistics Canada. *Fruit and Vegetable Consumption, 2011*. http://www.statcan.gc.ca/pub/82–625-x/2012001/article/11661-eng.htm. Updated June 2012. Accessed November 25, 2012.
74. Colley RC, Garriguet D, Janssen I, et al. Physical activity of Canadian children and youth: accelerometer results from the 2007 to 2009 Canadian Health Measures Survey. Statistics Canada. *Health Rep*. 2011;22(1). http://www.statcan.gc.ca/pub/82–003-x/2011001/article/11397-eng.pdf. Accessed November 25, 2012.
75. Kirkpatrick SI, Tarasuk V. Food insecurity is associated with nutrient inadequacies among Canadian adults and adolescents. *J Nutr*. 2008;138(3):604–612.
76. Marchand V; Canadian Paediatric Society, Nutrition and Gastroenterology Committee. Nutrition in neurologically impaired children. *Paediatr Child Health*. 2009;14(6):395–401.

Nutritional Epidemiology

17

Carine M. Lenders, MD, MS, ScD, Emily Keefe, RN, MSN, FNP, and Walter C. Willett, MD, DrPH

Nutritional epidemiology is the study of relationships between diet and disease. Although the effects of nutritional deficiencies have been recognized for centuries, the chemical structures of compounds responsible for these deficiencies have been determined only the past 100 years or so. Globally, the four most commonly recognized deficiencies in children include protein–energy malnutrition and iron, iodine, and vitamin A deficiencies. The importance of folate deficiency has also become apparent in the past two decades. Interventions have evolved beyond the treatment of deficiencies to the reduction of disease risk (e.g., food fortification with iodine, iron, and folate; zinc and vitamin A supplementation programs; and breastfeeding campaigns). More recently, the relationship between nutrition and disease has been expanded from that of deficiency to that of excess; the concept of *nutrition transition* has been developed. Countries undergoing modernization and westernization are faced with diseases associated with both undernutrition and overnutrition. Globally, overnutrition and related complications have become the most important nutritional problem.

In contrast to those of most adults, children's nutritional needs vary with time as they grow and mature; therefore, assessment of pediatric diet, nutrition, and associated diseases is more challenging than that of adults. The dietary choices of children depend on their food preferences and parental choice and control, food availability and cost, peer pressure, and advertising. Young children's ability to report their dietary intake is limited; thus, dietary assessment often requires a surrogate reporter. Despite these differences in approaches to assessment, current national

surveys conducted in the United States have shown overall similar dietary trends in adults and children aged 2–18 years.

The development of techniques to assess diet and breastfeeding, the refinement of anthropometric methods, and the identification of biochemical indicators of diet (biomarkers) specific to population groups have greatly facilitated the field of nutritional epidemiology. Information from basic research has been used at the population level, and new questions in epidemiologic studies have been brought to the laboratory bench and the clinical setting. The methods used to investigate the relationships among diet, nutrition, and diseases include descriptive, analytic, and experimental studies. The development of hypotheses often comes from ecologic observations, and prevalence rates and diet–biochemical relationships are typically based on cross-sectional studies. The assessment of etiologic relationships is usually performed using case-control studies and prospective studies, which are typically expensive and of long duration. Each type of study has its own benefits and pitfalls. Although the best evidence for causation comes from experimental studies such as randomized, double-blind trials, in many situations, it is not feasible to randomly assign subjects to diet for long periods of time.

Modern epidemiology suggests a potential interactive association between lifestyle, genetics, and the risk of many chronic diseases. In this updated overview of nutritional epidemiology, we briefly describe issues and approaches for assessing dietary intake, nutritional status, and physical activity, the application of different study designs, and considerations in the analysis and interpretation of data. Because of its vital importance to child health, we use the relationship between folate intake and incidence of neural tube defects (NTDs) to illustrate various approaches in nutritional epidemiology.

ASSESSMENT OF DIETARY INTAKE

Diet exposure can be measured either in terms of foods or as nutrient and nonnutrient components. The power to detect an association between diet and disease is usually greater when using nutrients (from all foods) rather than one food or food group to represent diet, and interpretation of associations is easier to explain biologically. However, food analyses are used frequently because people eat foods, not nutrients, and food can reflect the combined effects of multiple nutrients and differences in bioavailability of nutrients. Each dietary representation has its benefits and weaknesses, and the use of both methods provides the greatest insight.

An association between a food and disease can lead to the identification of a substance that is responsible for disease or its prevention. A classic example is that of scurvy. As recorded by Hippocrates, symptoms of scurvy were already recognized in ancient Egypt, Greece, and Rome. Later, a tree extract provided by North American natives cured scurvy among members of an expedition led by Cartier in the sixteenth century. Lind, in his *Treatise of the Scurvy* (1753),[1] documented that lemons and oranges also cured subjects with scurvy. However, the relationship between scurvy and vitamin C deficiency was made much later. Celiac disease provides another example.[2] Although descriptions of symptoms compatible with celiac disease were recorded in antiquity, the classic symptoms of celiac disease were defined last century to include chronic diarrhea, abdominal distention, and poor weight gain several months after the introduction of solid foods. The relationship between certain types of cereals and celiac disease was determined when the typical symptoms of pediatric celiac disease disappeared during the food shortages of World War II and relapsed after cereal supplies were restored.[3] Gluten, the major component of cereal proteins, was implicated.[4]

Intake of nutrients or foods is typically interrelated, which can result in difficulties in interpreting findings. For example, an inverse relationship exists between dietary fat and fiber intake among children.[5] Therefore, associations between food and disease could be attributable to foods either high in fiber or low in fat, or both. Sometimes multivariate analysis can be used to disentangle these associations. To minimize interrelations, investigators sometimes use food groups or account for the contributions of nutrient intake from various food groups. Also, "pattern analysis,"[6] based on factor or cluster analysis, groups together subjects with similar intakes of food. These patterns can then be evaluated in relation to disease risk.

DIETARY ASSESSMENT METHODS

Methods to measure dietary intake include 24-hour recall, food records, food frequency questionnaires (FFQs), diet histories, and observation (Table 17-1). New tools, including short questionnaires, interactive software, picture cards, and media (e.g., phone, mobile, camera), are being developed to target (e.g., linking kitchen scale, food bar codes, computer, and phone) specific nutrients (e.g., calcium, vitamin D, fluoride,

antioxidants) and foods (e.g., fruits and vegetables) and specific populations.[7–11] Other methods are being developed to assess healthful and unhealthful food and eating behaviors in 9- to 14-year-old children.[12,13] The validity and the reliability of dietary intake methods among 2- to 18-year-old children and adolescents from industrialized countries have been reviewed elsewhere.[11,14,15] Most validation studies have used observation, diet records, or repeated 24-hour recalls as reference standards. Assuming energy balance, the doubly labeled water method is considered the best method for validation of energy intake using traditional dietary assessment methods.

The doubly labeled water method is the first truly noninvasive means to measure total daily energy expenditure accurately in free-living humans and is based on measurement of CO_2 using a nonradioactive isotope. The technique was introduced in the 1950s, but applications started being made among adults in the 1980s. The total energy expenditure obtained from the doubly labeled water method can be accurate,[16] but large interlaboratory variability has been found in a blinded evaluation of multiple laboratories.[17] Also, the reproducibility of this method at an interval of several months or more has not been evaluated in free-living populations of children. Overall, food records and recalls are more accurate than other dietary assessment methods in school-age children.[11] A review by Burrows et al.[15] compared dietary assessment techniques with the doubly labeled water technique in children in different age groups. The findings suggest that the 3-day, 24-hour recall with parents as proxy reporters provided the best estimate for energy intake for children aged 4–11 years, while weighed food records for children aged 0.5–4 years and diet history for adolescents aged 16 years and older were optimal in these age groups.

TABLE 17-1 Advantages and Disadvantages of Dietary Assessment Methods

Method	Recording Time	Period	Inter-viewer Training	Subject Literacy	Affects DietIn-take	Memory	Subject Burden	Food Description	Automated Procedure	Budget
Recall	Short	Defined	High	No	No	Yes	No	Yes	Yes	Expensive
Food record	Defined	Defined	High	Yes	Yes	No	Yes	Yes	Yes	Expensive
Food frequency	Short	Long	None	Yes	No	Yes	Yes	No(ranks)	Yes	Relatively inexpensiveve
Histories	Long	Imprecise	High	No	No	Yes	Yes	Yes	No	Expensive
Observation	Long	Defined	High	No	No	No	No	Yes	No	Expensive

Studies also show that most dietary assessment methods provide better

estimates for the composition of the diet—after adjusting for energy intake—rather than absolute intake of nutrients or energy and for a group rather than individuals.[18] As a measure of validity, correlation coefficients of at least 0.5 between a new method and a standard method, such as many days of food records, are recommended for nutrients of interest in epidemiological studies.

24-Hour Recall

The 24-hour recall method is based on an interview, typically conducted by a trained dietary interviewer. It provides appropriate mean estimations for groups but not for individuals because of large day-to-day variation in individual intakes. Surrogate reporters are required for children who are young (preteen),[19] have mental delays, or are too sick to respond by themselves. Reporting problems can arise if parents spend less time with their children, for example, when they work outside of the household or have a large family.[18]

Twenty-four-hour recalls by children aged 2–18 years, or by surrogate reporters, have been compared to reference standards (observations, food records, and food chemical analysis) and summarized elsewhere.[14] In their review, McPherson et al. found correlation coefficients(r values) of 0.2–0.9 for energy intake and 0.4–0.6 for most nutrients compared with the standard methods.[14] The correlation coefficients for nutrients improved after adjusting for energy intake. Compared with the standard method, recalls tended to either overestimate or underestimate energy intake.[14] The extent of underreporting was greater in adolescents than in all other age groups[20,21] and among girls participating in a dietary intervention compared to control.[22] Food eaten varied from 68% to 84% when diet recall was compared with the validation standard.[14] The validity of the dietary recall improved when individual meals were compared rather than using 24-hour periods[14] and when school-aged children (8–10 years) were trained to record their food intake using diet records and then asked to recall their 24-hour food intakes using the diet records as memory prompts during interviews.[23] Baxter et al. found that the accuracy of dietary recall did not vary with gender or ethnicity.[24]

The number of days for which dietary recording is necessary to accurately identify specific nutrient intakes in children has been calculated by determining the day-to-day intraindividual variation in dietary intake data for infants aged 4–6 months[25] and preschool children.[26] Quandt obtained dietary information using 24-hour recall among 28 mothers for

their infants' diets on 4 random days over approximately 1 month.[25] In contrast to what is the case with adults, investigators found no effect of day of the week on nutrient variation in this age group. Interindividual variation exceeded intraindividual variation for all nutrients except vitamin C.

Stein et al. measured intraindividual day-to-day variation and tracking of nutrient intakes in a longitudinal study of 181 preschool children (93% Hispanic) aged 45–60 months recruited through a hospital-based pediatrics practice in New York.[26] In that study, 24-hour dietary recalls were administered four times in the first year and three times in the third year to children's mothers. Although the reliability of estimates (intraclass correlation coefficients) of energy and 9 nutrients obtained from a single administration of the dietary recall ranged from 0.15 to 0.38, the authors were able to show substantial tracking of underlying diets among preschool children over a 19-month period, despite day-to-day variation. Dietary methods for low-income populations have recently been reviewed elsewhere.[27]

Fisher et al. showed that body weight also influenced the accuracy of dietary reports made by children and their parents in a sample of 146 children aged 4–11 years using the doubly labeled water method to validate the energy intake measured using 24-hour recalls.[28]

Food Record

The food record method consists of a detailed diary of foods consumed over one or more days.[29] It provides appropriate mean values for groups but not for individuals, unless the number of days is substantial. The information is collected at the time of consumption so that memory is not a limitation. However, assessment is imprecise when dining out and can interfere with dietary intake. McPherson et al. pointed out that there were no specific validation studies of portion size collection, and none of the studies reviewed presented results on validity in reporting food items.[14] Underreporting in children's adult-assisted food records increased with age (9–12 years) when energy expenditure was measured with doubly labeled water. In that study, the investigators found differences in reporting by race but not by gender. African American children were significantly more likely to underreport energy intake than were white children.

Black et al. studied the day-to-day variation in energy intake of 48 breast-fed infants using four-day diet records at monthly intervals from age 1.5 to 7.5 months and of 37 fully weaned infants studied at 10, 12, 15, and

18 months of age.[30] Dietary variation was lowest in fully breast-fed infants, increased as solids were introduced, and approached values found for adults in the 18-month-old infants. Nelson et al. showed that with the exception of ascorbic acid, intakes were the lowest in the youngest age groups (<36 months of age) and rose through childhood to peak in adolescence and young adulthood.[31] In contrast to the homogeneity of the intragroup coefficient of variation (CVw) of specific nutrients among children and adults, the intergroup coefficient of variation (CV_b) varied largely by age group and gender, resulting in variance ratios that were relatively low for toddlers, high for children aged 5–17 years, and intermediate for adults. The lower toddler's variance ratios is best explained by their ability to adjust their energy intake at successive meals resulting in relatively constant within-subject daily energy intake.[32] Nelson et al. found that up to 4 years of age, seven days were adequate to estimate all macronutrients except type of fat.[31] For animal protein, the number of days required to achieve a level of accuracy of ranking at and above 0.9 was 18 days (Table 17-2).[31]

As determined by the doubly labeled water method, underreporting of energy intakes on children's assisted food records increased with age.[14,33] Using that same reference method, other investigators have also shown that estimations by food record can be made at the group, but not at individual, level. O'Connor et al. compared measurements of energy intake from diet records and total energy expenditure using the doubly labeled water method in 47 children aged 6–9 years.[34] Although the mean energy intake and the total energy expenditure were not significantly different, the authors showed no significant correlation between energy intake and total energy expenditure. The findings of this study confirm the notion that energy intake assessed by diet records is not representative of total energy expenditure at the individual level but that three-day food records could be used to estimate energy intake at the population level. Even longer periods of dietary records (e.g., 7 days) compared to the doubly labeled water method can underestimate energy intake, especially in overweight adolescents.[35]

Food Frequency Questionnaire

In contrast to food records and 24-hour recalls, FFQs are relatively inexpensive and aim to represent long-term intake. The food frequency method relies on a food list, and participants are asked to report the frequency of consumption of each food item; the design of these

questionnaires is described in detail elsewhere.[18] FFQs can be used to rank respondents by levels of intake. The period of time covered by the FFQ can be weeks, months, or years, and the questionnaires can be classified as quantitative (i.e., food models shown for usual portion size), semiquantitative (i.e., standard serving size used), or nonquantitative (only frequency assessed). Overestimation of intakes of energy and other nutrients was observed in studies using a validation standard (diet record or repeated 24-hour recall) when adults' portion size, rather than children's portion size, was used in children's studies.

TABLE 17-2 Number of Days Dietary Records Are Required for a Correlation Coefficient of 0.9 Between Observed and True Intakes

	Days for Aged Children 1–4 yr	Days for Boys Aged 5–15 yr	Days for Girls Aged 5–15 yr
Protein	5	6	15
Animal protein	6	7	18
Fat	7	8	12
Saturated	8	6	12
Polyunsaturated	15	18	28
Monounsaturated	11	9	14
Cholesterol	27	18	21
Carbohydrate	6	10	9
Folate	6	9	22

Source: Adapted from Nelson et al.[31]

When multiple 24-hour recalls, rather than just one, were used as the validation standard, investigators observed similar estimates for energy intake obtained with the 24-hour recalls and the FFQ, referred to as the Youth–Adolescent FFQ.[18] In that study, further adjustment for intraindividual variation in recalls improved the nutrient correlation coefficients. Inner-city sixth and seventh grade students demonstrated the ability to provide valid estimates of intake of calories and a variety of nutrients over one year. However, children in the fourth and fifth grades in inner-city schools experienced difficulties in completing the same FFQ.[36] This finding shows that surrogates are needed in older children from disadvantaged populations. A FFQ for low-income, 1- to 3-year-old Hispanic, African American, and white children who attended the WIC

program was recently found to have excellent reliability and acceptable validity.[37] Blum et al. also validated the use of a shortened version of the Youth–Adolescent FFQ in Caucasian and Native American children aged 1–5 years, with a parent or guardian filling out the questionnaire.[38] The average correlation was similar to that found in validation studies among adolescents and adults. Finally, the use of a 124-item FFQ has been found to be a reasonably valid method to capture adolescent diet recalled by young adults.[39]

In contrast to the accuracy of repeated 24-hour recalls[40] and weighed diet records,[21] when the doubly labeled water method was used as a standard, energy intake was overestimated by 50% in infants when FFQs were used.[41] Using the same doubly labeled water method as a standard in a group of older children (9–16 years), energy intake obtained from the Youth–Adolescent FFQ was underestimated. However, the authors showed that despite a wide range of variability in reporting accuracy across adiposity status and gender, this method estimated the mean energy intake of a group with appropriate accuracy.[42]

Diet History

Diet history provides information on typical meals, food intakes, and preparation of food using a questionnaire, an interview, or both. This method is more qualitative than FFQs; the interview can take between 1 and 2 hours. The studies in children have shown overestimation of nutrients and energy intakes in the first interview compared with overestimations in subsequent interviews.[29] In addition, Livingstone et al. validated the energy intake assessed by *seven-day* weighted food records and diet histories with that obtained using the doubly labeled water method in 78 subjects aged 3–18 years.[21] Food records underestimated energy intake, whereas diet histories tended to overestimate energy intake in most age groups if it is assumed that the results from the doubly labeled water method were without error.[17]

Observation

Trained personnel can observe patients in a variety of settings, especially schools. Food intake can be measured once or multiple times. The method is intrusive and is impractical in large studies. However, this method is often used as a validation standard for school-aged children and is reliable.[43]

SPECIAL CASE FOR DIETARY ASSESSMENT: BREASTFEEDING

Studies relating breastfeeding to illness are difficult to evaluate because of the use of a variety of definitions for breastfeeding,[44] selection bias,[45] confounding,[46] and reverse causality.[18] As reviewed below, few studies have addressed the reproducibility and validity of breastfeeding assessment methods. Several techniques have been used to evaluate the intake of breast milk among breastfeeding children. The doubly labeled water method is considered the gold standard for evaluating energy intake in breast-fed children.[47] Other techniques include pre- and postnursing infant weights, and the evaluation of the duration and frequency of breastfeeding using observation, recall, and record studies. The Centers for Disease Control and Prevention (CDC) has developed a comprehensive breastfeeding website with current updates and intervention tools (http://www.cdc.gov/breastfeeding/).

The use of doubly labeled water has been validated against periodic open-circuit respiratory gas exchange in infants.[48] Using double-labeled water, Butte et al. showed that energy requirements of infants and toddlers were 80% of recommendations and were lower in breast-fed than in formula-fed infants.[47] As a result, recommendations for energy intake have been decreased in infancy and early childhood and recommendations for physical activity provided.[49]

Test-Weighing Techniques

Conventional test-weighing techniques include weighing either infants or mothers before and after a known or unknown amount of breast milk intake. Brown et al. conducted validation studies of different test-weighing techniques among 64 infants before and after the consumption of a known amount of milk.[50] The measured bottle weight corresponded to 94.9% ± 13.2% of the estimated change in weight of the infant. These techniques allowed good estimates of breast milk consumption to be made, noting that the average amount of breast milk received declined with time from 632 g/d for the 5- to 12-month olds to 368 g/d for the 24- to 30-montholds.[50] Because of the intrusive nature of test weighing, it generally will not be directly useful in epidemiologic studies, but it could serve as a standard in validation studies of other methods.

Observation, Recalls, and Questionnaires

Piwoz et al. conducted a validation study of breastfeeding recall using observation as the reference standard.[51] They obtained 1574 single-day dietary intakes (12-hour observations plus 12-hour recall) and mothers' monthly reports of usual feeding practices. They demonstrated that single-day studies produced a different view of feeding practices compared to the mothers' reports. Exclusive breastfeeding was observed 25% more frequently among infants younger than 4 months than was self-reported. In contrast, self-reported formula consumption was 30% more frequent compared to observation data. Differences between reported and observed practices were related to mothers' age and education, number of children younger than 5 years in the home, and infant age and disease on the observation day. The investigators concluded that consumption of breast milk or formula during the past 24 hours should not be used alone as the basis for classifying infant feeding practices in surveys. Infant Breastfeeding Assessment Tool, the Mother Baby Assessment Tool, and the LATCH assessment tool (L for latch, A for audible swallowing, T for type of nipple, C for comfort [breast/nipple], and H for Hold [positioning]) were evaluated for reliability (6 months) and validity using videotapes of breastfeeding sessions.[52] Using food records and FFQs, the relative agreement for the frequency of breastmilk and study formula consumption was found to be very good.[53]

Most epidemiologic studies have used recalled or observed data to provide information on breastfeeding. Investigators have used information on the duration, frequency, and minutes of breastfeeding rather than test-weighting or isotopic methods to evaluate the relationship between breastfeeding and outcome (i.e., growth and disease), and some, but not all, studies have gathered information on additional sources of foods beside breastfeeding. For example, Zeitlin and Ahmed examined the relationship among breastfeeding, total energy intake, and growth of infants in Bangladesh using observation (minutes of breastfeeding) and 24-hour recall to estimate breast milk and other food intake in infants and children aged 5–23 months.[54] Beaudry et al. used a self-administered standardized questionnaire mailed to every mother 1 week before infants reached 6 months of age to evaluate past breastfeeding and weaning behaviors.[55] Vähätalo et al. found that a dietary interview method including a short FFQ was useful for ranking infant subjects according to the consumption frequency of breastmilk and infant/study formula, as well the amount of formula used.[56] However, the amounts of study formula were overestimated by the dietary interview as compared with two 48-hour

recalls.[56] Types of questions asked during the interview are reported by the authors in the methods section and the FFQ used in that study can be found in the appendix of that article. Finally, some authors have previously suggested that clarification of the intensity and duration of breastfeeding might help detect dose–response effects of breastfeeding. More recent information on infant feeding practice survey is available on the CDC website http://www.cdc.gov/ifps/questionnaires.htm.

ASSESSMENT OF GROWTH AND BODY COMPOSITION

The focus of many studies in nutritional epidemiology is body weight, but recent techniques have evolved to better characterize changes in body weight in terms of body composition. Chapter 4, "Body Composition and Growth," describes the theory and practice of many of these techniques, as well as what is known about developmental changes in body composition.

Because nutritional epidemiologic studies in pediatrics often employ a variety of body composition techniques, the relationships among them are of interest. Densitometry is typically considered the gold standard for a two-compartment model of fat and nonfat tissue in adults and depends on measurements of total body density. Despite the lack of data on fat and fat-free mass densities in children,[57] densitometry has been compared with anthropometric measurements involving weight, height, and skinfold thickness measurements (Table 17-3).[58–63] Likewise, anthropometric data based on weight and height indices have been compared with skinfold thickness measurements (Table 17-4).[64–66]

TABLE 17-3 Correlations Between Anthropometric Measurements and Total Body Densitometry

Author(s)	Sample	Weight	Height	Weight/ Height	Weight/ Height^2	Weight/ Height^3	Triceps Skinfold	Subscapular Skinfold
Parizkova[a]58	N = 62 white girls 12–16 yr	—	—	—	—	—	–0.74	–0.80
Seltzer et al.[b]59	N = 32 obese white girls 12–18 yr	–0.53	–0.23	—	—	—	–0.69	–0.59
Young et al. [c]60	N = 102 white girls 9–16 yr	–0.36	—	—	—	—	–0.73	—
	N = 41 prepubertal	–0.75	—	—	—	—	–0.82	—
	N = 15 pubertal	–0.69	—	—	—	—	–0.78	—
	N = 46 postpubertal	–0.42	—	—	—	—	–0.70	—
Harsha et al.[61]	White, 6–16 yr; *n* = 79 boys, *n* = 64 girls	–0.17/–0.52	0.31/0.03	—	—	—	–0.76/–0.75	–0.75/–0.80
	Black, 6–16 yr; *n* = 49 boys, *n* = 50 girls	–0.36/–0.59	0.25/0.14	—	—	—	–0.82/–0.82	–0.75/–0.87
	N = 242 boys and girls 6–16 yr	–0.32	0.18	—	—	—	–0.81	–0.76
Roche et al.[62]	N = 68 boys, n = 49 girls 6–12 yr	0.74/0.74	—	0.75/0.62	0.90/0.84	0.66/0.55	0.81/0.74	0.84/0.80
	N = 63 boys, n = 81 girls 13–17 yr	0.65/0.88	—	0.91/0.87	0.88/0.89	0.91/0.82	0.93/0.86	0.94/0.87
Maynard et al.[63]	N = 35 boys, n = 39 girls 8 yr	—	0.27/0.34	—	0.93/0.67	—	—	—
	N = 49 boys, n = 56 girls 9 yr	—	0.27/0.46	—	0.86/0.74	—	—	—
	N = 79 boys, n = 63 girls 10 yr	—	0.46/0.43	—	0.90/0.79	—	—	—
	N = 90 boys, n = 79 girls 11 yr	—	0.51/0.42	—	0.94/0.78	—	—	—
	N = 103 boys, n = 86 girls 12 yr	—	0.44/0.28	—	0.90/0.85	—	—	—
	N = 103 boys, n = 81 girls 13 yr	—	0.39/0.39	—	0.89/0.82	—	—	—
	N = 101 boys, n = 74 girls 14 yr	—	0.30/0.16	—	0.86/0.84	—	—	—
	N = 94 boys, n = 80 girls 15 yr	—	0.13/0.01	—	0.83/0.88	—	—	—
	N = 91 boys, n = 87 girls 16 yr	—	0.22/0.19	—	0.83/0.85	—	—	—
	N = 90 boys, n = 83 girls 17 yr	—	–0.09/0.07	—	0.90/0.90	—	—	—

Of note, the correlation coefficients between predictors of adiposity and total body densitometry tend to be positive when authors have transformed body density measurements using an equation

to estimate fat (as in Maynard or Roche) but tend to be negative when using direct measurements of hydrodensitometry (as in Horsha). In the Bogalusa Study, the authors used W/H^p rather than W/H^3. They calculated the optimal "*p*" for each category of gender, race, and age because they wanted to obtain an index that was independent of *H* during growth and early life development [$\log(W) = a + p \log(H) + e$].

[a]The relationship between body density and triceps skinfold for the nonobese adolescents was [body density = 1.128 − 0.78 log triceps skinfold].

[b]Arm circumference had the highest correlation with both weight ($r = 0.90$) and body density ($r = -0.63$). The authors showed body densities for their subjects similar to those shown by previous authors estimated from skinfold thickness measures [body density = 1.1516 − 0.09256 log triceps skinfold] and comparable to real body density measurements.

[c]Total skinfolds provided (at least 12 sites from the chin to the knee). Overall, correlation for triceps was −0.79 and −0.72 for subscapular.

TABLE 17-4 Correlations of Body Mass Indices With Skinfold Thickness Measures in Large Studies

Study	Sample	Skinfold Thickness	Weight/Height	Weight/Height^2	Weight/Height^3
Forsyth County Study[64]	N = 832, 5–12 yr boys	Triceps	0.72	0.81	0.81
	N = 835, 5–12 yr girls		0.73	0.80	0.64
	N = 66 boys/n = 80 girls 5 yr		—	0.24/0.68	—
	N = 98 boys/n = 86 girls 6 yr		—	0.53/0.71	—
	N = 107 boys/n = 121 girls 7 yr		—	0.67/0.66	—
	N = 110 boys/n = 116 girls 8 yr		—	0.79/0.75	—
	N = 137 boys/n = 144 girls 9 yr		—	0.78/0.84	—
	N = 110 boys/n = 88 girls 10 yr		—	0.82/0.81	—
	N = 89 boys/n = 83 girls 11 yr		—	0.89/0.86	—
	N = 115 boys/n = 118 girls 12 yr		—	0.79/0.81	—
Bogalusa Study[65]	N = 821 white boys 4–5 yr	Triceps/ subscapular	—	0.53/0.52	0.53/0.51
	N = 523 black boys 4–5 yr		—	0.50/0.44	0.51/0.44
	N = 838 white girls 4–5 yr		—	0.61/0.71	0.60/0.67
	N = 499 black girls 4–5 yr		—	0.63/0.60	0.63/0.56
	N = 1570 white boys 12–13 yr		—	0.72/0.86	0.76/0.81
	N = 925 black boys 12–13 yr		—	0.64/0.81	0.67/0.71
	N = 1462 white girls 12–13 yr		—	0.81/0.84	0.77/0.82
	N = 821 black girls 12–13 yr		—	0.83/0.84	0.81/0.81
NHANES III[66]	Boys/girls 2–5 yr	Average triceps and subscapular	0.67/0.73	0.68/0.71	0.61/0.66
	Boys/girls 6–11 yr		0.79/0.82	0.81/0.85	0.77/0.82
	Boys/girls 12–19 yr		0.79/0.83	0.81/0.85	0.79/0.84

TABLE 17-5 Characteristics of Body Composition Measurements in Children

Characteristics	Weight	BMI	Waist Circum-ference	Skinfold	Bioelectrical Impedance (BIA)	Densi-tometry	Isotopes[a]	Radiology[b]
Advantages								
Convenient	Excellent	Excellent	Excellent	Very good	Good	No	No	No
Reproducible	Excellent	Excellent	Excellent	Very good	Good	Very good	Very good	Very good
Fast	Excellent	Excellent	Excellent	Very good	Relatively	No	No	No
Expensive	No	No	No	No	No	Yes	Yes	Yes
Correlates with morbidity	Excellent	Excellent	Excellent	Excellent	Good	Good	Good	Excellent
Large studies	Good	Good	Excellent	Good	Good	Not good	Not good	Not good
Standard	Excellent	Excellent	Excellent	Excellent	Good	Very good	Very good	Very good (some lack)
Disadvantages								
Correlates with height	Highly	Not highly	Not highly	Not highly	No	No	No	No
Assess fat distribution	Poorly	Poorly	Poorly	Excellent	Poorly	Poor	Poor	Excellent
Equations	No	No	No	Many	Many	Many	Many	Many
Training	Some	Some	High	High	High	Very high	Very high	Very high
Radiation	No	No	No	No	No	No	No	Yes (CT)

[a]Isotope studies such as ^{40}K, doubly labeled water.

[b]Radiology studies such as computed tomography scan, magnetic resonance imaging, and ultrasonography.

Data on weight and height were also compared more recently not only for measurement of skinfold thickness but also for dual-energy x-ray absorptiometry (DXA) among children aged 2–19 years.[66] Compared with weight for height and the Rohrer's index (Wt/Ht^3), the Quetelet or body mass index (BMI; Wt/Ht^2) was identified as the better predictor of overweight (but not underweight) when average skinfold thickness measurements were used as standard. BMI was also shown to perform better than the Rohrer index when the standard for percent body fat or total fat mass was estimated using DXA in children aged 3–19 years. No differences were found between BMI and weight for height in detecting overweight or underweight.

As described in Table 17-5, anthropometric measurements are most useful in large-scale studies. A study of bioelectrical impedance analysis (BIA) and BMI in adults[67] suggested that BIA was not superior to BMI as a predictor of overall adiposity. Likewise, Eisenman et al. found that BIA had limited utility in estimating body composition in their validation study among 3- to 8-year-old children (age of adiposity rebound) using anthropometry (BMI, waist circumference, and skinfolds), BIA, and DXA

scan.[68] Recommendations from these studies are to use BMI, skinfolds, and waist circumference measurements in epidemiological studies.

Although BMI does not directly measure body fat, it is typically used to evaluate adiposity in adults and has been recognized as a useful predictor of adiposity in children and adolescents, which, in turn, also predicts risks for present or future medical complications ofobesity.[69] The weight indices are considered adequate when the correlation with percent body fat is high and the correlation with height is low. Although there is no perfect weight index of overweight and underweight—irrespective of height and width—BMI is a reasonable index from childhood to adulthood.[66] Transformation of weight and height is also discussed in the review by Gasser et al.[70] Frontini et al. further demonstrated that the association of weight to height index was not superior to BMI as an indicator of adiposity compared to skinfold thickness measurements as reference standards and related cardiovascular risk factors during childhood in the Bogalusa Study.[65]

Self-reported weight and height among adults is frequently used in nutritional epidemiology. In contrast to the findings in adult studies, results from adolescents aged 12–17 years in the Third National Health and Nutrition Examination Survey (NHANES III) showed that self-reported weights were missing in 40% of 12 year olds and 25% of 13 year olds. On the basis of their findings, the investigators did not recommend the use of self-reported weight and height as proxy measures of weight and height among adolescents younger than 14years.[71] In contrast to these findings, in a study of 72 overweight and obese children and adolescents aged from 8 to 18 years old, self-assessment of height, weight, and sexual maturation were not found to be valid proxy measures for a physician's assessment, although the reproducibility of self-report of height and weight was good or excellent.[72] The renewed interest in body composition, combined with the awareness of the growing obesity epidemic, has resulted in the need for improved reference data for body composition.

BIOCHEMICAL MARKERS OF DIETARY INTAKE

A biologic marker of dietary intake (or nutritional biologic marker) is defined as "any biological specimen that is an indicator of nutritional status with respect to intake or metabolism of dietary constituents."[73] A nutritional biological marker may thus be a biochemical, functional, genetic, or clinical index of status of a nutrient or other dietary component.

This section focuses on biochemical markers of dietary intake (biomarkers).

Biochemical indicators (biomarkers) of dietary intake—blood, urine, stool, and tissue nutrients—can reflect nutrient intake.[74] Biomarkers are important in nutritional epidemiology because they can be used as surrogates for dietary intake when the nutrient content in a food varies greatly, used to validate questionnaire-based assessment of dietary intake, and used as markers of compliance in intervention studies. They can also be useful as markers of disease, even if they reflect intake poorly; serum cholesterol provides a good example. Biomarkers can also be used to validate nutrient intake assessed by dietary assessment methods. When a biomarker needs to represent the intake of a specific nutrient in an epidemiologic study, the most important requirements are its sensitivity to changes in dietary intake and its ability to reflect the cumulative average intake over an extended period of time.[18] As opposed to nutrient intakes, relatively few foods have corresponding specific biomarkers of intake.

Sensitivity of a Biomarker

The sensitivity of a biomarker depends on how well the biomarker reflects true nutrient intake, which can vary with the chemical form of the nutrient. For example, the bioavailability is lower for dietary folate than for supplemental folate, so serum levels will differ with the form of folate. Serum sodium is an example of a biochemical measure biomarker that is not sensitive to intake because it is tightly regulated to remain within a specific range. The 24-hour urinary measurement of 3-methylhistidine is an example of a good biomarker for animal protein intake ($r = 0.77$).[75]

Specificity of a Biomarker

The lack of specificity of a biomarker of dietary intake may result in an association between the biomarker of dietary intake and health outcomes that is confounded by other dietary factors. An example is urinary alkylresorcinol metabolites and whole grain (wheat and rye) affected by vitamin C.[76] Another example is that of folate, which is present not only in fruit and vegetables but also in cereals. As a result, variation in plasma folate is not only associated with cereal but also associated with fruit and vegetable intake, and the extent to which plasma folate is a good biomarker of fruit and vegetable intake will differ by study population. Similarly, serum folate will not be a good indicator of fruit and vegetable intake if folate comes mostly from fortified foods. Variation in biomarkers

can also reflect genetic variation in metabolizing pathways or supplements.

Period Covered by a Biomarker

On the basis of the time period covered by a biomarker, there are two major types of biomarkers: recovery and concentration markers. For concentration markers, ranking of the individual's intake can be estimated based on biomarkers' values because they are correlated. Most markers are in this category; an example is transfat in adipose tissue. A recovery biomarker is a marker that is correlated to dietary intake for a specific period of time: an example is urinary nitrogen over a 24-hour period for protein intake.[77] A special case is that of predictive biomarkers, which are highly correlated with dietary intake like the recovery biomarkers, but, as opposed to concentration markers, only a fraction is recovered (e.g., sucrose plus fructose in 24-hour urine). As a result, predictive biomarker values can be used to estimate absolute intake.

Studies of chronic diseases require biomarkers that are less susceptible to short-term fluctuations. For example, serum folate is sensitive to changes in intake and modestly sensitive to temporary changes in folate metabolism. Data from the Framingham Study demonstrated a strong inverse correlation between plasma homocysteine and folate intake over long periods, assessed using FFQs.[78] Another example is that of plasma amino acids. There is evidence that plasma amino acids are indicators of dietary intake of amino acids if they are measured within two hours of a protein meal.[79] Plasma amino acid measurements in the postabsorptive state (at least eight hours without food consumption) are an indicator of dietary intake of amino acids when similar protein meals have been consumed for at least two weeks.[80] Although red blood cells membrane fatty acids were not found to be reliable as an independent measure of compliance for dietary saturated fatty acids intake in short-term studies,[81] red blood cell levels of the monounsaturated fatty acid, oleic acid, and the polyunsaturated fatty acids such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were more useful biomarkers during short-term intervention trials. Cholesterol ester appears to be the most suitable serum fraction to assess short-term dietary compliance.[82] Examples that reflect long-term nutrition status include toenail selenium and tissue fatty acid composition.

Source of Biomarker Measurements

Sources of biomarkers in humans include direct measurements of

nutrients, enzyme activity, products of the metabolic pathway, and challenge (tolerance) tests. The biomarker can be measured directly in body fluids or tissues such as plasma, serum, erythrocytes, adipose tissue, muscle, urine, stool, hair, and nails. For example, mean population intakes of iodine and sodium can be estimated by casual urine samples.[83] Another example is that of dietary fatty acids. Using dietary intake from 200 FFQs as well as tissue, whole blood, and plasma levels of fatty acids, the correlation coefficients for α-linolenic acid and linoleic acid was 0.38 and 0.43, respectively, between diet and whole blood, 0.51 and 0.52 between diet and adipose tissue, and 0.39 and 0.41 between diet and plasma. In this study, for α-linolenic and linoleic acid, high correlations were observed between whole blood and adipose tissue (r = 0.59 and 0.67, respectively) and between whole blood and plasma (r = 0.96 and 0.88, respectively).[84] Enzyme activity can be used to evaluate specific dietary nutrients (e.g., alkaline phosphatase for zinc, glutathione peroxidase for selenium, and erythrocyte transketolase for thiamin).

Plasma homocysteine is a sensitive, but not a specific, marker of folate, and 25-hydroxyvitamin D is a good marker for vitamin D deficiency. Although serum ferritin is a sensitive test of iron status and continues to be the leading single determination for individual iron status, it is not recommended in inflammatory states because it is an acute-phase reactant. Serum transferrin receptor is a promising alternative because it is more specific than ferritin. Another example of a nutrient that can be confounded by inflammation is serum betacarotene.[85] Finally, by-products of nutrients can be related to disease, such as protein carbonyl content, which is the most widely used marker of oxidative modification of proteins.[86] See Chapter 3: Laboratory Assessment of Nutritional Status for a detailed description for common nutritional biomarkers.

Factors Influencing Biomarker Measurements

The magnitude of the correlation between a biomarker and dietary intake depends mainly on bioavailability, homeostasis, range of intake, and determinants of the concentration other than intake (e.g., lifestyles, gender, BMI, genetics). Bioavailability of a nutrient can differ with processing of products. An example is that of adding fat to the preparation of vegetables because it increases the bioavailability of fat-soluble carotenoids. The mechanisms responsible for homeostasis may include nutrient absorption (e.g., iron), excretion of excess (e.g., vitamin C), and hormonal pathways (e.g., calcium). As a result, the relationships between concentrations and

intakes are rarely linear and may result in attenuation or plateau of the relationships with higher intakes. The biomarker may be a poor indicator of intake if the plateau phase is wide (e.g., retinol) or the range of intake is narrow and falls in the plateau phase. For vitamin A status, retinol concentration will not be useful unless the liver stores are either very depleted or highly saturated.[87] Biomarkers of long-term intake with large within-person variation are likely to result from a response to short-term fluctuations in intake or to changes in other determinants of its concentration (e.g., gender, body fat). Therefore, several measurements may be needed to improve prediction at an individual level and to account for population trends in intakes.

The relationship between dietary nutrients and biomarkers can be influenced by sample collection, storage techniques, seasonal timing, time of day, study design, contamination (e.g., zinc collected in ethylenediaminetetraacetic acid tubes), stability, storage temperature, and freeze-thaw cycles, as well as level of quality control.[18]

If these factors influencing biomarkers are not taken into account, misclassification of individuals with dietary intake may occur. When the factor is not associated with disease outcome, it contributes to random misclassification and thus attenuation of the true relationship between the biomarker and the disease outcome. When the factor is associated with disease, confounding may occur. An example of confounding is that of serum vitamin E (biomarker for vitamin E intake) and liver disease in preterm infants.

Another complication might be due to reverse causation, which is observed when a disease outcome affects the concentration of a biomarker of dietary intake. An example is cancer and nutrient concentration.

Biomarkers to Validate Dietary Assessment Methods

Biomarkers can be useful for validating other methods of dietary assessment such as FFQs, diet records, or recalls.[18,88] A major advantage is that the source of error in the biochemical measure and intake assessment should be independent. For example, good correlations between folate intake and serum or red blood cell folic acid provided important documentation about the assessment of dietary folate, despite concerns about bioavailability and stability in food processing. Sometimes biomarkers can detect biases in reporting.

Biomarker Limitations

Biomarkers of dietary intake are measures of nutrients and other food components if they are digested, absorbed, and transported in the extravascular system. However, these processes, metabolism in the intestinal bacteria, metabolism in the gut mucosa, metabolism or storage in the liver, and excretion, may affect the relationship between biomarker and intake. An example of anthropometric and dietary factors that may affect the relationship between a biomarker of dietary intake and disease outcome is that of alkylresorcinol metabolites.[76] Sex, BMI, and vitamin C intake were identified as determinants of the proportion of free and glucuronidated 3-(3,5-dihydroxyphenyl)-propanoic acid, and therefore investigators recommended that urine samples be deconjugated prior to quantification.

Analysis of diet and disease typically require the use of energy-adjusted intakes rather than absolute intakes and long-term intakes (see the section on Adjustment for Energy Intake). Because recovery biomarkers reflect absolute intake, a marker of energy such as the doubly labeled water method would be needed, but this measure is expensive and often not available. Therefore, when recovery biomarkers are used, multiple repeated assessments or adjustment for within-person variation in the biomarker is used to estimate long-term intakes. Limitations for use include the cost and participant burden for doubly labeled water assessments. Finally, recovery biomarkers are available for limited number of dietary factors. Although both recovery and concentration biomarkers can be used to validate or calibrate dietary instruments, correlation with a dietary measure are generally greater for recovery biomarkers and thus additional methods (e.g., methods of triads, which is a triangulation technique to calculate the validity coefficient of a food-frequency questionnaire) necessary with concentration biomarkers.

TRACKING DIETARY INTAKE

For adults, year-to-year dietary intakes, assessed by FFQs, show correlations of 0.6–0.7, with decreasing correlations over longer intervals,[18] but diet tracking varies according to age category among children. Although parents have a dramatic influence on their children's dietary intakes, young children's preferences are not necessarily similar to those of their parents; however, as children grow older, their preferences become more similar.[89] Energy-density manipulation studies show that children are able to adjust their daily energy intake when they focus on their own hunger and satiety cues rather than on their parents asking them

to “clean their plates.”[89]

There was substantial tracking of underlying diets in a study of preschool children (45–60 months old) over a 19-month period.[26] After correction for intraindividual variation, the correlations ranged between 0.2 and 0.6. The correlation between nutrient intake at age 2 and 4 was examined in the Bogalusa Study cohort of children aged 6 months to 4 years.[90] Total protein, animal protein, total sugar, sucrose, starch, total fat, saturated fatty acids, polyunsaturated fatty acids, and cholesterol showed correlations of 0.5–0.6 in children aged 2 years compared with children aged 4 years. At 2 years of age, 47%–65% of children in the top tertile for total fat, saturated fat, and cholesterol intake remained in the top tertile by age 4.

Tracking of diets has also been studied in older children and adolescents. Tracking of fruit and vegetable intakes (food records) among children in third grade showed correlations of 0.4–0.5 over 2 years.[91] Calcium and dairy intake (diet histories) from adolescence into adulthood (13–27 years old) showed a correlation of 0.4.[92] Using three 24-hour recalls per survey, a longitudinal study of 984 children aged 6–13 years tracked nutrient intakes over a 6-year period in China.[93] The authors concluded that even under conditions of rapid socioeconomic change, children were likely to maintain their dietary intake patterns from childhood into adolescence. The correlations between dietary intakes measured at a six-year interval ranged between 0.28 and 0.51 for macronutrients and major food groups (vegetable and fruit, meat, and edible oils).

Results from a longitudinal English study of 198 participants with two 3-day food diaries collected in 1980 and 2000 (at ages 12 and 33 years) showed that adolescent food intake was correlated with that in adulthood, but it was not a strong predictor. Dietary changes were influenced by variables including gender, location, and socioeconomic status.[94] However, these results could also reflect longitudinal societal changes over time. As an example, trends in food consumption patterns of 1584 ten-year old children at 7 separate cross-sectional surveys (65% Euro American and 35% African American) changed over time (1973–1994).[95] More recently, a study of 425 children aged 9–14 years in Sweden showed fair tracking of food groups and select nutrients in a 6-year period.[96] Those results were in line with previous studies reviewed in the literature.

TRACKING OF NUTRIENTS AND BIOMARKERS

Using 3-day dietary records, participants in the Penn State Young Women's Health Study showed weaker tracking of nutrients over a 6-year period among subjects who were 12 years of age at initiation of the study until they reached 18 years.[94] Although the year-to-year correlation coefficient for body weight over the six-year period of that study was 0.9, rank and correlation analyses showed low tracking among nutrients examined (fat, sugar, iron, and vitamin C). Another longitudinal study of tracking between 13 and 33 years of age in Amsterdam showed that tracking of nutrient intakes showed relatively low but significant stability coefficients for all macronutrients and micronutrients (0.28–0.52).[97] Overall, most nutrient tracking studies have shown that patterns of dietary intake are already present by 2 years of age. Although tracking varies with age, most studies show a low to modest correlation over time. Possible reasons for conflicting results of tracking could include study design, age groups under study, dietary instrument used to assess diet, socioeconomic background, and prevalence of overweight, among others.

Studies of tracking of diet and disease using biomarkers in pediatrics are few, except for those of cardiovascular disease. One example is that of diet and cardiovascular disease risk factors in the Bogalusa Study, carried out in Louisiana.[98] Dietary cholesterol changes correlated significantly with serum total cholesterol changes ($r = 0.42$) and low-density lipoprotein cholesterol($r = 0.50$) from 6 months to 4 years of age. The correlation between dietary intake and subscapular skinfold thickness measurements was 0.3–0.4 for total protein, total fat, starch, and energy.[98] In another longitudinal study from birth to 5 years of age, tracking of serum cholesterol was similar to that in older children and adolescents once the weaning process was completed.[99] The composition of fatty acids in blood has been shown to reflect the quality of dietary fat. Therefore, the long-term stability (tracking) of the serum (and hence dietary) fatty acid and composition of serum cholesteryl ester fatty acids were analyzed in a randomly selected population sample of 1029 Finnish boys and girls aged 3, 6, 9, 12, 15, and 18 years and again 3 years later. The correlations were high for all fatty acids; both linoleate and arachidonate had correlations of 0.6.[100]

Tracking of biomarkers from childhood to adulthood may need to be adjusted for change in BMI, however. In fact, recent meta-analysis shows little evidence that childhood obesity is an independent risk factor for adult cardiometabolic markers of chronic disease, morbidity, and mortality.[101]

These authors suggested that the lack of adjustment for adult BMI may explain these findings. Studies that adjust for adult BMI even show a weak negative association between childhood BMI and metabolic variables in adulthood, especially for those with low BMI status in childhood and obesity in adulthood.[102]

ASSESSMENT OF PHYSICAL ACTIVITY

Physical activity is related to both diet and chronic disease risk and may be both an important confounder and effect modifier. Physical activity is the most important factor in the variation in total energy expenditure among individuals and is thus highly relevant in dietary analysis. Physical activity is considered a behavior that can be defined as "any bodily movement produced by skeletal muscles that results in energy expenditure beyond resting expenditure" and is not synonymous of physical fitness, which includes "cardiorespiratory fitness, muscle strength, body composition, and flexibility."[103] Although physical fitness is influenced by physical activity behaviors, the determinants of physical fitness and physical activity may differ.[18,104] Measures of physical activity are either direct or indirect, with the majority of children and adolescent studies reporting low to moderate correlation coefficients between indirect and direct measures.[105] Besides direct observation, two methods are currently considered the gold standard for physical activity as they measure energy expenditure: the Doubly Labeled Water and Indirect Calorimetry.

Measures of energy expenditure such as heart rate monitors, pedometers, and accelerometers have become increasingly popular because they can be used with large groups of individuals. Accelerometers have many advantages, including the ability to measure frequency, duration, and intensity of physical activity, but they are fairly expensive. Pedometers are much simpler and less expensive but cannot distinguish between different intensities and forms of activity. Pedometers and accelerometers showed good correlations with oxygen consumption (r = 0.62–0.93) and direct observation (r = 0.80–0.97).[106] Although many studies focus on pedometers and accelerometers, self-reporting has been used in many studies. One method for short-term activity assessment is a diary and/or record of all physical activity performed or a physical activity log. A similar method is short-term recalls, which asks participants about their physical activity in the recent past, usually one day. The most commonly used self-reporting methods used in large epidemiologic studies include routine physical activity questionnaires. In these questionnaires,

participants are asked to provide information on the type, frequency, duration, and intensity of physical activity. Physical activity questionnaires are simple, inexpensive, and put little burden on the participant. However, they have been considered by some to be inappropriate for children younger than 12 years,[107] but can be complete with parental assistance.

A review of physical activity questionnaires was performed recently by Biddle et al.[108] Of the 20 instruments, 3 were recommended for use: the Youth Risk Behavior Surveillance Survey (YRBSS), the PAQ-C or PAQ-A, and the teen health questionnaire. The questionnaires are age dependent and are completed by the parent/guardian or the child. The YRBSS provides information on school and leisure time moderate to vigorous activity (past week or year), frequency of activity, and sedentary behaviors.[108] The PAQ-C targets children aged 8–14 years, while the PAQ-A targets children aged 14–20 years, and both have shown moderate validity.[109–142] These findings support the validity of the PAQ-C as a method of assessing older children's general physical activity levels. More recently, the pre-PAQ[110] and a playground activity assessment[111] were validated with accelerometer and showed promise in preschool-aged children.

The relationship between physical activity and fitness (or performance) is not clear in children and that between physical activity and sedentary behavior is weakly correlated. Measures of fitness[112] and measures of sedentary behaviors have been reviewed elsewhere. Although a relationship between physical activity and health-related physical fitness has been reported, a large part of the variability (80%–90%) is not explained.[113] Due to the fact that children's physical activity is characterized by rapid changes from rest to activity including vigorous intensities, there is recent evidence of a poor relationship between physical activity and fitness.[114] The leading criterion for measuring cardiorespiratory fitness is VO_2max (maximum volume of oxygen a muscle can use while engaged in dynamic exercise). VO_2max can be tested when a subject performs an activity such as running until exhaustion. Other methods of measuring cardiorespiratory fitness used in larger studies include heart rate monitoring, which is an excellent method to estimate energy expenditure but is less reliable in low-intensity activities and easily influenced by other factors. Several methods can be used at the same time in a study. Similar principles to those of dietary instruments can be applied in the study of physical activity in chronic disease, especially to correct for measurement errors.

EVALUATION OF DIET–DISEASE RELATIONSHIPS

The ability to detect an association between a dietary variable and disease requires adequate interindividual variation in that dietary factor.[18] This implies that the likelihood of detecting a risk associated with a specific food or nutrient among homogeneous populations who are characterized by little variation in the food or nutrient studied will be low. Dietary intake can vary with cultural background (e.g., among religious groups), type of foods consumed (e.g., vegetarianism), type of soil (e.g., soil with little selenium or iodine), food preparation (e.g., manioc with cyanide), physical activity, genetic factors, and disease state (e.g., lactose intolerance, heart disease). The detection of an association between food and disease or nutrient and disease can also be limited by errors in measuring dietary intake. An example is underreporting or overreporting of specific foods or energy intake. Variation in diet over time can be considered to be an error if long-term average intake for an individual is etiologically important. Specific data collection and statistical procedures can be applied to minimize the effects of error.

Dealing With Missing Dietary Data and Outliers

A FFQ typically covers a list of 100–150 foods. Individuals will often leave one or more questions blank, which has to be taken into consideration during data analysis. Food items left blank in a questionnaire were typically not or rarely consumed in a repeated survey.[115] Assuming that the blanks indicated lack of consumption of a food item, the correlation coefficients of nutrients between two surveys were 0.97–0.98.[115] When the number of foods left blank is large and the value for consumption of these item by responders who did not leave these items blank is high, imputation of missing data based on the consumption of that food item in the study population may be useful. A criterion of 70 blanks (out of about 130 items) has been proposed to exclude participants from analysis and was validated in the Health Professionals Follow-Up Study.[116] However, this criterion cannot be generalized to all situations.

Another common issue in dietary analysis is the identification of outliers. The use of total energy intake as a primary criterion of allowable range may be useful as it is physiologically fixed within a fairly narrow range. Underreporting is currently defined based on reported energy intake and predicted energy intake[117] and is common in dietary analysis.[118] Investigators from the Nurse Health Study have used an arbitrary

allowable range of 500–3500 kcal per day for women and 800–4000 kcal per day for men as exclusion criteria. Although values at both ends of this range are rarely correct, adjustment of nutrient intakes for total energy intake will largely compensate for overall underreporting or overreporting.

Dietary data are also highly sensitive to coding and data entry errors because nutrients are calculated from large numbers of foods. Even low error rates can result in serious misclassification of an individual for many nutrients. In other cases, food composition database errors may be observed. However, some values may just reflect unusual food intake patterns without obvious error. Whatever decisions are made to handle missing data or possible errors, it is important to confirm that any conclusions are not sensitive to the inclusion of a small number of extreme nutrient values.

Adjustment for Energy Intake

The major components of energy intake have been reviewed elsewhere[18] and include body size, growth, physical activity and fidgeting, and metabolic efficiency. Most of the variation in energy intake among subjects is attributable to differences in body size and physical activity. Although metabolic efficiency and fidgeting also vary among subjects, they are not measured in nutritional epidemiology because of the lack of practical measures. In addition, a large proportion of the variation in nutrient intake is attributable to total energy intake. Adjusting for total energy intake can be viewed as a measure of nutrient composition rather than as a measure of absolute intake. Even though the association between nutrients and predictors of total energy intake is complex, failure to adjust for energy intake can result in misleading conclusions. Thus, and for the same reasons that animal studies of nutrient effects are conducted under isocaloric conditions, taking total energy intake into consideration when evaluating the relationships between diet and disease is usually important. An exception could be when the outcome being evaluated is growth or weight gain. Because adjusting for total energy is important in nutritional epidemiology, especially when energy intake is related to disease, models have been proposed and are reviewed elsewhere (Table 17-6).[18]

Residual Model

Nutrient residuals are computed using a regression model with the absolute nutrient as the dependent variable and calories as the independent variable and then entered into a regression model with disease as the dependent

variable. This model is comparable to isocaloric metabolic studies, in which total energy intake is constant, except for the modification in one nutrient. In the case of macronutrient calories, this model can also be viewed as the caloric substitution of one macronutrient for a similar number of calories from other sources.

To make the interpretation of actual nutrient intake easier, a constant (typically the mean nutrient intake of the study population) is added to the residual values, which have a mean of zero and include negative values. When total energy intake is an important predictor of disease, total energy intake is added to the model, which already includes the nutrient-adjusted variable, as summarized in the second equation in the first model in Table 17-6. However, the magnitude of the relationship between total energy intake and disease can vary among studies because the contribution of foods to total energy intake can be different.

Standard Multivariate Model

In the standard multivariate model, both absolute nutrient intake and calories are considered independent variables in a regression model. Although the nutrient is adjusted for total energy intake in this model, the meaning has changed to calories independent of the nutrient. If we consider that total energy intake includes only protein, fat, and carbohydrate calories and the nutrient studied is protein, then energy intake corresponds to carbohydrate and fat calories.

Energy Partition Model

The energy partition model is very different from the two preceding models. This is not an isocaloric model. Using the same example of total energy intake being equivalent to calories from protein, carbohydrate, and fat, this model implies that we examine the relationship between disease and increasing amount of protein in the diet, while keeping term for other calories (including fat and carbohydrate) constant. Therefore, the relationship between nutrient and disease can still be confounded by total energy intake and could erroneously indicate an effect of protein on disease.

Multivariate Nutrient Density Model

In the multivariate nutrient density model, the nutrient density is calculated as the nutrient divided by the total energy intake in a regression model, in which a term for calories is added as a second independent variable. This

is an isocaloric model. This model can be interpreted as the relationship between the nutrient composition of the diet with disease when total energy intake is constant. Because nutrient densities are not highly correlated with total energy intake, the calories will retain their meaning of total energy intake.

Unlike in the residual model with calories and the standard multivariate model, in which the effect of the nutrient is considered similar in subjects consuming high- or low-energy diets, the nutrient density model can be useful when body size (and therefore total energy intake) varies widely among subjects. However, nutrient densities can result in an erroneous relationship if total energy intake is not included in the model. For example, when a nutrient has a weak correlation with total energy intake or a low variability and is subsequently divided by total energy intake, the nutrient can become highly correlated to total energy intake. Therefore, dividing by another variable does not necessarily mean that its effect is removed. As for absolute nutrient, nutrient densities can result in erroneous conclusions when total energy intake is associated with disease. Given the inclusion of the inverse of total energy intake in nutrient densities, nutrient densities tend to be associated with disease in the direction opposite that of total energy intake.

TABLE 17-6 Possible Models That Account for Total Energy Intake in Epidemiologic Studies

1. *Residual model*
   Disease = nutrient residual
   Disease = nutrient residual + calories
2. *Standard multivariate model*
   Disease = calories + nutrient
3. *Energy partition model*
   Disease = calories (nutrient)[a] + calories (other)[b]
4. *Multivariate nutrient density model*
   Disease = (nutrient/calories) + calories

[a]Calories (nutrient) corresponds to caloric intake from a specific nutrient.

[b]Calories (other) corresponds to calories from other nutrients than those under study.

Source: Adapted from Willet.[18]

It is clear from this discussion that the interpretation of energy intake depends on the other variables added in the models. Therefore, the addition of predictors of energy intake to a model will also modify its

interpretation. For example, if we assume energy balance, the interpretation of total energy intake in a model in which physical activity and body size were included would be that of metabolic efficiency. In another example, when residuals of weight on height (remove the effect of height on weight) and residuals of height on weight (remove the effect of weight on height) are included in a model with energy intake, height represents lean body mass, weight represents fat, and energy intake represents both physical activity and metabolic efficiency.[18]

The relationships between dietary intake and disease are complex. Although the interpretation of energy intake varies with the analytic method, the association between absolute nutrient intakes and disease should take into account total energy intake because the observed association can simply reflect that of total energy intake and disease. The presentation of various analytic approaches could further strengthen the findings of an association between diet and disease.

GENETICS IN DIETARY ANALYSIS

The response to diet varies between individuals because of interindividual variations in genetic, epigenetic, and metabolic phenotype markers. Genetic variation may influence dietary intake and nutrient metabolism and thus the utility of a dietary biomarker as an adequate marker of dietary exposures. Technological advancement in analysis at the level of the genome (large-scale DNA sequencing), gene expression (transcriptomics), protein translation (proteomics), and metabolite network and fluxes (metabolomics) has been key to understanding the relationships between genome and phenotype in nutrient metabolism. In nutrition epidemiology, methods that currently incorporate genetic factors to improve our understanding of the effects of diet on risk of disease include (1) gene–diet interactions; (2) Mendelian randomization, and (3) genetic characteristics of endpoints (outcomes).

Variants in DNA sequences are called “mutations” when their prevalence is <1% and “polymorphisms” when they are common. Numerous polymorphisms in the human genome account for differences in the functions of proteins, which contribute to a variety of dietary responses and increase in familial disease risk. The ability to identify DNA polymorphisms from a small amount of DNA has increased rapidly allowing for the evaluation of several million single-nucleotide polymorphisms (SNPs) on a single DNA chip. Variations that are in close proximity on a DNA strand are typically strongly correlated, and thus one

chip can account for a large number of common variants. As a result, genome-wide association studies (GWAS) have been conducted for most common diseases, only a few gene–diet interactions have been identified. A recent example of gene–diet interactions (n = 649) in pediatrics is the interaction of breastfeeding and SNPs in the *IL12RB1*, *TLR9*, and *TSLP* genes both individually and jointly on food sensitivity.[119] Davis et al.[120] reported an interaction between PNPLA3 genotype and carbohydrate intake on fatty liver disease. Similarly, Christensen's findings suggested an interaction of methylenetetrahydrofolate reductase (MTHFR) polymorphism and low folate status, which was associated with a greater risk for NTDs rather than either variable alone.[121] As more studies are being published, clinically relevant findings may have implications for physicians' office counseling and personalized dietary guidelines.

In principle, the analyses of gene–diet interactions are not different from typical analyses of subgroups and interactions. When an interaction is significantly associated with the health outcome, the sample study is stratified according to the genotype for further analysis. However, the sample size required to identify significant interactions is typically four times larger than that needed to test for main effects.[122] Some of the challenges of GWAS are due to the fact that the number of variables examined is greater than the number of study subjects and that investigators are testing interactions between SNPs and one or more environmental exposures, in addition to determining the main effect of each SNP.[123] The latter authors recommend jointly testing for main and interaction effects, testing for population stratification, and using longitudinal data with multiple measurements of the environmental exposure to reduce measurement error issues. Adequate data are a usual limitation.

A second method to measure genetic factors responsible for variation in dietary responses is Mendelian randomization. This method uses genetic variants as proxy measures of nutritional exposure. There are three levels of evidence using Mendelian randomization. The first level is identified as the simple association between an SNP and risk of disease, assuming the genetic effect on disease is conferred only through the nutrient or food being investigated. The second level is when the genotype and intake of the corresponding dietary factor are associated with the disease. Finally, the third level is when both the genotype and the corresponding dietary factor are associated with disease and the association with diet is modified by the genotype; this combination of evidence provides the strongest support for causality.

Qi et al. defines Mendelian randomization as a method that "combines genetic and classical epidemiological analysis to infer causality for environmental exposures, based on the principle of Mendel's law of independent assortment."[124] Mendelian randomization is a method that is less likely to be affected by confounding and reverse causation. As opposed to environmental exposures (e.g., nutrients), genetic variants are not generally associated with the wide range of behavioral, social, and physiological factors that can confound associations.[125] As an example, in Mendelian randomization, postmeiotic germline genetic variants related to alcohol intake or intermediate phenotypes will not be influenced by the onset of disease. Typically, genetic variants are proxy for long-term differences in exposure levels (over a lifetime) rather than short-term differences in exposure levels (over the study period). Limitations include the need for large sample sizes and the inability to completely remove confounding (e.g., the same gene may metabolize related molecules).[125] Another problem with using Mendelian randomization is that developmental compensation that may occur through a polymorphic genotype being expressed during fetal or early postnatal development. These may influence development in such a way as to buffer against the effect of the polymorphism. A more significant limitation of this approach occurs when a genotype has a weak effect on nutritional status. In this case, it is very difficult to establish an association between genotype and disease risk. Despite these limitations, results from the Mendelian randomization can provide evidence of causality comparable with those of randomized clinical trials (RCTs).[125] Finally, Mendelian randomization can be used to select potential candidates for dietary intervention to be tested in a RCT before implementation.

The third method for measuring genetic factors responsible for variation in dietary responses is the use of genetic analyses to characterize outcomes. This method uses genetic variants to identify specific subset of cancers that may have different etiologies, or as proxy measures of disease progression. This method is most useful in the study of cancer, where DNA is extracted from premalignant or malignant tissue using similar techniques as those for germline DNA (entire sequence or one basepair of a tumor). Clinicians are typically most interested in how changes from normal to malignant tissue relate to prognosis and treatment. Diet and other environmental factors may be associated with certain changes in DNA. The identification of a specific genetic change being associated with an exposure increases the likelihood of an association being causal.

Systems genetics is an emerging discipline that is complementary to

traditional methods that emphasize the function of single genes or proteins. This approach has been defined as "a novel population-based approach for identifying the genetic architecture of complex (Quantitative) traits and gene-environment interactions with emphasis on gene networks."[126] Systems genetics allow the identification of new relationships between DNA sequence variation and intermediate phenotypes (e.g., transcript abundance, metabolomics, and proteomics), environment (e.g., nutrient), and disease outcomes.[127]

Presentation of Data

Data for individual foods are usually collected as ordinal categorical or semicontinuous variables, but are typically presented as continuous variables. However, the traditional presentation of nutritional epidemiologic data is based on rate ratios and rate differences for levels of exposure resulting in the categorization of data.

Approaches used to classify dietary data includes quantiles, standard round-numbered cutoff points or cutoff points using, for example, the recommended dietary allowances.[18] Regardless of the method used, categories have great advantages in dietary analysis. First, the investigator can visualize directly how the actual numbers of cases and noncases vary by level of intake. Second, the use of multiple categories makes no assumptions about the dose–response relationship, thus helping visualize whether the relationship is linear. Third, the use of categories may decrease the influence of outliers. However, it is important point that cutoff points are determined a priori rather than to maximize statistical significance in primary analysis. Alternative cutoff points that best describe the biological relationship can sometimes be justified in secondary analysis.

Arguments can also be made for using continuous variables. First, the greatest statistical power is provided by a continuous variable if the function reasonably fits the data. Second, a crudely categorized variable used as a covariate may not fully account for the effect of that variable, resulting in possible residual confounding. This is especially concerning when the primary variable is highly correlated with the confounder (e.g., fat and total energy intake). Third, the use of continuous variables may facilitate comparisons among studies because a single relative risk is reported (e.g., relative risk for 100 mg of cholesterol per day) that does not depend on the distribution of the dietary factor in the particular population or on the studies' cutoff points. However, caution is warranted in making

quantitative comparisons among studies.

The presentation of a single relative risk and its confidence interval provides a more compact presentation of data than relative risks for several categories. An example is the presentation of the relative risks for one serving per day for each food when many foods need to be summarized. Tests for nonlinearity, such as the addition of a quadratic term, can be used to evaluate the presence of nonlinearity but require large sample sizes and may not always describe correctly the data. For example, the analysis by quintiles could show that a substantial difference in risk exists between the first and second quintile but not between the second through the fifth quintile. The test for a simple linear relationship using continuous data might not be significant (leading to a report of no association) or it might be significant but an added exponential term might be not significant (leading to the report of a linear relationship). The use of locally smoothed regression curves represents an approach that combines some of the advantages of both categorical and continuous analysis.

A few aspects of graphical presentations are particularly relevant to nutritional epidemiology, which include single variable effects, actual versus predicted relationships, joint effects, and locally smoothed regression curves and regression splines.[18]

Other Considerations in Dietary Analysis

Additional methods can be used to improve our understanding of the relationship between diet and disease outcome, which have been reviewed elsewhere.[18] In addition to intake of nutrients, analysis of food intake can be conducted to support the association between diet and disease outcome. The use of multivariate analysis is especially important to help identify whether the relationship between exposure and disease is due to another correlated factor. Repeated measure measurements of dietary intake provide opportunities for reducing the effect of measurement error and evaluate various possible temporal relationships between a dietary factor and a disease outcome. Examination of alternative temporal relationships can be useful to reduce the likelihood of missed associations and to provide insight in the pathogenesis of disease. Dietary pattern analysis may be complementary to traditional analysis of nutrients and food and occasionally reveal missed associations. To correct for error of measurements, data from validation and reproducibility studies can be used. Finally, pooled analysis of select diet and disease relationships can enhance our understanding of the effect of diet on disease.

TYPES OF STUDIES

The case of the relationship between folate deficiency and NTDs will be used to illustrate the contribution of a variety of study designs to the determination of a causal relationship between nutrient intake and an important health outcome. In addition, this example shows how the knowledge of a relationship between a nutrient and an outcome might not be sufficient to develop prevention interventions. Finally, it also demonstrates the value of nutritional epidemiology in establishing and evaluating public health policies.

Descriptive Epidemiology

The description of variation in rates of disease among different populations and locations, and over time, is generally called descriptive epidemiology. An environmental role in the development of NTDs was suspected from ecologic (differences in rates among geographic areas) and migration studies. The frequency of NTDs was higher in the northern United Kingdom and decreased when populations moved from high-prevalence areas in the United Kingdom to the United States. High prevalence of NTDs seemed to plague disadvantaged populations. For example, an epidemic of NTDs was observed during the Great Depression in Boston.[128] Another example comes from the decreasing rates of NTDs in Ireland over the course of one decade concurrent with the use of fortified breakfast cereals.[129]

Clinical Observations and Intervention Studies

Clinical observations often provide important leads for further study. The finding of a relationship between socioeconomic background and NTDs resulted in a study of mothers' and children's folate status in the United Kingdom. Smithells et al. performed blood tests to study micronutrients and found that folate, among others, was low among mothers whose infants had NTDs.[130] It was also observed that drugs interfering with folate metabolism were associated with the risk of NTDs.[131]

Risk of recurrence was examined in a nonrandomized prevention trial of multivitamin and mineral supplementation in England and suggested a beneficial effect of supplementation.[132] Supplementation started at entry to prenatal care for all women. Women who received supplements before six weeks of gestation (which corresponds to the cutoff point for neural tube closure) were further compared with women who received supplements later. Other smaller intervention studies showed nonsignificant lower risk

of NTDs among supplemented women. Overall, intervention studies showed a fourfold decrease in risk of NTDs among women taking a multivitamin, but the studies have been criticized because many of the data were from nonrandomized trials.

Case-Control and Cohort Studies of Vitamin Supplements

In case-control studies, previous diet or use of supplements by cases (or, in this example, mother of the case) is compared with that of controls. In cohort studies, nutrient data are collected before the occurrence or recurrence of disease; thus, these studies tend to take longer and are more expensive. However, cohort studies are less subject to methodologic bias.

Only four studies examined folate intake from the diet; others used information on supplements containing folic acid. Mulinare et al. used a birth defects registry to identify children born with NTDs and retrospectively asked about periconceptional multivitamin use (a period of up to 14 years before the interview).[133] The risk of NTDs was approximately 60% lower for women who reported taking supplements. In contrast, Mills et al. did not find a significant relationship between folate and NTDs.[134] Shaw et al. found a similar reduction in NTD risk when folate supplementation was started after three months of gestation, raising concerns about the causality between folate and NTDs.[135] Milunsky et al. conducted a prospective cohort study of multivitamin use and NTDs by telephoning mothers at the end of the first trimester of pregnancy to inquire about the type of supplement taken.[136] The findings showed a strong inverse relationship between multivitamin intake during the first 6 weeks of pregnancy and protection against NTDs.

In summary, all case-control and cohort studies except one supported a protective effect of periconceptional multivitamin use. It was unlikely that results observed in most studies were attributable solely to chance because of the variety of design and the choice of controls. However, confounding factors could have led to these findings. For example, NTDs occurred more often in populations of lower socioeconomic background, so that, in principle, socioeconomic background could have been a confounding factor. Also, the association between folate and NTDs might have been attributable to other vitamins in the multivitamin preparation.

Case-Control and Cohort Studies of Serum Folate

Daly et al. collected blood samples prospectively from more than 50,000

women attending their first antenatal clinic in Dublin maternity hospitals from 1986 to 1990.[137] They used a nested case-control design (sample from all cases and a matched sample of mothers who did not develop an NTD pregnancy) comparing early pregnancy blood folate concentrations in 81 women with pregnancies subsequently affected by NTDs and in a systematic sample of 247 control pregnancies. Folate levels were found to be lower for mothers who later had children who developed NTDs. The same authors found that a woman's risk of having a child with an NTD was associated with early pregnancy red blood cell folate concentrations in a continuous dose–response relationship.[137] However, in the absence of double-blind randomization, the social and reproductive differences between mothers who took the supplement and those who were considered part of the control group (mothers received supplements after the periconceptional period or refused to take the supplement) led to uncertainty as to the significance of the results.

Randomized Trials

The evidence for the relationship between folate intake and NTDs from randomized trials comes from two studies. One was on the *recurrence* of NTDs (history of previous pregnancy resulting in an NTD) conducted by the Medical Research Council (MRC) Vitamin Study Research Group. This was a randomized trial conducted in five European countries, Israel, and Canada.[138] The other trial was on the *occurrence* of NTDs (first pregnancy resulting in an NTD), conducted in Hungary.[139] In the MRC study of recurrence, women were randomized in a 2 × 2 factorial design to 4 mg of folate versus placebo or to other vitamins typically contained in multivitamins versus placebo. The trial was stopped early because of the findings of a 72% protection rate for folate. The Hungarian study used a multivitamin with trace elements and folate versus trace elements and vitamin C. This trial was also stopped because of a high occurrence of NTDs in the placebo group.

Implications

The findings from these studies led to policy responses that were controversial and unclear. A variety of strategies could have been used to improve the folate status of the population: (1) increase intakes of foods rich in folate, (2) recommend routine use of supplements, or (3) suggest the fortification of foods.

Although increasing intakes of fruits and vegetables rich in folate

would be beneficial in many respects, its effect on occurrence of disease would be minimal. Not only would large amounts of folate be necessary to increase plasma levels but also national programs to increase fruit and vegetable intake have not been particularly successful in the past. In addition, the bioavailability of folate in foods is not as good as that of supplemental folate.[140] Cuskelly et al. conducted a randomized trial of strategies to provide a daily intake of 0.4 mg of folate through either supplementation, food fortification, free provision of folate-rich foods or dietary advice regarding folate-rich foods compared with a control group over a three-month period.[141] Concentration of folate in red cells increased significantly only in the groups receiving folate supplements or folate-fortified foods. This study reinforces the concern of the effectiveness of dietary interventions in view of social and economic barriers.

The strategy that consisted of recommending folate supplementation to women during their reproductive years was supported by most epidemiologic studies and was endorsed by the CDC in 1992. To come up with recommendations, the percentage of NTDs in the population that is potentially preventable by supplementation with folic acid should be calculated. The preventable fraction (PF) is the proportion of cases that would not have occurred in the absence of exposure among the exposed population or the total population. The proportion of cases in a population that is not exposed to supplement is labeled P_1. Therefore, the percentage of NTDs in a population that are potentially preventable by supplementation with folic acid can be estimated as the preventable fraction (PF) multiplied by the proportion of cases in a population that were not already exposed to supplements (P_1): ($PF \times P_1$). The preventable fraction is (1−RR), where the relative risk is obtained from an observational study or a randomized trial. For example, in the prospective study by Milunsky et al., the relative risk was 0.27 and the proportion of cases that were unexposed—that is, the mother did not take folic acid supplements during the critical period—was 0.82. The population preventable fraction in this example would be equivalent to 0.60, which is calculated based on $(1-RR) \times P_1 = (1-0.27) \times 0.82$.[136] In 1992, the Centers for Disease Control estimated that 50% of cases of NTDs in the United States could be prevented by universal supplementation with folic acid among women in the reproductive ages. In absolute numbers that meant that about 1500 of the approximately 3000 cases of spina bifida or anencephaly that occurred annually in the United States would be prevented.

The CDC recommended the use of 0.4 mg of folate daily in women who planned a pregnancy and 4 mg for those who had a history of NTD pregnancies. In addition, folic acid supplement of 0.4–0.8 mg was recommended by the Preventive Medicine Task Force for women who might become pregnant[142]; failure of a physician to advise women appropriately might well be considered negligence if a child was affected with a NTD. Although multivitamin consumption was affordable for most, the timing of supplementation and compliance were of concern. Many institutions supported the notion of informing women about folate supplementation; failure to provide this information was considered negligent. Targeting supplementation to a population at risk was difficult because there were no clear affordable screening tools available to identify subjects at risk and the population as a whole seemed to be at risk, based on the study by Daly et al.[142]

Finally, the strategy of fortifying the food supply had been used long before with success in many countries (e.g., with iron and iodine). Fortification of the food supply would have an impact on the entire population and would not require ongoing educational campaigns. Nevertheless, the level of fortification and the type of foods to be fortified were the center of intense debate. The main concern was the masking of vitaminB_{12} deficiency. However, given that about 25% of the population of the United States consumes multivitamins with 0.4 mg of folate, the rare report of neurologic complications associated with vitamin $B_{12}$was reassuring. Nevertheless, the US Food and Drug Administration (FDA) considered that levels of folate consumption from food and supplements above 1 mg of folate daily were a matter of concern.

Although flour was a good candidate for fortification because it is consumed by most subjects in the population, the amount of consumption varies in the population. Therefore, simulations of food fortification of specific foods were performed by artificially increasing the folate content of products made of flour in existing food intake databases. Results from these simulations showed that folate intakes higher than 0.4 mg would have to be tolerated to increase folate intake of the entire population. Using this method, Oakley et al. showed that fortification with 0.35 mg of folate in 100 g of flour would increase the average intake by 0.25 mg, but 30% of women would still have a total folate intake below the recommended 0.4 mg and 15% of the women would have an intake above 1 mg per day (for most multivitamin users). Fortification with 0.14 mg of folate in 100 g of flour would result in an average increase of only 0.1 mg

per day.[143] The FDA recommended the use of 0.14 mg of folate in 100 g of flour; this was implemented in 1998. Since then, fortification of flour became mandatory in the United States, Canada, and Chile which substantially improved folate and homocysteine status while NTDs rates fell by between 31% and 78%.[144] As of November, 2011, 63 countries now have guidelines to fortify and 57 actually do fortify cereal grains of one kind or another.[145] The CDC has estimated that the spina bifida rate declined 25% from 1995 (prefortification) to 2000, and there was a further decline of 13% from 2000 to 2005.[146] In a review that included intervention studies with folic acid supplements, folate fortification programs, and prospective studies, Heseker et al. concluded that the minimal rate achievable by folate intake is approximately 5–8 per 10,000 pregnancies, which is only modestly below the United States rate after fortification.[147] Johnston has estimated that the reduction of NTDs in the United States after fortification (approximately 27%) could be increased substantially by increasing the amount of folic acid that is currently added to cereal grain.[148] However, the fortification of flour is considered suboptimal to prevent spina bifida and the debate continues.[149] Tragically, recommendations alone in other countries such as in France did not influence trends in NTD.[150]

The demonstration that folic acid plays a major role in the prevention of *recurrence and occurrence* of NTDs stimulated further research on the possible mechanisms underlying the relationship and a possible nutrient–gene relationship. The association of a polymorphism in a folic acid–metabolizing gene with the risk of NTD strongly supports the role of folate in the etiology of NTDs and could indicate a group with a higher folate requirement. A polymorphism in the 5,10-methylenetetrahydrofolate reductase gene is associated with a reduction of activity of that specific enzyme. An increase in the prevalence of this polymorphism was observed in infants born with NTDs and in their parents compared with controls.[151] The possible role of vitamin B_{12} in NTDs is still unclear, as reviewed elsewhere.[152]

The findings on folate and the risk of NTDs have given credibility to the search for other examples in which suboptimal intakes of nutrients could lead to high risk of chronic disease. The case of folate and NTDs was resolved in a relatively short time, in part because of the short interval (first 6 weeks of pregnancy) of exposure during which a nutritional imbalance could result in detrimental effects. In addition, the period between exposure and outcome was relatively short, the exposure could be

evaluated by dietary assessment techniques and biomarkers, safe randomized trials could be conducted, and the strength of the relationship was large. Although a causal relationship is typically based on randomized clinical trials, the case of folate and NTDs suggests that a combination of study approaches can provide sufficient evidence when randomized studies cannot be easily performed.

SUMMARY

Nutrition epidemiology has been highly successful in identifying the responsible factors in many diseases, including scurvy, pellagra, blindness, and spina bifida. Nutritional epidemiology can potentially contribute substantially to our understanding of diseases in children and to the relationship between diet during childhood and risk of chronic diseases and cancer. The latency period between dietary exposure and disease can be years rather than weeks. Support for the long-term effects of diet comes from studies of diabetes and cardiovascular disorders that led to the Barker hypothesis of fetal origin of chronic disease, as well as from studies of cancer. A variety of approaches will provide the best evidence on these relationships, particularly when used in conjunction with clinical, biochemical, and molecular data.

REFERENCES

1. Lind J. *A Treatise on Scurvy*. London, UK: A. Millar; 1753.
2. Farrell RJ, Kelly CP. Current concepts: celiac sprue. *N Engl J Med*. 2002;346:17180–17188.
3. Dicke WK. *Investigation of the Harmful Effects of Certain Types of Cereals on Patients With Coeliac Disease [Doctoral Thesis]*. Utrecht, The Netherlands: University of Utrecht; 1950.
4. Dicke WK, Weijers HA, Van de Kramer JH. The presence in wheat of a factor having a deleterious effect in cases of celiac disease. *Acta Paediatr*. 1953;42:34–36.
5. Nicklas TA, Myers L, Berenson GS. Dietary fiber intake of children: the Bogalusa Heart Study. *Pediatrics*. 1995;96:988–994.
6. Jacobson HN, Stanton JL. Pattern analysis in nutrition research. *Clin Nutr*.1996;5:249–253.
7. Illner AK, Freisling H, Boeing H, et al. Review and evaluation of innovative technologies for measuring diet in nutritional epidemiology. *Int J Epidemiol*. 2012;41(4):1187–1203.
8. Neuhouser ML, Lilley S, Lund A, Johnson DB. Development and validation of a beverage and snack questionnaire for use in evaluation of school nutrition policies. *J Am Diet Assoc*. 2009;109(9):1587–1592.
9. Townsend MS, Kaiser LL, Allen LH, et al. Selecting items for a food behavior

checklist for a limited-resource audience. *J Nutr Educ Behav*. 2003;35(2):69–77. Erratum in: *J Nutr Educ Behav*. 2003;35(4):224.
10. Thompson FE, Subar AF, Loria CM, et al. Need for technological innovation in dietary assessment. *J Am Diet Assoc*. 2010;110(1):48–51.
11. Rockett HR, Berkey CS, Colditz GA. Evaluation of dietary assessment instruments in adolescents. *Curr Opin Clin Nutr Metab Care*. 2003;6:557–562.
12. Feskanich D, Rockett HR, Colditz GA. Modifying the Healthy Eating Index to assess diet quality in children and adolescents. *J Am Diet Assoc*. 2004;104:1375–1383.
13. Isasi CR, Soroudi N, Wylie-Rosett J. Youth WAVE Screener: addressing weight-related behaviors with school-age children. *Diabetes Educ*. 2006;32:415–422.
14. McPherson RS, Hoelscher DM, Alexander M, et al. Dietary assessment methods among school-aged children: validity and reliability. *Prev Med*. 2000;31:S11–S33.
15. Burrows T, Golley RK, Khambalia A, et al. The quality of dietary intake methodology and reporting in child and adolescent obesity intervention trials: a systematic review. *Obes Rev*. 2010;13(12):1125–1138.
16. Schoeller DA. Recent advances from application of doubly labeled water to measurement of human energy expenditure. *J Nutr*.1999;129:1765–1768.
17. Roberts SB, Dietz W, Sharp T, et al. Multiple laboratory comparison of the doubly labeled water technique. *Obes Res*. 1995;3:3–13.
18. Willet W. *Nutritional Epidemiology. Monographs in Epidemiology and Biostatistics.* Vol 40. 3rd ed. New York, NY: Oxford University Press; 2013.
19. Baxter SD, Thompson WO, Litaker MS, et al. Low accuracy and low consistency of fourth-graders' school breakfast and school lunch recalls. *J Am Diet Assoc*. 2002;102:386–395.
20. Bandini LG, Schoeller HN, Cyr HN, et al. Validity of reported energy intake in obese and non-obese adolescents. *Am J Clin Nutr*. 1990;52:421–425.
21. Livingstone MBE, Prentice AM, Coward WA, et al. Validation of estimates of energy intake by weighed dietary record and diet history in children and adolescents. *Am J Clin Nutr*. 1992;56:29–35.
22. Harnack L, Himes JH, Anliker J, et al. Intervention-related bias in reporting of food intake by fifth-grade children participating in an obesity prevention study. *Am J Epidemiol*. 2004;160:1117–1121.
23. Weber JL, Lytle L, Gittelsohn J, et al. Validity of self-reported dietary intake at school meals by American Indian children: the pathways study. *J Am Diet Assoc*. 2004;104:746–752.
24. Baxter SD, Thompson WO, Davis HC, et al. Impact of gender, ethnicity, meal component, and time interval between eating and reporting on accuracy of fourth-graders' self-reports of school lunch. *J Am Diet Assoc*. 1997;97:1293–1298.
25. Quandt SA. Variation in nutrient intake of infants and its implications for collecting reliable dietary intake data. *Am J Phys Anthropol*. 1987;73:515–523.
26. Stein AD, Shea S, Basch CE, et al. Variability and tracking of nutrient intakes of preschool children based on multiple administrations of the 24-hour dietary recall. *Am J Epidemiol*. 1991;134:1427–1437.
27. Vucic V, Glibetic M, Novakovic R, et al. Dietary assessment methods used for low-income populations in food consumption surveys: a literature review. *Br J Nutr*. 2009;101(S2):S95–S101.
28. Fisher JO, Johnson RK, Lindquist C, et al. Influence of body composition on the accuracy of reported energy intake in children. *Obes Res.* 2000;8:597–603.
29. Thompson FE, Byers T. Dietary assessment resource manual. *J Nutr*. 1994;124:2245S–317S.

30. Black AE, Cole TJ, Wiles SJ, et al. Daily variation in food intake of infants from 2 to 18 months. *Hum Nutr Appl Nutr*. 1983;37:448–458.
31. Nelson M, Black AE, Morris JA, et al. Between- and within-subject variation in nutrient intake from infancy to old age: estimating the number of days required to rank dietary intakes with desired precision. *Am J Clin Nutr*. 1989;50:155–167.
32. Birch LL, Johnson SL, Andresen G, et al. The variability of young children's energy intake. *N Engl J Med*. 1991;324:232–235.
33. Champagne CM, Baker NB, DeLany JP, et al. Assessment of energy intake underreporting by doubly labeled water and observations on reported nutrients intakes in children. *J Am Diet Assoc.* 1998;98:426–430.
34. O'Connor J, Ball EJ, Steinbeck KS, et al. Comparison of total energy expenditure and energy intake in children aged 6–9 y. *Am J Clin Nutr*. 2001;74:643–649.
35. Bratteby LE, Sandhagen B, Fan H, et al. Total energy expenditure and physical activity as assessed by the doubly labeled water method in Swedish adolescents in whom energy intake was underestimated by 7-d diet records. *Am J Clin Nutr*. 1998;67:905–911.
36. Field AE, Peterson KE, Gortmaker SL, et al. Reproducibility and validity of a food frequency questionnaire among fourth to seventh grade inner-city school children: implications of age and day-to-day variation in dietary intake. *Public Health Nutr*. 1999;2:293–300.
37. Klohe DM, Clarke KK, George GC, et al. Relative validity and reliability of a food frequency questionnaire for a triethnic population of 1-year-old to 3-year-old children from low income families. *J Am Diet Assoc*. 2005;105:727–734.
38. Blum RE, Wei EK, Rockett HR, et al. Validation of a food frequency questionnaire in Native American and Caucasian children 1 to 5 years of age. *Matern Child Health J*. 1999;3:167–172.
39. Maruti SS, Feskanich D, Rockett HR, et al. Validation of adolescent diet recalled by adults. *Epidemiology*. 2006;17:226–229.
40. Johnson RK, Driscoll P, Goran MI. Cross-calibration of energy intake by 24-hour recall against total energy expenditure by doubly labeled water in young children. *J Am Diet Assoc*. 1996;96:1140–1144.
41. Kaskoun MC, Johnson R, Goran MI. Comparison of energy intake by food frequency questionnaire with total energy expenditure by doubly labeled water in young children. *Am J Clin Nutr*. 1994;60:43–47.
42. Perks SM, Roemmich JN, Sandow-Pajewski M, et al. Alterations in growth and body composition during puberty. iv. Energy intake estimated by the youth-adolescent food-frequency questionnaire: validation by the doubly labeled water method. *Am J Clin Nutr*. 2000;72:1455–1460.
43. Simons-Morton BG, Forthofer R, Huang IW, et al. Reliability of direct observation of school-children's consumption of bag lunches. *J Am Diet Assoc*. 1992;92:219–221.
44. Raisler J, Alexander C, O'Campo P. Breastfeeding and infant illness: a dose-response relationship? *Am J Public Health*. 1999;89:25–30.
45. Habicht JP, DaVanzo J, Butz WP. Does breastfeeding really save lives, or are apparent benefits due to biases? *Am J Epidemiol*. 1986;123:279–290.
46. Fawzi WW, Herrera MG, Nestel P, et al. A longitudinal study of prolonged breastfeeding in relation to child undernutrition. *Int J Epidemiol*. 1998;27:255–260.
47. Butte NF, Wong WW, Hopkinson JM, et al. Energy requirements derived from total energy expenditure and energy deposition during the first 2 y of life. *Am J Clin Nutr*. 2000;72:1558–1569.

48. Jones PJ, Winthrop AL, Schoeller DA, et al. Validation of doubly labeled water for assessing energy expenditure in infants. *Pediatr Res*. 1987;21:242–246.
49. Brooks GA, Butte NF, Rand WM, et al. Chronicle of the Institute of Medicine physical activity recommendation: how a physical activity recommendation came to be among dietary recommendations. *Am J Clin Nutr*. 2004;79:921S–930S.
50. Brown KH, Black RE, Becker S, et al. Consumption of foods and nutrients by weanlings in rural Bangladesh. *Am J Clin Nutr*. 1982;36:878–889.
51. Piwoz EG, Creed de Kanashiro H, Lopez de Romana G, et al. Potential for misclassification of infants' usual feeding practices using 24-hour dietary assessment methods. *J Nutr*. 1995;125:57–65.
52. Riordan JM, Koehn M. Reliability and validity testing of three breastfeeding assessment tools. *J Obstet Gynecol Neonatal Nurs*. 1997;26:181–187.
53. Andersen LF, Lande B, Arsky GH, et al. Validation of a semi-quantitative food-frequency questionnaire used among 12-month-old Norwegian infants. *Eur J Clin Nutr*. 2003;57:881–888.
54. Zeitlin MF, Ahmed NU. Nutritional correlates of frequency and length of breastfeeds in rural Bangladesh. *Early Hum Dev*. 1995;41:97–110.
55. Beaudry M, Dufour R, Marcoux S. Relation between infant feeding and infections during the first six months of life. *J Pediatr*. 1995;126:191–197.
56. Vähätalo L, Barlund S, Hannila ML, et al. Relative validity of a dietary interview for assessing infant diet and compliance in a dietary intervention trial. *Matern Child Nutr*. 2006;2:181–187.
57. Lohman TG. Applicability of body composition techniques and constants for children and youths. *Exerc Sports Sci Rev*. 1986;14:325–357.
58. Parizkova J. Total body fat and skinfold thickness in children. *Metabolism*. 1961;10:794–807.
59. Seltzer CC, Goldman RF, Mayer J. The triceps skinfold as a predictive measure of bone density and body fat in obese adolescent girls. *Pediatrics*. 1965;36:212–218.
60. Young CM, Sipin SS, Roe DA. Body composition of preadolescent and adolescent girls. *J Am Diet Assoc*. 1968:67:25–31.
61. Harsha DW, Frerichs RR, Berenson GS. Densitometry and anthropometry of black and white children. *Hum Biol*. 1978;50:261–280.
62. Roche AF, Siervogel RM, Chumlea C, et al. Grading body fatness from limited anthropometric data. *Am J Clin Nutr*. 1981;34:2231–2238.
63. Maynard LM, Wisemandle W, Roche AF, et al. Childhood body composition in relation to body mass index. *Pediatrics*. 2001;107:344–350.
64. Michielutte R, Diseker RA, Corbett WT, et al. The relationship between weight-height indices and the triceps skinfold measure among children age 5 to 12. *Am J Public Health*. 1984;74:604–606.
65. Frontini MG, Bao W, Elkasabany A, et al. Comparison of weight-for-height indices as a measure of adiposity and cardiovascular risk from childhood to young adulthood: the Bogalusa Heart Study. *J Clin Epidemiol*. 2002;54:817–822.
66. Mei Z, Grummer-Strawn LM, Pietrobelli A, et al. Validity of body mass index compared with other body-composition screening indexes for the assessment of body fatness in children and adolescents. *Am J Clin Nutr*. 2002;75:978–985.
67. Willett K, Jiang R, Lenart E, et al. Comparison of bioelectrical impedance and BMI in predicting obesity-related medical conditions. *Obesity (Silver Spring)*. 2006;14:480–490.
68. Eisenmann JC, Heelan KA, Welk GJ. Assessing body composition among 3- to 8-year-old children: anthropometry, BIA, and DXA. *Obes Res*. 2004;12:1633–1640.
69. Dietz WH, Robinson TN. Use of the body mass index (BMI) as a measure of

overweight in children and adolescents. *J Pediatr*. 1998;132:191–193.
70. Gasser T, Ziegler P, Seifert B, et al. Measures of body mass and of obesity from infancy to adulthood and their appropriate transformation. *Ann Hum Biol*. 1994;21:111–125.
71. Himes JH, Faricy A. Validity and reliability of self-reported stature and weight of US adolescents. *Am J Hum Biol*. 2001;13:255–260.
72. Lee K, Valeria B, Kochman C, et al. Self-assessment of height, weight, and sexual maturation: validity in overweight children and adolescents. *J Adolese Health*. 2002;39:346–352.
73. Potischman N. Biologic and methodologic issues for nutritional biomarkers. *J Nutr*. 2003;133(S3):875S-880S.
74. Gibson RS. *Principles of Nutritional Assessment*. 2nd ed. New York, NY: Oxford University Press; 2005.
75. McKeown-Eyssen GE, Yeung KS, Bright-See E. Assessment of past diet in epidemiologic studies. *Am J Epidemiol*. 1986;124:94–103.
76. Marklund M, Landberg R, Andersson R, et al. Alkylresorcinol metabolism in Swedish adults is affected by factors other than intake of whole-grain wheat and rye. *J Nutr*. 2012;142(8):1479–1486.
77. Bingham SA. Biomarkers in nutritional epidemiology. *Public Health Nutr*. 2002;5:821–827.
78. Rader JI. Folic acid fortification, folate status and plasma homocysteine. *J Nutr*. 2002;132:2466S-2470S.
79. Bounous G, Kongshavn PAL. Influence of protein type in nutritionally adequate diets on the development of immunity. In: Friedman M, ed. *Absorption and Utilization of Amino Acids*. Vol II. Boca Raton, FL: CRC Press; 1989:219–233.
80. Sanchez A, Hubbard R. Dietary protein modulation of serum cholesterol: the amino acid connection. In: Friedman M, ed. *Absorption and Utilization of Amino Acids*. Vol II. Boca Raton, FL: CRC Press; 1989:247–273.
81. Poppitt SD, Kilmartin P, Butler P, et al. Assessment of erythrocyte phospholipid fatty acid composition as a biomarker for dietary MUFA, PUFA or saturated fatty acid intake in a controlled cross-over intervention trial. *Lipids Health Dis*. 2005;4:30.
82. Baylin A, Campos H. The use of fatty acid biomarkers to reflect dietary intake. *Curr Opin Lipidol*. 2006;17:22–27.
83. Ovesen L, Boeing H. The use of biomarkers in multicentric studies with particular consideration of iodine, sodium, iron, folate and vitamin D. *Eur J Clin Nutr*. 2002;56:S12–S17.
84. Baylin A, Kim MK, Donovan-Palmer A, et al. Fasting whole blood as a biomarker of essential fatty acid intake in epidemiologic studies: comparison with adipose tissue and plasma. *Am J Epidemiol*. 2005;162:373–381.
85. Erlinger TP, Guallar E, Miller ER III, et al. Relationship between systemic markers of inflammation and serum betacarotene levels. *Arch Intern Med*. 2001;161:1903–1908.
86. Chevion M, Berenshtein E, Stadtman ER. Human studies related to protein oxidation: protein carbonyl content as a marker of damage. *Free Radic Res*. 2000;33:S99–S108.
87. Russell RM, Krasinski SD, Dawson-Hughes B. Indices of fat-soluble vitamin states. *Clin Nutr*. 1984;3:161–168.
88. Bingham S, Luben R, Welch A, et al. Epidemiologic assessment of sugars consumption using biomarkers: comparisons of obese and nonobese individuals in the European prospective investigation of cancer Norfolk. *Cancer Epidemiol*

Biomarkers Prev. 2007;16(8):1651–1654.
89. Birch LL, Davidson KK. Family environmental factors influencing the developing behavioral controls of food intake and childhood overweight. *Pediatr Clin N Am.* 2001;48:893–907.
90. Nicklas TA, Webber LS, Berenson GS. Studies of consistency of dietary intake during the first four years of life in a prospective analysis: Bogalusa Heart Study. *J Am Coll Nutr*. 1991;10:234–241.
91. Resnicow K, Smith M, Baranowski T, et al. Two-year tracking of children's fruit and vegetable intake. *J Am Diet Assoc*. 1998;98:785–789.
92. Welten DC, Kemper HC, Post GB, et al. Longitudinal development and tracking of calcium and dairy intake from teenager to adult. *Eur J Clin Nutr*. 199;51:612–618.
93. Wang Y, Bentley ME, Zhai F, et al. Tracking of dietary intake patterns of Chinese from childhood to adolescence over a six-year follow-up period. *J Nutr*. 2002;132:430–438.
94. Lake AA, Mathers JC, Rugg-Gunn AJ, et al. Longitudinal change in food habits between adolescence (11–12 years) and adulthood (32–33 years): the ASH30 Study. *J Public Health (Oxford)*. 2006;28:10–16.
95. Nicklas TA, Elkasabany A, Srinivasan SR, et al. Trends in nutrient intake of 10-year-old children over two decades (1973–1994): the Bogalusa Heart Study. *Am J Epidemiol*. 2001;153:969–977.
96. Patterson E, Wärnberg J, Kearney J, Sjöström M. The tracking of dietary intakes of children and adolescents in Sweden over six years: the European Youth Heart Study. *Int J Behav Nutr Phys Act*. 2009;11(6):91.
97. Bertheke Post G, de Vente W, Kemper HC, Twisk JW. Longitudinal trends in and tracking of energy and nutrient intake over 20 years in a Dutch cohort of men and women between 13 and 33 years of age: the Amsterdam growth and health longitudinal study. *Br J Nutr*. 2001;85(3):375–385.
98. Berenson GS, Srinivasan SR, Nicklas TA. Atherosclerosis: a nutritional disease of childhood. *Am J Cardiol*. 1998;82:22T–29T.
99. Kallio MJ, Salmenpera L, Siimes MA, et al. Tracking of serum cholesterol and lipoprotein levels from the first year of life. *Pediatrics*. 1993;91:949–954.
100. Moilanen T, Viikari J, Rasanen L, et al. Three-year tracking of serum fatty acids in Finnish boys and girls. *Atherosclerosis*. 1987;67:191–197.
101. Lloyd LJ, Langley-Evans SC, McMullen S. Childhood obesity and risk of the adult metabolic syndrome: a systematic review. *Int J Obes (Lond)*. 2012;36(1):1–11.
102. Park MH, Falconer C, Viner RM, Kinra S. The impact of childhood obesity on morbidity and mortality in adulthood: a systematic review. *Obes Rev*. 2012;13(11):985–1000.
103. Thompson PD, Buchner D, Pina IL, et al. Exercise and physical activity in the prevention and treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). *Circulation.* 2003;107:3109–3116.
104. Sallis JF. Measuring physical activity environments: a brief history. *Am J Prev Med*. 2009;36(4 S):S86–S92.
105. Adamo KB, Prince SA, Tricco AC, et al. A comparison of indirect versus direct measures for assessing physical activity in the pediatric population: a systematic review. *Int J Pediatr Obes*. 2009;4(1):2–27.

106. Sirard JR, Pate RR. Physical activity assessment in children and adolescents. *Sports Med*. 2001;31(6):439–454.
107. Pate RR, Freedson PS, Sallis JF, et al. Compliance with physical activity guidelines: prevalence in a population of children and youth. *Ann Epidemiol*. 2002;12(5):303–308.
108. Biddle SJH, Gorely T, Pearson N, Bull FC. An assessment of self-reported physical activity instruments in young people for population surveillance: project ALPHA. *Int J Behav Nutr Phys Act*. 2011;8:1.
109. Kowalski KC, Crocker PRE, Faulkner RA. Validation of the physical activity questionnaire for older children. *Pediatr Exerc Sci*. 1997;9(2):174–186.
110. Dwyer GM, Hardy LL, Peat JK, Baur LA. The validity and reliability of a home environment preschool-age physical activity questionnaire (Pre-PAQ). *Int J Behav Nutr Phys Act*. 2011;8:86.
111. Burdette HL, Whitaker RC. A national study of neighborhood safety, outdoor play television viewing and obesity in preschool children. *Pediatrics*. 2005;116:657.
112. Tremblay MS, LeBlanc AG, Kho ME, et al. Systematic review of sedentary behaviour and health indicators in school-aged children and youth. *Int J Behav Nutr Phys Act*. 2011;8:98.
113. Katzmarzyk PT, Malina RM, Song TM, Bouchard C. Physical activity and health-related fitness in youth: a multivariate analysis. *Med Sci Sports Exerc*. 1998;30(5):709–714.
114. Blaes A, Baquet G, Fabre C, et al. Is there any relationship between physical activity level and patterns, and physical performance in children? *Int J Behav Nutr Phys Act*. 2011;8:122.
115. Michels KB, Willett WC. Self-administered semiquantitative food frequency questionnaires: patterns, predictors, and interpretation of omitted items. *Epidemiology*. 2009;20:295–301.
116. Rimm EB, Giovannucci EL, Stampfer MJ, et al. Reproducibility and validity of a expanded self-administered semiquantitative food frequency questionnaire among male health professionals. *Am J Epidemiol*. 1992;135:1114–1126.
117. Anonymous. Human Energy Requirements. Scientific Background Papers From the Joint FAO/WHO/UNU Expert Consultation. October 17–24, 2001. Rome, Italy. *Public Health Nutr*. 2005;8(7A):929–1228.
118. Subar AF, Kipnis V, Troiano RP, et al. Using intake biomarkers to evaluate the extent of dietary misreporting in a large sample of adults: the OPEN study. *Am J Epidemiol*. 2003;158(1):1–13.
119. Hong X, Wang G, Liu X, et al. Gene polymorphisms, breast-feeding, and development of food sensitization in early childhood. *J Allergy Clin Immunol*. 2011;128(2):374–381.
120. Davis JN, Lê KA, Walker RW, et al. Increased hepatic fat in overweight Hispanic youth influenced by interaction between genetic variation in PNPLA3 and high dietary carbohydrate and sugar consumption. *Am J Clin Nutr*. 2010;92(6):1522–1527.
121. Christensen B, Arbour L, Tran P, et al. Genetic polymorphisms in methylenetetrahydrofolate reductase and methionine synthase, folate levels in red blood cells, and risk of neural tube defects. *Am J Med Genet*. 1999;84(2):151–157.
122. Smith PG, Day NE. The design of case control studies: the influence of confounding and interaction effects. *Int J Epidemiol*. 1984;13:356–365.
123. Engelman CD, Baurley JW, Chiu YF, et al. Detecting gene-environment interactions in genome-wide association data. *Genet Epidemiol*. 2009;33(S1):68S–73S.

124. Qi L, Cornelis MC, Zhang C, et al. Genetic predisposition, Western dietary pattern, and the risk of type 2 diabetes in men. *Am J Clin Nutr*. 2009;89:1453–1458.
125. Smith GD. Use of genetic markers and gene-diet interactions for interrogating population-level causal influences of diet on health. *Genes Nutr*. 2011;6(1):27–43.
126. Churchill GA. Recombinant inbred strain panels: a tool for systems genetics. *Physiol Genomics*. 2007;31:174–175.
127. Kalupahana NS, Moustaid-Moussa N. Overview of symposium "Systems Genetics in Nutrition and Obesity Research". *J Nutr*. 2011;141(3):512–514.
128. MacMahon B, Yen S. Unrecognized epidemic of anencephaly and spina bifida. *Lancet*. 1971;i:31–33.
129. Kirke PN, Molloy AM, Daly LE, et al. Maternal folate and vitamin B12 are independent risk factors for neural tube defects. *Q J Med*. 1993;86:703–708.
130. Smithells RW, Sheppard S, Schorah CJ. Vitamin deficiencies and neural tube defects. *Arch Dis Child*. 1976;51:944–950.
131. Rhoads GG, Mills JL. The role of the case-control study in evaluating health interventions. Vitamin supplementation and neural tube defects. *Am J Epidemiol*. 1984;120:803–808.
132. Smithells RW, Sheppard S, Schorah CJ, et al. Apparent prevention of neural tube defects by periconceptional vitamin supplementation. *Arch Dis Child*. 1981;56:911–918.
133. Mulinare J, Cordero JF, Erikson JD, et al. Periconceptional use of multivitamins and the occurrence of neural tube defects. *JAMA*. 1988;260:3141–3145.
134. Mills JL, Rhoads GG, Simpson JL. The absence of a relationship between the periconceptional use of vitamins and neural tube defects. *N Engl J Med*. 1989;321:430–435.
135. Shaw GM, Schaffer D, Velie EM, et al. Periconceptual vitamin use, dietary folate and the occurrence of neural tube defects in California. *Epidemiology*. 1995;6:219–226.
136. Milunsky A, Jick H, Jick SS, et al. Multivitamin/folic acid supplementation in early pregnancy reduces the prevalence of neural tube defects. *JAMA*. 1989;262:2847–2852.
137. Daly LE, Kirke PN, Molloy A, et al. Folate levels and neural tube defects: implications for prevention. *JAMA*. 1995;274:1698–1702.
138. Medical Research Council Vitamin Study Research Group (MRCVSRG). Prevention of neural tube defects: results of the MRCVS. *Lancet*. 1991;338:131–137.
139. Czeizel AE, Dudas I. Prevention of the first occurrence of neural tube defects by conpericonceptual vitamin supplementation. *N Engl J Med*. 1992;327:1832–1835.
140. Gregory JF. The bioavailability of folate. In: Bailey LB, ed. *Folate in Health and Disease*. New York, NY: Marcel Dekker; 1995:195–235.
141. Cuskelly GJ, McNulty H, Scott JM. Effect of increasing dietary folate on red-cell folate: implications for prevention of neural tube defects. *Lancet*. 1996;347:657–659.
142. Daly LE, Kirke PN, Molloy A. et al. Folate levels and nural tube defects: Implication for prevention. *JAMA* 1995;274:1698–702.
143. Oakley GP, Adams MJ, Dickinson CM. Urgent need to increase folic acid for everyone, now. *J Nutr*. 1996;126:751S–755S.
144. Eichholzer M, Tonz O, Zimmermann R. Folic acid: a public health challenge. *Lancet*. 2006;367:1352–1361.
145. Flour Fortification Initiative. Flour fortification initiative: a public-private-civic

investment in each nation. http://www.ffinetwork.org/. Accessed June 6, 2014.
146. Martin JA, Hamilton BE, Sutton PD, Ventura SJ, Menacker F, Kirmeyer S, Munson ML; Centers for Disease Control and Prevention National Center for Health Statistics National Vital Statistics System. Births: Final data for 2005. *Natl Vital Stat Rep*. 2007;56(6):1–103.
147. Heseker HB, Mason JB, Selhub J, et al. Not all cases of neural-tube defect can be prevented by increasing the intake of folic acid. *Brit J Nutr*. 2009;102:173–180.
148. Johnston RB. Will increasing folic acid in fortified grain products further reduce neural tube defects without causing harm?: consideration of the evidence. *Pediatr Res*. 2008;63:2–8.
149. Brent RL, Oakley GP Jr. The Food and Drug Administration must require the addition of more folic acid in “enriched” flour and other grains. *Pediatrics*. 2005;116:753–755.
150. Stoll C, Alembik Y, Dott B. Are the recommendations on the prevention of neural tube defects working? *Eur J Med Genet*. 2006;49:461–465.
151. Van der Put NM, Steegers-Theunissen RP, Frost P, et al. Mutated methylenetetrahydrofolate reductase as a risk factors for spina bifida. *Lancet*. 1995;346:1070–1071.
152. Groenen PM, van Rooij IA, Peer PG, et al. Marginal maternal vitamin B12 status increases the risk of offspring with spina bifida. *Am J Obstet Gynecol*. 2004;191:11–17.

Drug Therapy and the Role of Nutrition

18

Kathleen M. Gura, PharmD, BCNSP, FASHP, FPPAG and Lingtak-Neander Chan, PharmD, BCNSP

The nutritional status of patients and the constituents of their diet can significantly impact the pharmacokinetic (i.e., absorption, distribution, metabolism, transport, and elimination) and pharmacodynamic (e.g., affinities to receptors or tissues) properties of drugs.[1] Body weight and/or surface area are also important determinants of optimal dosages, and abnormal body composition may impact proper drug dosing. Various nutritional components can affect gastrointestinal (GI) motility, blood flow rates, gastric secretions, and enzymatic activity, ultimately affecting biotransformation and disposition of drugs. The magnitude of these effects, however, is highly variable and patient specific, confounded by many other factors, and often difficult to predict. Susceptibility to drug-induced nutrient deficiency appears to be greater during periods of increased caloric demand (i.e., during growth, pregnancy, and lactation). When developing a therapeutic plan for the pediatric patient, it is important that practitioners consider the interactions that occur between nutritional status, age, disease state, and drug action. This chapter discusses the developmental changes that occur in the pharmacokinetics and pharmacodynamics of drugs and presents a discussion on the impact of nutrition on these factors. Figure 18-1 illustrates the relationship between nutritional factors and drug therapy. By definition, pharmacokinetics is the exposure of drugs in the body over a period of time, including the processes of absorption, distribution in tissues,

metabolism, and elimination. This contrasts with pharmacodynamics, which is the study of the biochemical and physiologic effects of drugs and the mechanisms of their actions, including the correlation of actions and effects of drugs with their chemical structure, as well as the effects on the actions of another drug or nutrient. Figure 18-2 provides an example of the age-dependent variation in pharmacokinetic and pharmacodynamic properties. It should be remembered, however, that in the pediatric patient, in addition to true pharmacokinetic and pharmacodynamic differences, some alterations in drug response may be due to perception as medications are often not adequately studied in children of different ages and different disease states. Thus, there may be difficulties in assessing small but significant differences in medication response because limitations associated with conventional outcome measures.

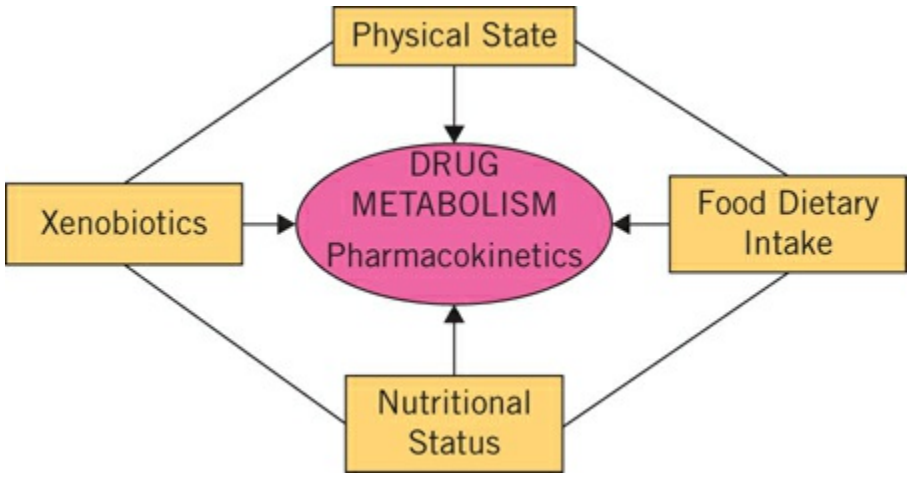


Figure 18-1 Relationship between nutritional factors and drug therapy.

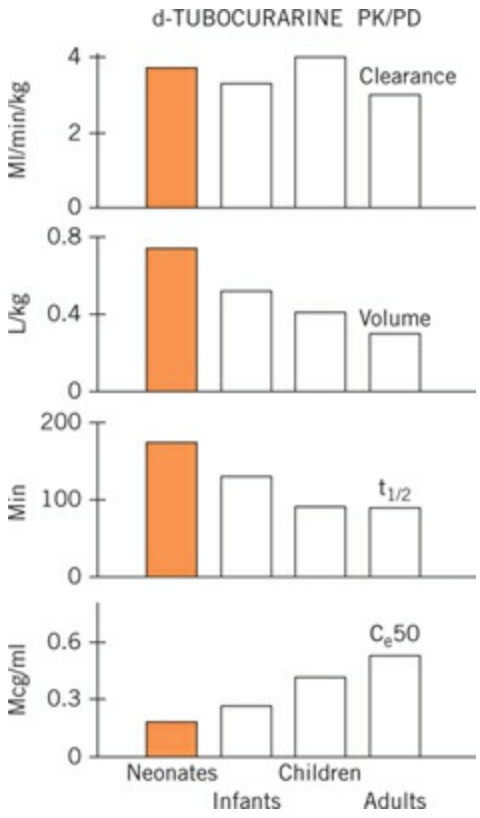


Figure 18-2 Age-dependent variability in the pharmacodynamic and pharmacokinetic properties of *d*-tubocurarine. In this example, neonates require significantly lower plasma concentrations of *d*-tubocurarine to achieve 50% paralysis in comparison to older children and adults, suggesting that neonates have increased receptor sensitivity. The larger volume of distribution in the neonate may also account for the prolonged half-life although plasma clearance is comparable in all four age groups. (Adapted from Fisher et al.[257])

DEVELOPMENTAL CHANGES IN BODY COMPOSITION

Unlike the adult, the body composition of a pediatric patient is undergoing constant changes due to the tremendous growth and development. Children should not be thought of as "little adults," because there are a number of developmental aspects that must be considered when approaching pediatric therapeutics. In fact, during the first decade of life, growth is erratic and does not follow a linear path, thus making simplified dosing approaches inappropriate.[2] Drug–receptor interactions, ontogenetic changes in

receptor number, receptor affinity, receptor–effector coupling, and receptor modulation and regulation change as the child grows.[3] Moreover, during infancy and childhood, the proportions of body weight contributed by fat, protein, intracellular water, and extracellular water change dramatically. In the full-term neonate, total body water comprises approximately 70%–80% of body weight.[4] By age 5 months, this decreases to 60%. From infancy to young adulthood, extracellular water decreases, although the percentage of total body water does not change significantly. In addition, neonates have reduced muscle mass, lower concentrations of albumin, and less intracellular fluid than adults.[5] The change in body water content may significantly affect the volume of distribution of drugs, especially those that are highly hydrophilic. Changes in serum protein (e.g., albumin) may alter the free concentration of certain drugs and thus change the patient's pharmacodynamic response. During the second year of life, fat mass is reduced with a corresponding increase in protein mass. For additional information, refer to Chapter 4, which reviews the differences in body composition in premature infants, neonates, and adults.

Moreover, epigenetic factors influenced by the nutritional state of the mother in the periconceptional period can later influence DNA methylation in the infant. Supplementation with vitamin B_{12}, folic acid, and some amino acids during pregnancy can alter DNA methylation that can offer some protection against infection and immune response.[6]

As the child grows, there is a corresponding change in liver and kidney size, with each organ reaching maximum relative size for weight in the 1- to 2-year-old child, when the capacity for drug metabolism and elimination is high.[3] For example, in the term infant, the glomerular filtration rate (GFR) is approximately 2–4 mL/min/1.73 m^2, while a premature infant may be as low as 0.6–0.8 mL/min/1.73 m^2, and adult values are achieved within the first 8–12 months of life.[2] In the infant and young child, body surface area is greatest relative to body mass in comparison with the older child or young adult. As a percentage of adult norms, intestinal length in infants and children exceeds other anthropometric measurements.[2] Furthermore, the absorptive surface of small intestine is proportionately greater, whereas GI transit time is shorter.

During adolescence, there is an approximately 25% increase in height, whereas weight nearly doubles.[3] In prepubertal males and females, lean body mass, skeletal mass, and body fat per unit body weight are similar, but, by adulthood, women have twice as much fat relative to total body

weight in comparison with adult men.

Recently, research has begun to focus on the relationship between genes (including genetic polymorphisms in drug-metabolizing enzymes and transport proteins) and related systems, developmental, changes, and lactation. Similarly, acute-phase reactions may alter the disposition of certain amino acids and cofactors which in turns can change the functions and/or expression of these enzymes and transporters. For example, the presence of an acute infection or renal failure can substantially alter iron status, which results in changing the metabolic activities of a major drug-metabolizing enzyme, cytochrome P450 (CYP) 3A4.[7]

FACTORS THAT AFFECT DRUG KINETICS

The aforementioned differences in body composition in the pediatric population are an important consideration when determining the pharmacokinetics of a given medication (Table 18-1). Nutrients and drugs delivered via the GI tract need to go through an absorption phase prior to reaching the systemic circulation and the sites of action. Absorption from the GI lumen into the hepatic portal vein and subsequently to the systemic circulation is a series of complex processes including the dissolution of the solid dosage form, passing of the chyme along the GI tract (i.e., gastric emptying and intestinal transit), passive diffusion, active transport, and presystemic metabolism of the compounds. Each of these processes alone may affect the pharmacokinetics of the drug.

GASTRIC EMPTYING AND INTESTINAL TRANSIT

The oral route continues to be the most widely used route of administration in the pediatric patient. As a result, intraluminal pH in different areas of the GI tract can impact medication stability, dissolution rate of solid dosage forms, and possibly the extent of drug absorption in some cases. Neonates typically have a higher gastric pH (>4), and thus, acid-labile drugs such as penicillin G are more bioavailable and lower doses can be used while weakly acidic drugs such as phenobarbital require much larger oral doses to reach therapeutic levels.[2] With the exception of a few acidic drugs (e.g., aspirin), maximal absorption of most drugs and nutrients take place in the small bowel. Therefore, gastric emptying and intestinal transit time have a significant impact on the rate and magnitude of the oral absorption of the drugs and certain nutrients.

Table 18-1 Factors Associated With Differences in Pharmacokinetic/Pharmacodynamic Parameters in Neonates versus Adults
Altered receptor sensitivity
Enhanced intramuscular absorption
Greater blood–brain barrier permeability
Hepatic enzyme immaturity/limited enzyme capacity
Higher gastric pH
Immature renal function
Increased percutaneous absorption
Larger liver:body weight ratio
Limited protein-binding capacity
Reduced fat mass
Slower GI absorption/increased gastric emptying time

Gastric emptying time changes in early postnatal life and is likely one of the many explanations for erratic drug absorption rate and oral bioavailability in infants and young children.[6] During infancy, the rate of drug absorption is slower than in older children. As the intestinal tract matures, antral contractions improve and with it a marked increase in gastric emptying. Similarly, intestinal motor activity increases in amplitude and frequency as well as duration of contractions.[2] In addition, gastric acid secretion is low in infants and does not reach the normal adult gastric pH until 3–7 years of age.[8]

The lack of developed physiologic regulation of GI motility and gastric acidity during infancy and early childhood suggests that the oral route may not be a reliable route for optimal drug delivery. For instance, delayed absorption and/or decreased bioavailability have been observed for acetaminophen, phenytoin, and phenobarbital in young children.[9]

As the child continues to grow, GI motility increases by both neuronal and hormonal input.[10] Vagal stimulation increases upper gastric contraction and decreases pyloric sphincter and duodenal contractions. As a result, gastric emptying is promoted, and the time it takes for the drug or nutrient to reach the small intestine is shortened. For drugs that are absorbed in the small intestine, this action can result in shortening the time of reaching peak plasma concentration and may promote faster onset of drug action. In addition, because of the decrease in intestinal motility, more complete absorption of the drug may be achieved. Anticholinergic

drugs (e.g., sedative antihistamines, phenothiazines, tricyclic antidepressants) can reverse these trends by counteracting the effect of the vagus nerve.

PRESYSTEMIC CLEARANCE

Presystemic clearance, also known as first-pass effect, refers to the removal of orally ingested compounds prior to reaching the systemic circulation. Presystemic effect occurs primarily in the intestine and the liver, whereas the stomach has only a minor role.[11] Type III alcohol dehydrogenase, for example, is present in the gastric mucosa and is responsible for the activation of some biogenic amines and steroids.

Many drug-metabolizing enzymes and active transport proteins are present in the intestinal epithelial tissues. For example, CYP3A4 isoenzyme is present in the small bowel and plays a role in the regulation of oral bioavailability of a large number of medications. Induction or inhibition of the enzyme in the gut by nutrients can affect the oral bioavailability of drugs. Certain nutrients are known to affect the enzymatic activity of intestinal CYP3A4 and change the pharmacokinetics of the object drugs. Grapefruit juice is a classic example of an intestinal CYP3A4 inhibitor.[11] Its action is discussed in further detail below. Water-soluble vitamin E (d-alpha-tocopheryl polyethylene glycol succinate) has also been found to increase oral absorption of cyclosporine.[12] The primary mechanism appears to involve inhibition of an intestinal efflux protein, P-glycoprotein (P-gp) instead of intestinal CYP enzymes.[13]

HEPATIC METABOLISM

The metabolism of a drug involves different steps once it has been absorbed from the GI tract. As drugs are metabolized, they first undergo biotransformation (i.e., phase I metabolism), which results in the formation of a more polar compound by oxidation, reduction, hydroxylation, and so forth. This results in either an activation of the drug or a deactivation. The second phase (i.e., phase II reaction) is a synthetic process, involving conjugation of the polar compound with endogenous molecules such as glucuronic acid, sulfate, glutathione, glycine, and acetate, resulting in a more hydrophilic compound that is more suitable for excretion into bile or urine.[14] It is important to point out that some drugs do not have to go through phase I metabolism prior to undergoing phase II metabolism. Lorazepam, for example, undergoes phase II metabolism (glucuronidation) by UDP-glucuronosyltransfersae (UGT) 2B7 without any phase I reaction

involved. The resultant conjugated metabolite is then excreted renally. An enzyme system responsible for the metabolism of most nutrients and drugs is the CYP enzyme super family, principally located in the endoplasmic reticulum of the hepatocytes and enterocytes, which is somewhat unique in its ability to use a wide range of substrates.

The maturation of phase I and phase II enzymes varies among individuals. Limited studies suggest that fetal and neonatal CYP, the most important phase I enzyme in metabolizing drugs and biogenic amines and steroids, has about 50%–70% of the adults' activity and continues to mature throughout childhood. Although some children may have fully matured CYP enzyme activities as early as 6 months of age, it may take up to 12 months for others.[8] For example, the serum half-life of theophylline, a bronchodilator primarily metabolized by CYP1A2 and CYP3A4, is significantly longer in neonates and younger infants than in adults. Similarly, *N*-demethylation of diazepam, a CYP2C19-mediated pathway, is also significantly slower in infants. There is no well-documented dietary factor known to promote the maturation of these enzymes during the prenatal period and infancy. Table 18-2 describes the impact of various nutrients on Phase I/mixed function oxygenase metabolism.

RECTAL ADMINISTRATION

Rectal route of drug administration is an alternative way of drug delivery when the enteral route is not preferable. Unlike the small bowel venous mesentery, the rectal capillary blood supply collects blood directly into the inferior vena cava instead of the hepatic portal vein. Although certain phase II enzymes are present in the colon (e.g., UGT1A9), there is no strong evidence to suggest the presence of drug-metabolizing enzyme in the rectum.[15] Hence, presystemic metabolism of the administered drug is prevented. Therefore, compared with oral administration, the relative bioavailability of the drug is usually higher with rectal administration.[16] Drugs that are more commonly delivered rectally in children include anticonvulsants (e.g., diazepam), antiemetics (e.g., trimethylbenzamide), and sedative agents (e.g., chloral hydrate).

Table 18-2 Impact of Nutrients on Phase1/Mixed Function Oxygenase Metabolism

Nutrient	Effect on Metabolism	Proposed Mechanism
Ascorbic acid	Deficiency: ↓	Alterations in CYP450 and CYP 450

	Excess: ↑ CYP450 activity	reductase via changes in expression of specific CYP isoenzymes depending on deficiency or excess state
Carbohydrates	Excess: ↓	May be due to ↓protein or inhibition of CYP 450 via ↓ in supporting enzyme components
Fats	High intake saturated fatty acids: ↓ High intake polyunsaturated fatty acids: ↑ Activity and induction of CYP enzymes Deficiency: ↓ Excess: ↑	Decreased CYP activity may be due to requirement of PUFAs in the B-postion of phosphatidylcholine (lecithin), which is an essential component of the CYP system
Iron	Deficiency: ↑↓ Excess: ↑ Microsomal lipid peroxidation	↑ Lipid peroxidation may lead to damage to the integrity of the system
Protein	Deficiency: ↓ Rate of metabolism Excess: ↑ Rate of metabolism	↓ Protein synthesis ↓ Synthesis of other elements (i.e., hormones) involved in enzyme induction
Pyridoxine	Deficiency: ↓	Impaired protein synthesis ↓ Synthesis of heme
Riboflavin	Deficiency ↑↓ depending on severity	↓ Reductase activity but ↑ CYP450 activity such that drug metabolism may be altered (↑↓)
Thiamine	Deficiency: ↑ CYP450 activity Excess: ↓ (both reductase and CYP 450)	↑ Activity of specific CYP450 isoenzymes and perhaps other enzymes in deficiency due to an unknown mechanism Excess may be due to decreased substrate binding
Vitamin E	Deficiency: ↓	May be due to a reduction in antioxidative mechanisms (e.g., protection of the lecithin component thus the activities of CYP450 and reductase are unaffected)

Source: Adapted from Raiten.[258]

DRUG–NUTRIENT INTERACTIONS

By definition, a food–drug interaction is one that is the result of a chemical, physical, physiological, or pathological relationship between a medication and a nutrient.[17] They are clinically relevant if the interaction alters the therapeutic response of the medication: either decreasing the

bioavailability of the drug (and increasing the risk of treatment failure) or by increasing is bioavailability (and increasing the risk of toxicity).

There are four basic classifications of drug–food interactions that are based on their nature and mechanism.[18]

1. *Type I ex vivo bioinactivations*. These are interactions that occur between the nutrient and the drug, usually in the delivery device, via biochemical or physical reactions. Examples include: complexation, hydrolysis, neutralization, oxidation, and precipitation.
2. *Type II*. These interactions impact absorption by modifying the bioavailability of the medication. The causative agent may alter the function of the enzymes or transport systems that are responsible for drug biotransformation.
3. *Type III*. These interactions are involved in the systemic disposition of the drug and occur after the medication or nutrient has been absorbed from the GI tract and has entered the systemic circulation. This can result in changes in tissue distribution, transport, or penetration to a specific organ or tissue.
4. *Type IV*. In this type of interaction, the elimination or clearance of the drug or nutrient is impacted. It may modify either the renal or the enterohepatic elimination process.

DRUG AND NUTRIENT TRANSPORT SYSTEM

Oral drug absorption across the GI tract is highly dependent on its lipophilic properties and its affinity for membrane transporters. Since the most drugs are most efficiently absorbed in the small intestine, transport protein present in the enterocytes play a pivotal role in facilitating the absorption of many drugs.[19] Although most transport proteins facilitate absorption by increasing the intraluminal uptake of compounds, some transporters efflux molecules already absorbed in the cytoplasm of the enterocyte back into the intestinal lumen, thus decreasing the bioavailability of certain compounds. This is believed to be an intrinsic protective mechanism by the host to minimize xenobiotic exposure. P-gp is a representative of this type of efflux system.[20] P-gp belongs to the family of adenosine triphosphate–binding cassette (ABC) transporters and is an efflux protein widely distributed in normal tissues, including the intestinal epithelium, renal tubule, liver, and blood–brain barrier. Hepatic

transporters are membrane proteins that facilitate nutrient and drug transport into the cell via uptake transporters or pump out toxic entities via canalicular transporters.[21] Although it is known that foods, in particular fruits and vegetables that contain photochemicals have the potential to alter drug absorption, research continues to be limited. Recent studies, however, suggest that phytochemicals can serve as substrates and modulators of specific members of the superfamily of ABC transporting proteins (i.e., P-gp, MRP1, MRP2, and BCRP). For example, isothiocyanates, a class of chemotherapeutic agents derived from cruciferous vegetables (i.e., broccoli, cabbage, watercress) may influence the pharmacokinetics of substrates of these transporters and interact with ABC efflux transporters.[22] CYP3A4 and P-gp are the two most important limiting factors in regulating the oral bioavailability of drugs.[23] It is estimated that CYP3A4 is responsible for the metabolism of more than 50% of all medications.[24] For instance, cyclosporine is a known substrate for both CYP3A4 and P-gp. After oral administration, absorption of cyclosporine across the epithelium in the small intestine is limited by P-gp efflux and prehepatic CYP3A4 metabolism. The P-gp-mediated efflux of cyclosporine from the intestinal cell back into the lumen enables the CYP3A4 enzymes another opportunity to metabolize the drug when it again enters the enterocyte. These mechanisms explain why the oral bioavailability remains poor despite improvement of its formulation to microemulsion.[25] When cyclosporine is coadministered with a CYP3A4 and P-gp inhibitor, such as ketoconazole, erythromycin, or diltiazem, significantly higher maximum blood cyclosporine concentration is observed, indicating an increase in absolute oral bioavailability. Oral bioavailability can be increased by over 40% with concomitant administration of water-soluble vitamin E.[12] The mechanism is mediated through intestinal P-gp inhibition by the formulation. It appears that polyethylene glycol, the surfactant incorporated in the liquid formation, instead of vitamin E is the inhibitor of P-gp.[13,26]

BINDING INTERACTIONS BETWEEN FEEDINGS AND DRUGS REVISITED

Feeding and food intake are common causes of drug–nutrient interactions. Enteral feeding is the preferred method of providing nutrition support and also allows easy access for administering drugs to patients who are unable to swallow. However, enteral feeding formulas have been implicated in a

number of drug–nutrient interactions.[27] Moreover, in children with chronic medical conditions receiving tube feedings, nutrient deficiencies are also a concern because multiple medications and complicated feeding regimens can result in a decrease intake due to feedings being held or due to drug interactions.[28] For example, the oral absorption of warfarin, tetracycline, fluoroquinolone antibiotics, thyroid supplements, and phenytoin is decreased with concomitant enteral feeding.[29,30] Possible explanations include a decrease in the anticoagulant effect of warfarin caused by increased vitamin K absorption from the enteral formulas or binding of warfarin by a protein component of the enteral formula.[29] It has also been reported that the acidic nature of most enteral formulas alters the solubility of phenytoin. Phenytoin, a weak acid, is not readily absorbed in acidic conditions and absorption is optimal when it is given in the ionized state. Furthermore, when phenytoin is given with nasogastric feeds, it may become bound to the tubing when it is nonionized in a low pH environment.[31] Drug–nutrient interactions through chelation of divalent and trivalent cations has also been proposed (e.g., enteral feeding formulas or dairy products interact with fluoroquinolones).[25] A case of scurvy was reported in a child who received long-term enteral tube feedings using a commercial product. Scurvy occurred due to the degradation of the ascorbic acid in the formula due to prolonged storage after mixing and from the addition of an iron supplement that further catalyzed the reaction.[32]

Unlike medications given via the oral route, whenever a medication is delivered via feeding tube, tube placement and device characteristics must also be considered. Medications may adhere to the sides of the feeding tube, and thus not be delivered to the patient, or may obstruct the tube in the case of an oral solid not properly pulverized and diluted prior to administration. In some instances, medications are best absorbed in the "fasted" state, requiring that feedings be held so as to optimize absorption. Intragastric administration of a medication may delay gastric emptying rates. Administering solid dosage forms of medications (e.g., crushed tablets) to patients with jejunostomy, nasojejunal, or nasoduodenal feeding tubes present an additional challenge as these devices bypass the stomach where medications are dissolved and diluted with gastric contents. Due to the narrow bore of the devices and direct administration of medication into the small intestine, liquid dosage forms of medications (e.g., oral liquid, suspension, syrup) must be used. When administering oral dosage forms, several factors should be considered, however, including the osmolality of

the drug, its viscosity, and particle size. Medications that are hypertonic and not diluted prior to administration into the small bowel can result in dumping. The osmolality of stomach secretions is approximately 300 mOsm/kg and many liquid medications exceed this value significantly, resulting in an osmotic-induced diarrhea if given in its undiluted stated. Whenever a hypertonic medication is given undiluted, gastric emptying rates are altered, and there is a flux of water and electrolytes into the small bowel and the absorptive capacity of the small bowel is overwhelmed. Proper dilution of the medication can prevent this flux and improved medication as well as nutrient absorption. Moreover if the feeding tube does terminate in the jejunum, pH is an important consideration to assure optimal dissolution of certain dosage forms.

EFFECTS OF MALNUTRITION ON DRUG KINETICS

It has become increasingly recognized that nutritional status is capable of modifying the pharmacologic effect of a medication. Both malnutrition and obesity can substantially alter drug pharmacokinetics and pharmacologic responses by causing functional and structural alterations in organs that directly affect drug disposition.[33] These changes can affect the absorption, distribution, metabolism, and elimination of a medication that can ultimately affect the therapeutic or toxic response of the drug. Moreover, interindividual and intraindividual variations in the pharmacokinetic responses to a medication can further complicate interpreting the actual impact of altered nutritional status on drug disposition. The variation can be 3- to over 20-fold, depending on genetic (e.g., genetic polymorphism) and environmental factors, patient variables, and underlying disease.[33] Pediatric patients, of course, have a higher incidence of undernutrition (see Chapters 14 and 15). Moreover, several epidemiological studies have demonstrated that children with malnutrition have a high prevalence of bacteremia, pneumonia, and urinary tract infections.[34] The pathologic changes seen in malnutrition can impact the pharmacokinetics of drugs in all phases of disposition within the body, suggesting that the degree of malnutrition can determine the body's response to a particular drug. The integrity of the intestinal mucosa is dependent on a continuous intake of adequate nutrition as the turnover of enterocytes in the small intestine takes two to three days and in the colonocyte of the large intestine three to five days.[35] Even short-term

malnutrition can potentially have a negative impact on drug absorption and metabolism, a risk that increases significantly in patients who go without food for more than 5 days due to a decline in mucosal integrity. Physiologic changes of protein–energy malnutrition (PEM) may result in alterations in the absorptive capacity of the GI tract, body fluid status, cardiac output, GFR, and plasma protein concentrations, as well as hormonal and metabolic changes. Therapeutic drug levels may be altered as a result of malnutrition-associated tissue receptor alterations.[36] It is conceivable that the risk of toxicities owing to a drug or its metabolites is greater in malnourished patients, with a subsequent risk of morbidity or mortality. This suggests that close drug therapy monitoring and modification of dosage in malnourished patients are imperative. Drugs with narrow therapeutic indices or narrow dose-response curves (i.e., phenytoin, theophylline) are particularly susceptible as even small changes in absorption can become significant.

Relatively little is known about the handling of drugs in malnourished children despite the fact that the combination of malnutrition and accompanying disease (typically infection) in need of treatment is a common pediatric problem in many parts of the word. It is possible that the high morbidity and mortality so characteristic of malnutrition may be enhanced by adverse drug reactions.[37]

ABSORPTION

Little research has been done on the effect of malnutrition on drug absorption. O'Doherty et al. observed that phenytoin absorption was slow and erratic in PEM, although the peak concentration and time to reach peak concentration did not differ significantly from controls.[38] Mehta et al. reported similar findings with acetaminophen disposition in children suffering from PEM.[39] Like O'Doherty et al., they observed that the absorption rate constant was not altered in malnutrition. This contrasts with work by Raghuram and Krishnaswamy, indicating that tetracycline absorption was significantly reduced in subjects with malnutritionand pellagra but not in patients with vitamin B complex deficiency or in patients with severe anemia.[40]

DISTRIBUTION

The important serum proteins that bind to drug molecules and biogenic

amines and lipids include albumin, alpha 1-acid glycoprotein, sex hormone–binding proteins, and lipoproteins. Only free, unbound drug molecules are pharmacologically active. Because the amount of serum protein changes over the first few years of life, the pharmacokinetics and pharmacodynamics of a drug may change. The changes in circulating plasma proteins can impact the distribution of highly bound drugs. In neonates and young infants, a relative reduction in total plasma proteins can result in increasing the availability of active drug due to an increase in the free fraction of drug.[2] The serum concentration of alpha 1-acid glycoprotein is lower in children. On the other hand, the concentrations of certain fetal serum proteins are higher in infants and may lead to relatively unique pharmacokinetic changes in this population. For example, in neonates, the presence of fetal albumin is associated with reduced binding capacity to weak acids and, this, along with a corresponding increase in bilirubin and free fatty acids, resulting in a high free fraction of highly protein bound drugs such as phenytoin.[2]

Drugs can distribute into various body compartments such as the intracellular fluids, extravascular space, lean body tissue, and adipose tissue. Alterations in body composition, particularly the presence of edema, can influence the plasma clearance of drug by changing the medication's volume of distribution.[41] Many medications, once in the systemic circulation, become bound to plasma protein, such as albumin, globulins, and lipoproteins. These binding proteins play an important role in the intravascular transport of the drugs to the target organs. In the bloodstream, drugs that are not bound to plasma proteins are free and are able to exert their pharmacologic response. Malnutrition can alter the rate of tissue protein synthesis as well as the concentration of plasma proteins. The extent of drug–protein binding depends on the concentration of plasma bind proteins as well as the physicochemical properties of the medication. In malnutrition, albumin and lipoprotein synthesis is reduced, whereas globulin and a1-acid glycoprotein synthesis is increased.[25] Drugs extensively bound to a1-acid glycoprotein, such as propranolol, have a decreased percentage of drug unbound in malnutrition (i.e., decreased in free fraction), resulting in a lesser amount of active drug available to exert a therapeutic response. Other drugs, such as metronidazole, have no significant change in volume of distribution between malnourished and well-fed children.[42] In the case of parenteral penicillin, no significant difference in C_{max} was noted when penicillin was administered intramuscularly to malnourished versus eutrophic individuals, suggesting

that the rate of penicillin absorption from the muscle mass to the systemic circulating is unchanged. On the other hand, in patients with marasmus and kwashiorkor, there was a trend toward a lower volume of distribution for penicillin, suggesting that overdosing could be a concern in some patients if the typical pediatric dosing strategy is used.[43]

METABOLISM

In chronic starvation, the body is able to adapt and various processes are altered to protect or maintain enzyme activities.[44] In fact, in chronic starvation, enzyme activity may even increase. Endocrine tissue is typically affected in semistarvation as many hormones serve as substrates for drug-metabolizing enzymes.[45] For example, elevations in free cortisol are often seen in malnutrition that may enhance the metabolism of contraceptive steroids in malnourished women, thus increasing the risk of contraceptive failure in that population.[46]

CLEARANCE

Hepatic clearance of drugs may be affected by several major physiologic changes that occur during malnutrition. Protein deprivation or malnutrition may result in a reduction in cardiac function with a subsequent decreased perfusion of the liver and kidneys.[47] Typically, hepatic drug clearance is determined by three independent factors: hepatic blood flow, the amount of free fraction of the drug in blood, and hepatic clearance of the unbound drug. Diminished hepatic blood flow can reduce the clearance of drugs with high extraction ratios (i.e., those in which hepatic clearance of the unbound drug depends on hepatic blood flow but not on changes in protein binding). Presystemic metabolism can become altered. Increases in the unbound fraction of a drug owing to hypoalbuminemia provide more available drug for metabolism, resulting in higher clearance (i.e., rate of drug removal from the body) and lower serum concentrations.[47]

The kidney is extensively involved in drug elimination. Renal elimination includes the processes of glomerular filtration, active tubular secretion, and passive tubular excretion. Drugs or their metabolites that are primarily filtered and excreted renally may be affected by nutritional status. Dietary protein increases renal blood flow, GFR, and renal tubular function.[48] Severe PEM is associated with decreased GFR and renal blood flow.[49] When renal perfusion is reduced, less drug is available to be

filtered by the tubules. However, because plasma protein binding is also reduced, more free drug becomes available for renal excretion, thus further reducing plasma drug concentrations. Medications that have decreased renal elimination in severely malnourished patients include penicillins, tetracyclines, cefoxitin, aminoglycosides, and methotrexate.[33]

Refeeding an undernourished patient can often increase the systemic clearance of a medication, necessitating a dosage adjustment to maintain efficacy. For example, with theophylline, the volume of distribution decreases and the rate of elimination gradually increases in malnourished patients given a dextrose-based parenteral nutrition (PN) solution for at least two days.[50] The protein component of enteral or PN, however, appears to be the major macronutrient enhancing systemic clearance of affected drugs in patients transitioned from the “unfed” to the “fed” state. Lares-Asseff et al. confirmed these findings in their study investigating the pharmacokinetics of metronidazole in severely malnourished children.[42] On the basis of clearance data, they recommended that the daily maintenance doses for pediatric patients with severe malnutrition should be 60% less of the usual pediatric dose to achieve and maintain a therapeutic plasma concentration of metronidazole.

DRUGS IN KWASHIORKOR

Kwashiorkor is associated with edema resulting from increases in total body water, extracellular fluid volume, and plasma volume along with a decrease in intracellular water.[51] Lares-Asseff et al. studied the effects of nutritional status of children with autoimmune disease on the disposition of aspirin and its metabolites.[52] The results suggested that a decrease in the hydrolysis and oxidative reaction of the metabolic pathway of aspirin (ASA) and its metabolites occurs in children with PEM with juvenile rheumatoid arthritis. This suggests that the hepatic elimination of salicylates may be altered by disorders of nutritional state.[53]

Based on these and other studies, it is thought that low protein intake results in a negative nitrogen balance that decreases drug metabolism, whereas drug metabolism in patients receiving adequate total calories but with less than optimal protein intake is not significantly impacted.[54]

DRUGS IN MARASMUS

Unlike kwashiorkor, marasmus can be considered as an adaptation to an

insufficient energy intake. There is decreased total body water, reduced intracellular water, increased extracellular fluid, and increased plasma volume.[33] Gentamicin, which distributes predominantly to extracellular fluids, has an increased volume of distribution in malnourished patients compared with well-nourished patients.[55] Antipyrine, a drug model commonly used as an index of hepatic drug-metabolizing capacity, distributes primarily in total body water and has been shown to have no variation in the apparent volume of distribution in malnourished children.[56] Likewise, in patients treated with metronidazole, a medication that also has a large volume of distribution, no difference in volume of distribution was observed between malnourished and nutritionally rehabilitated children.[42] A pharmacokinetic study by Treluyer et al. focused on the impact of human global PEM on quinine metabolism.[57] In that study, they reported that the metabolism of quinine is increased in children with global malnutrition, suggesting that the dosing interval in these children be reduced to obtain the same therapeutic quinine levels as seen in well-nourished children.

Interestingly, the initiation of antiretroviral therapy (ART) in HIV-infected children with marasmus can precipitate the onset of kwashiorkor.[58] Several mechanisms have been proposed. It is thought that some immune competence is necessary to develop the edema associated with kwashiorkor. Second, immune reconstitution inflammatory syndrome may occur soon after ART is started. Finally, the edema that results may be due to refeeding syndrome that may be the result of ART-associated appetite stimulation. It may also be simply a manifestation of ART toxicity in severely malnourished children. Current guidelines recommend initial therapeutic feedings for HIV-infected children with severe malnutrition followed by 50%–100% increased energy intake for the first 6–10 weeks of ART.[59]

In summary, normal or increased drug metabolism occurs in mild to moderate cases of malnutrition, whereas decreased metabolism is seen in severe cases of malnutrition. Given the unique needs of the malnourished ill child, special guidelines have been created for the use of antimicrobial agents.[60] A recent systematic review of antibiotic use in children with severe acute malnutrition (SAM) has attempted to validate these guidelines.[34] On the basis of the evidence to date, the authors support the use of broad-spectrum antibiotics in this population but that well-designed randomized controlled trials are required to further address concerns surrounding the routine use of antibiotics in uncomplicated SAM, with

consideration for HIV status.

OBESITY

Obesity (i.e., body mass index [BMI] ≥ 95th percentile for age and sex) results in altered body composition in which there is both an increased proportion and an absolute amount of adipose tissue as well as an increase in lean body mass, blood volume, cardiac output, and organ size.[61] A crude measure of obesity is the BMI, a person's weight (in kilograms) divided by the square of their height (in meters). A BMI equal to 25 or more is considered overweight; an individual with a BMI of 30 or more is generally considered obese. Obesity is linked with physiological changes that can impact pharmacokinetic parameters of many medications, but for most drugs, dosing recommendations do not take into account the need for dosing adjustments due to size measures.

In children, obesity can result in an expansion of blood volume, resulting in increased stroke volume and cardiac output. There seems to be no differences in albumin binding of drugs although increases in CYP2E1 activity and phase II conjugation activity have been observed.[62] Nevertheless, the increased in CYP2E1 activity appears to be down-regulated with weight loss. An increased abdominal fat can also lead to increased intra-abdominal pressure and impact gastric emptying.[63] Drug distribution depends on body composition and may be altered in obese patients. Absorption of drugs evaluated to date appears to be unchanged owing to obesity, although the data are very limited.[64] Severely obese patients who have undergone bariatric surgery for weight loss are more likely to experience altered drug absorption that may affect the clinical responses to therapy. Generally speaking, malabsorption of drugs is more likely to occur with the primary malabsorptive procedures such as jejunoileal bypass and pancreatobiliary diversion. A commonly performed surgical procedure for weight loss is the Roux-en-Y gastric bypass (RYGB), which is a primary restrictive procedure with mild malabsorptive component. Studies aimed to investigate the impact of RYGB on pharmacokinetics are very limited. Existing data suggest that the impact appears to be minimal with the current procedure when the Roux-limb (bypass limb) is not more than 150 cm. In addition, the bioavailability of orally administered liquid medications appear to be unaffected by RYGB. These early data suggest that if oral absorption is a concern in a patient with RYGB or similar upper GI surgical procedures, liquid dosage forms of medications are preferred.

The lipophilicity of a drug determines the extent to which obesity influences the volume of distribution and ultimately whether dosing should be based on actual or adjusted body weight. Highly lipophilic drugs, such as lidocaine, thiopental, phenytoin, verapamil, and most benzodiazepines, have an increased volume of distribution in severely obese patients.[51] Modest increases in volume of distribution have also been reported for aminoglycosides, heparin, ibuprofen, methylxanthines, prednisolone, and vancomycin, suggesting that an adjusted body weight rather than actual body weight be used to avoid toxicity.[64] However, the most accurate approach to adjust for the excess body mass is unknown and appears to be different depending on the characteristics of individual compounds. Therapeutic drug monitoring is recommended when applicable to optimize therapy.

Although the protein binding of acidic drugs is unchanged, the free fraction of basic drugs may be decreased with obesity.[64] Similarly, changes in hepatic drug clearance are variable. Phase 1 reactions and phase I acetylation appear to be unaffected by obesity, but the phase II glucuronidation and sulfation pathways are enhanced. This may result in the shortened duration of action of oral contraceptives and may increase the risk of oral contraceptive failure.[65] Obesity may also affect systemic clearance of highly extracted drugs such as aminoglycosides and unmetabolized procainamide.[64] Both glomerular filtration and tubular secretion also appear to be increased.[40] Renal clearance of drug that mediated by both glomerular filtration and active secretion, such as digoxin and cimetidine, are relatively unchanged in obesity.[64] Table 18-3 provides a examples of dosing strategies for medications used in obese patients.

Aminoglycosides are distributed within the extracellular fluid compartment. Early dosing recommendations suggested that initial dosing be based on ideal body weight (IBW) as it was thought that the drug distributed only into lean body mass. Schwartz et al., however, have since determined that when the volume of distribution is corrected for total body weight, it is significantly smaller when compared with normal-weight subjects.[66] The distribution of aminoglycosides into excess body weight is estimated to be about 40% of that distributed into ideal body tissue. The authors concluded that initial dosing of aminoglycosides in obese patients be determined by adding 40% of the excess weight to the patient’s ideal body weight, with subsequent dosage adjustments based on serum drug levels and clinical status.

Example: For a 7-year-old child, 41 kg, 135 cm, gentamicin 2.5 mg/kg/dose intravenously every 8 hours is ordered:

1. Determine IBW[16]

$$\text{IBW} = \frac{\text{ht}^2 \times 1.65}{1000} = \text{IBW kg, height cm}$$

$$\text{IBW} = \frac{135^2 \times 1.65}{1000} = 30 \text{ kg}$$

2. Determine dosing weight (DW): DW = IBW + (actual body weight [ABW] – IBW) × 0.4 DW (in kg), ABW is actual body weight (in kg)
   DW = 30 kg + (41 – 30) × 0.4 DW = 34.4 kg
3. Calculate gentamicin dose using DW: mg/kg/dose × DW = dose in mg 2.5 mg/kg/dose × 34.4 kg = 86 mg intravenously every 8 hours

Table 18-3 Suggested Dosing Strategies for Medications in Obese Patients

Fixed Doses	Ideal Body Weight	Adjusted Ideal Body Weight	Total Body Weight	Othe
Caspofungin Clarithromycin Co-trimoxazole Linezolid Meropenem Metronidaozole Metoprolol Piperacillin/tazobactam Posaconazole	Acyclovir Filgrastim (G-CSF) Fludarabine Methotrexate Methylprednisolone	Aminoglycosides Amphotericin B, liposomal Busulfan Cyclosporine Phenytoin	Enoxaparin Vancomycin	Ciprof

Source: Adapted from Ref.62.

A similar controversy exists for optimizing theophylline dosing in obese patients and whether actual or ideal body weight should be used to calculate the initial theophylline loading dose. This is attributable to conflicting data suggesting that variations in distribution volume may be the result of differences in the degrees of obesity within study populations.[67] Visram et al. suggested that the differences in theophylline volume of distribution based on ideal body weight and actual body weight increases as the degree of obesity increases.[68] They recommended that theophylline dosing in patients with mild to moderate obesity be based on actual body weight but that ideal body weight be used when initiating

therapy in severely or morbidly obese patients.

Hepatic drug metabolism may also be altered in patients with nonalcoholic steatohepatitis (NASH),[42] including enhanced glucuronidation and sulfonation, causing a faster drug excretion compared with normal-weight subjects.[64] CYP2E1 expression and activities are increased in obese patients with nonalcoholic fatty liver disease, including NASH.[69] Compounds that are metabolized by CYP2E1 include fatty acids, ketones, and ethanol. It appears that the presence of an increase amount of fatty acids, such as in the case of NASH, leads to an induction of this enzyme. The metabolic pathways of fatty acids and other xenobiotics may lead to the release of free radicals, which can cause lipid peroxidation and liver injury, including mitochondrial damage. These changes are partially responsible for the hepatic inflammation and fibrosis sometimes observed in patients with NASH.[70] Patients with fatty liver may also be more susceptible to the toxic effects of drugs owing to impaired metabolism.[71]

EFFECT OF DIETARY MANIPULATION ON DRUG KINETICS

Modifying the intake of specific foods may influence the clinical response of a medication. Populations at particular risk to an adverse event associated with a drug–nutrient interaction include those with chronic disease requiring use of multiple drugs and those requiring specialized nutritional support or some evidence of malnutrition.

Several dietary factors can alter the rate of drug metabolism. Model drugs, such as antipyrine, theophylline, and acetaminophen, have often been used in these studies. Both food and fluids can alter the rate and extent of drug absorption. These alternations in response may occur as a result of influences on gastric pH, gastric emptying time, intestinal motility, and mesenteric and hepatic portal blood flow or biliary flow, or the activities of the enzymes and transport proteins in the gut.[72] Direct physicochemical interactions with dietary components can also alter the absorption of susceptible agents.[73] Interactions such as the binding of the medication with metal ions, solubilization of the drug in dietary fat, or adsorption of the drug to insoluble dietary components may occur.[74]

Dietary changes can alter the expression and activity of hepatic drug-metabolizing enzymes.[75] This can lead to alteration in the systemic elimination kinetics of medications metabolized by these enzymes, although the impact of this change is typically minimal.[75] The rate of drug

metabolism can be accelerated by drugs themselves or by a variety of dietary factors, such as protein supplementation or inclusion of cruciferous vegetables or charcoal-broiled meats in the diet.[76] It has also been reported that charcoal-broiled beef induces the metabolism of antipyrine and theophylline, reducing the half-lives of these drugs by 20%.[77] These effects were related to hepatic enzymatic induction caused by charcoal-broiled beef that contains large quantities of polycyclic aromatic hydrocarbons (PAHs).[78] PAHs are potent inducers of CYP1A2, the primary metabolizing enzyme for theophylline and antipyrine. Conversely, low-protein, high-carbohydrate diets and various vitamin and mineral deficiencies can reduce levels of drug-metabolizing enzymes and consequently the rate of drug metabolism, so that the serum drug concentrations decline much more slowly resulting in increased drug potency.[76]

In general, when orally administered medications are taken with meals, the rate rather than the extent of absorption is more significantly affected. Food affects drug absorption by enhancing gastric blood flow in conjunction with delayed gastric emptying or by altering dissolution. Food can increase, decrease, or have no effect on the absolute systemic availability of a medication.[79] Concomitant food ingestion reduces the absorption of drugs such as ampicillin, penicillin, and isoniazid.[33] Conversely, food may actually enhance the absorption of other medications, including diazepam, lithium, carbamazepine, ziprasidone, and griseofulvin. Table 18-4 lists medications whose absorption is altered by food. In most cases, changing the rate of absorption of a drug alone without affecting the total amount absorbed should not affect its efficacy.

The composition of the meal will alter splanchnic blood flow. Blood flow can be doubled by a high-protein liquid meal and slightly reduced by a liquid glucose meal.[80] The significance of this effect on splanchnic flow is important for those medications with high hepatic extraction.[74] Continued meal intake, especially food with high fat content, will also slow the rate of gastric emptying, which may subsequently cause a delay in drug absorption from the GI tract. Changes in gastric emptying are related not only to the physicochemical properties of the drug but also to the type of meal itself. Hot meals, highly viscous solutions, or those rich in fat have the most significant affect in slowing down gut motility.[74] Melander et al. reviewed the impact of food on the presystemic clearance of drugs.[81] They observed that meals commonly enhanced presystemic clearance of lipophilic basic drugs (e.g., propranolol, amiptriptyline) but

rarely altered the clearance of drugs that were lipophilic acids (e.g., salicylic acid, penicillin). Alternatively, food may reduce presystemic clearance of some lipophilic basic drugs via transient, complex effects on splanchnic-hepatic blood flow. Furthermore, repeated intake of specific nutrients (e.g., protein) and food contaminants (e.g., aromatic hydrocarbons) can enhance presystemic drug clearance by enzyme induction.

Table 18-4 Examples of Medications Whose Absorption Is Affected by Food

Drug Absorption Reduced/Delayed by Food	Drug Absorption Enhanced by Food
Ampicillin	Albendazole
Aspirin	Amiodarone
Atenolol	Atazanavir
Azithromycin	Atorvastatin
Calcium salts	Atovaquone
Captopril	Carbamazepine
Cefaclor	Cerivstatin
Cefixime	Chlorothiazide
Cephalexin	Cefuroxime
Ciprofloxacin	Clarithromycin
Dapsone	Clofazimine
Didanosine	Cyproheptadine
Digoxin	Diazepam
Doxycycline	Efavirenz
Dirithromycin	Etravirine
Erythromycin stearate	Erythromycin estolate
Famciclovir	Erythromycin ethyl succinate
Flecainide	Fluvastatin
Fluoride	Gabapentin
Indinavir	Ganciclovir
Isoniazid	Griseofulvin
Loratidine	Guanfacine
Metronidazole	Hydralazine
Naficillin	Hydrochlorothiazide
Nalidixic acid	Hydroxychloroquine
Penicillin G or V	Isotretinoin
Phenobarbital	Itraconazole
Phenytoin	Ivermectin
Rifampin	Ketoconazole
Sucralfate	Lithium
Tacrolimus	Lovastatin
Tetracycline	Mebendazole
Thioridazine	Methylphenidate
Voriconazole	Metoprolol
Zafirlukast	Nelfinavir

	Nitrofurantoin Pravastatin Propanolol Propoxyphene Rilpivirine Ritonavir Saquinavir Simvastatin Spironolactone Tenofovir Tipranavir Valganciclovir Zalcitabine

Source: Adapted from Gura.[259]

In some instances, dietary manipulation can act as a therapeutic strategy. Nutt et al. reported that large neutral amino acids and the medication levodopa compete for transport from the plasma to the brain and may be partly responsible for the "on-off" phenomenon often seen in patients treated with levodopa for Parkinson's disease.[82] By reducing protein intake, patients who failed to respond to a dosage adjustment in their levodopa may see clinical improvement.

IMPACT OF SPECIFIC NUTRIENTS ON DRUG KINETICS

Until recently, most practitioners dismissed the possibility that dietary substances could significantly alter medication response by affecting intestinal transporter and metabolizing enzymes. This is based on the commonly believed misconception that the absorption of most drugs is a passive process and that the role of the intestine in drug elimination is minimal.[75] Since the report of the interaction between grapefruit juice and several medications was described, this premise has changed, and the role of the diet on drug performance is being re-evaluated.[83]

Carbohydrates

The impact of carbohydrates on drug metabolism is conflicting. It is known that high carbohydrate diets may induce the expression of several glycolytic and lipogenic hepatic enzymes,[84] but some suggest that carbohydrates have little impact on drug metabolism.[85] Kappas et al., however, noted that antipyrine and theophylline metabolism decreased in carbohydrate-supplemented diets but increased in the protein-enriched

diet, suggesting that carbohydrates and protein have opposite effects on oxidative drug metabolism.[76] Although many medications are often given to children in a sugar syrup, little research has been done on its effect on disposition and action. Animal studies by Sonawane et al. suggested that dietary carbohydrates and fat may significantly influence the hepatic drug-metabolizing enzymes.[86] It has been hypothesized that these changes may occur owing to alteration in the phospholipid composition of endoplasmic reticulum or by limiting the supply of cofactor(s) necessary for optimal functioning of CYP and UGT.

Protein

Several investigators have reported that medications that undergo extensive first-pass effect, such as propranolol, metoprolol, and lidocaine, can have enhanced bioavailability after a high-protein meal owing to enhanced hepatic blood flow. High-extraction drugs can then rapidly pass through the liver, allowing higher drug concentrations in the systemic circulation.[75,81] A decrease in dietary protein depresses creatinine clearance and renal plasma flow.[87] In situations where there is low protein intake, careful attention is needed to avoid toxicity secondary to delayed drug clearance. Specific dietary proteins can also impact a response to a medication. One of the classic examples is that of the monoamine oxidase inhibitor (MAO-I) drug class and the amino acid tyramine that is contained in aged cheeses, pickled/smoked meats, fermented foods, and red wines. Tyramine is an indirect sympathomimetic amine that releases norepinephrine from the adrenergic neurons, causing a significant pressor response. Typically, tyramine is metabolized by the enzyme monoamine oxidase before any significant increases in blood pressure are seen. If the enzyme is blocked, however, severe and potentially fatal increases in blood pressure can occur when tyramine-rich foods are ingested. Although MAO-Is are not used routinely as antidepressants, other medications, such as the oxazolidinone antibiotic and linezolid, also have MAO-I properties and patients should avoid ingesting large amounts of tyramine while being treated with this antibiotic.[88]

Dietary protein also affects the renal tubular transport of certain compounds, although the mechanism by which this occurs is still not understood. The binding of dietary proteins to a drug may underscore changes in bioavailability after a protein meal. For example increases in both the maximum concentration (C_{max}) and area under the curve (AUC) are seen in patients receiving gabapentin. This enhanced absorption was

attributed to trans-simulation, a carrier-mediated process in which increased intestinal luminal amino acid concentrations result in an upregulation and/or increased activity of the L-amino acid transporter.[89]

Dietary Fat

Lipids are an essential part of cell membrane structure and are involved in many of the normal enzymatic activities located within the cell membrane.[71] Diets that are deficient in fat or essential fatty acids decrease the activity of the enzyme systems responsible for the metabolism of nutrients.[90] Plasma-free fatty acid levels become elevated after consumption of a high-fat meal, increasing the potential to become bound to plasma albumin, and subsequently displace albumin bound drugs, increasing the risk of drug toxicity.[85] Dietary fats along with food-stimulated secretions (e.g., bile salts) may facilitate the solubilization and dispersion of lipophilic compounds. This may contribute to a reduction in the extent of first-pass metabolism due to enhanced splanchnic blood flow.[91] Ingestion of diets high in fat has been associated with the induction of CYP2E1. The extent to which this enzyme is upregulated is dependent upon the type of fat. Polyunsaturated fats such as corn and menhaden oils appear to have the greatest influence in comparison to lard or olive oils. This can result in enhanced peroxidation of the PUFA substrates and contribute to free radical production.[92] The rate of gastric emptying is also influenced by the fat content of a meal. Fat retards gastric emptying to a greater degree than does protein or carbohydrate.[93]

The effect of dietary fat on drug absorption, however, depends on the route of the drug absorption, either portal or lymphatic. For medications absorbed via the lymphatic route, dietary fat enhances the absorption of the dissolved drug while poorly bioavailable lipophilic drugs absorbed by the portal route have their absorption enhanced by improved drug dissolution. Conversely, lipophilic medications that have good bioavailability will less likely to be impacted by a high-fat meal. Hydrophilic medications are probably not impacted significantly when co-administered with fatty meals.[94]

The impact of food on the absorption of cyclosporine is controversial. There are conflicting reports, depending on the formulation studied, stating that food impairs, enhances, or does not affect cyclosporine compared with the fasting state.[95] Similarly, high-fat meals may impact sirolimus bioavailability. When taken with a high-fat meal, the rate of sirolimus absorption is slower and results in a greater AUC in comparison to when

taken in the fasted state. This contrasts with tacrolimus in which a 71% mean decrease in C_{max} and a 39% mean decrease in AUC was seen immediately following its administration after a high-fat meal relative to when the same dose was given in the fasted state.[96]

The antiviral agent zidovudine is also impacted by dietary fat. When administered orally, its absorption is reduced when the drug is taken with a high-fat meal in comparison with when taken in the fasted state.[97] It is recommended that zidovudine be taken on an empty stomach to achieve peak serum concentrations. Similarly, treatment failures can occur if the antitubercular drug cycloserine is taken with a high-fat meal due to decreased absorption.

Minerals

In most instances, mineral deficiencies (i.e., zinc, iodine, magnesium, and potassium) have been associated with a decrease in drug oxidation and drug clearance.[98] Low dietary intake of iron, however, has been associated with an increase in some CYP functions and a decrease in other degradative activities, the reasons for which are not fully apparent. Foods fortified with multivalent minerals, such as calcium, are presenting a new challenge. Any medication carrying a warning to avoid milk, dairy products, or nutritional supplements can be affected by calcium-fortified orange juice.[35] As a class, antibiotics are the most susceptible to chelation and adsorption by fortified cereals, calcium-fortified orange juice or protein supplements. Patients need to be counseled that consuming fortified foods may decrease the clinical efficacy and increase the risk of treatment failure.[99]

Vegetables

Diets rich in vegetables and fruit may also impact the response to medications. Both serve as sources of trace minerals that are contained in metalloenzymes, including several antioxidants. Many plants contain flavonoids, isothiocyanates, and allyl sulfides that are potent modulators of the cytochrome monoxygensease system.[100]

Phyotochemicals are linked with the modulation of a variety of metabolic pathways. The most frequently sources include cruciferous vegetables, citrus juices, and spices. Dietary supplements and herbs are also associated with this category. There are five major families of phytochemicals: carotinoids (e.g., beta carotene, lycopene), alkaloids, phenolics (including flavonoids, coumarins, tannins), nitrogen compounds,

and sulfur compounds (e.g., isothiocyanates, allylic sulfur).[101] Recent research has focused on how vegetables and fruits can influence a variety of enzymatic pathways. Typically induction of these enzyme systems is rapid and plateaus within 5 days of continued daily ingestions of the food with the enzyme-inducing capacity.[101]

Cruciferous vegetables, including brussels sprouts, cabbage, turnips, broccoli, cauliflower, and spinach, contain indols that induce aryl-hydrocarbon hydroxylase enzyme activity as well as the conjugation of phenacetin and acetaminophen.[102,103] In one study, patients fed a diet of Brussels sprouts and cabbage had a 50% lower serum phenacetin level in comparison with the same subjects fed a control diet that contained none of the enzyme-inducing vegetables.[103] A later study investigated the effects of cruciferous vegetables on acetaminophen conjugation.[102] The researchers found that the test diet of cruciferous vegetables enhanced acetaminophen glucuronide conjugation as evidenced by a 16% decrease in the AUC, a 17% increase in metabolic clearance, and an increased plasma acetaminophen glucuronide-to-acetaminophen ratio.

Cruciferous vegetable are not the only plants that can impact drug response. Potatoes, tomatoes, and eggplant contain natural insecticide compounds called solanaceous glycoalkaloids that even in small amounts may greatly slow the metabolism of muscle relaxants and anesthetic agents such as suxamethonium, mivacurium, and cocaine. Cooking does not reduce them, and they may remain in the body for several days after ingestion. Solanaceous glycoalkaloids inhibit butyryl cholinesterase, which breaks down many anesthetic agents, and cetylcholinesterase, which breaks down acetylcholine. When these enzymes are inhibited, patients receiving medications metabolized by them may take longer to recover fromanesthesia.[104]

Grapefruit Juice and Drug Interactions

Bailey et al. discovered that grapefruit juice, which was used as a taste-masking agent for alcohol, caused a two- to threefold increase in oral absorption of the calcium channel blocker felodipine.[105] This finding has subsequently led to the intensive investigation of grapefruit juice and drug interactions. This interaction is the classic example of drug–nutrient interaction exclusively caused by inhibition of intestinal CYP3A4. Oral absorption pharmacokinetic studies of CYP3A4 substrates, such as cyclosporine or felodipine, consistently showed that grapefruit juice increased the oral bioavailability of these agents. Interestingly, the plasma

half-lives of most of the drugs studied were not affected, suggesting that the systemic clearance or hepatic metabolism of these drugs was unchanged by grapefruit juice. It was later determined that this interaction occurs in the enterocytes but not in the hepatocytes. Furthermore, Lown et al. showed that repeated consumption of grapefruit juice inhibits not only the intestinal CYP3A4 activity but also the expression of this gene in the enterocytes.[11] As grapefruit juice inhibits intestinal CYP3A4 expression and thus decreases the presystemic metabolism of certain drugs, the bioavailability of the affected agents will remain increased until the expression of the CYP3A4 gene returns to baseline. This suggests that mere separation of the administration time between grapefruit juice and the potential interacting drugs cannot prevent this interaction.

Grapefruit products also interact with uptake transporters such as organic anion-transporting polypeptides (OATPs).[106] OATPs are uptake transporters distributed in the liver, small intestine, kidney, and brain. In the small intestine, OATPs are expressed on the apical surface of enterocytes, facilitating in the intestinal absorption of orally administered medications. Thus, nutrients can alter OATP activity that can impact the bioavailability of a drug that relies on influx transport systems for its intestinal absorption. Fexofenadine was the first OATP substrate identified to interact with grapefruit juice. Inhibition of OATP1A2 transport by flavonoids in grapefruit (naringin) lowered oral fexofenadine bioavailability significantly.[107] Similar results were observed with orange juice, which contains the flavonoid hesperidin. This decrease in oral bioavailablity among OATP1A2 substrates impacts both hydrophilic and renally excreted drugs such as atenolol and ciprofloxacin, more than lipophilic agents. Medications with greater polarity and which are eliminated mainly by excretion are more dependent on uptake transporters rather than passive diffusion. Thus, OATP1A2 substrates, which are excreted renally unchanged, may undergo significant decrease in oral bioavailability if given concurrently with orange or grapefruit juice.[108] Conversely, the oral bioavailability of a medication such as imatinib, an OATP1A2 substrate that is mainly metabolized by CYP3A4, is increased in the presence of grapefruit juice because CYP3A4 inhibition is more dominant than OATP1A2 inhibition as it relates to imatinib pharmacokinetics.

As the aforementioned examples illustrate, the clinical significance of these interactions depends on the pharmocokinetic and toxicity profiles of the co-administered medication. Rather than empirically reducing the dose of the affected drugs to avoid toxicity, it is more advisable to suggest

patients avoid grapefruit juice if they are taking an interacting drug since the magnitude of interaction is inconsistent among individuals and difficult to predict. In some centers, grapefruit juice has been removed from the institutional formulary to minimize the risk of this interaction.

HIGH-FIBER DIETS

Dietary fiber and other bulk-forming compounds may interfere with the GI absorption of a medication.[109] The bioavailability of digoxin is reduced significantly when given with a fiber-rich meal, with almost half of the dose sequestered in or bound to the fiber.[110] Similar effects have been reported with lithium salts and lovastatin.[111] Stewart reported that several patients with recurrent major depression who had been successfully treated with tricyclic antidepressants became refractory to treatment after beginning a high-fiber diet.[112] Nutrient absorption can also be negatively impacted in the presence of a high-fiber diet. Zinc absorption in the intestine is reduced by the binding of zinc to a number of materials, including phytate, which is found in high-fiber diets.[113]

COOKING METHODS

Drug metabolism can also be enhanced by the ingestion of charcoal-broiled beef. Patients fed such a diet saw a significant drop in serum phenacitin levels in comparison to the same subjects fed a control diet. It has also been reported that charcoal-broiled beef induced the metabolism of antipyrine and theophylline, reducing the half-lives of these drugs by 20%.[114] It has been postulated that these effects were related to the large quantities PAHs present in charcoal-broiled beef.

VEGETARIANISM

Drug metabolism among vegetarians will vary dramatically depending on the protein intake. Most research has focused on Asian vegetarians in which the half-lives of drugs that underwent significant hepatic metabolism (e.g., antipyrine, acetaminophen, phenacetin) were significantly longer than in nonvegetarians.[115] When similar studies were conducted in white vegetarians, they found no significant difference in half-life between the vegetarians and nonvegetarians.[116] Protein quality may contribute to these findings as the authors found that protein intake

between the white vegetarians and the nonvegetarians was similar.

Vegetarian diets are also associated with lower circulating concentrations of sex steroids hormones, increased fecal excretion of estrogens, and different hormonal profiles in comparison to individuals consuming an omnivorous diet.[117] Vegetable intake may influence total body estrogen load via the modulation of CYP enzymes involved in estrogen metabolism. CYP 13C, found in cruciferous vegetables, can increase estrogen hydroxylation.[118]

VERY LOW CALORIE DIETS

In diets involving severe protein–energy restriction, such as extreme slimming diets, the metabolism of drugs may be affected in one of two ways. First, tissue protein is catabolized and used as an energy source, thus reducing the availability of amino acids for protein synthesis, which in turn reduces the amount of enzymes available for drug metabolism. Second, endogenous substrates derived from carbohydrate and protein such as glucuronide, sulfate, and glycine could also compete for the tissue needs for these nutrients and that of the drug metabolism/detoxification.[119] Eating behaviors, especially among dieters that omit or severely restrict whole categories of foods, have a negative impact on micronutrient status. Diets that eliminate all animal foods provide almost no cyanocobalamin and have been associated with other vitamin deficiencies including vitamin C. Moreover, skipping meals and fad diets to lose weight frequently compromise micronutrient intake. It should be routinely assumed that it is extremely difficult to meet all the requirements at intakes of less than 1200 calories per day. An analysis of 11 published weight loss diets showed that none provided 100% of the US Recommended Dietary Allowance (RDA) for representative vitamins.[119]

In a review by Anderson, the author suggests that the metabolic improvements seen after obese patients receive very low calorie diets (VLCDs) are not different from those obtained by equally large weight loses promoted by other methods.[120] Patients with VLCD weight loss have improved hyperinsulinism as a result of a reduction in basal insulin production as well as enhanced hepatic insulin extraction. Moreover, it is thought the weight loss through VLCD lowers the hepatic glucoronidation of drugs leading to higher plasma concentrations of the affected drugs.[121] Despite this, however, the fatty liver may still be more susceptible to drug toxicity.[122] For example, several reports have suggested that caloric

restriction may contribute to acetaminophen hepatotoxicity by altering drug metabolism although under controlled conditions, Schenker et al. saw no impact on food restriction of acetaminophen metabolism.[123]

OTHER DIETARY RESTRICTIONS

In addition to caloric restriction, restriction of other dietary components can also impact drug response. In patients who have sodium restricted diets, there is an increased risk of acute renal failure if these same patients are given concomitant angiotensin-converting enzyme inhibitors or nonsteroidal anti-inflammatory agents. There is enhanced nephrotoxicity in patients who are sodium depleted and are given cyclosporine or tacrolimus. Sodium restriction can also increase the renal tubular absorption of lithium, leading to toxicity. Patients receiving aminoglycosides, amphotericin B, cisplatin, or radiocontrast media in conjunction with a low sodium diet have an increased risk for hemodynamic nephrotoxic and ischemic acute renal failure. For reasons still not known, the efficacy of calcium channel blockers is reduced in patients on a sodium-restricted diet.[124]

IMPACT OF BEVERAGE TYPE ON DRUG BIOAVAILABILITY

To most practitioners, the term beverage refers to any drinkable liquid other than plain water. They are typically classified as caffeinated, alcoholic, milk-based, mineral waters, or fruit/vegetables juices.[125] Depending on the type of fluid taken with a medication, drug absorption may be impacted. Mixing drugs with fruit juices or other beverages to mask their taste may impact absorption due to changes in gastric pH, which in turn can affect dissolution of solid dosage forms and formulations with a pH-sensitive coating. Dairy products decrease the absorption of tetracyclines and reduce their bioavailability due to the formation of insoluble chelates between the drug and the calcium present in the beverage. Similar decreases in bioavailability were noted when fluoride tablets are taken with milk. Tannins present in teas may impair iron absorption. Alcoholic beverages reduce the absorption of folic acid, cyanocobalamin, and magnesium. Soft drinks, such as colas, may decrease the drug absorption for a variety of reasons. The phosphoric acid and sugar present in these drinks can slow gastric emptying, and the tendency to

serve them chilled may also reduce the rate of blood flow within the intestines. Moreover, the carbonation may increase mixing and possibly motility. Interestingly, the acidic pH of cola beverages can be used to optimize clinical responses of both ketoconazole and itraconazole when administered as solid dosage forms in patients with gastric hypochlorhydria, such as those patients with AIDS gastropathy. Acidic beverages improve dissolution of tablets and capsules, thus can improve and stabilize oral bioavailability if these drugs are ones that are poorly soluble in higher pHs.[125] The effects of grapefruit juice on drug disposition are discussed separately.

PARENTERAL NUTRITION

Lack of oral nutrient intake during PN leads to mucosal atrophy of the bowel along with a reduction in gastricbiliary, pancreatic, and intestinal secretions.[126] Bacterial overgrowth can result in a progressive decline in intestinal function owing to impaired motility and depressed enzyme activity. This may alter the rate and extent of absorption of specific nutrients as well as various drugs. Decreases in nutrient absorption include fat, iron, peptides, and vitamins A and B_{12} as well as drugs such as chloramphenicol, chloroquine, tetracycline, and rifampin.[47]

PN has been observed to decrease hepatic drug and xenobiotic metabolism in animals and humans.[127,128] Drug metabolism may be altered due to jejunal and ileal mucosal hypoplasia and hypofunction as parenterally administered nutrients bypass the intestine. Studies using antipyrine as a marker have shown that PN regimens containing amino acids have higher antipyrine clearance than regimens consisting mainly of dextrose.[127] Burgess et al. investigated the response of PN regimens on antipyrine clearance.[128] Patients receiving a postoperative 2000 kcal PN regimen providing all nonprotein calories as dextrose showed a 34% reduction of mean antipyrine clearance after 7 days of PN compared with unfed controls. This effect was seen also in patients receiving a 1600 kcal dextrose-based regimen. More over, in patients receiving a 2000 kcal PN regimen in which 500 kcal was provided as lipid, mean antipyrine clearance was not significantly different from that of the unfed control group. This study suggested that hepatic CYP1A activity may be affected by different PN regimens.

EFFECTS OF DRUGS ON NUTRIENT STATUS

Drugs can alter the use of many nutrients by a variety of mechanisms, both specific and nonspecific. Malabsorption syndromes may occur from direct toxic effects of the medication to the intestinal mucosa, inhibition of enzymes, binding of bile and fatty acids, alteration of dietary ions, or alteration of GI pH. Indirect, nonspecific effects of a drug may manifest themselves as a decrease in appetite, ultimately leading to a decrease in food intake.

Although the concept of drug–nutrient interactions is not a new one, only recently has it reached the forefront in the medical world. Malnutrition, as well as the composition of the diet, can affect the disposition of a medication's half-life.[129] In addition to traditional dietary components, nonnutrient ingredients, such as food additives, preservatives, antioxidants, sweeteners, flavoring, and coloring agents, can interact with a medication. Natural products, such as plant food groups containing alkaloids, flavonoids, and other compounds, can behave in the body in the same manner of that a drug and, consequently, can be involved in a variety of interactions and are potentially toxic if they are consumed or accumulate in the body.[130] Drug–nutrient interactions are typically defined as situations that result from chemical, physical, physiologic, or pathophysiologic relationships between nutrients and drugs.[131] Infants and adolescents, like the elderly, are at particular risk for drug–nutrient interactions. Several factors may influence the possible interactions: first, their nutrient needs are typically higher; second, the systems for detoxification of nutrients may be incomplete; and third, adolescents (especially females) tend to restrict their diets and thus are unable to meet the actual recommended intakes for a variety of micronutrients.[132] Furthermore, the minimum dietary requirement may be insufficient when a patient is under psychological and pathologic stresses, as in the case of pregnancy or athletic training. In pathologic states, the requirement for some micronutrients may be increased.

Drugs may potentially cause vitamin deficiency ranging from subclinical deficiency to potentially fatal sequelae. The clinical intensity of the interaction in many cases may depend on the nutritional status of the patient. Patients with borderline intake of vitamins or those in poor nutritional health appear to be at a greater risk of developing symptomatic vitamin deficiency states. Other factors include the type of drug treatment, dose, and duration of therapy, as well as the age of the patient. For example, individuals on a normal diet rarely become vitamin K deficient,

although malnourished patients, regardless of cause, often have moderate or significant deficiency, often with prolonged bleeding times.[132,133] Cohen et al. reported that hospitalized patients may become vitamin K deficient within 7–10 days after admission, with those at greatest risk being those who were previously malnourished and had received antibiotics for 7 or more days.[134] Adolescents, due to periods of rapid growth, are particularly sensitive to micronutrient status. Even a typical diet may not meet their high nutrient demands necessary for growth. Intake of folate and vitamin A tend to be less than optimal.[126]

ABSORPTION OF NUTRIENTS

There are also many secondary mechanisms that can interfere with nutrient absorption. Medications may alter gastric or intestinal secretion, pancreatic exocrine function, or hepatic bile secretion. For example, H_2 antagonists and proton pump inhibitors inhibit gastric acid production. Chronic use of these medications can significantly decrease the bioavailability of the dietary source of vitamin B_{12}.[135]

The direct systemic effect of a drug on one nutrient may have secondary effects on another nutrient. Isoniazid and cimetidine inhibit the hydroxylation of vitamin D in the liver and kidney, and phenytoin and phenobarbital promote the breakdown of vitamin D metabolites, each resulting in a functional deficiency of vitamin D and secondary impairment of calcium absorption.[136] Neomycin, colchicine, and para-aminosalicylic acid may damage intestinal mucosa and destroy intestinal villi and microvilli, resulting in an inhibition of brush border enzymes and intestinal transport systems.[137] Nonsteroidal anti-inflammatory agents can cause multiple small hemorrhages of the intestinal mucosa leading to iron-deficiency anemia and decreased absorption of vitamin C.[138] Folic acid deficiency and macrocytic anemia can occur in patients on chronic aspirin therapy, especially if their diet is low in dietary sources of folate.[139]

Laxative abuse has also been implicated with malabsorption of vitamin D and calcium. Use of irritant laxatives, such as phenolphthalein and bisacodyl, may damage intestinal epithelial cells and impair colonic reabsorption, resulting in steatorrhea, protein-losing enteropathy, and decreased absorption of glucose, potassium, calcium, and vitamin D.[140] Other stool regimen medications, such as docusate, alter electrolyte transport and can cause hypomagnesemia owing to GI losses and failure of colonic reabsorption.[141] Orally ingested mineral oil can coat ingested food

particles along with the surface of the intestines. This forms a mechanical barrier to the digestion and absorption of nutrients. Mineral oil also increases gastric motility, which reduces the time required to adequately absorb ingested nutrients.[142] Furthermore, studies have shown that mineral oil, especially when taken at mealtime or during the postprandial absorptive period of optimal nutrient absorption, can reduce the absorption of vitamin D.[142]

Alterations in vitamin and mineral absorption can also occur owing to chelation or precipitation with a medication. Cholestyramine, a basic anion-exchange resin, binds bile salts and impairs the absorption of fat-soluble vitamins, vitamin B_{12}, folic acid, and the minerals such as calcium, iron, and zinc.[143] Aluminum- and magnesium hydroxide–containing antacids may form nonabsorbable phosphate in the gut lumen, resulting in hypophosphatemia along with anorexia and secondary syndromes of hypomagnesemia and osteomalacia.[136]

Alteration in GI pH may also cause drug-induced malabsorption. Drugs that increase gastric pH (e.g., H_2 antagonists) can theoretically decrease the breakdown of fat necessary to complex with calcium, thus reducing gut absorption.[144] Antacids can also reduce the bioavailability of riboflavin and folic acid as well as copper and iron, all of which depend on a low pH.[145] In some cases, the effect of drugs on nutrient malabsorption may be indirect. Proton-pump inhibitors (e.g., omeprazole, lansoprazole) suppress gastric acid release and induce a dose-dependent achlorhydric state. The increase in gastric pH reduces the amount of cobalamin liberated from diet. This, in turn, limits the binding of cobalamin to intrinsic factor in the jejunum. Chronic use of this class of drugs may lead to malabsorption of cobalamin in some patients. These drugs do not affect the bioavailability of crystalline vitamin B_{12} which is commonly used in vitamin supplements. Thus, as in the case of RYGB surgery recipients, oral vitamin supplementation may be necessary and is an effective approach to minimize the risk of developing anemia in patients receiving chronic acid reduction therapy.[146]

METABOLISM OF NUTRIENTS

Drugs may affect nutrient metabolism by several methods. They may inhibit the essential intermediary metabolism of a nutrient, usually a vitamin, or promote the catabolism of the nutrient. Medications with these properties may be used therapeutically, as in the case of coumarin

anticoagulants or methotrexate. In other cases, this may be an unwanted side effect, as in the case of pyridoxine antagonism seen in isoniazid and hydralazine use. Isoniazid and hydralazine can result in pyridoxine deficiency by the inhibition of pyridoxal kinase. Both compounds deplete pyridoxine stores and consequently the neurotransmitter gamma-aminobutyric acid, resulting in seizures. Administration of pyridoxine in cases of isoniazid overdose eliminates seizures and metabolic acidosis, which often occur.[147]

Many medications induce drug metabolism enzymes leading to greater activity of these enzymes, increasing the demand for their vitamin cofactor, and, with chronic drug therapy and marginal nutrient intakes, can precipitate signs of deficiency of several vitamins, especially folic acid. Triamterene can inhibit dihydrofolate reductase, resulting in megaloblastosis.[148] The 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitor class of antihyperlipidemic agents (i.e., statins) can lower plasma concentration of endogenous ubiquinone (coenzyme Q), a cellular antioxidant.[149]

Patients receiving chronic anticonvulsant therapy are at particular risk of developing metabolic bone disease as a result of these types of reactions.[150] Several mechanisms have been suggested. One suggests that drugs such as phenytoin, a potent inducer of multiple CYP enzymes, stimulate the catabolism of vitamin D to produce an inactive metabolite since the biotransformation of vitamin D is mediated by numerous CYP-dependent pathways.[151] Although not well proven, another theory suggests that phenytoin, alone or in combination with phenobarbital, interferes with vitamin K metabolism, with a corresponding elevation in serum osteocalcin levels without a-carboxyglutamate (Gla) residues, resulting in metabolic bone disease because osteocalcin-containing Gla residue is necessary for normal bone mineralization.[152]

Patients treated with cephalosporin antibiotics may develop hemorrhagic states owing to drug-induced vitamin K deficiency.[153] It is thought that these antibiotics block the vitamin K reductase, which is necessary for vitamin K activation.[154] They may also block carboxylation of vitamin K–dependent peptides to yield Gla residues that are required for calcium binding in the conversion of vitamin K–dependent proenzymes to their active state, which are needed in the coagulation cascade.[154]

Some drugs have been linked with subnormal serum folate levels, probably owing to weakly bound folate to serum proteins that can be easily displaced by drugs.[155] The mechanism for this interaction has yet to be

described, but it appears that there is a decrease in serum binding of methyltetrahydrofolate caused by the drug itself or one of its metabolites.[156]

Folate deficiency secondary to long-term phenytoin therapy is a common occurrence; approximately 75% of patients taking anticonvulsants have low serum folic acid levels.[157] However, progression to megaloblastic anemia is relatively rare.[158] Supplementation of folic acid with as littleas 1 mg per day may lead to a significant decrease in serum phenytoin concentrations in 15%–50% of the patients.[159] Although the exact mechanism is unknown, pharmacokinetic analysis of phenytoin suggests that folic acid may increase the affinity of the metabolic enzyme(s) involved in the elimination of the phenytoin without causing overall enzymatic induction.[159,160] To avoid potential folate deficiency and subsequent fluctuations in serum phenytoin levels, practitioners should routinely supplement all patients with folate when phenytoin therapy is initiated.[159]

Like phenytoin, sulfasalazine, an anti-inflammatory agen tused to treat Crohn's disease and juvenile rheumatoid arthritis, is one of the leading causes of drug-induced folic acid deficiency in children.[160] Megaloblastic anemia, caused by folate deficiency, has been reported in patients receiving high doses of sulfasalazine for prolonged periods.[161]

Aspirin has also been linked with folate deficiency. Serum folate levels are known to be low in many patients with rheumatoid arthritis.[160] It is thought that aspirin alters the transport of folate by competing for binding sites on serum proteins.

Methotrexate, a chemotherapeutic agent that is also used to treat psoriasis, rheumatoid arthritis, and Crohn's disease, limits the availability of methyl groups derived from one-carbon metabolism by inhibiting competitively a key enzyme in the intracellular folate metabolism.[162] Methotrexate functions as an antimetabolite, reversibly inhibiting dihydrofolate reductase.

Valproic acid (VPA), an antiepileptic agent, has been associated with folate deficiency. Like other agents, the mechanism is not fully understood, although it is thought that VPA causes an alteration in the methionine cycle.[163] This results in an elevation of tetrahydrofolate levels and a reduction of both formulated forms of folate. This could also explain VPA's role as a teratogen.[164] In addition, it has been established that VPA may induce L-carnitine deficiency. Patients who develop VPA-induced carnitine deficiency typically experience hyperammonemia syndrome with

no other abnormality in the liver function tests. If diagnosed early, the clinical presentation, which includes altered mental status or encephalopathy, can be reversed by L-carnitine supplementation.[165]

DRUG EFFECTS ON NUTRIENT TRANSPORT

There are several mechanisms by which a drug can impact nutrient transport and then lead to nutritional deficiency. A nutrient can become displaced from plasma protein binding sites by a medication, leading to an increase in the renal excretion of the nutrient, or a drug may compete with a nutrient such that intracellular use of the nutrient is impaired.[166] This may occur when folate stores become depleted in patients receiving therapeutic doses of aspirin.[166]

Table 18-5 Examples of Drug–Nutrient Interactions

Medication/Drug Class	Nutrient	Interaction
5-ASA derivatives	Folic acid	Decreased serum folate levels
Albuterol	Glucose	Hyperglycemia (in diabetics)
Aminoglycosides	Potassium Magnesium Sodium Calcium	Increased urinary excretion of potassium and magnesium and may also deplete sodium and calcium
Amphotericin	Masium Potassium Sodium	Electrolyte wasting
Aspirin	Folic acid	Decreased serum folate levels
Bisacodyl	Glucose	Decreased absorption
Calcium carbonate	Iron	Decreased iron absorption
Captopril	Potassium	May cause hyperkalemia
Chloramphenicol	Protein Riboflavin Vitamin B_6 Vitamin B_{12}	Decreased protein synthesis
Cholestyramine	Fat MCT Vitamins A,D, E, K Vitamin B_{12}, folate, calcium Glucose, xylose	Decreased absorption

Cimetidine	Vitamin B_{12}	Depletion of vitamin B_{12} stores
Cisplatin	Magnesium	Magnesium depletion
Colchicine	Vitamin B_{12} Vitamin A Folate Potassium Sodium	Causes intestinal mucosal damage, decreases vitamins A and B_{12} absorption
Corticosteroids	Glucose	Hyperglycemia
Digoxin	Bran fiber Calcium Magnesium Potassium	Decrease drug absorption Arrhythmias Hypomagnesemia or hypokalemia may enhance toxic effect of digoxin; arrhythmias
Ethambutol	Copper Zinc	Copper and zinc depletion
Fluconazole	Potassium	Hypokalemia
Fluorouracil	Thiamine	Inhibits conversion of thiamine to thiamine pyrophosphatase; increase thiamine requirements
Foscarnet	Calcium Magnesium Phosphorus Potassium	Electrolyte wasting
Furazolidine	Tyramine-rich foods	Increased risk of hypertensive effects if used chronically or at high doses with tyramine-rich foods
Furosemide	Calcium Magnesium Potassium Sodium	Electrolyte depletion
Gemfibrozil	Vitamin E Ubiquinone	Decreased absorption
Insulin	Dextrose	Increased insulin requirements
H-2 antagonists (cimetidine, ranitidine, famotidine)	Vitamin B_{12} Zinc	Depletion of vitamin B_{12} stores May deplete zinc
Isoniazid	Pyridoxine Tyrosine-rich foods Histamine-rich foods Tyramine-rich foods	Vitamin B_6 antagonism Blocks conversion of tyrosine to niacin May cause headaches, itching, redness, chills, hypotension Isoniazid has some MAO-I activity; tyramine-rich foods may cause hypertensive crisis
Linezolid	Tyramine-rich foods	Linezolid has some MAO-I properties; tyramine-rich foods may cause

		hypertensive crisis
Lithium	Sodium	Increased sodium intake decreases lithium's effectiveness; decreased sodium intake may increase lithium toxicity
Metformin	Coenzyme Q10 Folic acid Cobalamin	Depletes stores
Methotrexate	Folic acid	Methotrexate may cause folate deficiency; folate supplementation may decrease methotrexate effects
Mineral oil	Fat-soluble vitamins	Decreased absorption of fat-soluble vitamins with chronic use
Neomycin	Fat-soluble vitamins MCT Cobalamin Sodium Glucose Lactose Sucrose Xylose	Impaired absorption
Nonsteroidal anti-inflammatory agents	Potassium	Hyperkalemia in patients with renal disease or receiving potassium supplements or potassium-sparing diuretics
Orlistat	Vitamin E Vitamin D Beta carotene	Reduced absorption
Oral contraceptives	Ascorbic acid Folic acid Pyridoxine	Ascorbic acid, folic acid, and vitamin B_6 requirements increased
Paraaminosalicylic acid (PAS)	Vitamin B_{12} Folate Calcium Iron Magnesium	Decreased absorption of cobalamin, folate, calcium, iron, magnesium
Penicillamine	Acidic foods/beverages	Decreased penicillamine absorption/inactivation
Penicillin	Acidic foods/beverages	Penicillin inactivation
Phenobarbital	Ascorbic acid Vitamin D	Decreased ascorbic acid absorption Interferes with vitamin D metabolism
Phenytoin	Folic acid Pyridoxine Vitamin D	High-dose folic acid may antagonize phenytoin effects; phenytoin may cause folate deficiency resulting in megaloblastic anemia

		May antagonize phenytoin Interferes with vitamin D metabolism
Primidone	Folic acid	May cause folic acid deficiency leading to megaloblastic anemia
Proton pump inhibitors	Iron Cobalamin Magnesium	Decreased absorption
Pyrimethamine	Folic acid	Decreased serum folate levels
Sevelamer	Folic acid Vitamin D Vitamin E Vitamin K	Decreased levels
Spironolactone	Sodium Potassium	May cause hyponatremia May cause hyperkalemia
Sulfasalazine	Folic acid	Inhibits the absorption of folic acid
Terbutaline	Glucose	Hyperglycemia (in diabetics)
Theophylline	Caffeine Carbohydrates Charcoal-broiled meat Protein	Increases theophylline side effects Decreased carbohydrate intake may decrease plasma half-life of theophylline May decrease theophylline half-life Increased protein intake may decrease plasma half-life of theophylline
Thiazide diuretics	Magnesium Potassium Sodium	Increased electrolyte wasting
Triameterene	Folic acid	May cause folic acid depletion
Trimethoprim	Folic acid	May cause folic acid depletion
Valproic acid	Carnitine	May cause carnitine deficiency with hyperammonemia
Warfarin	Vitamin K Onions, garlic Vitamin E	May inhibit warfarin response; increase dose needed Excessive amounts may increase the fibrnolytic activity of warfarin May enhance anticoagulant effect of warfarin
Zidovudine	Carnitine Folic acid	May cause carnitine deficiency May cause megaloblastic anemia
Zonisamide	Biotin Vitamin B_1 Vitamin B_2 Niacin Pyridoxine Vitamin B_{12} Vitamin K	May deplete stores

Excretion

Thiamin deficiency can occur in patients treated with chronic loop diuretic therapy. Seligmann et al. reported that patients with congestive heart failure treated with long-term furosemide therapy had increased urinary excretion of thiamin, leading to frank deficiency over time that may have contributed to poor cardiac performance.[167] Similar findings were reported by Brady et al., who observed laboratory evidence of thiamin deficiency in congestive heart failure patients being treated with loop diuretics on a chronic basis.[168]

Drug-Induced Fluid and Electrolyte Imbalance

Several widely used medications can affect electrolyte balance, as summarized in Table 18-5. Sodium balance is altered by thiazide diuretics and the neuroleptic carbamazepine.[169,170] Patients treated long term with carbamazepine have been reported to develop the syndrome of inappropriate antidiuretic hormone (SIADH), resulting in hyponatremia and water retention.[170] In those patients, hyponatremia develops owing to the enhanced renal conservation of water. Similarly, patients receiving excessive diuretic therapy with fluid loss replaced with excessive water are prone to developing SIADH.[169] Conversely, certain medications can cause hypernatremia as a result of water loss and subsequent dehydration. Hypernatremia, secondary to excessive lactulose therapy for hepatic encephalopathy or constipation, is a common drug-induced cause of this disorder.[171]

Renal wasting of potassium, resulting in hypokalemia, has been associated with thiazide and loop diuretics, corticosteroids, amphotericin B, and antipseudomonal penicillins.[172] Insulin and inhaled β2-agonists (e.g., albuterol) can cause a shift of potassium from the extracellular to the intracellular spaces.

Potassium-sparing diuretics (e.g., spironolactone), angiotensin-converting enzyme inhibitors (e.g., enalapril), heparin, and trimethoprim can cause hyperkalemia.[172] A variety of mechanisms are involved. Trimethoprim has weak diuretic properties with potassium-sparing activity.[173] Heparin can suppress aldosterone, leading to sodium wasting and potassium retention. Patients with renal insufficiency or diabetes mellitus appear to be more susceptible to heparin-induced hyperkalemia.[174]

The impact of medications on phosphorus balance is important in patients receiving nutritional support as the synthesis of new cells

increases the need for phosphorus. Patients already at risk for refeeding syndrome are particularly susceptible to the effects of drugs known to decrease available phosphorus stores.[175] Drugs such as antacids and sucralfate can alter the absorption of phosphorus from theGI tract by binding to dietary phosphate, thus preventing its absorption. Conversely, patients with renal dysfunction are at risk for development of hyperphosphatemia owing to the inherent phosphate content present in the phospholipid emulsifiers in intravenous fat emulsion or clindamycin phosphate injection.[175]

As discussed previously, medications such as isoniazid and cimetidine can inhibit the hepatic and/or renal hydroxylation of vitamin D, leading to impaired calcium absorption.[176] Odes et al. concluded that even short-term therapy with cimetidine altered vitamin D metabolism in humans.[176] The researchers came to this conclusion after studying 25-hydroxyvitamin D_3 levels for 30 days in patients treated with cimetidine.

Chronic corticosteroid use can cause a net negative calcium balance and increased bone resorption owing to suppressed intestinal absorption of calcium in conjunction with increased renal calcium and phosphate excretion and a subsequent decrease in renal tubular calcium resorption, resulting in bone osteopenia.[136,177] Even frequent use of inhaled steroids has been associated with this response.[178]

Hypomagnesemia as a result of renal wasting can occur in patients treated with loop diuretics, thiazide diuretics, amphotericin B, aminoglycosides, cisplatin, or cyclosporine. Cisplatin-induced hypomagnesemia is dose and duration dependent.[179] Cisplatin induces hypomagnesemia by reducing magnesium reabsorption in the ascending loop of Henle and the distal tubule.[180] Forastiere et al. reported that the total exposure of free platinum contributes to direct injury and renal toxicity.[181] Carboplatin, an antineoplastic with a chemical structure similar to cisplatin, has been reported to have a lower incidence of hypomagnesemia.[182]

Renal wasting of magnesium is also common in patients on prolonged courses of high doses of aminoglycosides.[136,183] Aminoglycosides can inhibit the proximal tubular transport of magnesium in the kidney, predisposing patients with already low intakes of magnesium to hypomagnesemia.[183]

If left untreated, hypomagnesemia will ultimately lead to hypocalcemia. Magnesium deficiency can induce a transient hypoparathyroidism by reducing the secretion of parathyroid hormone

(PTH) and a blunted PTH response. This results in an inhibition of the hypocalcemic feedback loop.[179] Other agents, such as aluminum salts, also suppress PTH secretion, resulting in hypocalcemia. Treatment for hypocalcemia induced by hypomagnesemia involves correcting the hypomagnesemia first and then managing the magnesium losses. In some cases, calcium supplementation may be unnecessary.[179]

TRACE ELEMENTS

Zinc is essential for the function of hundreds of enzymes such as dehydrogenases, aldolases, and peptidases and is involved in a variety of metabolic processes.[184] The formation and activation of zinc-dependent enzymes are regulated by zinc tissue levels.[185] Zinc metalloenzymes are responsible for structural integrity at the cellular level and for the regulation of various aspects of RNA and DNA metabolism.[185] Zinc deficiency limits the activity of these enzymes, resulting in decreased cell replication and tissue growth and repair.

Zinc interacts with several other nutrients. At supplemental doses, zinc may impair copper absorption.[186] Zinc deficiency can also negatively impact vitamin A metabolism by impairing the mobilization of retinol from the liver and altering retinal visual pigment metabolism, which may contribute to the night blindness seen in zinc deficiency.[187] It is thought that zinc deficiency may result in abnormal dark adaptation and/or age-related macular degeneration.[188]

Patients receiving PN are at risk for developing zinc deficiency. Zinc balance during PN depends on the infusion of zinc in amounts sufficient to offset urinary and GI losses that occur independent of zinc intake.[189] Premature infants receiving PN will experience a progressive decline in plasma zinc levels despite 14 days of continuous treatment, reflecting their high metabolic demands.[189]

GLUCOSE

Patients with diabetes mellitus or others with insulin resistance (e.g., severe infections, catabolic stress) are susceptible to the effects of medications known to impact glucose metabolism. Thiazide diuretics, corticosteroids, and cyclosporine have all been associated with inducing hyperglycemia in susceptible patients.[190] Hyperglycemia occurs in approximately 20% of patients treated with pentamidine.[191] Moreover,

protease inhibitors have been recognized as a cause of hyperglycemia.[192] The long-acting somatostatin analog octreotide has been shown to inhibit insulin secretion and may result in a transient deterioration in glucose tolerance on initiation of therapy.[193] In most cases, alteration of carbohydrate metabolism does not appear to be a problem. Table 18-6 lists some of the more common medications that alter glucose metabolism or response.

Table 18-6 Medications That Alter Glucose Metabolism/Response

Medication/Drug Class	Response
Acarbose	Hypoglycemia
ACE inhibitors	Hypoglycemia
Acetazolamide	Hyperglycemia
Acritretin	Hyperglycemia
Amiodarone	Hyperglycemia
Amprenavir	Hyperglycemia
Arginine	Hyperglycemia
Chloroquine	Hypoglycemia
Clofazimine	Hyperglycemia
Corticosteroids	Hyperglycemia
Clonidine	Hyperglycemia
Cyproheptadine	Hyperglycemia
Diazoxide	Hyperglycemia
Didanosine	Hyperglycemia
Emtricitabine	Hyperglycemia
Epinephrine	Hyperglycemia
Ethionamide	Hypoglycemia
Etravirine	Hyperglycemia
Fosamprenavir	Hyperglycemia
Fosphenytoin	Hypoglycemia
Glucagon	Hyperglycemia
Glycerol	Hyperglycemia
Haloperidol	Hyperglycemia/hypoglycemia
Imatinib	Hyperglycemia
Indinavir	Hyperglycemia
Isotretinoin	Hyperglycemia
Lamivudine	Hyperglycemia

L-Asparaginase	Hyperglycemia
Lithium	Hyperglycemia
Loop diuretics	Hyperglycemia
Megestrol	Hyperglycemia
Metformin	Hypoglycemia
Micofungin	Hyperglycemia/hypoglycemia
Nelfinavir	Hyperglycemia
Nicotinic acid	Hyperglycemia
Oral contraceptives	Hyperglycemia
Octreotide	Hyperglycemia/hypoglycemia
Paricalcitol	Hypoglycemia
Penacillamine	Hypoglycemia
Pentamidine	Hyperglycemia/hypoglycemia
Phenytoin	Hyperglycemia
Posaconazole	Hyperglycemia
Proton pump inhibitors	Hyperglycemia/hypoglycemia
Quinine	Hypoglycemia
Ritonavir	Hyperglycemia
Saquinavir	Hyperglycemia
Sertraline	Hyperglycemia
Sirolimus	Hyperglycemia/hypoglycemia
SSRIs	Hypoglycemia
Tacrolimus	Hyperglycemia
Valganciclovir	Hyperglycemia
Zalcitabine	Hyperglycemia

Hypoglycemia is the most common metabolic abnormality associated with pentamidine therapy.[194] The mechanism responsible for this adverse effect may involve a direct cytolytic effect on pancreatic beta cells, resulting in insulin release and hypoglycemia and a subsequent insulin deficiency owing to loss of beta cell function.[195] Eventually, however, pentamidine-induced pancreatic beta cell damage may lead to insulin deficiency and result in hyperglycemia, although this is considerably less frequent than hypoglycemia.[195]

Drugs or foods that have the ability to induce a rapid release of insulin can increase the risk of hypoglycemia, especially if taken with alcohol. Beta-blockers such as propranolol can inhibit glycogenolysis and, during periods of vigorous exercise, induce hypoglycemia.[196]

FAT

With increased awareness of lipid abnormalities and coronary artery disease, drug-induced lipoprotein abnormalities must be considered; especially in patients in whom no other causes of dyslipidemia exist. Some of the most common causes of secondary dyslipidemia are medications. When a drug is used for a short period only, practitioners need to be aware of the effects on the patient's lipoprotein profile versus the chance that an underlying dyslipidemia has been exacerbated. Drugs used chronically may be more problematic as they may predispose the patient to atherosclerosis.[197] Table 18-7 lists medications associated with dyslipidemia.

Protease inhibitors interfere with some proteins involved in fat metabolism (i.e., cytoplasmic retinoic acid–binding protein type 1).[198] Protease inhibitor binding to low-density lipoprotein (LDL) receptor–related proteins (LRPs) impair hepatic chylomicron uptake and triglyceride clearance by the endothelial LRP–lipoprotein lipase complex. The resulting hyperlipidemia contributes to central fat deposition and insulin resistance.[198] It is also thought that protease inhibitors may also disrupt steroid hormone production, leading to lipodystrophy.[199] See Chapter 49 for a full discussion of HIV lipodystrophy.

L-Asparaginase has been reported to cause abnormalities in lipid metabolism, ranging from hypocholesterolemia and hypotriglyceridemia to hypercholesterolemia and hypertriglyceridemia. Parsons et al. suggested that this alteration in lipid metabolism is caused by structural changes in the high-density lipoprotein (HDL) particles from high density to lower density that is reflected in an altered ratio of lipid to protein.[200] The authors suggested that prior to the initiation of L-asparaginase therapy, HDL-cholesterol particles are protein rich but gradually become lipid rich because of an asparaginase-associated reduction in protein synthesis. They also concluded that modifications in asparaginase therapy are not necessary, although close monitoring is recommended for patients whose triglyceride levels exceed 2000 mg/dL when the risk of pancreatitis is increased.

In addition to asparaginase-associated lipid abnormalities, patients being treated for acute lymphoblastic leukemia often receive corticosteroids that may alter lipid and lipoprotein metabolism by increasing hepatic cholesterol synthesis.[200] Corticosteroids increase the frequency of hypercholesterolemia and hypertriglyceridemia with elevations in LDL and HDL levels, often in a dose-related manner.[197]

Females appear to be more susceptible than males to these changes. Switching to alternate-day therapy may reduce lipoprotein levels in some patients.[197]

Orally administered estrogens decrease serum LDL and increase serum HDL levels in a dose-related manner.[197] Moreover, these agents are known to increase serum triglyceride levels by 30%–87%.[197] It has been suggested that low-dose estrogens be used to minimize elevations in triglyceride levels without compromising their favorable impact on LDL and HDL levels.[197] Interestingly, other routes of estrogen administration (topical, intradermal, intramuscular) tend not to have as pronounced an effect on lipoproteins, perhaps because these routes bypass first-pass metabolism through the liver, thus having a smaller impact on hepatic protein synthesis.[197,201] Progestins, often used in combination with estrogens in various oral contraceptive products, can also affect lipoprotein levels. Generally, progestins elevate LDL levels and decrease triglyceride, HDL, and very LDL levels, correlating with the androgenic activity associated with specific progestins.[197] Second- and third-generation progestins (e.g., levonorgestrel, desogestrel, and norgestimate) when used in combination with low-dose estrogen, do not have any appreciable alterations in lipid profiles or at best mild increases in serum cholesterol, LDL, and triglycerides.[201]

Table 18-7 Medications Associated With Dyslipidemia

Medication	Total Lipids	Total Cholesterol	Triglycer-ides	Low-Density Lipoprotein	High-Density Lipoprotein	Very Low-Density Lipoprotein
Abacavir			↑			
Adalimumab		↑				
Amiodarone		↑	↑			
Amprenavir		↑	↑			
Anabolic steroids				↑	↓	
Atazanavir		↑				
Beta-blockers			↑			
Calcitriol		↑				
Cholestyramine			↑			
Colchicine		↓				
Colesevelam			↑			
Cyclosporine	↑	↑	↑	↑		
Danazol				↑	↓	
Dexrazoxane			↑			
Didanosine			↑			
Disopyramide		↑	↑			
Efavirenz		↑	↑		↑	
Emtricitabine			↑			
Enoxaparin	↑					
Ergocalciferol		↑				
Estrogen			↑			
Etravirine		↑	↑	↑		
Fluconazole		↑	↑			
Fosamprenavir		↑	↑			
Glucocorticoids	↑	↑	↑	↑	↓	↑
Idinavir			↑			
Interferons			↑			↑
Isotretinoin		↑	↑		↑	
Itraconazole			↑			
L-Asparaginase			↑			
Miconazole (IV)	↑	↑				
Mycophenolate		↑				
Nelfinavir	↑					
Nevirapine			↑		↑	
Oral contraceptives			↑			
Paclitaxel			↑			
Phenothiazines		↑			↓	
Progestins			~	↑	↓	↓
Propofol	↑					
Retinoids (vitamin A)			↑	↑	↓	↑
Rilpivirine		↑	↑	↑		
Risperidone			↑			
Ritonavir		↑	↑			
Sirolimus		↑	↑			
Sodium phenylbutyrate		↑				
Succimer		↑				
Tacrolimus				↑		↑
Tipranavir		↑	↑			
Valproic acid						↑
Vitamin E		↑	↑			

Like estrogens, anabolic steroids can also cause profound, dose-related

effects on lipoprotein metabolism. Reductions in HDL levels in conjunction with an elevation in LDL levels are often seen in patients using these agents.[197] The exact mechanism has not yet been defined, but caution is advised in using these agents in patients prone to dyslipidemia or atherosclerosis.

GASTROINTESTINAL ADVERSE EFFECTS

Many medications can adversely affect the function of the GI tract. Most are minor and resolved over time without any specific intervention. Others, however, can be significant and resulting in severe GI illness (e.g., esophagitis, colitis) that can negatively impact the tolerance to an oral or tube fed diet. Drug-induced esophagitis can occur with doxycycline capsules, iron sulfate tablets, extended-release potassium chloride tablets, and tetracycline.[202] Typically, this occurs in adolescents who drink inadequate amounts of fluid with their medications, or patients with left atrial enlargement or cardiomegaly as the heart impinges on the esophagus, thus increasing the transit time of the medication in the esophagus. Switching to a liquid medication or administering the problematic drug with plenty of water often helps alleviate the situation.

DRUGS AND APPETITE

Impairment of nutritional status owing to drug use often results in drug-induced nutritional deficiencies in those cases in which the medication results in appetite suppression and decreased food intake. Often these signs and symptoms of nutrient deficiencies are nonspecific and may mimic those of other diseases and conditions.

Drugs can reduce food intake through a variety of mechanisms. Drugs that affect appetite (Table 18-8) may do so by either a central or a peripheral effect, including loss of appetite, inducing sedation, or evoking adverse response when food is ingested.[203] The primary effect typically centers around appetite suppression, a centrally acting mechanism that includes the catecholaminergic (e.g., dextroamphetamine), dopaminergic (e.g., levodopa), serotoninergic (e.g., fenfluramine), and endorphin (e.g., naloxone) modulators, which may all act to suppress appetite.[204] Peripherally acting mechanisms that can indirectly suppress appetite include those agents that inhibit gastric emptying (e.g., levodopa) or bulking agents (e.g., methylcellulose).

A secondary response may also occur when an adverse response to

food caused by the drug results in a loss of appetite. The emetic center, located within the brainstem, is easily stimulated by the action of many drugs. These include drugs that cause nausea and vomiting (e.g., digoxin toxicity), drugs causing a loss of taste (e.g., penicillamine), drugs causing stomatitis (e.g., fluorouracil), and hepatotoxic agents (e.g., alcohol).[203]

Altered taste perception can also lead to a decrease in nutrient intake. Medications such as angiotensin-converting enzyme inhibitors, allopurinol, amiodarone, baclofen, lithium, metronidaozole, and terbinafine have all been linked with causing taste perversions. Taste is regulated by chemosensory nerves that respond to stimulation chemicals by direct receptor binding, opening ion channels, or secondary messenger channels using nucleotides or phosphorylated inositol.[205] Medications that disrupt these cellular processes can cause a loss or distortion in taste. Similarly, dry mouth due to reduced saliva production can alter ion concentrations between saliva and plasma that results in decreased taste sensation. For example, anticholinergic drugs such as antihistamines and tricyclic antidepressants can cause dry mouth as a side effect that can lead to anorexia.

Another way in which medications can cause anorexia is through depletion of various nutrients. High doses of aluminum- or magnesium-containing antacids can result in phosphate depletion, leading to muscle weakness and anorexia.[206] Similarly, loop diuretics can lead to depletion of sodium, potassium, and magnesium, which can result in anorexia.[207] Drugs known to deplete folate, such as phenytoin, sulfasalazine, and trimethoprim, can result in weight loss and anorexia.[208] Penicillamine, which induces zinc depletion, can lead to diminished taste acuity and possibly decreased food intake.[209] Ironically, even a nutrient can induce anorexia. Belle and Halpern reported that patients taking relatively large doses of niacin for hyperlipidemia experienced GI symptoms that resulted in poor appetite and moderate weight loss.[210]

Zinc supplements have been used to treat drug-induced hypogeusia with mixed results. Although zinc supplementation has been used successfully to manage hypogeusia in patients undergoing dialysis, Others, however, did not see similar findings when patients with acetazolamide-induced taste disturbances were given zinc supplements.[211]

Table 18-8 Examples of Medications That Affect Appetite

Drugs That Decrease Appetite	Drugs That Stimulate Appetite

5-ASA derivatives
5HT3 receptor antagonists
Abacavir
Acetazolamide
Aluminum hydroxide
Amantidine
Aminoglycosides
Amphetamines
Artemether and lumefantrine
Asparaginase
Atomoxetine
Atovaquone
Azathioprine
Budesonide
Carboplatin
Caspofungin
Cidofaovir
Cisplatin
Cholestyramine
Chloroquine
Clofazimine
Clonidine
Colestipol
Dactinomycin
Dapsone
Digoxin
Enfurvitide
Ethionamide
Ethambutol
Ethosuximide
Flecainide
Fluorouracil
Furosemide
Gemfibrozil
Griseofulvin
Hydralazine
Hydroxychloroquine
Hydroxyurea
Imatinib
Indinavir
Itraconazole
Lamivudine
Lamotrigine
Levetiracetam
Lopinavir and ritonavir
Metformin
Methenamine
Methotrexate
Methylcellulose
Metronidazole
Micafungin
Benzodiazepines
Bupropion
Clemastine
Corticosteroids
Cyproheptadine HCl
Dronabinol
Gabapentin
Insulin
Levothyroxine
Lithium
Megestrol acetate
Oral contraceptives
Phenothiazines
Tricyclic antidepressants

Mineral oil Mycophenolate Nelfinavir Nitrofurantoin Penicillamine Penicillins Pentamidine Posaconazole Primaquine Proton pump inhibitors Quinolones Rifabutin Rifampin Rilpivirine Ritonavir Sibutramine Spironolactone SSRIs Stavudine Sulfasalazine Tenofovir Tetracycline Tiagabine Tipranavir Thiazide diuretics Topiramate Valganciclovir Voriconazole Zalcitabine Zidovudine Zonisamide
Source: Adapted from Gura.[259]

In some cases, weight gain may be an undesirable side effect of a medication. Antiepileptic drugs (AEDs), both old and new, have weight changes associated with their use. AEDs associated with marked weight gain include carbamazepine, gabapentin, VPA, and vigabatrin. Both lamotrigine and lecvetiracetam are considered weight-neutral, while felbamate, topiramate, and zonisamide have been associated with significant weight loss.[212]

APPETITE ENHANCERS

Appetite enhancers are useful in reversing the anorexia of disease, in particular cancer cachexia and HIV wasting. Several medications are frequently used, although none have yet become a standard of therapy.

The appetite-boosting properties of corticosteroids have been well established.[213] Their impact on weight gain seems to be short term, however. Moertel et al. suggested that patients treated with dexamethasone saw appetite improvement after two weeks of treatment, but the effect disappeared by week 4.[214] Some attribute the improved appetite to the mood-enhancing properties of steroids rather than a specific effect on appetite.[215] The antihistamine cyproheptadine has become one of the most commonly used appetite stimulants in pediatric patients, particularly those with anorexia nervosa. It is a potent serotonin antagonist. By decreasing brain serotonin levels, appetite is enhanced and food intake is increased, resulting in weight gain.[216] Megestrol acetate is a drug with antiestrogen properties whose original indication was in the treatment of breast cancer. It was one of the first drugs used to treat HIV wasting. It is a powerful appetite stimulant, and weight gain seen with its use is substantial, although it tends to be mostly fat, with minimal increases in lean body mass.[217] Megestrol also lowers testosterone levels in males, which may explain its minimal increases in lean body mass. Adrenal suppression has also been reported with long-term megestrol use; thus, abrupt discontinuation should be avoided.[218] Megestrol acetate also has been used to stimulate appetite and promote weight gain in a limited number of patients with cachexia associated with neoplastic disease. Although the exact mechanism of action has not been determined, it has been suggested that megestrol and/or its metabolites may, either directly or indirectly stimulate appetite, resulting in weight gain, or may alter metabolic pathways by interfering with the production or response of mediators such as cachectin, a hormone that inhibits adipocyte lipogenic enzymes.[219]

Dronabinol is a derivative of marijuana that is primarilyused in pediatric patients as an antiemetic but has some use in treating anorexia, primarily in HIVwasting.[220] Like megestrol, it can stimulate appetite as well as interest in food. Its effects on mental status, however, limit its usefulness.

Because cytokine effects on appetite can contribute to the anorexia of disease, anticytokine monoclonal antibodies and receptor antagonists may have a role in inhibiting cytokine action.[221] Some agents such as the corticosteroids inhibit the transcription of interleukin (IL)-1, tumor necrosis factor (TNF), and other cytokinases. Other agents appear to act by binding a mitogen-activating protein kinase necessary for the translation of messenger RNAs.[221] Fish oil supplements that are rich in omega-3 fatty acids decrease the anorectic effect of IL-1 and TNF.[222] Unfortunately, the

fishy taste associated with these supplements makes them a less-than-desirable option.

APPETITE SUPPRESSANTS

Although, for long-term success, weight loss takes a great deal of self-control to resist the many internal and external cues to eat, appetite suppressants can augment weight loss by reducing the hunger drive, thereby assisting the patient to adhere to a restricted caloric diet.[223] Currently, the major class of drugs in use for the adjunctive treatment is the centrally acting appetite suppressants. This class is further subdivided into those that act on the noradrenergic nervous system and those that act on the serotoninergic nervous system.

Given their high potential for abuse and numerous side effects, the noradrenergic agents (e.g., dextroamphetamine) are no longer routinely used in weight loss management unless other therapies have been proven ineffective. These agents block the reuptake of dopamine and norepinephrine from the synapse, thus increasing the amount available in the cerebral cortex and reticular activating system.[223]

The neurotransmitter serotonin is responsible for appetite suppression and satiation. Serotonin, as part of a complex negative feedback loop, accumulates in certain areas of the brain in response to feeding.[223] Sertraline, paroxetine, and fluoxetine inhibit serotonin reuptake at presynaptic spaces. They have few central stimulatory effects. Lorcaserin, the newest agent in the class, exerts its appetite suppressing effect by binding to serotonin 5-HT(2C) receptors.[224]

Other agents that have the potential to cause anorexia and may have a potential role in weight loss management are the thermogenic drugs.[225] These include endogenous hormones (i.e., insulin, thyroid hormone) and sympathomimetic agents with alpha- or beta-adrenergic properties.[225] Currently, exogenous thyroid has no role in obesity management owing to its adverse effects on protein breakdown, cardiovascular complications, and bone mineralization.[226]

VITAMIN AND MINERAL SUPPLEMENTATION/HYPERVITAMINOSIS

Megavitamin therapy has been advocated in the treatment of various disease states, including schizophrenia, cancer, and the common cold.[227] A

megadose is generally considered to be 10 or more times the recommended dietary allowance.[227] Although excess water-soluble vitamins are excreted and usually cause few problems, side effects have been reported in cases of excessively high doses.[228] Dietary supplementation can also impact response to medications. If used carelessly, vitamin and mineral supplementation alone or as part of a fad diet can potentiate or exacerbate nutrient–nutrient interactions, as summarized in Table 18-9 and Table 18-10.

Drug-metabolizing enzyme systems have been shown to be highly dependent on vitamin C status. Houston showed that normal individuals receiving supplemental ascorbic acid had a substantial increase in antipyrine clearance,[229] whereas Beattie and Sherlock reported that vitamin C deficiency was partly responsible for impaired drug clearance in patients with liver disease.[230] When ascorbic acid is taken in excessive amounts, enzyme saturation occurs, and the vitamin acts as a chemical and enters into nonenzymatic reactions.[231] Ascorbic acid is a strong reducing agent and is able to impact other dietary components as well as alter drug disposition.[231] Vitamin C has been shown to significantly affect the regulation of several key CYP enzymes, including CYP3A, some of which can be decreased in times of vitamin C deficiency.[232] Ironically, scurvy, the main deficiency state associated with inadequate dietary intake of ascorbic acid, a relatively rare condition today, can occur in infants born to mothers taking large doses of vitamin C.[231] Both the mother and fetus increase their metabolic destruction of ascorbic acid after maternal ingestion of large doses of ascorbic acid. At birth, any dietary ascorbic acid ingested by the infant is degraded rapidly, leading to a deficiency state. Reports of acute scurvy have been reported in infants breast-fed by mothers who had ingested more than 400 mg vitamin C daily during pregnancy.[233] A similar situation can occur in adults dependent on large doses of vitamin C. Therefore, it is recommended that patients receiving megadoses of vitamin C not be abruptly stopped but rather tapered by 10%–20% until discontinued or on maintenance doses. Other complications associated with megadoses of ascorbic acid include vitamin B_{12} deficiency. Herbert and Jacob reported that even ascorbic acid doses as little as 250 mg may destroy up to 81% of cyanocobalamin in a moderate vitamin B_{12}–containing meal and up to 25% in a vitamin B_{12}–rich meal.[234] To blunt the intensity of this interaction, the authors suggested that ascorbic acid be taken two or more hours after meals. Oxalate stones may occur in susceptible patients receiving doses of greater

than 4 g/d of ascorbic acid. Auer et al. reported that a hematuria and calcium oxalate dihydrate crystal formation in a 25-year-old male 8 days after ingesting 4 g of ascorbic acid daily resolved 5 days after discontinuing ascorbic acid supplementation.[235]

Table 18-9 Examples of Nutrient–Nutrient Interactions

Nutrient	Interaction
Calcium	High calcium intake interferes with fluoride, magnesium, phosphorus, and iron absorptionCalcium absorption is potentiated by vitamin D, reduced by dietary oxalates Excessive protein intake may increase calcium losses
Chromium	A high-sugar diet may increase urinary chromium losses
Copper	High intake may impair zinc absorption
Cysteine	May chelate with copper in parenteral nutrition solutions
Iron	Iron salts might decrease gastrointestinal zinc resorption; impairs manganese absorption Soy products, including soy-based infant formula, significantly reduce absorption of iron Vitamin C enhances absorption of iron Calcium salts may reduce iron absorption Vitamin A deficiency increases liver storage of iron
Magnesium	High magnesium intake may impair calcium absorption, decreases phosphorus absorption
Manganese	Calcium and iron salts decrease manganese absorption Zinc enhances manganese absorption
Phosphorus	High intake decreases magnesium absorption
Potassium	Decreases cyanocobalamin absorption
Thiamine	High carbohydrate diet or intravenous dextrose may increase thiamine requirement
Zinc	High zinc intake may impair copper absorption as well as interfere with absorption and metabolism of iron and vitamin A Zinc may increase manganese absorption Iron salts may impair zinc absorption
Vitamin A	Megadoses may interfere with iron, iodine, copper, calcium absorption; can also interfere with absorption of ascorbic acid, vitamin K, vitamin E, and vitamin D
Vitamin C	Increases iron absorption, impairs copper absorption, interferes with cyanocobalamin absorption
Vitamin D	Reduced intake impairs calcium and phosphorus absorption and utilization
Vitamin E	Inhibits the reticulocyte and hemoglobin response to iron

Table 18-10 Examples of Vitamin–Drug Interactions

Nutrient	Medication	Interaction
Ascorbic acid (vitamin C)	Coumarin anticoagulants	Shortens prothrombin time, may antagonize warfarin response
	Gentamicin	Acidifies urine, decreases efficacy of gentamicin
	Iron salts	Increased iron absorption; intermittent use may cause contraceptive failure
	Oral contraceptives	Megadoses (>2 g/d) may reduce therapeutic response of tricyclic antidepressants
	Tricyclic antidepressants	Ascorbic acid can increase the absorption of aluminum hydroxide
	Aluminum hydroxide	
Folic acid	Fosphenytoin, phenobarbital, phenytoin, primidone	Reduces fosphenytoin, phenobarbital, phenytoin, primidone bioavailability, may antagonize anticonvulsant action of these agents
	Pryimethamine	Reduces pyrimethamine efficacy
	Raltitrexed	Folic acid may diminish the therapeutic effect of raltitrexed
Niacin	Adrenergic blockers, aspirin, HMG-CoA reductase inhibitors (“statins”), isoniazid	Enhances vasodilation, may cause orthostatic hypotension, aspirin decreases flushing seen with niacin use; increased risk of statin-associated myopathy/rhabdomyolysis with concurrent use; increased niacin requirement seen with isoniazid use
	Bile acid sequestrants	Bile acid sequestrants may decrease the absorption of niacin
Pyridoxine (vitamin B6)	Barbiturates, levodopa, fosphenytoin, phenytoin	Reduced barbiturate response, reduces levodopa response; megadoses may impair fosphenytoin/phenytoin effect, cause decrease in phenytoin levels
	Fosphenytoin	Pyridoxine may increase the metabolism of fosphenytoin. This is most apparent in high pyridoxine doses (e.g., 80–200 mg daily)
	Altretamine	Pyridoxine may diminish the therapeutic effect of altretamine. Specifically when altretamine is used in combination with cisplatin the response duration may be diminished
Vitamin A	Aluminum hydroxide, coumarin anticoagulants, isotretinoin, oral contraceptives	Reduced vitamin A absorption; megadoses of vitamin A can enhance anticoagulant response, increase risk of bleeding; isotretinoin competes with vitamin A; increases serum vitamin A levels
Vitamin D	Aluminum hydroxide	Vitamin D analogs may increase the absorption of aluminum may be increased, leading to increased serum aluminum concentrations
	Digoxin	
	Sucralfate	Vitamin D may cause hypercalcemia, leading to

		arrhythmias Vitamin D may increase the serum concentration of sucralfate. The absorption of aluminum from sucralfate may be increased, leading to an increase in the serum aluminum concentrations
Vitamin E	Courmarin anticoagulants Vitamin A Iron salts Cyclosporine	Potentiates anticoagulant response Increases serum vitamin A levels Inhibits the reticulocyte and hemoglobin response to iron therapy Vitamin E may decrease serum concentration of cyclosporine
Vitamin K	Coumarin anticoagulants	Antagonizes anticoagulant response

Source: Adapted from Gura.[259]

The daily consumption of vitamin B_6 (pyridoxine) in doses of 50–500 mg is not unusual given the currently available strengths of over-the-counter products. High doses, however, may have a toxic action on the nervous system.[236] Sensory neuropathies have been reported in adults who received daily doses of 2 g of pyridoxine over a four-month period or as soon as two months when daily doses of 5 g were ingested.

Megadoses of folic acid given to patients on long-term phenytoin therapy exhibit seizures and electroencephalogram abnormalities.[237] It appears that folic acid supplementation increases phenytoin liver metabolism; studies suggest increased phenytoin parahydroxylation while another suggests increased O-methylation of the catechol metabolite.[238] Another study concluded that folic acid does not stimulate phenytoin metabolism, but rather it changes the rates of elimination.[239]

Trace elements such as manganese can also impact the absorption of other nutrients such as iron. In patients with iron deficiency, there is a greater absorption of manganese. When iron supplementation occurs, there is a corresponding decrease in the amount of manganese absorbed with a greater amount appearing in the feces. This interaction appears to be due to competition for similar binding and absorption sites of (nonheme) iron and manganese.[240]

Hypervitaminosis A and D has been described in children, but the occurrence is rare.[241,242] Both are life threatening on an acute basis; vitamin A may cause increased intracranial pressure, whereas vitamin D may cause fatal hypercalcemia. Infants and young children are especially susceptible to excessive doses of vitamin A. Acute toxicity has been reported in infants receiving doses of 50,000–100,000 µg (166,666–

333,333 IU) retinol as palmitate. Individual variation is significant, however. Field studies have used periodic massive doses of vitamin A, with thousands of 1- to 6-year-old children receiving 200,000 IU of retinyl palmitate (60,000 µg retinol), with approximately 1% exhibiting signs of intolerance that disappeared within hours of administration.[243] Chronic toxicity seen in infants and young children usually results from daily doses of 10,000–50,000 µg (33,333–166,666 IU) for several months.[244] Symptoms gradually disappear on discontinuation. The American Academy of Pediatrics recommends that daily vitamin A supplementation doses of more than 3000 µg (10,000 IU) for young children be used only under medical supervision.[245]

Kutuzova and DeLuca have described the impact of vitamin D and drug metabolism. They reported that 1,25 dihydroxy vitamin D3 regulates the genes responsible for the production of enzymes (e.g., CYP3A4) that are responsible for detoxification in the intestine although the clinical significance has yet to be realized.[246] In typical doses, however, vitamin D is used to prevent and treat rickets.[247] Serious poisoning can occur with excessive doses. Reports of sustained daily intake of as little as 1800 IU in children has been associated with adverse events.[248] Excessive vitamin D leads to increased absorption of calcium from the GI tract and enhanced bone resorption, with a subsequent loss of renal concentrating ability. Children typically will present with anorexia, vomiting, polyuria, or irritability associated with the increased serum calcium concentrations.[249] It should be noted, however, that very high doses of vitamin D have been effective in some patients with hereditary vitamin D–resistant rickets (HVDRR).[250] HVDRR accompanied by alopecia reflects a more severe form of vitamin D receptor resistance and is rarely responsive to vitamin D. Initial ergocalciferol therapy for children with vitamin D–resistant rickets is 1000–2000 µg/d (40,000–80,000 IU) with phosphate supplement. The daily dose is increased at 3- to 4-month intervals in 250–500 µg (10,000–20,000 IU) increments.[251]

In 1992, an outbreak of hypercalcemia owing to vitamin D intoxication was noted. In the case series, eight patients suffered vitamin D toxicity that was traced to excessive amounts of the vitamin in milk produced by a single dairy.[252] These were the first reported cases of hypervitaminosis D from commercial food products in the United States since fortification became commonplace in the 1930s. A second study summarized the findings of an analysis of 42 containers of milk and 10 cans of infant formulas from supermarkets in five states.[253] Nearly two-thirds of the milk

container samples had less than 80% of the stated amount, whereas 4 of the 42 samples had over 120% of the labeled vitamin D content. The 10 infant formulas tested had vitamin D levels ranging from 111 IU for every 100 kcal to 250 IU per 100 kcal, exceeding the maximum permitted by the US Food and Drug Administration (FDA) regulation of 100 IU per 100 kcal. FDA-compliant studies on both dairy products and infant formulas compiled over a 10-year period also showed some variance in vitamin D levels but not to the extent that would constitute a health risk.[254]

Vitamin K–rich foods (i.e., green leafy vegetables, cabbage, broccoli), when intake is irregular, can alter response to anticoagulants such as warfarin. When ingested in high amounts, these foods can be associated with anticoagulant failure, resulting in the need for higher doses. Similarly, when patients who have been stable on warfarin suddenly decrease their intake of these vitamin K–rich foods, they can put themselves at risk for bleeding because they are suddenly receiving too large a warfarin dose. Warfarin creates a partial deficiency of the active form of vitamin K involved in the posttranslational modification factors (II, VII, IX, X, and proteins C and S).[255] Other sources of vitamin K, such as intravenous fat emulsions that are prepared from soybean or safflower and soybean oils, contain phytosterols that contain vitamin K.[256] Warfarin resistance has been reported in patients taking warfarin who also received intravenous fat emulsions.[256] This interaction presumably occurs because of the vitamin K content of the intravenous fat emulsion. Frequent monitoring of the international normalized ratio and warfarin dosage adjustments are imperative in these patients.

CONCLUSION

The human diet is highly heterogeneous in its composition, method of preparation, quantity, and time of consumption. These variations may alter the body's responses to drugs. Consequently, pharmacokinetic and pharmacodynamic responses as a result of diet vary widely in subjects based on age, gender, culture, genetic makeups, and economic status. Even within the same individual, seasonal variations will occur that impact dietary habits and ultimately drug-related effects. Although each factor may play a small role by itself, a much larger synergistic effect could occur when combined with other dietary factors, as well as genetic and environmental factors.

REFERENCES

1. Waler-Sack I, Klotz U. Influence of diet and nutritional status on drug metabolism. *Clin Pharmacokinet*. 1996;31:47–64.
2. Kearns GL, Abdel-Rahman SM, Alander SW, et al. Developmental pharmacology–drug disposition, action, and therapy in infants and children. *N Engl J Med*. 2003;349(12):1157–1167.
3. Yaffee SJ, Sonawane BR. Drug therapy and the role of nutrition. In: Walker WA, Watkins JB, eds. *Nutrition in Pediatrics*. 2nd ed. Hamilton,Ontario, Canada: BC Decker; 1997:320–334.
4. Skaer TL. Dosing considerations in the pediatric patient. *Clin Ther*. 1991;13:526–544.
5. Soldin OP, Soldin SJ. Review: therapeutic drug monitoring in pediatrics. *Ther Drug Monit*. 2002;24:1–8.
6. Christian P, Black RE. Food, micronutrients, and birthoutcomes. *JAMA*. 2012;307(19):2094–2096.
7. Pai AB, Norenberg J, Boyd A, et al. Effect of intravenous iron supplementation on hepatic cytochrome P450 3A4 activity in hemodialysis patients: a prospective, open-label study. *Clin Ther*. 2007;29(12):2699–2705.
8. Stewart CF, Hampton EM. Effect of maturation on drug disposition in pediatric patients. *Clin Pharm*. 1987;6:548–564.
9. Loebstein R, Koren G. Clinical pharmacology and therapeutic drug monitoring in neonates and children. *Pediatr Rev*. 1998;19:423–428.
10. Berseth CL. Gastrointestinal motility in the neonate. *Clin Perinatol*. 1996;23:179–190.
11. Lown KS, Bailey DG, Fontana RJ, et al. Grapefruit juice increases felodipine oral availability in humans by decreasing intestinal CYP3A protein expression. *J Clin Invest*. 1997;99:2545–2553.
12. Pan S, Lopez RR, Sher LS, et al. Enhanced oral cyclosporine absorption with water-soluble vitamin E after liver transplantation. *Pharmacotherapy*. 1996;16:59–65.
13. Chan L, Humma LM, Schriever CA, et al. Vitamin E formulation affects digoxin absorption by inhibiting P-glycoprotein (P-gp) in humans. *Clin Pharmacol Ther*. 2004;75(2):P95.
14. Guengerich FR. Influence of nutrients and other dietary materials on cytochrome P450 enzymes. *Am J Clin Nutr*. 1995;61:651S-658S.
15. Strassburg CP, Strassburg A, Kneip S, et al. Developmental aspects of human hepatic drug glucuronidation in young children and adults. *Gut*. 2002;50:259–265.
16. van Hoogdalem E, de Boer AG, Breimer DD. Pharmacokinetics of rectal drug administration, part I. General considerations and clinical applications of centrally acting drugs. *Clin Pharmacokinet*. 1991;21:11–26.
17. Santos CA, Boullata JI. An approach to evaluating drug-nutrient interactions. *Pharmacotherapy*. 2005;25(12):1789–1800.
18. Rodríguez-Fragoso L, Martínez-Arismendi JL, Orozco-Bustos D, et al. Potential risks resulting from fruit/vegetable-druginteractions: effects on drug-metabolizing enzymes and drug transporters. *J Food Sci*. 2011;76(4):R112–R124.
19. Reuss L. One-hundred years of inquiry: the mechanism of glucose absorption in the intestine. *Annu Rev Physiol*. 2000;62:939–946.
20. Kim RB. Drugs as P-glycoprotein substrates, inhibitors, and inducers. *Drug Metab Rev*. 2002;34:47–54.
21. Li P, Wang GJ, Robertson TA, Roberts MS. Liver transporters in hepatic drug disposition: an update. *Curr Drug Metab*. 2009;10(5):482–498.

22. Telang U, Ji Y, Morris ME. ABC transporters and isothiocyanates: potential for pharmacokinetic diet-drug interactions. *Biopharm Drug Dispos*. 2009;30(7):335–344.
23. Ito K, Kusuhara H, Sugiyama Y. Effects of intestinal CYP3A4 and P-glycoprotein on oral drug absorption—theoretical approach. *Pharm Res*. 1999;16:225–231.
24. Kimura Y, Ito H, Ohnishi R, Hatano T. Inhibitory effects of polyphenols on human cytochrome P450 3A4 and 2C9 activity. *Food Chem Toxicol*. 2010;48(1):429–435.
25. Chan L-N. Drug-nutrient interactions in transplant recipients. *JPEN J Parenter Enteral Nutr*. 2001;25:132–141.
26. Collnot EM, Baldes C, Wempe MF,et al. Influence of vitamin E TPGS poly(ethylene glycol) chain length on apical efflux transporters in Caco-2 cell monolayers. *J Control Release*. 2006;111(1–2):35–40.
27. Singh BN. Effects of food on clinical pharmacokinetics. *Clin Pharmacokinet*. 1999;37:213–255.
28. Skelton JA, Havens PL, Werlin SL. Nutrient deficiencies in tube-fed children. *Clin Pediatr (Phil)*. 2006;45(1):37–41.
29. Penod LE, Allen JB, Cabacungan LR. Warfarin resistance and enteral feedings: 2 case reports and a supporting in vitro study. *Arch Phys Med Rehabil*. 2001;82:1270–1273.
30. An Yeung SCS, Ensom MHH. Phenytoin and enteral feedings: does evidence support an interaction? *Ann Pharmacother*. 2000;34:896–905.
31. Fleisher D, Sheth N, Kou JH. Phenytoin interaction with enteral feedings administered through nasogastric tubes. *JPEN J Parenter Enteral Nutr*. 1990;14(5):513–516.
32. Gorman SR, Armstrong G, Allen KR, et al. Scarcity in the midst of plenty: enteral tube feeding complicated by scurvy. *J Pediatr Gastroenterol Nutr*. 2002;35(1):93–95.
33. Mayhew SL, Christensen ML. Pharmacokinetic alterations in malnutrition and obesity. *Hosp Pharm*. 1993;28:836–837.
34. Lazzerini M, Tickell D. Antibiotics in severely malnourished children: systematic review of efficacy, safety and pharmacokinetics. *Bull World Health Organ*. 2011;89(8):594–607.
35. McCabe BJ. Prevention of food-druginteractions with special emphasis on older adults. *Curr Opin Clin Nutr Metab Care*. 2004;7(1):21–26.
36. Mehta S. Drug disposition in children with protein energy malnutrition. *J Pediatr Gastroenterol Nutr*. 1983;2:407–417.
37. Erikson M, Catz C,Yaffe S. Effect of weanling malnutrition upon hepatic drug metabolism. *Biol Neonate*. 1975;27:339–351.
38. O'Doherty DS, O'Malley WE, Denckla MB. Oral absorption of diphenylhydantoin as measured by gas liquid chromatography. *Trans Am Neurol Assoc*. 1969;94:318–319.
39. Mehta S, Nain CK, Yadav D, et al. Disposition of acetaminophen in children with protein calorie malnutrition. *Int J Clin Pharmacol Ther Toxicol*. 1985;23:311–315.
40. Raghuram TC. Krishnaswamy K. Tetracycline absorption in malnutrition. *Drug Nutr Interact*. 1981;1(1):23–29.
41. Salako LA, Sowun A, Akibami FO. Pharmacokinetics of quinine in African children suffering from kwashiorkor. *Br J Clin Pharmacol*. 1989;28:197–201.
42. Lares-Asseff I, Cravioto J, Santigo P, Perex-Ortiz B. Pharmacokinetics of metronidazole in severely malnourished and nutritionally rehabilitated children.

Clin Pharmacol Ther. 1992;51:42–50.
43. Bolme P, Gebre-Ab T, Hadgu P, et al. Absorption and elimination of penicillin in children with malnutrition. *Ethiop Med J*. 1980;18(4):151–157.
44. Krishnaswamy K. Nutrients/non-nutrients and drug metabolism. *Drug Nutr Interact*. 1985;4:235–247.
45. Fishman J, Bradlow HL. Effect of malnutrition on the metabolism of sex hormones in man. *Clin Pharmacol Ther*. 1977;22:721–727.
46. Prasad KVS, Rao BSN, Sivakumar B, et al. Pharmacokinetics of norethinodrone in Indian women. *Contraception*. 1979;20:77–90.
47. Boudreaux JP, Hayes DH, Mizrahi S, et al. Use of water-soluble E to enhance cyclosporine absorption in children after liver transplant. *Transplant Proc*. 1993;25:1975.
48. Park GD, Spector R, Kitt TM. Effect of dietary protein on renal tubular clearance of drugs in humans. *Clin Pharmacokinet*. 1989;17:441–451.
49. Alleyne GA. The effect of severe protein calorie malnutrition on renal function of Jamaican children. *Pediatrics*. 1967;39:400–411.
50. Cuddy PG, Bealer JF, Lyman EL, Pemberton B. Theophylline disposition following parenteral feeding of malnourished patients. *Ann Pharmacother*. 1993;28:836–837.
51. Brinksman GI, Bowie MD, Friis-Hansen B, Hansen JDL. Body water composition in kwashiorkor before and after loss of edema. *Pediatrics*. 1965;36:94–103.
52. Lares-Asseff I, Flores-Perez J, Juarez-Olguin H, et al. Influence of nutritional status on the pharmacokinetics of acetylsalicylic acid and its metabolites in children with autoimmune disease. *Am J Clin Nutr*. 1999;69:318–324.
53. Johansson V, Portinsson S, Akesson A, et al. Nutritional status in girls with juvenile chronic arthritis. *Hum Nutr Clin Nutr*. 1986;40:57–67.
54. Feldman CH, Huthinson VE, Pigpenger CE, et al. Effect of dietary protein and carbohydrate on theophylline metabolism in children. *Pediatrics*. 1980;66:956–962.
55. Zarowtiz B, Pilla A, Popovich J. Expended gentamicin volume of distribution in patients with indicators of malnutrition. *Clin Pharm*. 1990;9:40–44.
56. Naran RK, Mehta S, Mathur BS. Pharmacokinetics study of antipyrine in malnourished children. *Am J Clin Nutr*. 1977;30:1979–1982.
57. Treluyer JM, Roux A, Mugnier C, et al. Metabolism of quinine in children with global malnutrition. *Pediatr Res*. 1996;40:558–563.
58. Amadi B, Kelly P, Mwiya M, et al. Intestinal and systemic infection, HIV, and mortality in Zambian children with persistent diarrhea and malnutrition. *J Pediatr Gastroenterol Nutr*. 2001;32(5):550–554.
59. World Health Organization. *Guidelines for an Integrated Approach to the Nutritional Care of HIV-Infected Children (6 Months–14 Years)*. Geneva, Switzerland: WHO Press; 2009.
60. World Health Organization. *Management of Severe Malnutrition: A Manual for Physicians and Other Senior Health Workers*. Geneva, Switzerland: WHO; 1999.
61. Abernathy DR, Greenblatt DJ. Drug disposition in obesehumans: an update. *Clin Pharmacokinet*. 1986;11:199–213.
62. Langebrake C, Bernhardt F, Baehr M, et al. Drug dosing and monitoring in obese patients undergoing allogenic stem cell transplantation. *Int J Clin Pharm*. 2011;33(6):918–924.
63. Strauss SG, Lynn AM, Bratton SL, Nespeca MK. Ventilatory response to CO_2 in children with obstructive sleep apnea from adenotonsillar hypertrophy. *Anesth Analg*. 1999;89(2):328–332.

64. Blouin RA, Kolpek JH, Mann HJ. Influences of obesity on drug disposition. *Clin Pharm*. 1987;6:706–714.
65. Holt VL, Scholes D, Wicklund KG, et al. Body mass index, weight, and oral contraceptive failure risk. *Obstet Gynecol*. 2005;105(1):46–52.
66. Schwartz SN, Pazin GJ, Lyon JA, Ho M. A controlled investigation of the pharmacokinetics of gentamicin and tobramycin in obese patients. *J Infect Dis*. 1978;138:499–505.
67. Traub SL, Johnson CE. Comparison of methods of estimating creatinine clearance in children. *Am J Hosp Pharm*. 1980;37:195–201.
68. Visram N, Friesen EG, Jamali F. Theophylline loading dose in obese patients. *Clin Pharm*. 1987;6:188–189.
69. Emery MG, Fisher JM, Chien JY, et al. CYP2E1 activity before and after weight loss in morbidly obese subjects with nonalcoholic fatty liver disease. *Hepatology*. 2003;38(2):428–435.
70. Lieber CS. CYP2E1: from ASH to NASH. *Hepatol Res*. 2004;28(1):1–11.
71. Corcoran GB, Salazar DE. Obesity as a risk factor in drug-induced organinjury. IV. Increased gentamycin nephrotoxicity in the obese overfed rat. *J Pharmacol Exp Ther*. 1989;248:17–22.
72. Evans AM. Influence of dietary components on the gastrointestinal metabolism and transport of drugs. *Ther Drug Monit*. 2000;22:131–136.
73. Fleisher D, Li C, Zhou Y, et al. Drug, meal and formulation inter-actionsinfluencing drug absorption after oral administration. Clinical implications. *Clin Pharmacokinet*. 1999;36:233–254.
74. Vessell ES. Complex effects of diet on drug disposition. *Clin Pharmacol Ther*. 1984;36:285–296.
75. Ioannides C. Effect of diet and nutrition on the expression of cytochrome P450. *Xenobiotica*. 1999;29:109–154.
76. Kappas A, Anderson KE, Conney AH, et al. Influence of dietary protein and carbohydrate on antipyrine and theophylline metabolism in man. *Clin Pharmacol Ther*. 1976;20:643–653.
77. Kappas A, Alvares AP, Anderson KE, et al. Effect of charcoal broiled beef on antipyrine and theophylline metabolism. *Clin Pharmacol Ther*. 1978;23:445–450.
78. Conney AH. Pharmacological implications of microsomal enzyme induction. *Pharmacol Res*. 1967;19:317–366.
79. Welling PG, Lyons LL, Craig WA, Trochta GA. Influence of diet and fluid on bioavailability of theophylline. *Clin PharmacolTher*. 1975;17:475–480.
80. Schwartz JG, McMahan CA, Green GM, Phillips WT. Gastric emptying in Mexican Americans compared to non-Hispanic whites. *Dig Dis Sci*. 1995;40:624–630.
81. Melander A, Danielson K, Schersten B, Wahlin E. Enhancement of the bioavailability of propranolol and metoprolol by food. *Clin Pharmcol Ther*. 1977;22:108–112.
82. Nutt JG, Woodward WR, Hammerstad JP, et al. The “on-off” phenomenon in Parkinson’s disease. Relation to levodopa absorption and transport. *N Engl J Med*. 1984;310:483–488.
83. Bailey DG, Spence JD, Munoz C, et al. Interaction of citrus juices with felodipine and nifedipine. *Lancet*. 1991;337:268–269.
84. Corthesy-Theulaz I, den Dunnen JT, Ferre P, et al. Nutrigenomics: the impact of biomics technology on nutrition research. *Ann Nutr Metab*. 2005;49(6):355–365.
85. Williams L, David JA, Lowenthal DT. The influence of food on the absorption and metabolism of drugs. *Med Clin North Am*. 1993;77:815–829.

86. Sonawane BR, Coates PN, Yaffe SJ, et al. Influence of dietary carbohydrates (alpha-saccharides) on hepatic drug metabolism in male rats. *Drug Nutr Interact*. 1983;2:7–16.
87. Anderson KE. Influences on diet and nutrition on clinical pharmacokinetics. *Clin Pharmacokinet*. 1988;14:325–346.
88. Cantarini MV, Painter CJ, Gilmore EM, et al. Effect of oral linezolid on thepressor response to intravenous tyramine. *Br J Cln Pharmacol*. 2004;58(5):470–475.
89. Singh BN. Effects of food on clinical pharmacokinetics. *Clin Pharmacokinet*. 1999;37(3):213–255.
90. Guengerich FB. Effects of nutritive factors on metabolic processes involving bioactivation and detoxification of chemicals. *Annu Rev Nutr*. 1984;4:207–231.
91. Zimmerman JJ, Ferron GM, Lim HK, Parker V. The effect of a high-fat meal on the oral bioavailability of the immunosuppressant sirolimus (rapamycin). *J Clin Pharmacol*. 1999;39(11):1155–1161.
92. Murray M. Altered CYP expression and function in response to dietary factors: potential roles in disease pathogenesis. *Curr Drug Metab*. 2006;7(1):67–81.
93. Singh BN. Effects of food on clinical pharmacokinetics. *Clin Pharmacokinet*. 1999;37(3):213–255.
94. Zhi J, Rakhit A, Patel IH. Effects of dietary fat on drug absorption. *Clin Pharmacol Ther*. 1995;58(5):487–491.
95. Tan KKC, Trull AK, Uttridge JA, et al. Effect of dietary fat on the pharmacokinetics and pharmacodynamics of cyclosporine in kidney transplant recipients. *Clin Pharmacol Ther*. 1995;57:425–433.
96. Zimmerman JJ, Ferron GM, Lim HK, Parker V. The effect of a high-fat meal on the oral bioavailability of the immunosuppressant sirolimus (rapamycin). *J Clin Pharmacol*. 1999;39(11):1155–1161.
97. Unadkat JD, Collier AC, Crosby SS, et al. Pharmacokinetics of oral zidovudine (azidothymidine) in paients with AIDS when administered with and without a high fat meal. *AIDS*. 1990;4:229–232.
98. Bidlack WR, Brown RC, Mohan C. Nutritional parameters that alter hepatic drug metabolism, conjugation, and toxicity. *Fed Proc*. 1986;45(2):142–148.
99. Wallace AW, Victory JM, Amsden GW. Lack of bioequivalence when levofloxacin and calcium-fortified orange juice are coadministered to healthy volunteers. *J Clin Pharmacol*. 2003;43(5):539–544.
100. Lampe JW. Health effects of vegetables and fruit: assessing mechanisms of action in human experimental studies. *Am J Clin Nutr*. 1999;70(3 suppl):475S-490S.
101. Sreerama L, Hedge MW, Sladek NE. Identification of a class 3 aldehyde dehydrogenase in human saliva and increased levels of this enzyme, glutathione S-transferases, and DT-diaphorase in the saliva of subjects who continually ingest large quantities of coffee or broccoli. *Clin Cancer Res*. 1995;1(10):1153–1163.
102. Pantuck EJ, Pantuck CB, Anderson KE, et al. Effect of Brussels sprouts and cabbage on drug conjugation. *Clin Pharmaco Ther*. 1984;35:161–169.
103. Pantuck EJ, Pantuck CB, Garland WA, et al. Stimulatory effect of Brussels sprouts and cabbage on human drug metabolism. *Clin Pharmacol Ther*. 1979;25:88–95.
104. Tanne JH. Food and drugs alter response to anaesthesia. *BMJ*. 1998;317(7166):1102.
105. Bailey DG, Spence JD, Edgar B, et al. Ethanol enhances the hemodynamic effects of felodipine. *Clin Invest Med*. 1989;12:357–362.
106. Seden K, Dickinson L, Khoo S, Back D. Grapefruit-drug interactions. *Drugs*.

2010;70(18):2373–2407.
107. Bailey DG, Dresser GK, Leake BF, Kim RB. Naringin is a major and selective clinical inhibitor of organic anion-transporting polypeptide 1A2 (OATP1A2) in grapefruit juice. *Clin Pharmacol Ther*. 2007;81(4):495–502.
108. Han HK. Role of transporters in drug interactions. *Arch Pharm Res*. 2011;34(11):1865–1877.
109. D'Arcy PF. Nutrient-drug interactions. *Adverse Drug React Toxicol Rev*. 1995;14:233–254.
110. Reissell P, Manninen V. Effect of administration of activated char-coal and fibre on absorption, excretion and steady state blood levels of digoxin and digitoxin. Evidence for intestinal secretion of the glycosides. *Acta Med Scand*. 1982;668(suppl):88–89.
111. Richter WO, Jacob BG, Schwandt P. Interaction between fibre and lovastatin. *Lancet*. 1991;338:706.
112. Stewart DE. High-fiber diet and serum tricyclic antidepressant levels. *J Clin Psychopharmacol*. 1992;12:438–440.
113. Champagne ET. Low gastric hydrochloric acid secretion and mineral bioavailability. *Adv Exp Med Biol*. 1989;249:173–184.
114. Kappas A, Alvares AP, Anderson KE, et al. Effect of charcoal broiled beef on antipyrine and theophylline metabolism. *Clin Pharmacol Ther*. 1978;23:445–450.
115. Muclow JC, Caraher MT, Henderson DB, Rawlins MB. The effect of individual dietary constituents on antipyrine clearance in Asian immigrants. *Br J Clin Pharmacol*. 1979;7:416–417.
116. Brodie MJ, Boobis AR, Toverud EL, et al. Drug metabolism in white vegetarians. *Br J Clin Pharmacol*. 1980;9:523–525.
117. Rose DP. Dietary fiber, phytoestrogens, and breast cancer. *Nutrition*. 1992;8(1):47–51.
118. Michnovicz JJ, Bradlow HL. Induction of estradiol metabolism by dietary indole-3-carbinol in humans. *J Natl Cancer Inst*. 1990;82(11):947–949.
119. Alonso-Aperte E, Varela-Moreiras G. Drugs-nutrient interactions: a potential problem during adolescence. *Eur J Clin Nutr*. 2000;54(suppl 1): S69–S74.
120. Anderson KE. Influences of diet and nutrition on clinical pharmacokinetics. *Clin Pharmacokinet*. 1988;14(6):325–346.
121. Sonne J, Dragsted J, Loft S, et al. Influence of a very low calorie diet on the clearance of oxazepam and antipyrine in man. *Eur J Clin Pharmacol*. 1989;36(4):407–409.
122. Corcoran GB, Salazar DE. Obesity as a risk factor in drug-induced organ injury. IV. Increased gentamicin nephrotoxicity in the obese overfed rat. *J Pharmacol Exp Ther*. 1989;248(1):17–22.
123. Schenker S, Speeg KV Jr, Perez A, Finch J. The effects of food restriction in man on hepatic metabolism of acetaminophen. *Clin Nutr*. 2001;20(2):145–150.
124. Bennett WM. Drug interactions and consequences of sodium restriction. *Am J Clin Nutr*. 1997;65(2 suppl):678S–681S.
125. Singh BN. Effects of food on clinical pharmacokinetics. *Clin Pharmacokinet*. 1999;37(3):213–255.
126. Bistrian BR, Blackburn G,Vitalie J, et al. Prevalence of malnutrition in general medical patients. *JAMA*. 1976;235:1567–1570.
127. Pantuck EJ, Pantuck CB, Weissman C, et al. Effects of par-enteral nutritional regimens on oxidative drug metabolism. *Anesthesiology*. 1984;60:534–536.
128. Burgess P, Hall RI, Bateman DN, Johnston ID. The effect of total parenteral nutrition on hepatic drug oxidation. *JPEN J Parenter Enteral Nutr*. 1987;11:540–

543.
129. Knapp HR. Nutrient-drug interactions. In: Ziegler EF, Filer LJ, eds. *Present Knowledge in Nutrition*. Washington, DC: ILSI Pres; 1996:540–546.
130. Roe DA. Drug-folate interrelationships: historical aspects and current concerns. In: PiccianoMF, Stokstad ELP, Gregory JF III, eds. *Folic Acid Metabolism in Health and Disease*. New York, NY: Wiley-Liss; 1990:91–120.
131. Chan L-N. Redefining drug-nutrient interactions. *Nutr Clin Pract*. 2000;15:249–252.
132. Alonso-Aperte E, Varela-Moreiras G. Drugs-nutrient interactions: a potential problem during adolescence. *Eur J Clin Nutr*. 2000;54:S69–S74.
133. Streiff RR. Anemia and nutritional deficiency in the acutely ill hospitalized patients. *Med Clin North Am*. 1993;77:911–918.
134. Cohen H, Scott D, Mackis IJ, et al. The development of hypoprothrombinema following antibiotic therapy in malnourished patients with low serum vitamin K levels. *Br J Haematol*. 1988;68:63–66.
135. Termanimi B, Gibril F,Sutliff V, et al. Effect of long-term gastric acid suppressive therapy on serum vitamin B12 levels in patients with Zollinger-Ellison syndrome. *Am J Med*. 1998;104:422–430.
136. Gura KM, Couris RR. Drug-induced bone disease. *US Pharmacist*. 2002;27:HS43–HS57.
137. Dobbins WO III, Herrero BA, Mansbach CM. Morphologic alterations associated with neomycin induced malabsorption. *Am J Med Sci*. 1968;255:63–77.
138. Black DA, Fraser CM. Iron deficiency anaemia and aspirin use in old age. *Br J Gen Pract*. 1999;49:729–730.
139. Zvaifler NJ, Rath CE. Interrelationship of rheumatoid arthritis, folic acid, and aspirin. *Blood*. 1971;38:405–416.
140. Tolstoi LG. Nutritional problems related to stimulant laxative abuse. *Hosp Pharm*. 1988;23:564–573.
141. Schindler AM. Isolated neonatal hypomagnesaemia associated with maternal overuse of stool softener [Letter]. *Lancet*. 1984;ii:822.
142. Clark JH, Russell GJ, Fitzgerald JF, et al. Serum beta-carotene, retinol, and alpha-tocopherol levels during mineral oil therapy for constipation. *Am J Dis Child*. 1987;141:1210–1212.
143. Thomas FB, McCullough FS, Greenberger NJ. Inhibition of the intestinal absorption of inorganic and hemoglobin iron by cholestyramine. *J Lab Clin Med*. 1971;78:70–80.
144. Aou S, Ma J, Shiramine K, Hori T. The stomach is the etiologicorgan for immobilization-induced hypocalcemia in rats. *Am J Physiol*. 1993;265:376–379.
145. Walan A, Strom M. Metabolic consequences of reduced gastric acidity. *Scand J Gastroenterol Suppl*. 1985;111:24–30.
146. Ruscin JM, Page RL II, Valuck RJ. Vitamin B(12) deficiency associated with histamine(2)-receptor antagonists and a proton-pump inhibitor. *Ann Pharmacother*. 2002;36(5):812–816.
147. Romero JA, Kuczler FJ Jr. Isoniazid overdose: recognition and management. *Am Fam Physician*. 1998;57:749–752.
148. Corcino J, Waxman S, Herbert V. Mechanism of triamtereneinduced megaloblastosis. *Ann Intern Med*. 1970;73:419.
149. Palomaki A, Malminiemi K, Solakivi T, Malminiemi O. Ubiquinone supplementation during lovastatin treatment: effect on LDL oxidation ex vivo. *J Lipid Res*. 1998;39:1430–1437.
150. Williams C,Netzloff M, Folkerts L, et al. Vitamin D metabolismand

anticonvulsant therapy:effect of sunshine on incidence of osteomalacia. *South Med J*. 1984;77:834–836.
151. Hahn TJ, AvioliLV. Anticonvulsant-drug-induced mineral disorders. In: Roe DA, Campbell TC, eds. *Drugs and Nutrients: The Interactive Effects*. New York, NY: Marcel Dekker; 1984;409–427.
152. Petty SJ, O'Brien TJ, Wark JD. Anti-epileptic medication and bone health. *Osteoporos Int*. 2007;18:129–142.
153. Reddy J, Bailey RR. Vitamin K deficiency developing in patients with renal failure, treated with cephalosporin antibiotics. *N Z Med J*. 1980;92:378–379.
154. New example of vitamin K-drug interactin [Editiorial]. *Nutr Rev*. 1984;42:161–163.
155. Lambie DG, Johnson RH. Drugs and folate metabolism. *Drugs*. 185;30:145–155.
156. Labadarios D, Dickerson JW, Park DV, et al. The effects of chronic drug administration on hepatic enzyme inductions and folate metabolism. *Br. J. Clin. Pharmacol*. 1978;5(2):167–73.
157. Mallek HM, Nakamoto T. Dilantin and folic acid status. Clinical implications for the periodontist. *J Periodontol*. 1981;52:255–259.
158. Rivey MP, Schottelius DD, Berg MJ. Phenytoin-folic acid: a review. *Drug Intell Clin Pharm*. 1984;18:292–301.
159. Lewis DP, Van Dyke DC, Wilhite LA, et al. Phenytoin-folic acid interaction. *Ann Pharmacother*. 1995;29:726–735.
160. Seligmann H, Potasman I, Weller B, et al. Phenytoin-folic acid interaction: a lesson to be learned. *Clin Neuropharmacol*. 1999;22:268–272.
161. Hoshino J, Sugawara K, Ishikawa S, et al. A case of ulcerative colitis with folate deficient megaloblastic anemia induced by sulfasalazine. *Nihon Shokakibyo Gakkai Zasshi*. 1999;96:840–845.
162. Selhub J, Seyoum E, Pomfret EA, Zeisel SH. Effects of choline deficiency and methotrexate treatment upon liver folate content and distribution. *Cancer Res*. 1991;51:16–21.
163. Wegner C, Nau H. Alteration of embryonic folate metabolism by valproic acid during organogenesis: implications for mechanism of teratogenesis. *Neurology*. 1992;42(suppl5):17–24.
164. Ubed N, Alonso E, Martin-Rodriguez JC, et al. Valproic induced developmental modifications may be partially prevented by coadministration of folinic acid and S-adenosylmethionine. *Int J Dev Biol*. 1996;40(suppl 1):291S-292S.
165. Ohtani Y, Endo F, Matsuda I. Carnitine deficiency with hyperammonemia associated with valproic acid therapy. *J Pediatr*. 1982;101:782–785.
166. Branda RF, Nelson NL. Inhibition of 5-methyltetrahydrolic acid transport by amphipathic drugs. *Drug Nutr Interact*. 1981;1:45–53.
167. Seligmann H, Halkin H, Rauchefleisch S, et al. Thiamine deficiency in patients with congestive heart failure receiving long-term furosemide therapy: a pilot study. *Am J Med*. 1991;91:151–155.
168. Brady JA, Rock CL, Horneffer MR. Thiamine status, diuretic medications, and the management of congestive heart failure. *J Am Diet Assoc*. 1995;95:541–544.
169. Sonneblick M, Friedlander Y, Rosin AJ. Diuretic-induced severe hyponatremia. Review and analysis of 129 reported patients. *Chest*. 1993;103:601–606.
170. Lahr MB. Hyponatremia during carbamazepine therapy. *Clin Pharmacol Ther*. 1985;37:693–696.
171. Nelson DC, McGrew WR, Hoyumpa AM. Hypernatremia and lactulose therapy. *JAMA*. 1983;249:1295–1298.
172. Tolstoi LG. Drug-induced hyperkalemia. *Hosp Pharm*. 1996;31:221–228.

173. Velazquez H, Perazella MA, Wright FR, Ellison DH. Renal mechanism of trimethoprim-induced hyperkalemia. *Ann Intern Med*. 1993;119:296–301.
174. Oster JR, Singer I, Fishman LM. Heparin-induced aldosterone suppression and hyperkalemia. *Am J Med*. 1995;98:575–586.
175. Brown GR, Greenwood JK. Drug- and nutrition-induced hypophosphatemia: mechanisms and relevance in the critically ill. *Ann Pharmacother*. 1994;28:626–632.
176. Odes HS, Fraser GM, Krugliak P, et al. Effect of cimetidine on hepatic vitamin D metabolism in humans. *Digestion*. 1990;20:251–262.
177. LoCascio V, Bonucci E, Imbinbo B, et al. Bone loss in response to long term glucocorticoid therapy. *Bone Miner*. 1990;8:39–51.
178. Isreal E, Banerjee TR, Fitzmaurice GM, et al. Effects of inhaled glucocorticoids on bone density in premenopausal women. *N Engl J Med*. 2001;345:941–947.
179. al-Ghamdi SM, Cameron EC, Sutton RA. Magnesium deficiency: pathophysiologic and clinical overview. *Am J Kidney Dis*. 1994;24:737–752.
180. Maviachak V, Coppin CM, Wong NL, et al. Renal magnesium wasting and hypocalcemia in chronic cis-platinum nephropathy in man. *Clin Sci*. 1988;75:203–207.
181. Forastiere AA, Belliveau JF, Goren MP, et al. Pharmacokinetic and toxicity evaluation of five-day evaluation of five day continuous infusion versus intermittent bolus cisdiamminedichloroplatinum III) in head and neck cancer patients. *Cancer Res.* 1988;48:3869–3874.
182. Ferra C, Berlanga JJ, Gallardo D, et al. Mitoxantreone, etoposide, carboplatinum and Ara-C combination therapy in refractory and relapsed acute leukemia. *Leuk Lymphoma*. 2000;39:583–590.
183. Shetty AK, Rogers NI, Mannick EE, Aviles DH. Syndrome of hypokalemic metabolic alkalosis and hypomagnesemia associated with gentamicin therapy: case reports. *Clin Pediatr*. 2000;39:529–533.
184. Arlette JP. Zinc and the skin. *Pediatr Clin North Am*. 1983;30:583–596.
185. Prasad AS. Clinical, biochemical and pharmacological role of zinc. *Annu Rev Pharmacol Toxicol*. 1979;2:393–426.
186. Prasad AS, Brewer GJ, Schoomaker EB, et al. Hypocupremia induced by zinc therapy in adults. *JAMA*. 1978;240:2166–2168.
187. Solomons NW. *Zinc and Copper in Human Nutrition: Nutrition in the 1980's: Constraints on Our Knowledge*. New York, NY: Alan R. Liss; 1981.
188. Ugarte M, Osborne NN. Zinc in the retina. *Prog Neurobiol*. 2001;64:219–249.
189. Michie DD, Wirth FH. Plasma zinc levels in premature infants receiving parenteral nutrition. *J Pediatr*. 1978;92:798–800.
190. Pandit MK, Burke J, Gustafson AB, et al. Drug-induced disorders of glucose tolerance. *Ann Intern Med*. 1993;118:529–539.
191. Shopbell JM, Hopkins B, Shronts EP. Nutritional screening and assessment. In: Gotschlich MM, ed. *ASPEN Core Curriculum*. Dubuque, IA: Kendall/Hunt; 2001:107–140.
192. Kaufman MG, Simionatto C. A review of protease inhibitor-induced hyperglycemia. *Pharmacotherapy*. 1999;19:114–117.
193. Plewe G, Beyer J, Krause U, et al. Long-acting and selective suppression of growth hormone secretion b somatostatin analogue SMS 201–995 in acromegaly. *Lancet*. 1984;ii:782–784.
194. O'Brien JG, DongBJ, Coleman RL, et al. A 5-year retrospective review of adverse drug reactions and their risk factors in human immunodeficiency virus-infected patients who were receiving intravenous pentamidine therapy forpneumocystis

carinii pneumonia. *Clin Infect Dis*. 1997;24:854–859.
195. Coyle P, Carr AD, Depczynski BB, et al. Diabetes mellitus associated with pentamidine use in HIV-infected patients. *MedJ Aust.* 1996;165:587–588.
196. Mills GA, Horn JR. Beta-blockers and glucose control. *Drug Intell Clin Pharm*. 1985;19:246–251.
197. Henkin J, Como JA, Oberman A. Secondary dyslipidemia: inadvertent effects of drugs in clinical practice. *JAMA*. 1992;267:961–968.
198. Korach M, Leclercq P, Peronnet F, Leverve X. Metabolic response to a C-glucose load in human immunodeficiency virus patients before and after antiprotease therapy. *Metab Clin Exp*. 2002;51:307–313.
199. Jain RG, Furfine ES, Pedneault L, et al. Metabolic complications associated with antiretroviral therapy. *Antiviral Res*. 2001;51:151–177.
200. Parsons SK, Skapek S, NeufeldEJ, et al. Asparaginase-associated lipid abnormalities in children with acute lymphoblastic leukemia. *Blood*. 1997;89:1886–1895.
201. Baker VL. Alternatives to oral estrogen replacement. Transdermal patches, percutaneous gels, vaginal creams and rings, implants, other methods of delivery. *Obstet Gynecol Clin North Am*. 1994;21:271–297.
202. Bonavina L, Demeester TR, McChesney L, et al. Drug-induced esophageal strictures. *Ann Surg*. 1987;206:173–183.
203. Welling PG. Effects of food on drug absorption. *Annu Rev Nutr*. 1996;16:383–415.
204. Leong RW, Chan FK. Drug–induced side effects affecting the gastrointestinal tract. *Expert Opin Drug Sat*. 2006;5(4):585–592.
205. Mason P. Important drug-nutrient interactions. *Proc Nutr Soc*. 2010;69(4):551–557.
206. Herzog P, Holtermuller KH. Antacid therapy—changes in mineral metabolism. *Scand J Gastroenterol Suppl*. 1982;75:56–62.
207. KinzieBJ. Management of the syndrome of inappropriate secretion of antidiuretic hormone. *Clin Pharm*. 1987;6:625–633.
208. Dreizen S, McCredie KB, Keating MJ, Andersson BS. Nutritional deficiencies in patients receiving cancer chemotherapy. *Postgrad Med*. 1990;87:163–167.
209. Knudsen L, Weismann K. Taste dysfunction and changes in zinc and copper metabolism during penicillamine therapy for generalized scleroderma. *Acta Med Scand*. 1978;204:75–79.
210. Belle M, Halpern MM. Oral nicotinic acid for hyperlipidemia with emphasis on side effects. *Am J Cardiol*. 1985;2:449–452.
211. Heyneman CA. Zinc deficiency and taste disorders. *Ann Pharmacother*. 1996;30:186–187.
212. Sheth RD, Gidal BE. Optimizing epilepsy management in teenagers. *J Child Neurol*. 2006;21(4):273–279.
213. Wilcox JC, Car J, Shaw J, et al. Prednisolone as an appetite stimulant in patients with cancer. *BMJ*. 1984;288:27.
214. Moertel CG, Schutt AJ, Reitemeier RJ, Hahn RG. Corticosteroid therapy of preterminal gastrointestinal cancer. *Cancer*. 1974;33:1607–1609.
215. Bruera E, Roca E, Cedaro L, et al. Action of oral methylprednisolone in terminal cancer patients: a prospective randomized double-blind study. *Cancer Treat Rep*. 1985;69:751–754.
216. Nobel RE. Effect of cyproheptadine on appetite and weight gain in adults. *JAMA*. 1969;209:2054–2055.
217. Oster MH. Megestrol acetate in patients with AIDS and cachexia. *Ann Intern Med*.

1994;121:400–408.
218. Leinung MC, Liporace R, Miller CH. Induction of adrenal suppression by megestrol acetate in patients with AIDS. *Ann Intern Med*. 1995;122:843–845.
219. McCarthy HD, Crowder RE, Dryden S, et al. Megestrol acetate stimulates food and water intake in the rat: effects on regional hypothalamic neuropeptide-y concentration. *Eur J Pharmacol*. 1994;265:99–102.
220. Plasse TF, Gorter RW, Krasnow SH, et al. Recent clinical experience with dronabinol. *Pharmacol Biochem Behav*. 1991;40:695–700.
221. Hellerstein MK, Meydani SN, Meydani M, et al. Interleukin-1-induced anorexia in the rat. Influence of prostaglandins. *J Clin Invest*. 1989;84:228–235.
222. Mulrooney HM, Grimble RF. Influence of butter and of corn, coconut and fish oils on the effect of recombinant human tumour necrosis factor alpha in rats. *Clin Sci*. 1993;84:105–107.
223. Silverstone T, Goodalle E. Centrally acting anorectic drugs: a clinical perspective. *Am J Clin Nutr*. 1992;55:211s-214s.
224. Martin CK, Redman LM, Zhang J,et al. Lorcaserin, a 5-HT(2C) receptor agonist, reduces body weight by decreasing energy intake without influencing energy expenditure. *J Clin Endocrinol Metab*. 2011;96(3):837–845.
225. Landsberg L, Young JB. Sympathoadrenal activity and obesity: physiological rationale for the use of adrenergic thermogenic drugs. *Int J Obes Relat Metab Disord*. 1993;17(suppl 1):S29–S34.
226. Uzzan B, Campos J, Cucherat M, et al. Effects on bone mass of long term treatment with thyroid hormones: a metaanalysis. *J Clin Endocrinol Metab*. 1996;81:4278–4289.
227. Ban TA. Pharmacotherapy of schizophrenia with special reference to megavitamin therapy. *Can J Psychiatr Nurs*. 1973;14:6–9.
228. Dipalma JR. Vitamintoxicity. *Am Fam Physician*. 1978;18:106–109.
229. Houston JB. Effect of vitamin C supplements on antipyrine disposition in man. *Br. J. Clin. Pharmacol*. 1977;4(2):236–9.
230. Beattie AD, Sherlock S. Ascorbic acid deficiency in liver disease. *Gut*. 1976;17:571–575.
231. Herbert VD. Megavitamin therapy. *J Am Pharm Assoc*. 1977;17:764–766.
232. Slain D, Amsden JR, Khakoo RA, et al. Effect of high-dose vitamin C on the steady-state pharmacokinetics of the protease inhibitor indinavir in healthy volunteers. *Pharmacotherapy*. 2005;25(2):165–170.
233. Cochrane W. Overnutrition in prenatal and neonatal life. *Can Med Assoc J*. 1965;93:893–899.
234. Herbert V, Jacob E. Destruction of vitamin B12 by ascorbic acid. *JAMA*. 1974;230:241–242.
235. Auer BK, Auer D, Rodgers AL. Relative hyperoxaluria, crystalluria and haematuria after megadose ingestion of vitamin C. *Eur J Clin Invest*. 1998;28:695–700.
236. Schaumburg H, Kaplan J, Winderbank A, et al. Sensory neuropathy from pyridoxine abuse. *N Engl J Med*. 1983;309:445–448.
237. Ch'ien LT, Krumdieck CL, Scott CW Jr, Butterworth CE Jr. Harmful effect of megadoses of vitamins: electroencephalogram abnormalities and seizures induced by intravenous folate in drug-treated epileptics. *Am J Clin Nutr*. 1975;28(1):51–58.
238. Makki KA, Perucca E, Richens A. Stimulation of drug metabolism by folate replacement in folate-deficient epileptic patients. *Br J Clin Pharmacol*. 1980;9(3):304P-305P.

239. Andreason PB, Hansen JM, Skovsted L, et al. Folic acid and the half-life of diphenylhydantoin in man. *Acta Neurol Scand*. 1971;47:117–119.
240. Rodriguez-Matas MC, Campos MS, Lopez-Aliaga I, et al. Iron-manganese interactions in the evolution of iron deficiency. *Ann Nutr Metab*. 1998;42(2):96–109.
241. Nako Y, Fukushima N, Tomomasa T, et al. Hypervitaminosis D after prolonged feeding with a premature formula. *Pediatrics*. 1993;92:862–864.
242. Gamble JG, Ip SC. Hypervitaminosis A in a child from mega-dosing. *J Pediatr Orthop*. 1985;5:219–221.
243. Arroyave G, Bauemfeind JC, Olson JA, Underwood BA. Selection of intervention strategies. In: *Guidelines for the Eradication of Vitamin A Deficiency and Xeropthalmia. A Report of the International Vitamin A Consultative Group*. Washington, DC: The Nutrition Foundation; 1976:1–8.
244. Jeghers H. MarroH. Hypervitaminosis A: its broadening spectrum. *Am J Clin Nutr*. 1958;6:335–372.
245. Yaffe SJ, Filer LJ Jr. Use and abuse of vitamin A. *Pediatrics*. 1971;48:655–656.
246. Kutuzova GD, DeLuca HF. 1,25-Dihydroxyvitamin D3 regulates genes responsible for detoxification in intestine. *Toxicol Appl Pharmacol*. 2007;218(1):37–44.
247. Chesney RW. Requirement and upper limits of vitamin D intake in the term neonate, infant and older child. *J Pediatr*. 1990;116:19–66.
248. Haddad JG. Vitamin D solar rays, the Milky Way or both? *N Engl J Med*. 1992;326:1213–1215.
249. British Pediatric Association. Hypercalcemia in infants and vitamin D. *BMJ*. 1956;2:149–155.
250. Malloy PJ, Pike JW, Feldman D. The vitamin D receptor and the syndrome of hereditary 1,25-dihydroxyvitamin D-resistant rickets. *Endocrine Rev*. 1999;20:156–188.
251. Taketomo CK, Hodding JH, Kraus DM. *Pediatric Dosage Handbook*. 8th ed. Hudson, Cleveland, OH: Lexi-Comp Inc.; 2001.
252. Blank S, Scanolon KS, Sinks TH, Falk H. An outbreak of hypervitaminosis D associated with overfortification of milk from a home-delivery dairy. *Am J Public Health*. 1995;85:656–659.
253. Jacobus CH, HolickMF, Shao Q, et al. Hypervitaminosis D associated with drinking milk. *N Engl J Med*. 1992;326:1173–1177.
254. US Food and Drug Administration, Center for Food Safety and Applied Nutrition. FDA mailgram, May 4, 1992.
255. Stirling Y. Warfarin-induced changes in procoagulant and anticoagulant proteins. *Blood Coagul Fibrinolysis*. 1995;6:361–373.
256. Lutomski DM, Palascak JE, Bower RH. Warfarin resistance associated with intravenous lipidadministration. *JPEN J Parenter Enteral Nutr*. 1987;11:316–318.
257. Fisher DM, O'Keeffe C, Stanski DR,et al. Pharmacokinetics and pharmacodynamics of *d*-tubocurarine in infants, children, and adults. *Anesthesiology*. 1982;57(3):203–208.
258. Raiten DJ. Nutrition and pharmacology: general principles and implications for HIV. *Am J Clin Nutr*. 2011;94(6):1697S-1702S.
259. Gura KM. Drug-nutrient interactions. In: Hendricks KM, Duggan C, eds. *Manual of Pediatric Nutrition.* 5th ed. Hamilton, Ontario, Canada: BC Decker; 2012.

Nutrition and Gene Expression

19

Mona Bajaj-Elliott, PhD and Ian R. Sanderson, MA, MSc, MD, FRCP, FRCPCH

Altering the expression of genes has become a rapidly developing area of research in medicine. The realization that gene expression is important in a wide range of diseases, and not just in inherited disease, has resulted in recognition of the whole field of gene expression as one that may bring new therapeutic options. Although more recent attention has focused on the benefits of altering gene expression by inserting or silencing genetic material by a variety of vectors, the expression of genes can also be altered by other means, most notably by changing the molecular environment that cells inhabit. Utilizing the natural responses of a cell to changes in its surroundings offers a new and amenable way to alter the expression of its genes. Many ways of altering these surroundings can be proposed, but no single act alters the environment of the cells of the body more than the ingestion of food. Thus, the future of nutrition as a therapeutic tool may lie in its potential for influencing gene regulation.

In recent years, epigenetic modifications have also emerged as a mechanism by which nutrition may modulate downstream metabolic traits.[1] Studies have begun to identify and associate nutritional risk factors with diseases such as metabolic disorders and predisposition to obesity with changes in epigenetic patterns. This chapter will examine this emerging field and will lay down some concepts that may prove useful in establishing the scientific basis from which future treatments may develop.

The survival of a child to reproductive age and beyond requires an ability to respond to external demands. Every organ in the body is attuned to this need. Many organ systems have two levels of response to external

changes. There is a rapid response, often occurring within seconds of a new stimulus; for example, contraction of muscle fibers following a neuronal impulse or the breakdown of glycogen by the liver during hypoglycemia. Such responses do not involve changes in gene expression. The cells maintain themselves in a state of readiness, responding rapidly to external stimuli by altering protein/ion activity. Behind this immediate response, there lie other slower, but more lasting, responses that require genetic control. For example, exercise on a regular basis leads not only to muscle mass increase but change in activity of the attendant enzymes that serve the increased metabolic need also occurs. Similarly, regular exposure of the liver to drugs induces the expression of enzymes that catalyze their breakdown.

There are few external stimuli on a child more important than his or her nutritional environment. The metabolic processes underlying the rapid response of cells to nutritional variations have long been documented in humans and other mammals.[2] However, the mechanisms whereby gene expression changes in response to nutritional stimuli have till recently remained poorly understood in humans or indeed in any multi-organ animal. Progress in the study of nutrient-gene interactions in eukaryotic cells has been slower for two main reasons. First, the molecular mechanisms controlling gene expression are more complex than in prokaryotes,[3] and second, it is more difficult to identify the metabolites of nutrients that may be responsible for inducing such changes. This review will therefore cover some of the recent advances in the study of nutrition and gene expression in the human and, where necessary, in other mammals.

Nutritional changes ultimately impinge on most cells in the body; however, it is the epithelium of the gastrointestinal (GI) tract that first encounters any variation in nutrient intake. Much of this chapter will therefore concentrate on how nutritional factors can alter the expression of genes in intestinal epithelial cells (IECs). The relevance of nutrient-gene interactions to human physiology will also be stressed. Finally, because manipulating nutritional intake may be a way of treating disease in children, the review will discuss nutritional therapy in childhood in the light of its effects on gene expression.

PHYSIOLOGIC IMPORTANCE OF NUTRITIONAL REGULATION IN GENE EXPRESSION

The effect of nutrients on gene expression may have different implications in different individual situations (Table 19-1). First, genes may be up-regulated to better utilize the supply of a particular nutrient when it is scarce. Transporters of nutrients and the enzymes that metabolize them are examples of proteins that may be induced by nutrients. Second, the expression of genes required for the storage of a particular nutrient may be altered according to that nutrient's abundance. Third, nutrients regulate the secretion of hormones that control the homeostasis of metabolic processes. For example, insulin synthesis increases after increased carbohydrate intake to maintain glucose homeostasis. Finally, food is part of our external environment and, as such, represents a challenge to the cells that come into intimate contact with it. This challenge is met in the main by the epithelial cell lining the gastrointestinal tract.[4] The ability of these cells to alter the expression of their genes with changes in food intake is one of the ways by which the intestinal epithelium can dominate the intestinal environment.

TABLE 19-1 Physiological Importance of Nutritional Regulation of Gene Expression

To satisfy the nutritional needs of an organ
Managing storage fuels required by other organs
Production of hormones essential to whole body metabolism
Direct interaction with the body's external environment along the gastrointestinal tract

Certain fundamental characteristics are found in the mechanisms that underlie each of these different aspects of nutrient-gene interactions. They include (Figure 19-1) a specific interaction between the cell and a particular nutrient (sensing) and a pathway by which such an interaction may translate into alterations in gene expression (signal transduction). We have little understanding of these mechanisms at present. However, some aspects of the molecular biology of these two functions will be examined later in this review.

Genes Controlling the Nutritional Requirements of Individual Organ Systems

Survival of cells depends on their ability to extract nutrients and other essential molecules from their surroundings, and they synthesize proteins

specifically for this purpose. Broadly speaking, two categories of genes are involved: those genes that express proteins that transport nutrients across cell membranes and those genes that express enzymes that metabolize nutrients. In both cases, genes are regulated purely to satisfy the requirements of an organ system. The cells are changing their phenotype to their own advantage, rather than to suit the needs of the body as a whole.

A good model of how cells may respond to their nutritional environment for their own needs is the utilization of glutamine.[5] Glutamine is the most abundant amino acid in milk and it also serves as a major fuel for the large intestine. Lactation promotes glutamine utilization (for milk synthesis and fuel provision) interestingly lactation induces a mild catabolic state in which skeletal muscle proteins are degraded to provide the additional glutamine required. One may speculate that glutamine supplementation during this crucial period is likely to be beneficial for both, the mother and the child's well-being.[6]

Glutamine availability modulates muscle turnover, and glutamine is an important source of amino-nitrogen for muscle cells.[7] Glutamine and glutamate are transported into cells by sodium-coupled transporters. For experimental purposes, rat skeletal muscle can be grown as primary cell cultures. These cells exhibit an up-regulation of the inward transport of both glutamine and glutamate when they are deprived of an exogenous glutamine supply, [8–10] in addition the activity of glutamine synthetase (GS) simultaneously increases. This enzyme catalyzes the addition of an amino group to the carboxyl moiety of the glutamate molecule to form glutamine. Two different nutrients, glutamine and glutamate, can therefore ultimately satisfy the glutamine requirements of the muscle cell through separate pathways, each of which is under genetic control. This system exemplifies how different nutrients can cross-stimulate the expression of a related set of proteins: removal of glutamine or glutamate from the cell medium results in an enhancement of the transport of both amino acids. Not only does lack of glutamine and glutamate enhance the expression of their own transport proteins, but deprivation of either substrate will also up-regulate the transporter of the other.[8] A study of the kinetics of transport 10 demonstrated that withdrawal of glutamine (or glutamate) enhanced the maximum rate at which the amino acid is transported into the cell (Vmax) and did not alter the affinity of the transporter for its amino acid. This suggests that deprivation resulted in an increase in the production of the number of transporters available, an effect that occurs at the transcriptional level. Studies in muscle-specific GS knockout mice indicate that GS is dispensable under "fed" conditions, however, becomes

vital to mounting an adaptive response during fasting by contributing to ammonia detoxification and urea synthesis highlighting how local nutritional status may impact on metabolic functions.[11]

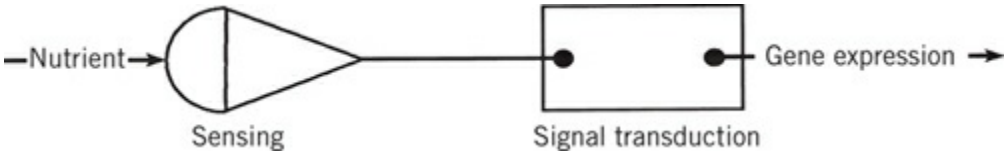


Figure 19-1 The regulation of gene expression by nutrients requires two generic mechanisms. First, molecules (either receptors or enzymes) within the cell must recognize the nutrient. Second, this interaction must initiate a sequence of events which ultimately alters the transcription or translation of genes. These functions of sensing and signal transduction have been depicted in this cartoon using electronic symbols.

In recent years, there has been an explosion in our understanding of the physiology of glutamine transporter systems.[12] At least four distinct families have been identified to date which include the Na+-dependent glutamine transporter, system N (SN1), system A (ATA1, ATA2), system ASC/B and system y (+)L, the latter encoding for Na+-independent glutamine transporter genes.[13,14] Molecular characterization of these systems suggest specific roles in glutamine efflux and uptake,[14] however, the exact mechanism(s) by which transport regulates gene expression remain unclear.

In vivo, glutamine homeostasis is to a large extent determined by the activities of glutamine synthetase (GS) and glutaminase.[15] Both enzymes exhibit regulation of gene expression at both transcriptional and post-transcriptional levels. During stress and sepsis, glutamine production is enhanced primarily due to greater GS activity in the skeletal muscle, beneficial effects of glutamine during sepsis include improvement in intestinal microcirculation with reduced leukocyte activation.[16]

Although glutamine regulates the expression of health- promoting proteins in the gut epithelia,[17] overall the glutamate/glutamine circuit constitutes only a minor part of nutrient homeostasis in vivo. Glucose, on the other hand, is central to human metabolism. Glucose, galactose, and fructose are the major dietary saccharides and the derangement of glucose handling in diabetes mellitus constitutes the most common condition

requiring nutritional advice. Intensive research in the last four decades has revealed the great complexity involved in glucose transport. Glucose is transported into enterocytes primarily by the sodium-glucose co-transporter (SGLT-1). Two Na+ ions must first bind to the transporter, and then one molecule of glucose or galactose may be actively transported into the cell. To date, 12 members of the SGLT-1 family in humans have been identified on the basis of sequence homology within the human genome.[18] Efforts to curb the ever burgeoning epidemics of diabetes and obesity has made dietary manipulation and therefore the SGLT family an attractive therapeutic target.[19] One must also not forget that oral rehydration therapy (ORT) developed to treat diarrheal dehydration is intimately linked with carbohydrate digestion. The therapy relies on the ability of SGLT1 to co-transport water without affecting Na+ or glucose uptake. In addition to SGLT proteins, facilitative-diffusion glucose transporter (GLUT) proteins catalyze glucose transport into cells. To date, 14 mammalian GLUT proteins had been identified, each a separate gene product with unique transport properties[20] GLUTs 1, 3, and 4 can transport glucose and galactose but not fructose, whereas GLUT-2 can transport all three whereas GLUTs 5 and 7 recognize fructose.[20–23] The GLUT family can be divided into three groups: class I (the previously known glucose transporters GLUT1-4), class II (GLUT-5, 7, 9, 11), and class III (GLUT-6, 8, 10, 12, and the myo-inositol transporter HM1T1). The various members exhibit both tissue and sub-cellular specificity, which most likely represents another level of regulation. Analysis of GLUT protein levels in patients with non-insulin- dependent diabetes mellitus revealed a 4-5-fold induction in SGLT-1, GLUT-2 and -5 protein levels, highlighting dysregulation in monosaccharide transport.[18] Identity of mechanism(s) by which these essential dietary carbohydrates directly affect their own uptake (via availability of GLUT and SGLT proteins) is most urgent if we are to understand the marked increase in the incidence of childhood obesity and diabetes facing the developed world.[24,25]

Genes Regulating the Storage of Nutrients

The storage of nutrients is essential to provide energy in times of fasting, and the mechanisms that control this function must be regulated by nutrient intake to be effective. The two most important organs involved in nutrient storage are the liver and adipose tissue. Both cell types transport glucose and other nutrients and convert them into fat; in the case of the liver, glucose is also converted to glycogen. Insulin is the major hormone

that promotes glucose utilization and induces storage. Its sensitive response to dietary intake complicates the study of the direct effects of nutrients on genes that regulate storage. The direct effects of glucose in particular, on the expression of transporters and metabolic enzymes have to be dissected from the effects of insulin on these same genes. Genome-wide association studies (GWAS) in recent years have highlighted a plethora of candidate genes that regulate and exquisitely fine-tune glucose homeostasis.[24] Exploring association between genotype and clinical phenotype will no doubt contribute to improved personalized medicine in the very near future.[24–26]

Research in the last decade has led to dramatic improvement in our understanding of the molecular mechanism(s) by which glucose directly modulates lipogenic enzymes. In 2001, Kawaguchi and colleagues identified a hepatic transcription factor carbohydrate response element-binding protein (ChREBP), which is regulated at two different levels - nuclear entry and DNA-binding via phosphorylation events mediated by glucose and cAMP.[27] Earlier studies focused on the three main control points of glucose metabolism before it enters the mitochondrion as pyruvate. These are the enzymes – glucokinase (GK), 6-phosphofructokinase (PFK), and L-type pyruvate kinase (PK). Fructose is also metabolized to pyruvate, bypassing the first two enzymes. PK is therefore a regulatory enzyme in the metabolism of both monosaccharides. Greater complexity in glucose-pyruvate biochemical axis was recently highlighted by the fact that the M2 isoform of PK tends to be predominant in cancer cells where it diverts glycolytic flux away from the mitochondria to serine and glycine biosynthesis.[28] Manipulating these cellular pathways offers an attractive therapeutic strategy for metabolic disorders and oncology.

Both in vitro and in vivo studies have identified a second critical transcription factor, sterol regulatory element binding protein (SREBP) in insulin-mediated transcriptional control of genes involved in glucose, fatty acid, and triglyceride metabolism.[29] -Three mammalian SREBP isoforms (SREBP-1a, SREBP-1c, and SREBP-2) have distinct and yet overlapping lipogenic transcriptional programs. SREBP-1a activates fatty acid and cholesterol synthesis, SREBP-1c fatty acid synthesis, and SREBP-2 cholesterol synthesis and uptake (for a recent update, see Ref. 30). The involvement of at least two transcription factors (ChREBP and SREBP) for optimum lipogenesis may reflect the degree of molecular complexity required for coordinated integration of hormonal and nutritional signals in

regulating energy requirements of a cell.[31]

Insulin has a major effect on the activity of the three glycolytic enzymes via transcriptional and/or posttranslational modifications.[31] Glucokinase mRNA expression is induced by insulin via an IRS-1-PI-3-kinase-dependent pathway.[32] PI-3-kinase and p44/p42 mitogen-activated protein kinases (MAPK) activities play a role in the transient activation of PK by insulin.[33] However, glucose itself independently alters the expression of pyruvate kinase by increasing the transcription of pyruvate kinase mRNA.[34] Glucose similarly enhances the expression of fatty acid synthase (FAS), a key enzyme in lipogenesis.[31] Glucokinase regulates the flow of substrate to pyruvate kinase. Because glucokinase requires insulin for its activity, it has been difficult to study the independent effects of glucose and its metabolites on pyruvate kinase expression. Nevertheless, two separate approaches directly demonstrate the effects of glucose on pyruvate kinase expression. The first approach was to demonstrate that the effects of insulin on transcription of pyruvate kinase mRNA were glucose-dependent, thus identifying glucose as a separate variable.[34–36] The second approach examined a particular hepatoma cell clone (mhAT3F) whose glucokinase (which is insulin-dependent) had been replaced, through spontaneous mutation, by hexokinase (which is not insulin-dependent). This allowed the effects of pyruvate kinase to be examined in the absence of insulin.[36] Several signaling pathways that may play a key role in glucose-dependent L-PK gene transcription have now known (for a recent review, see Ref. 37). The glucose-response elements in the promoter region of the L-PK gene are known to interact directly or indirectly with various families of transcription factors including the Sp1 family and the upstream stimulatory factors (USFs).[38] The complex interactions between the various factors (UFSs, COUP-TF II, ChREBP) is likely to act as a "glucose-sensing" mechanism allowing the cell to respond appropriately to continuous variation in its nutritional status.[37]

Nutrient Stimulation of Hormone Secretion

Insulin

In children, hormones are secreted into the circulation in response to nutritional stimulation for two main reasons: (*a*) to effectively store nutrients in appropriate storage molecules; and (*b*) to signal to tissues that substrate for growth is available to coordinate the cellular manifestations of growth (cell proliferation, differentiation, and controlled cell death) with the influx of nutrients. These two actions are mutually compatible,

and a number of hormones and growth factors serve both functions to varying degrees. Insulin, for example, is mainly a storage hormone, but it is also able to stimulate growth; the increased weight of babies born to diabetic mothers is due to increased fetal insulin acting as a growth hormone. Similarly, factors primarily responsible for growth, such as insulin-like growth factor-1 (IGF-1), which mediates many of the actions of growth hormone, are able to vary the rate of glucose uptake into cells.

The secretion of insulin by the pancreatic islets is tightly controlled by glucose concentrations in the blood. Alterations in gene expression play no part in these rapid responses to glucose. Regulatory proteins within islet cells are ready to respond to increases in glucose. Release of insulin from storage granules occur following calcium influx, which in turn is probably induced by minor increases in ATP from the metabolism of glucose and other nutrients. Circulating hormones also affect insulin release. The quick-acting mechanisms that do not involve control lie beyond the scope of this review. Again, the reader is referred to several excellent reviews on the subject.[39–41]

In addition to these rapid responses, glucose also exerts a more lasting effect on insulin production through increased translation and transcription of the insulin gene. This allows a child to adapt to longer periods of starvation or carbohydrate repletion. Glucose enhances the rate of translation of pro insulin mRNA to protein by three methods.[42] It increases the transfer of initiated insulin mRNA from free to membrane-bound ribosome. It also decreases the rate of pausing of the pro insulin mRNA as it passes along the ribosome. Lastly, glucose directly stimulates the cellular machinery involved in the elongation of proteins in pancreatic islet cells.[43,44] Long-term changes in insulin production are also mediated by transcription. Rats fasted over 4 days have low insulin mRNA concentrations, which return to normal on re feeding.[45,46] These changes control the ability of the pancreas to secrete insulin during dietary manipulations, and they enable the islet cell to adapt to long-term dietary changes.

Paired box 6 (Pax6) is a transcription factor belonging to the paired-box gene family. Mice heterozygous for Pax6 mutations develop diabetes suggesting Pax6 also regulates glucose metabolism.[47] Pax6 controls the expression of a serine protease convertase responsible for processing of pro insulin to its bioactive form; suggesting that individuals with Pax6 mutations may exhibit perturbed insulin-driven glucose homeostasis. This is indeed the case as a common variant upstream of the PAX6 gene has

been shown to influence islet function in man.[48] At present, the potential role and function of PAX6-mediated pro- insulin processing during fasting remains unclear.

IGF-1 and IGF-Binding Proteins (IGFBPs)

Although we do not fully understand the mechanisms where by children whose nutritional intake is reduced beyond a certain point stop growing, it is certainly true that nutrient intake has a significant effect on final adult height. This is common knowledge in developed countries with large immigrant populations from the developing world. The children in the immigrant communities grow up to be taller on average than their parents, whose childhoods may have been spent in lands where food was less abundant. Poor linear growth also occurs in childhood diseases such as cystic fibrosis. In inflammatory diseases, such as Crohn's disease or juvenile rheumatoid arthritis, the characteristic rate of growth may, in part, be related to poor nutritional intake[49] as well as to the direct effects of intestinal inflammation.[50,51]

IGF-1 and II are a family of polypeptide growth factors whose functions include mediation of growth hormone action, stimulation of insulin activity and autocrine regulators of cellular proliferation.[52,53] IGF-1 has been shown to be a key player in regulating organ and body growth during postnatal development both in rodents and humans.[52–54] There is now compelling evidence that IGF-1 secretion may be the point at which regulation of growth is controlled both in health and disease.[55,56] Although IGF is an important modulator of GH action, GH itself does not correlate with depressed linear growth under nutritional restriction[56] or in children with active inflammation.[57] Alterations in tissue responses to growth hormone may therefore mediate the effects of nutrition on growth. This has resulted in two separate lines of investigation:

1. The effect of nutritional alterations on expression of the GH receptor[58–60] and
2. The study of nutritional factors on IGF-1 and IGF-2 secretion in association with secretion of IGFBPs.

Nutritional status is a major determinant of IGF-1 mRNA expression in liver and non-hepatic tissues. Fasting, caloric restriction animal models studies all show a decrease in circulatory IGF-1 levels,[60,61] although circulating levels of GH remain elevated. Although fasting reduces the binding of GH to its receptor, down-regulation of the GH receptor does not

occur with dietary restriction,[62] and it is believed that neither of the above are principle causes of reduced IGF-1. Further, administration of IGF-1, but not GH stimulates mucosal hyperplasia in surgically stressed rats with intestinal atrophy induced by hypocaloric total parenteral nutrition.[63] Low protein intake studies in a liver-specific IGF-1 deficient mouse model led to a decrease in non-hepatic IGF-1 secretion into the circulation, with an increase in GH levels. Increased IGFBP-3 mRNA was also observed which supports the hypothesis that the binding protein may contribute to greater sequestration of locally synthesized IGF-1.[64] These studies support the view that the splenic GH/IGF-1 axis responds to nutritional stress to maintain tissue homeostasis.

Regulation of IGF mRNA expression is complex. The rat IGF-1 gene contains six exons spanning 100 kb of DNA. Exons 1 and 2 encode alternative amino terminal sequences of the IGF-1 protein. The gene has two distinct promoter regions upstream of exons 1 and 2, which can be separately activated.[65] Post-transcription, alternative splicing results in further heterogeneity of the IGF-1 mRNA. Furthermore, there are two different terminal poly A addition sequences in exon 6, adding to the general complexity. However, these myriad possibilities result in only three different-sized mRNA transcripts: a 3.8-kb, a 4.2-kb, and an 8-kb transcript. The importance of these different mRNAs to IGF-1 production and to growth awaits further investigation, but they are of interest because several studies have demonstrated that different forms of malnutrition affect the transcripts differently.[51,52,66]. Total energy restriction induces a coordinate decrease in all three forms of mRNA. In contrast, protein restriction results in a profound decrease in the 8-kb fragment with less effect on the two smaller fragments.[51,66] These observations open up two questions of further interest: (1) How do different nutrients differentially affect gene expression in a particular family of genes? (2) How do these differences in gene expression translate into differences in a child's final phenotype? We will be returning to similar questions when we consider the small intestinal epithelium, where nutritional variations are greater than those seen in the circulation. Corresponding changes in IGF-1 have been seen in childhood illness. IGF-1 concentration correlates with increased intake and subsequent growth of children with Crohn's disease, whereas growth has no correlation to growth hormone production.[51] Children with CF frequently manifest hormonal abnormalities that may contribute to malnutrition. Interestingly in a study by Bucuvalas and colleagues, no effect of exogenous IGF-1 on linear growth in prepubertal children with

CF was found although an increase in glucose/insulin ratio was observed.[67]

In extracellular fluids IGF-1 circulate bound to IGFBPs, of which six have been cloned and characterized.[52,53] IGFBP-3 is the main carrier of circulating IGF-1 and in the presence of an acid-labile subunit (ALS), a bio-inactive ternary complex is maintained during circulation. The complex allows greater control on IGF-1 bioavailability.[68] IGFBP-3 may also modulate IGF-1-stimulated DNA synthesis.[69] The biological function of each binding protein is under intense investigation.[70] Protection from the hypoglycemic effects of IGF-1 has also been suggested as a function. Nutritional factors alter the circulating levels of IGFBPs. There is an inverse correlation of the expression of binding proteins to that of IGF-1. Protein restriction results in an increase of IGFBP-1, which falls on refeeding. However, IGFBP-1 is sensitive to insulin, and it is not clear how nutrients in the liver alter their expression directly. Interestingly, IGFBPs are predominantly secreted by Kupffer cells, unlike IGF-1, which is synthesized by hepatocytes.[62] The difference between these two cell types in terms of their location and their expression of surface receptors could allow for subtle regulation of IGF action in the face of nutrients and other factors in the circulation, such as cytokines. Significant advances in recent years have provided the basic understanding required for development of IGFBPs as disease therapies. Mecasermin rinfabate (iPLEX™, a complex between IGF-1 and IGFBP-3 intact proteins) is currently in clinical trials for the treatment of conditions associated with poor growth, improved insulin sensitivity has also been reported in response to the drug.[71]

Effect of Intestinal Contents on Genes in the Epithelium

The gastrointestinal (GI) tract is the only mucosal surface that comes into contact with nutrients before they are absorbed. In addition, the large intestine is the natural habitat for trillions of bacteria, collectively referred as the microbiota. The microbiota generally constitutes families of bacteria that exist in a commensal/symbiotic relationship with the host. One can thus appreciate the unique challenges that are faced by the GI mucosal immune system. Simply, it has to exhibit tolerance toward ingested food antigens and its own microbiome, and yet at the same time be able to mount effective immunity toward enteropathogens such as *Enteropathogenic E. coli*, *Salmonella* and *Campylobacter* sps, and *Clostridium difficile*, infectious agents responsible for majority of the cases

of bacterial gastroenteritis worldwide. The luminal contents of the GI tract may therefore constitute a complex milieu containing live/dead bacteria, their byproducts (such as short-chain fatty acids[SCFAs], toxins) and food antigens digested to a varying degree (Figure 19-2). The dissociation between nutrients ingested and the changes observed in the bowel lumen becomes greater the further one proceeds down the GI tract. The contents of the distal colon are completely different from food, although even here they are affected to some extent by dietary intake. This relationship between ingested nutrients and the local environment of the lumen is a separate issue from the impact of that local environment on gene expression of the surface epithelium.

The GI tract consists of a single-cell epithelial lining that protects the underlying mucosa. It has been traditional to assume that gene expression in the small intestinal epithelial cells (IECs) is preprogrammed and that their expression is not influenced by events in the lumen. However, this view is being revised. An alteration in epithelial cell phenotype secondary to nutritional factors offers several advantages (Table 19-2). First, the intestine could adapt to absorb nutrients more effectively if specific digestive enzymes and IEC transporters were up-regulated in response to a particular nutrient. Second, as all mammals begin life on mother's milk, the opportunity exists for breast milk to influence the development of non-immune and immune components of the GI tract through actions of its own constituents. Third, if the genes affected in the IEC were immunologically important, the epithelial lining could mediate changes further afield by signaling information and modulating the mucosal immune system. Each of these areas is likely to represent important physiologic mechanisms that have implications for child health.

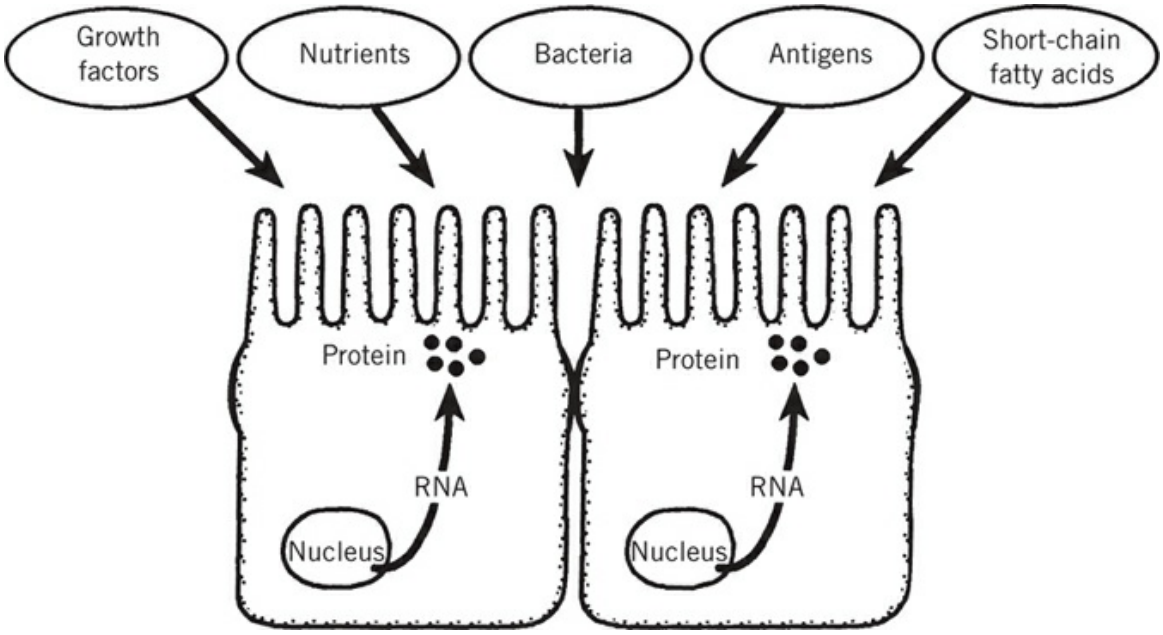


Figure 19-2 The luminal factors that influence gene expression in the enterocyte. Factors can affect individual genes or they may influence enterocyte differentiation and new cell lineages.

TABLE 19-2 Relevance of Dietary Regulation of Enterocyte Gene Expression

Intestinal adaptation
Influence of maternal breast milk
Signaling from lumen to mucosal immune system

Polarity of Epithelia

The cell types that we have discussed earlier in this chapter do not depend on a separation of functions to different cellular poles. For example, the pancreatic islet receives signals from the entry of glucose at any point on the plasma membrane and the insulin response is not affected by the site of glucose entry. Insulin can also be released in any direction, eventually diffusing into the circulation. Cells that form epithelia are different in that they exhibit polarity. This separation between the apical side (bordering the lumen in the case of the intestinal epithelium) and the basolateral side is central to IEC activity. This property is well recognized in the field of intestinal transport. Ions,[72–74] small molecules,[73–75] and macromolecules

[73,74,76] are all transported differently across the apical membrane than across the basolateral membrane (Figure 19-3). It is the polarity of the epithelium that gives direction to the movement of these substances across the epithelium into or out of the body. But this property must also be considered when one is examining all aspects of IEC function. Polarity is also of fundamental relevance in the study of nutrient-gene interactions in the intestinal epithelium. The polarity of the IEC distinguishes the two major mechanisms by which nutrients (and other luminal factors) affect genes: (1) a direct effect on enterocytes and (2) an indirect effect mediated through hormones, growth factor, and cytokines (Figure 19-3). As with the effects of insulin on target tissues, the mechanism(s) of action of direct (bacterial) and indirect (cytokines) mediators on the epithelium are beyond the scope of this chapter. The reader is again referred to excellent reviews on how epithelial cell expression can be regulated by a multitude of agents.[77,78]

The apical membrane mediates the direct effects of luminal factors on the epithelium. The functions of nutrient recognition (sensing) and signal transduction (see Figure 19-1) will therefore lie between the apical membrane and the apparatus of gene expression. The exact molecular mechanisms involved require further investigation. For purposes of investigation they can be considered as a series of black boxes connecting the lumen and gene expression (Figure 19-4). These black boxes contain the mechanisms that regulate gene expression and constitute the way by which molecules in the lumen "cross-talk" with the nucleus of the enterocyte. Figure 19-4 depicts the pathway linking single factors in the lumen to the expression of a single cell in a single gene. This is, of course, a gross oversimplification. As we have mentioned, an alteration in diet may alter different factors at the cell surface, and any one of these factors may have an effect on several different genes through different pathways (Figure 19-5). In addition method(s) by which a particular protein may be expressed is not necessarily always due to the induction of a single gene in cells along the crypt-villus axis. It can also be due to changes in the state of differentiation of the cells in the epithelium, the expression of a particular gene being just one component of that change. Lastly, alterations in luminal contents may not necessarily affect epithelial cells that are already formed along the epithelium, but instead result in the generation of new lineages of cells that express the gene arising from the crypt.[79] In this case, the mechanisms linking the lumen to gene expression would lie in the stem cell at the base of the crypt, rather than in the individual enterocyte itself.

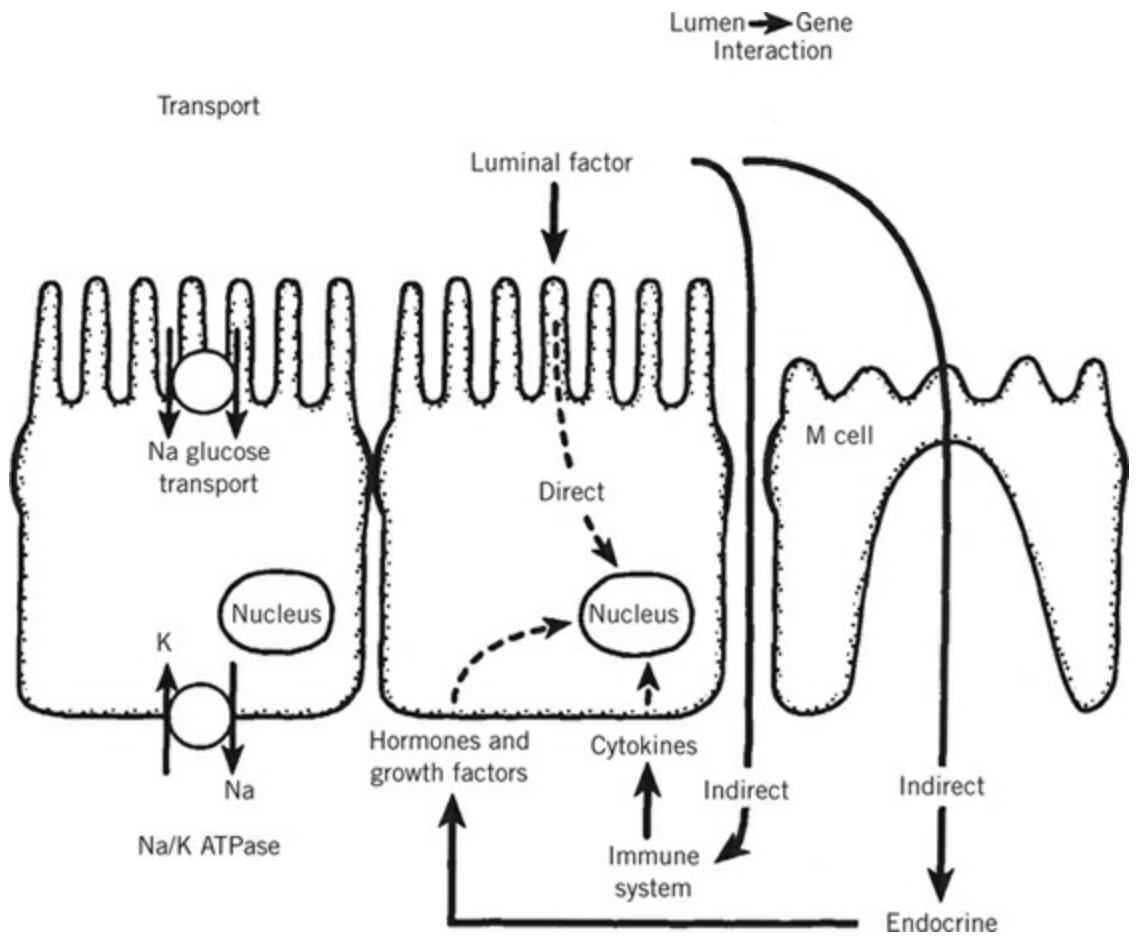


Figure 19-3 Dietary changes may affect gene expression directly (being mediated through the apical membrane) or indirectly (through the basolateral membrane).

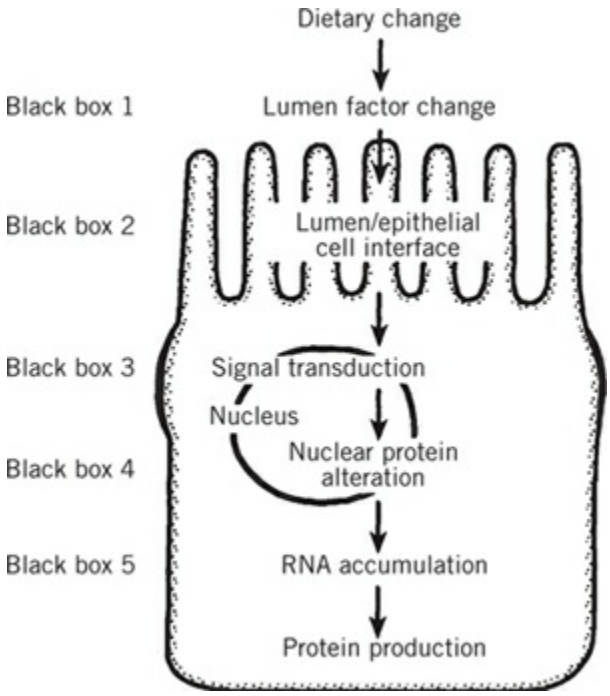


Figure 19-4 The pathway connecting dietary alterations to changes in gene expression is not well understood. It is likely that the pathway is mediated by a series of steps which at present we can only regard as black boxes. Different luminal events will influence gene expression through different series of black boxes.

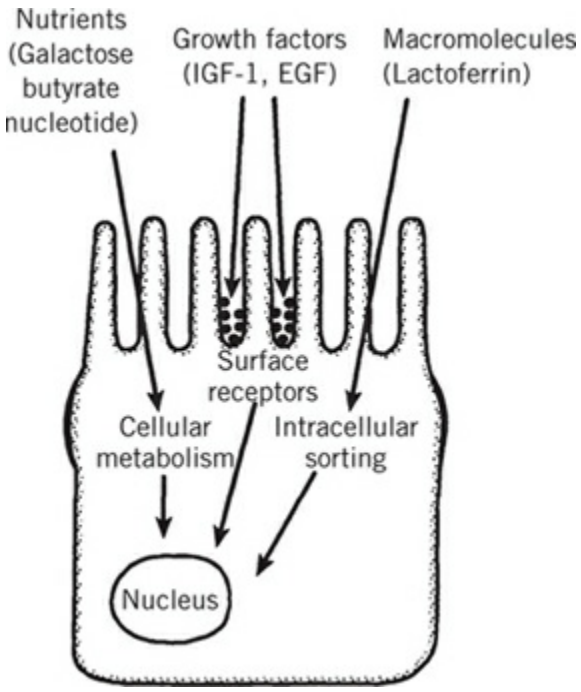


Figure 19-5 Cross talk through the apical membrane will depend on the particular signal involved. IGF, insulin-like growth factor; EGF, epidermal growth factor.

Adaptation of Enterocytes to the Nutritional Environment

The efficient utilization of food requires digestion, followed by absorption of digestion products and their release into the circulation. Adaptation is, by definition, the up-regulation of one or all of these functions in the presence of a relevant nutrient. Adaptive mechanisms are more likely to be associated with foods whose intake varies between different individuals. The similarity here with prokaryotic cells is clear. Bacterial gene regulation was discovered in the study of enzymes that metabolized nutrients whose abundance varied widely in the environment, like lactose or tryptophan. For children, of course, there are many foods whose abundance can vary, but to illustrate this section we have chosen two: dietary fat and sucrose. These two examples demonstrate different aspects of adaptation. Fat digestion is primarily controlled by enzymes secreted from the pancreas and gastric glands, whereas digestion and absorption of sucrose is controlled entirely by the enterocyte itself.

Sucrose Intake

The intake of sucrose can vary greatly between individual human beings. This is especially true when one compares individuals from different times in history. Until the industrial revolution, sucrose was present in the diet only when brought from the East as part of the spice trade, and its availability was limited. The introduction of plantations in the Caribbean in the 18th century, coupled with the industrial process of refining, resulted in an explosion of sucrose intake. In England in 1700, the estimated per capita daily consumption in adults was 4.5 g, rising to 37 g in 1800. By 1968, it had reached 140 g daily, which is around 15% of the total energy derived from food. The reader is recommended to a recent update by Stephens and colleagues where the authors summarize information available on the carbohydrate intake of children below 4 years of age.[80] Sucrose is digested by the epithelial cell brush border, where it is hydrolyzed to fructose and glucose by sucrase-isomaltase (SI). Fructose is absorbed by the carrier GLUT family members 18 and glucose by the specific sodium-dependent glucose transporter. Both sugars exit the cell through the basolateral membrane by facilitated diffusion via GLUT-2.

The up-regulation of any of these proteins by their substrate would constitute a nutrient-gene interaction.[81]

The brush border membrane (BBM) disaccharidases represent the final step in the digestion of carbohydrates.[81] Their activity can affect the glycemic response to oral carbohydrate.[82] Importantly, low levels of hydrolase activity in the face of carbohydrate in the intestine result in osmotic diarrhea.[83] A mechanism whereby expression of these enzymes could be linked to sugar intake would therefore have clear benefits. SI is one of the three β-glucosidases (together with maltase and trehalase). The remaining disaccharidase, lactase, is a β-galactosidase. Although sucrose digestion is the focus of our attention here, one must not forget that the major source of carbohydrate disaccharide in a new born is of course lactose from milk. Lactose is cleaved by lactase-phlorizin hydrolase (LPH) activity into glucose and galactose. LPH activity is crucial, particularly for those who are solely dependent on their mother's milk for nourishment.

In humans, BBM enzyme activity correlates with morphogenesis, as it associates with the formation of enterocytes.[84] Proximal-to-distal enzyme activity gradients are established early in gestation along the length of the small intestine. In addition, crypt-villous gradients are also seen with LPH activity which is highest at the villous tip, while maximal SI activity resides in the mid-villous region.

In humans, a marked increase in SI activity occurs just prior to birth, soon after however there is a sharp decline to levels comparable to those found in early gestation. As sucrase is not generally a major component of a neonate's diet, the role of high SI activity at birth remains ill-defined. SI is composed of two subunits sucrase is anchored to the microvillus membrane by the isomaltase subunit both subunits are synthesized as a single protein that is later cleaved.[85] The regulation of the induction of sucrase is not simple. The timing of its appearance during ontogeny in rodents was elegantly shown to be independent of luminal contents.[86] When the investigators transplanted isografts into animals 5 days younger than the donors, sucrase appeared earlier in the transplanted intestine than in the host. Therefore luminal factors in this case did not induce sucrase nor did circulating factors in the host (whose intestine would have been susceptible to them). This evidence suggests that sucrase is controlled by an internal biologic clock within the intestine itself. However, luminal contents can have a marked effect on the activity of sucrase, which is separate from this internal mechanism: (1) early feeding with sucrose will induce precocious sucrase induction in rodents[83]; (2) reduction of

carbohydrate intake results in a reduction of sucrase activity even when caloric intake is unchanged[87]; (3) feeding with glucose instead of sucrose reduces brush border hydrolase activity[88]; and (4) refeeding sucrose after a period of abstinence leads to rapid recrudescence of the hydrolase activity.[89] Increasing the intake of sucrose of mice from 2 per cent to 55 per cent of total energy intake had differing effects on the enterocyte, depending on the position of the cell in the small intestine.[90] Sucrase activity increased in the duodenum and upper jejunum but not in more distal segments of the intestine. Not surprisingly, the cells showing greatest alterations were those from the mid villus of the proximal intestine, with lesser increases in the villus tip and crypt/villus junction.

The control of sucrase activity is complex and depends on events within the enterocyte in addition to external influences. Sucrase activity depends mainly on events in the sucrase promoter since expression of growth hormone mirrors that of sucrase in transgenic animals containing a sucrase promoter-GH construct.[91] The expression of sucrase may therefore reflect nuclear protein-promoter interactions.[92] Evolutionarily conserved cis-acting elements in the promoter region have been found to interact specifically with transcription factors such as GATA-4 and Cdx proteins.[93–95] Further combinatorial interactions between HNF-1α protein, GATA-4 and Cdx-2 may play a critical role in the temporal and spatial regulation of the SI gene expression during post-natal development.[94,95] Post-transcriptional events may also likely to influence enzymatic activity, for example, reduction in lactase activity following weaning does not necessarily correlate with lactase mRNA but may be regulated during translation.[96]

Glucose Transporter Expression

It is worth noting that most of our understanding of nutrient transport has been gained from rodent studies. Unlike humans, rodent gut is immature at birth, and studies investigating gut maturation in these systems have provided invaluable information pertinent to preterm gut health in humans. Glucose transport is detected early in development and swallowing of amniotic fluid allows direct nutrient acquisition. Glucose enters the IEC primarily by active transport *via* a sodium-coupled glucose transporter, SGLT1 and GLUT2.[20,22,97] It is generally accepted that glucose (and galactose) uptake at the apical surface of the enterocyte is mediated by SGLT1 and exit is mediated *via* GLUT2 on the basolateral surface. Glucose entry via the SGLT1-GLUT2 axis is an exquisite example of how

the polarity of the IEC impacts on tissue homeostasis.

Direct transport studies undertaken before the identification of sodium-dependent transporters showed that glucose uptake increased with glucose feeding.[98] This increase was not due to changes in the affinity of glucose to the enterocyte (K_m), but to an increase in the maximum uptake (V_{max}) indicating an increase in the production of proteins that transport glucose. *In-vitro* and *in-vivo* studies indicate that JAK-2 mediated signalling can enhance SGTL1 expression at the apical surface of IECs with minimal impact on affinity clearly suggesting that this pathway may play a crucial role in maintaining glucose homeostasis in health and disease.[99] Modulation of SGLTI expression by dietary carbohydrates has also been observed in the human intestine.[97] Elegant studies by Wright and colleagues have recently provided a molecular, structural basis for SGTL1 function.[100] Several distinct conformational changes come into play upon ion (Na+) and substrate (glucose) binding. For detailed description, reference[100] is recommended.

Mechanisms involved in the regulation of glucose transport are complex and have been difficult to characterize as two distinct time-scales of dietary carbohydrate exposure are now known to play a role[101] Glucose transporters can respond within an hour to glucose[102] and also exhibit distinct changes in gene expression over a period of 1-3 days to a high carbohydrate diet.[103] Intestinal glucose transport mechanism(s) are also subject to diurnal changes adding further complexity to the system. Interestingly, impact of nutrients on induction of enzymes and transporters in mammalian cells show significant differences when compared to prokaryotes. First, the activity of transporters in bacteria increase by 100- to 1,000-fold in contrast the nutrient transporters in mammals show smaller variation with diet. For example, feeding mice with diets containing high levels of either D-glucose or non-metabolizable analogues of D-glucose results in only a two to three-fold increase in SGLT1 activity.[104] Second, in bacteria, the cell that receives the stimulus is the same cell that exhibits alterations in gene expression. In mammalian intestine, this appears not to be the case with the glucose transporter.[105] Sequential elution of different fractions of epithelial cells was used to distinguish different cells along the crypt-villus axis of mice.[106] Increase in carbohydrate intake resulted in enhanced expression of the glucose transporter (as measured by phlorhizin binding). Importantly, changes were observed in the crypts before they were seen in the villi. Similarly when carbohydrate was stopped, it was in the crypts that the reduction in

transporter was first observed. Thus crypt cells respond to changes in diet before villus cells, indicating that the changes in the expression of the glucose transporter were effected by new cell lineages from stem cells. It is the stem cell producing the new cell lineage that therefore must contain the machinery for detection of carbohydrate. The resulting cell population that ascends along the crypt-villus axis expresses this change. This separates carbohydrate detection temporally and spatially from glucose transporter expression. It is not known whether other genes in the intestine follow a similar pattern. Identification of molecular mechanisms underlying carbohydrate detection and signal transduction are currently under active investigation in several laboratories (see Figure 19-1).[107,108]

Sucrose induces an increase in the enzyme that hydrolyzes it (SI) and in SGLT1 that allows a greater influx into the enterocyte across a concentration gradient. This absorptive system is central to the efficacy of oral rehydration solution in the treatment of gastroenteritis and cholera.[22] The greater the number of transporters, the more sodium and fluid that can be transported into a child during dehydration. Up-regulation of SGLT1 has therefore a potential therapeutic role in alleviating the loss of sodium and fluid with glucose solutions. In this context, a drug that may potentiate JAK2 signalling leading to increase in SGTL1 expression is a viable option.[99] Better understanding of the mechanisms regulating this process would define to what extent and over what time period sodium and fluid transport could be maximized in children with gastroenteritis.

Intake of Dietary Fat

The intake of fat as a component of total energy varies considerably between different children. Fat consumption has been increasing in children from developed countries and has accelerated over the last 100 years. Contemporary comparison of fat intake between nations now shows large variations. For example, Perissé et al. reviewed countries for the Food & Agriculture Organization of the United Nations (FAO), and showed that between wealthy and non-wealthy countries, fat intake varies from about 12% of total energy intake to over 40%.[109] The explosion in the rate of increase in obesity clearly highlights the impact of recent changes in our dietary habits. As obesity begins in childhood, preventative measures early in life need to be in place if we are to reduce the burgeoning health and economic burden.[110,111] It is not only healthy children who have seen a change in fat intake, children with cystic fibrosis have been given much larger amounts of fats. This is because increased calorie intake, particularly that of fat, results in improvements in

respiratory function.[112,113] Much of this fat is digested in children with cystic fibrosis by the use of ingested enzyme preparations given with meals. However, lipases secreted in the gastrointestinal tract proximal to the duodenum (gastric lipase) may also play a prominent role. It is important, therefore, to document whether secretion of this enzyme varies with fat. Knowledge of mechanism(s) underlying this possible adaptive effect may lead to therapeutic optimization of enzyme production and better utilization of fat in children who are continually under threat of malnutrition.

Gastric Lipase

Human milk is rich in lipids and yet, surprisingly, the enzymes required for its digestion are expressed in low levels in the newborn.[114,115] The physiology of lipid digestion varies considerably between neonates and adults; a recent article by Abrahamse and colleagues offers excellent insight into this topic.[116] Gastric lipase accelerates the digestion of triglycerides in human milk,[117] by releasing free fatty acids which enhance pancreatic lipase and colipase activities. The actions of gastric lipase are magnified in children with little pancreatic lipase activity, as in cystic fibrosis. Newborn babies also have relatively low pancreatic lipase activity[114,115] and the relative importance of gastric lipase is exaggerated in the premature neonate.[118,119] There are significant biochemical differences between pancreatic and gastric lipase, most significant of which is the difference in pH required for maximal activity.[120,121] In addition, gastric lipase activity requires no bile salt or colipase. In the human, the gastric mucosa secretes all the preduodenal lipase.[122,123] In the rodent, the tongue is the most significant source, and here the equivalent enzyme is termed lingual lipase. There may also be some lingual production in the preterm human infant because lipase activity has been detected in esophageal pouches of infants with esophageal atresia with no communication to the stomach.[119] The rabbit predominantly secretes its preduodenal lipase from the gastric mucosa, making the rabbit a more appropriate model than the rodent in which to study the effect of dietary influence on this enzyme.[124]

Gastric lipase activity is more susceptible than pancreatic lipase to dietary changes. Increasing the dietary fat intake in rabbits from 2.7%– to 6% of diet for two weeks was sufficient to induce a 100% increase in gastric lipase activity.[124] This increase had no effect on pancreatic lipase production. Indeed, pancreatic lipase activity was only enhanced when the

fat intake was increased to 12% of diet, and then the increase was only modest, a 10% increase in pancreatic lipase activity. Studies in adult human volunteers have shown similar data with a doubling (5.7-9.95 U/mL) of gastric lipase after changing from a low-fat to a high-fat diet.[125] The mechanisms that detect the amount of fat intake (see Figure 19-1) need further examination, but triglyceride composition had no effect on the adaptive response in the rabbit.[124] This implicates a metabolite of fatty acid breakdown, which is not specific for either saturated or unsaturated fatty acids, as the cellular agent that initiates changes in gene expression. The signal may not have to come directly from the lumen of the GI tract because there is preliminary evidence, in neonates at least, indicating that increasing lipid intake parenterally results in enhanced gastric lipase activity. This suggests that signals from the serosal side of the IECs may be possible in this case.

Gastric lipase is therefore under the control of dietary signals. Although we do not at present understand the mechanisms that underlie this phenomenon, their elucidation may eventually allow us to manipulate gastric lipase expression when it is clinically important, as in the nutrition of cystic fibrosis and in the oral intake of the newborn.

Fatty Acid Uptake

Fatty acid (FA) and cholesterol uptake in the GI tract are major pathways involved in maintenance of lipid homeostasis.[126] FA uptake is facilitated by the presence of fatty acid transporter proteins (FATPs). These include (1) FABPpm, a 40 kDa plasma membrane protein homologous to the mitochondrial enzyme aspartate aminotransferase; (2) FAT/CD36, an 88 kDa membrane glycoprotein, also known as the Class B scavenger receptor; and (3) FATP4, which promotes FA uptake and activation by trapping FA inside the cell in their CoA-thioester forms prior to mitochondrial-driven beta oxidation.[127,128] The fatty-acid binding protein (FABP)-2 regulates the concentration of free FA in intestinal cells. It binds saturated and unsaturated FA with similar affinity and aids in transporting FA from the plasma membrane to the endoplasmic reticulum where enzymatic modifications ensue. Epidemiological studies investigating potential association of polymorphisms at codon 54(A/T) in FABP2 gene and insulin sensitivity and type 2 diabetes have been recorded.[129] The review by Rubin and colleagues provides a comprehensive analysis of the data available on FABP2 and its role in FA processing.[130] Ex vivo model studies of fetal jejunum explants implicate FABP2 polymorphism to

specifically influence small intestinal lipid absorption with minimal effect on glucose uptake or metabolism.[129]

A third fatty acid transporter (FAT; CD36) associated with long chain fatty acid as its ligand has recently been implicated to play a role in the pathogenesis of insulin resistance. CD36 -/- mice exhibited enhanced insulin responsiveness on a high-starch, low fat diet.[131] In a separate study, however, CD36 knockout mice were shown to absorb triacylglycerol and FA as well as the wild-type littermates.[132] Although evidence suggests that CD36 plays a role in FA uptake in adipose tissue, its role in the intestine requires clarification.[133]

Breast Milk and Gene Expression

Nutritional and immunological protection of the infant are the primary roles of breastfeeding, properties that are reviewed in greater detail in other accompanying chapters. Nevertheless, the cells of the GI tract are the first cells of the child to encounter breast milk, and it is not fanciful to suppose that breast milk may influence the development of these cells. A number of factors in milk can affect the expression of genes in the intestinal epithelium, particularly those genes that are associated with enterocyte differentiation. Milk-derived growth factors such as epithelial growth factor (EGF), milk fat globule-EGF factor 8 (a major protein associated with milk fat globule membrane; the protein is composed of EGF and factor VIII-like domains), vascular endothelial growth factor (VEGF), and transforming growth factor-β (TGF-β) have all been reported to have potent effects on the immature gut.[134–137] Although the exact effect of EGF in breast milk on the development of the intestinal epithelium depends on the age of the subject, its effect on enterocyte growth and development is not surprising. Less expected, perhaps, is the demonstration that other breast milk components such as nucleotides also affect these same genes.[138,139] Although our current understanding of metagenomic crosstalk between diet, the host microbiome and the GI mucosal immune system is still in its infancy, studies have begun to provide tantalizing clues as to how these various players impact on each other. Shwartz and colleagues compared these interactions in breast-fed versus bottle-fed neonates at 3 months of age and found that breastfeeding promotes functional changes that allow for optimal adaption to extrauterine life.[140] Herein, we describe, lactoferrin, as an example. We will examine what is known about its interactions with the intestinal epithelium, and how this knowledge may be increased in the future.

Lactoferrin

Lactoferrin, an 80kD iron-binding glycoprotein of the transferrin family, is a major component of mammalian colostrum and milk.[141,142] It is also found in mucosal secretions and neutrophil granules.[143] Lactoferrin has bactericidal properties and is a source of iron in breast-fed infants.[144] The protein has been found to have diverse biological functions including facilitating iron absorption, host defense, regulation of cell proliferation and differentiation, modulating the immune system and embryonic development.[145,146]

Specific binding sites for lactoferrin have been observed in a variety of tissues and cell types including immune cells, bone, brain, liver, and the GI tract.[147] The piglet model is considered to be an excellent model for human infants as the GI physiology and function are similar.[148] In neonatal pigs (1 week of age) lactoferrin receptor (LfR) expression is low on brush-border membranes and predominant expression is considered to be intracellular.[149] Increase in duodenum LfR does occur between 1 and 3 weeks of age with minimal changes in the jejunum suggesting site-specific modulation of lactoferrin action. To gain greater insight into the role of lactoferrin-LfR axis in iron metabolism in the prenatal and postnatal periods, a recent review by Lipinski and colleagues is recommended.[150] Lactoferrin is relatively resistant to proteolytic degradation in human infants this stability through transit in the gut suggests it has the potential of interacting directly with the intestinal epithelium.[151] Emerging data indicates that lactoferrin can indeed be taken up by intestinal epithelial cell-lines leading to changes in cell signaling and proliferation status.[152] The lactoferrin protein consists of a bactericidal domain (lactoferricin), which can be generated in vitro by pepsin digestion. This domain comprises of residues 1-45 of human lactoferrin and 17-41 of bovine lactoferrin and exhibits antimicrobial activity against a variety of pathogens.[153,154] An understanding of the actions of lactoferrin/lactoferricin at the molecular level are particularly relevant as some artificial milk formulas now contain added bovine lactoferrin.[154]

There is a school of thought that proposes that the main function of lactoferrin in vivo is not to transport iron into the epithelium, but to scavenge iron (and possible other substances) in the intestinal lumen. This unbound iron might otherwise cause free radical-mediated damage to sensitive tissues.[145,146] Accordingly, lactoferrin would have no direct effect on the intestinal epithelium, and only indirectly interact with it through the sequestration of other substances in the lumen. These luminal

factors, if left free in solution, may have an impact on enterocyte gene expression. An example of this is lactoferrin-mediated effects on microbial lipopolysaccharide (LPS) reactivity, thereby modulating the effect of this moiety on the host.[155] However, a purely luminal role for lactoferrin is difficult to reconcile as emerging studies suggest that once internalized, lactoferrin translocates to the nucleus where it may modulate gene transcription by mechanisms that currently remain unclear.[156]

Because lactoferrin is secreted by neutrophils, its effect on immune and non-immune cell gene expression has been closely studied. These observations have resulted in some intriguing information on lactoferrin function, which may be relevant to the actions of breast milk lactoferrin on epithelia. Lactoferrin interacts avidly with nucleic acids in a test tube.[157–159] He and Furmanski have determined the specific DNA sequence recognized by lactoferrin.[160] Iron-saturated lactoferrin exhibits greater affinity for the DNA sequence than the iron-unsaturated form, suggesting conformation a crucial factor in determining DNA-protein interaction. Lactoferrin has also been shown to modulate promoter activity of genes in granulocytes.[161]

So, taken together, evidence does support the notion that lactoferrin from mother's milk is taken up by receptor-mediated endocytosis allowing the protein to impact on epithelial gene expression in some way. How this is achieved and what benefits this would have for the developing infant are questions that future research may well answer. Previous work from our laboratory indicated that lactoferrin affects the proliferation and differentiation of intestinal epithelial cells, and that this effect depends on the degree of saturation by iron.[159] It is possible that these changes in enterocyte gene expression are mediated by mechanisms similar to those outlined by He and Furmanski in white cells.[160]

Signaling From the Intestinal Lumen to the Mucosal Immune System

The function of the small intestinal epithelium is to absorb nutrients from the environment while providing a barrier to the external world.[162,163] The barrier is not complete, as the intestine allows macromolecules to be sampled and actively absorb nutrients. The success of the intestine in achieving the balance between these competing aims was the critical step in the evolution of multi-organ animals. The ability of the intestine to monitor the environment is a key element in this process in which the mucosal immune system plays an important part. Curiously, until recently,

the possibility that epithelium may respond to luminal factors (and signal their presence to the mucosal immune system) had been substantially ignored. The epithelium has mainly been regarded as a passive barrier with points of selective filtration (Figure 19-6A), surveillance of the luminal contents occurring after this filtration has taken place. Increased appreciation of the impact of the resident microbiome in recent years has highlighted how critical the epithelial immune barrier function is to intestinal health.[164]

The epithelia express Pattern recognition receptors (PRRs), receptors that detect not only microbial signature motifs but also recognize endogenous host danger signals; referred as danger-associated molecular motifs (DAMPs). Toll-like receptors is one PRR family, members of this family reside in appropriate cellular compartments awaiting detection of microbial presence.[165,166] In recent years, a central role for intracellular PRRs the Nucleotide-like proteins (NLRs) in epithelial immune defense has emerged.[4,164,167] Initially, NLRs were considered to regulate inflammation via apoptosis; however, in the last decade, several members of the NLR family have been found to form components of a large protein platform referred as the “inflammasome” complex. Activation of the inflammasome can be triggered by NLR members such as NLRP1, NLRC4, NALP3, and NALP6 as they can sense exogenous and endogenous DAMPs. Once activated, through a series of protein associations, the final step of the complex formation involves the recruitment of procaspase-1, an enzyme that undergoes auto-proteolysis releasing active caspase-1. This enzyme is now known to be responsible for cleaving pro-IL-1β and pro-IL18 to their bioactive form. Both cytokines not only exert cytoprotective effects on the intestinal epithelia but can also modulate adaptive immune responses as the former is involved in differentiation of Th-17 cells while the latter is as known as an interferon-gamma inducing factor.[168,169] In addition to regulating cytokine release, the inflammasome can also activate a novel form of cell death called “pyropotosis”. Due to space constraints, these functions will not be discussed further, instead excellent reviews by Flavell and co-workers and Nunez and colleagues are recommended for deeper understanding of how the inflammasome function (both in non-hematopoietic and hematopoietic compartments) may contribute to GI homeostasis in health and disease.[164,167]

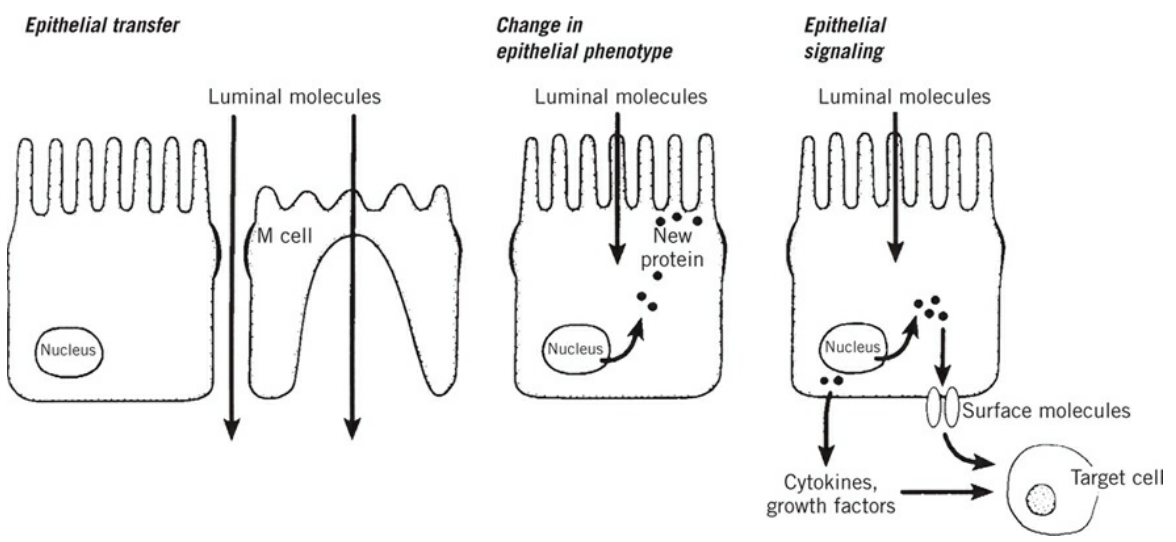


Figure 19-6 Possible interactions of molecules from the intestinal lumen with the epithelium. Traditionally the epithelium has been seen as a selective barrier to molecules, admitting those required for energy intake or immunosurveillance and excluding others (A). However, nutrients can alter the phenotype of the epithelium to adapt to changing nutritional needs (B). When changes in the intestinal lumen induce new proteins that interact with mucosal immune cells, the epithelium can act as a membrane signaling information from the lumen to the immune system (B).

The epithelial cell can also express proteins that may interact with immunocytes within the intestine. These include surface molecules such as class II major histocompatibilty complex and secretory chemokines and cytokines, thus modulating T cell function by direct and indirect mechanisms. This means that the epithelial cell must recognize (or "sense") the nutrient or other luminal factor, and a mechanism must then "transduce" this recognition into molecular changes that can directly alter gene expression (see Figures 19-1 and 19-4). The main difference in molecular terms from the adaptive response described earlier (Figure 19-6B) is that the resultant proteins escape from the compass of the epithelium and interact with receptors on target cells (Figure 19-6C).

Signaling, by transducing molecular interactions with the epithelium into secretion of messenger proteins, allows luminal factors to influence immune responses while maintaining full epithelial barrier integrity (Figure 19-6C). Although the absorption of luminal factors (Figure 19-6A) is an important method by which the mucosal immune system surveys the

intestinal lumen,[4] it requires the entry of molecules through the epithelial barrier. This provides a potential source for invasion by pathogens. The poliovirus, for example, enters by this means, exploiting the normal uptake of macromolecules. The ability of the epithelium to signal information to the mucosal immune system without the need for a physical pathway from the lumen to the serosal surface offers obvious advantages in terms of epithelial protection.

The notion of epithelial signaling (Figure 19-6C) implies that the epithelium can modulate the level of immune activity in the mucosal immune system according to the environment of the intestinal lumen. Mucosal immune responses play a large part in gastrointestinal diseases, particularly Crohn's disease, ulcerative colitis, and celiac disease. The milieu of the intestinal lumen could therefore be manipulated for therapeutic purposes, by alterations in diet.

Genes of Immunologic Importance in the Intestinal Epithelium

An increasing list of immunologically active proteins are now known to be synthesized by the intestinal epithelium (Table 19-3), and these originally were thought to be produced only by other cell types. Human diseases are associated with the increased production of some of these proteins in the circulation and the gastrointestinal tract. We and others have shown that the expression of these proteins in the epithelium is influenced by dietary factors.[170]

Cytokines

An interesting example of a cytokine secreted by the epithelium is IL-8. This protein is small (8,000 D) but has a long half-life in vivo relative to other cytokines, making it a feasible candidate for long-lasting control over mucosal immune responses. IL-8 is a member of a super family of cytokines, "chemokines," which have chemo tactic properties, recruiting as well as activating a number of immunologic cell types. It is a potent chemo tactic factor for neutrophils, and T lymphocytes and may be important in recruiting them into the lamina propria during GI infection and inflammation Invasion by neutrophils, in particular, is a hallmark of Crohn's disease, a condition in which IL-8 production is enhanced.[171] IL-8 also stimulates the release of superoxide radical in neutrophils as well as other potential mediators of damage in intestinal inflammation. Furthermore, it increases the permeability of vascular endothelium to albumin resulting in tissue edema. Its production in intestinal epithelial

cells has been documented in response to bacterial invasion. *Clostridium difficile* infection can result in pseudo membranous colitis in susceptible individuals, a colitis that is exemplified by a marked neutrophil influx causing a cytokine storm that leads to extensive tissue pathology. Animal and human studies suggest excessive IL-8 release most likely contributes to the presence of neutrophils and resultant pathology.[172] We generated MIP-2 (mouse chemokine similar to human IL-8) transgenic mice where the chemokine expression was confined to the small intestinal and colonic epithelium by the use of fatty-acid binding protein promoter.[173] The epithelium from the first generation of the founder showed effects on both neutrophil and lymphocyte recruitment. In the small intestine where the FABPI promoter is active, the neutrophil recruitment expressed as myeloperoxidase activity was significantly greater. In the distal colon where the promoter is inactive, there was no effect. These studies show that the epithelial can, through the release of chemokines, alter the mucosal immune function in vivo. Further analysis revealed increased lymphocyte infiltration ($P < 0.05$), in addition to neutrophils. There was also a doubling of the number of intraepithelial (IEL) lymphocytes. The increase in IELs was due to an increase in both αβ and γδ lymphocyte populations. Interestingly, these cells express CXCR2, the receptor for MIP-2. These studies highlight the potential recruitment capacity of a single chemo tactic activity of the in vivo situation, however, is likely to be more complex with simultaneous alteration of various immune regulators.

TABLE 19-3 Examples or Immunologically Relevant Genes Expressed by Intestinal Epithelial Cells

Cytokines	
IL-1β	Experimental colitis
IL-6	Isolated epithelium Rat IEC-6 cell line with IL-1
IL-8	Colon carcinoma cell lines
MIP-2	Rat IEC-6 cell line with IL-1
Acute-phase respondents	Colon carcinoma cell lines
Surface Molecules	
Class II	Sections of rat small intestine
MHC invariant chain	Freshly isolated mouse epithelial cells, IEC-6 cells
Growth Factors	

IGF-I	Colon carcinoma cell lines
IGFBPs	Colon carcinoma cell lines

Evidence suggests that various epithelial-derived immune regulatory molecules can be altered by dietary factors.[4,174] n-Butyrate is a short-chain fatty acid produced by the metabolism of normal intestinal bacteria whose levels in the intestine vary with diet. Newborn babies have very low butyrate levels in either the small or large intestine. Butyrate levels rise steadily reaching adult levels by 2 years of age. Lionetti and colleagues studied the effect of diet on fecal butyrate levels by comparing the fecal microbiota of children from Europe and a rural African village Burkina Faso (BF).[175] As the dietary habits of the latter population have not changed since the introduction of agriculture, the effect of diet was amenable to investigation. The BF children showed a significant enrichment in Bacteroidetes, with a unique abundance of the genus *Prevotella* and *Xylanibacter*, microbes that can hydrolyse cellulose and xylan. In addition, BF children also showed greater butyrate levels compared to EU children, providing compelling evidence of the impact of diet on butyrate levels.[175,176]

Butyrate levels clearly depend on the type of bacteria in the gut and how much substrate is available for butyrate production. Butyrate levels therefore reflect events in the intestinal lumen, and it is no surprise that butyrate alters intestinal epithelial cell signaling. Butyrate increases IL-8 secretion in enterocytes while simultaneously down-regulating another chemo tactic cytokine, macrophage chemo tactic protein-1 (MCP-1).[177,178] Butyrate-induced histone acetylation is involved in the regulation of chemokine gene expression.[179] Unlike IL-8, which attracts neutrophils, MCP-1 attracts macrophages and monocytes. It is constitutively secreted by the intestinal epithelium, whereas IL-8 is not. These observations are consistent with events in the whole intestine in vivo where, in healthy tissue, neutrophils are absent and macrophages abound. With increasing butyrate levels, IL-8 secretion is induced but MCP-1 production is depressed. Such “chemokine switching” in response to butyrate may well alter the population of immunologic cells in the lamina propria. Thus, butyrate may affect both the number and types of cells in inflamed tissue through changes in chemokine production of epithelium during inflammation.

Antimicrobial Peptides

Several families of epithelial-derived antimicrobial peptides are known to survey the gut lining in health and disease.[180,181] The expression of the β-defensin family and the single human cathelicidin, LL-37 has been observed in enterocytes (the expression of α-defensin family is confined only to Paneth cells). Several studies have shown dynamic changes in the expression of these peptides during infectious and inflammatory episodes implicating them in the front line of host defence in the GI tract.[180,181]

Class II MHC and Invariant Chain Expression

The initiation of an immune response to protein antigen normally requires the help of T lymphocytes. Activation of T cells in turn depends on the processing and presentation of peptides by an antigen-presenting cell (APC).[182] Class II MHC heterodimers (Ia in the rodent) are the molecules that present the processed exogenous antigen to the T cell receptor. In addition to classic APCs (dendritic cells, macrophages, B lymphocytes), a number of cell types express class II MHC (Ia antigen) including intestinal epithelial cells, highlighting the enterocyte as a potential APC. There is evidence that both the small intestinal and colonic epithelium may function as an APC.[183,184] The requirement for cell viability and an intact cytoskeleton suggests pinocytosis as a major mechanism for active antigen uptake by intestinal epithelial cells (IECs), however, passive diffusion and adsorption may also contribute to the process. Processing and presentation of exogenous antigen requires an additional protein, the invariant chain (Ii).[185] Diet has a marked effect on the expression of class II MHC and its associated invariant (Ii) chain in the mouse intestinal epithelium. The expression of class II MHC and Ii mRNA is developmentally regulated[186] in the epithelium (unlike its expression in the lamina propria, where it is expressed from before birth). In addition, the timing of expression can be altered by delaying the age of weaning from mother's milk to normal chow.[187] Expression of class II MHC and Ii chain is apparent three-four days after weaning on chow. However, weaning onto an elemental diet (which contains chemically synthesized amino acids, simple sugars, and fats) did not induce the expression of class II MHC or invariant chain (Figure 19-7). Thus, the expression of these genes in the intestinal epithelium is influenced by dietary manipulation in vivo. The fact that elemental diets used in these experiments were the same as those administered therapeutically in Crohn's disease lends support to the idea that alterations in gene expression may by a significant tool in the treatment of disease (see below).

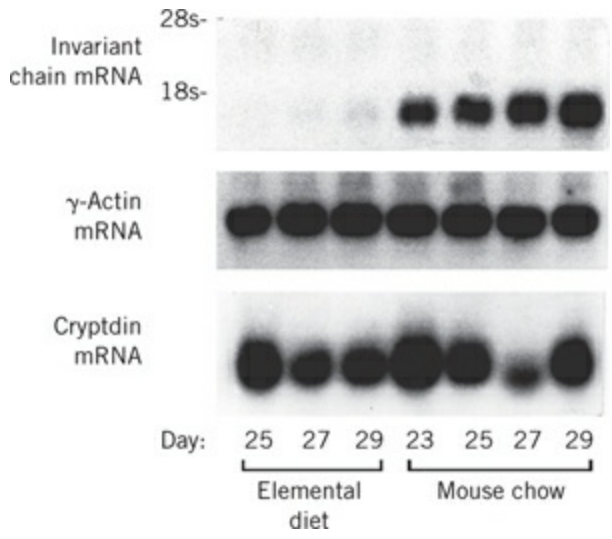


Figure 19-7 Dietary factors alter the expression of Class II MHC and invariant chain expression in intestinal epithelial cells. Northern blot of RNA from epithelial cells taken from individual mice of a single split litter weaned at day 17 onto elemental diet or Purina mouse chow. The blot was probed with invariant chain cDNA and auto radio graphed for 48 hours. Mice were examined at day 23, 25, 27, and 29. Blots were also probed with γ-actin and cryptdin cDNA to verify uniformity of enterocyte extraction. (Reproduced from Sanderson IR et al.[186])

Insulin-Like Growth Factor Binding Proteins (IGFBPs)

Immune responses depend not only on the activation of T cells, but also on the ability of these T cells to proliferate. Earlier sections of this chapter described the importance of IGF-1 in growth. However, IGF-1 is also an agent that affects cells of the immune system.[188] IGF-1 exerts a range of effects on T cell physiology. The growth factor induced a marked increase in the gene and protein expression of both CD25 and IL-2 and modulated T cell proliferation via both autocrine and paracrine mechanisms.[189] Treatment of adult mice with recombinant human IGF-1 induces striking modifications in lymphocyte number and function. Fourteen days of treatment with IGF resulted in increases in both CD4+ T cells and splenic B cells. Mitogenic responses of T and B cells were also enhanced, demonstrating that IGF-1 increases lymphocyte numbers and activity. As discussed earlier, IGFBPs modulate the actions of IGF-1. Because cultured intestinal epithelial cell,[190] secrete IGFBPs, it is possible that the epithelium can influence the proliferation of activated B cells by this

means. Furthermore, studies have shown that nutritional factors affect the production of IGF binding proteins in cell lines in vitro.[191] Therefore, not only may nutritional factors influence IGF/IGFBP secretion by the liver into the circulation, resulting in changes in growth of the whole individual, but nutrients may also affect the IGF/IGFBP system in the intestinal epithelium, leading to alterations in the proliferation of the local immune system.

Nutrient Recognition and Signal Transduction

The mechanisms where by a nutrient (or other factor in the intestinal lumen altered by dietary change) results in changes in RNA transcription are not well understood (see Figure 19-1). It is likely that increases in transcription are the consequence of alterations binding nuclear proteins to the promoter of the particular gene, but the steps by which a cell recognizes a change in that nutrient will differ for every nutrient examined; so too will the mechanisms that trans duce this signal into alterations of nuclear protein binding. In the enterocyte we have seen the variety of mechanisms that might occur simply by appreciating the many factors that may alter when the diet is changed (see Figure 19-2). Different theoretic mechanisms of interaction exist between the lumen and epithelial cells. These mechanisms differ, depending on the exact luminal factor involved (see Figure 19-5). Small nutrients such as butyrate or galactose are likely to enter the cell and affect gene expression through their respective metabolic pathways. Growth factors and lactoferrin involve specific receptors on the surface of the epithelium, and large proteins may enter the cell and alter gene expression as they are sorted. Some complex entities may affect more than one pathway. Bacteria for instance may interact through all three pathways: they could influence enterocyte metabolism; they may produce factors or components of their cell walls (e.g., endotoxin), which may interact with surface receptors; and the macromolecules from which they are composed may enter enterocyte compartments and be sorted inside the cell. Each pathway may influence aspects of nuclear function. In addition, indirect actions of cytokines produced from immune cells may also play a role.

Although sensing and signal transduction mechanisms (see Figure 19-1) in the epithelium require further investigation, there are some data on the mechanisms by which glucose regulates the expression of pyruvate kinase (in the liver) and insulin (in pancreatic islet cells). Three key questions relate to the effect of glucose on the expression of these two proteins: (1) what are the glucose metabolite(s) that set in train the effects

on transcription? (2) how is binding of nuclear proteins to gene promoter elements induced? (3) what is the sequence of DNA in the promoter that confers glucose sensitivity on the gene in question (the so-called nutrient response element)?

In practice, this last question is the easiest to answer because DNA sequences can be easily manipulated in the laboratory. Promoters linked to reporter genes allow researchers to directly measure promoter activity. This was first examined in the pyruvate kinase gene in response to glucose. The actual sequence in the pyruvate kinase promoter was determined by mutational analysis of the promoter and its effects on reporter activity. The glucose response elements in the pyruvate kinase gene and in the insulin I gene were discovered by this means. To determine the glucose response element (GlRE) in the pyruvate kinase gene promoter, promoter DNA sequences were transiently transfected into hepatocytes in culture,[192] and were also examined in transgenic mice.[193] Mutations induced into the DNA demonstrated a glucose response element occurring twice in the pyruvate kinase promoter. This motif is known as an E box because of its resemblance to a similar sequence in a promoter of the adenovirus, which had been given this name. This same DNA sequence also occurs in other genes that regulate glucose metabolism, most notably the FAS gene, whose protein product catalyzes one of the later steps in the conversion of carbohydrate to fat, and one that is up-regulated when glucose is abundant in the diet.

The rat insulin gene also contains an E box, which is termed the Far element. Paradoxically this element does not confer glucose sensitivity to the insulin gene. This attribute is conferred by a sequence 10 bases away termed the FLAT element. We now understand some of the features of the signal transduction from nutrient to nuclear function in the insulin gene. MacFarlane and colleagues[194] have identified a nuclear factor, insulin upstream factor-1 (IUF-1), which binds to the insulin promoter. There was a high degree of IUF-1 binding in cells incubated with 20 mmol/L glucose, but this was abolished in cells incubated in 3 mmol/L glucose. In addition, this protein-DNA binding depended on the phosphorylation of IUF-1. Phosphorylation therefore alters a factor in the initiation of transcription of at least one glucose-sensitive gene. It is particularly interesting that phosphorylation is the signal that transduces glucose intake to induce insulin transcription, because it is also phosphorylation that directly affects the activity of enzymes in the Emden-Meyerhoff pathway. Thus, in this system phosphorylation acts as a common transduction mechanism, responsible for both the quick and slow responses to glucose uptake.

Identification of metabolites derived from nutrients responsible for the change in nutrient gene expression is an active area of research. Glucose-6-phosphate may possibly be the metabolite that activates the glucose response element in the FAS[195] and pyruvate kinase genes. This was demonstrated by comparing the effects of the two glucose analogs, 2-deoxyglucose (2dG) and 3-O methyl glucose (3OMeG). Neither analog can be metabolized. However, 2dG can be phosphorylated, but not 3OMeG; 2dG stimulated transcription, but 3OMeG did not. Therefore, only the phosphorylated sugar was able to stimulate the transduction mechanisms that resulted in transcription, which it did without further metabolism. Whether this phosphate bond provides the energy to phosphorylate a nuclear factor to bind to DNA is not known, but it is an interesting possibility.

NUTRIENT EFFECTS ON BACTERIAL POPULATIONS IN THE INTESTINE

This chapter has already highlighted the possibility that nutrients act on bacterial metabolism, and that the consequent fermentation products modulate gene regulation in the intestine.[196] However, bacteria themselves through the actions of their structural components act on gene expression. This is most evident in the effects on pattern recognition receptors (PRRs) (reviewed in Ref. 197). Many distinct bacteria carry similar pathogen associated molecular patterns (PAMPs). This makes it difficult to understand how changing the microbiota through diet could have a major impact on innate immune signaling. Recently,[198] innate lymphoid cells (ILCs) have been shown to express class II MHC (but not T or B cell receptors). Furthermore, they present antigens to the adaptive immune system, limiting commensal-bacteria T cell responses. Thus, the varying structures that make up bacteria can be sensed in detail. It has now become relevant to ask how dietary changes affect bacterial populations as an intermediary to its effects on intestinal gene expression.

While studies on how diet affects bacterial populations have been conducted for some time, it is only since the development of high throughput bacterial genome sequencing (bases on the DNA that codes for the 16S rRNA in the past 5-10 years.[199] Although this is a relatively new field, interesting concepts have emerged. First, it is likely that every person has a distinct detailed microbiota, that those genes that the microbes express perform similar functions. This is analogous to other ecosystems:

tropical rain forests in different parts of the world support differing plant species, but the functions that the plants undertake, for example, photosynthesis, transpiration, and so forth, are similar to each.[200] Second, different human beings may not lie on a continuum of varying bacterial species, but particular groups of bacteria may preferentially segregate together. These groups of distinct bacteria have been termed enterotypes.[201] This clustering appears to be independent of nationality, sex, age, or body mass index. However, diet may be a determinant. Using dietary interventions and 16S sequencing to characterize fecal samples from[98] individuals, Wu and co-workers[202] examined the effects of long term diets. Enterotypes were strongly associated with long-term diets. In particular, protein and animal fat predominantly supported Bacteroides-dominated enterotypes, while carbohydrates supported enterotypes with a large Prevotella component. Small changes in the microbiome are detectable within 24 hours, but enterotypes are more stable, requiring sustained dietary changes for over 10 days for a change.

-Within pediatrics, the effect of the microbiome on the transcriptome was studied in breast fed babies and compared to bottle fed babies. Using multivariate analysis, differences in the microbiota correlated with difference with the transcriptome.[203] However, these effects can be explained by bacterial metabolic products (as the authors suggest, and as described earlier in this chapter) rather than the actions of bacterial components directly. Data such as these exemplify how relatively undeveloped our current knowledge is of this complex area. Future work will increase our understanding of the effects of the varied structures.

CLINICAL AND THERAPEUTIC IMPLICATIONS OF NUTRIENT-GENE INTERACTIONS

The role of diet in diseases of many organs has become an area attracting attention from medical scientists and the general public. However, the relationship between dietary factors and the health of most organs has been difficult to examine, other than by using epidemiology.[204] Occasionally, data have emerged suggesting that particular diets benefit certain disease states. For example, renal failure was shown to be less progressive in rats whose protein intake had been restricted,[205] but even this has been difficult to substantiate in children.[206] Furthermore, it is not easy to see how the protein load has its effect. We have no knowledge of which cell types in the kidney may be affected, let alone how their phenotype may be altered

by protein intake. On the other hand, in the small intestine, the relation between diet and gastrointestinal function is clearer.

In clinical practice, one of the most substantial changes in the content of the intestine occurs before and after parenteral nutrition. The effects of such changes have profound consequences for the small intestinal epithelium in human subjects (see Figure 19-6B). Intestinal brush border hydrolase activity is diminished during parenteral feeding. Moreover, the ability of the mucosa to form a barrier is compromised. Moore and colleagues studied 74 patients who had suffered abdominal trauma randomized to receive either enteral or parenteral nutrition.[207] Those fed parenterally had more episodes of infective complications (pneumonia and abdominal abscesses) than those fed enterally. Calorie and nitrogen intake was equivalent in the two groups. As it is thought that the main source of sepsis following trauma is the gastrointestinal tract, this study may be evidence that enteral feeding directly affects the ability of the injured patient to contain bacteria within the gastrointestinal lumen.

The effect of parenteral nutrition on the morphology of the intestine in animals has long been known.[208] Considerable attention has been paid to the question of which enteral components are responsible for maintaining normal bowel morphology and function. No clear answer has yet emerged. For example, Spector and colleagues observed that luminal infusion of 30% dextrose or 5% amino acids were both effective in restoring intestinal weight, protein content, and DNA content after a period of intravenous alimentation.[209] Amino acids were more potent on a weight-for-weight basis or on a molar basis than dextrose. More recently, the changes in the intestine seen in animals fed via parenteral nutrition have been compared with the changes seen during weaning.[82] Intestinal length, mucosal mass, DNA, protein, and disaccharidase activities were significantly lower in animals sustained by intravenous nutrition when compared to normally weaned controls. However, restoration of luminal feeding resulted in a return to normal of the intestine in the TPN-fed group.

There are now many reports of the relationship of intestinal function to parenteral nutrition.[210] However, a study of the interaction between luminal content and the molecular control of intestinal cell protein synthesis is only just beginning, and this relationship is likely to provide the key to our understanding of how enter al feeds maintain a healthy epithelium.

It is also likely that epithelial signaling (see Figure 19-6C) may have important therapeutic implications for diseased intestine. First, the contents of the intestinal lumen are directly amenable to therapeutic manipulation,

whereas the environment that surrounds other, more internal, organs is maintained in strict homeostasis by a plethora of mechanisms. Second, although we do not know exactly how the local environment surrounding the epithelial cell is changed by diet (see Figure 19-2), dietary manipulations are an important aspect of management of gastrointestinal disease states.

The activity of Crohn's disease is greatly influenced by altering the contents of the intestinal lumen. Inflammation subsides in patients who are fed parenterally and receive no oral nutrients and in those receiving elemental diets. (See Enter al Products in Appendix.) These chemically defined diets induce remission as effectively as high-dose oral corticosteroids in both adults and children.[211] The return of inflammatory changes to normal in the intestine has been documented indirectly by the use of markers of intestinal permeability[212] and scanning of labeled neutrophils, and directly by examining the mucosa at endoscopy with biopsies.[213]

It has been hypothesized that a reduction in antigen load in the intestine may underlie the efficacy of elemental diets in Crohn's disease[211] in a fashion similar to that in cow's milk-sensitive enteropathy and celiac disease. However, the switch from normal diet to an elemental diet alters many constituents of the intestinal milieu apart from food antigens (see Figure 19-2). In particular, bacterial populations (and therefore bacterial products) are profoundly altered by dietary alteration. Moreover, giving whole protein in the form of liquid diets (polymeric diets) is also an effective treatment for Crohn's disease.[213] Removal of protein antigen is therefore not essential for the efficacy of the formula diet. It is more likely that the diets are altering many factors in the local environment of the intestinal lumen (see Figure 19-2). The concept of genes in the epithelium signaling events to the mucosal immune system is a first step to understanding the basis of how such events may occur because it seeks to identify molecular pathways from the intestinal lumen, through enterocytes to the mucosal immune system and other target cells beyond the epithelium. Certainly, this would be in keeping with data presented earlier (see Figure 19-7) showing that class II MHC and invariant chain expression in the epithelium are related to dietary factors. There was no expression in epithelial cells of the mice weaned onto an elemental diet,[187] and this diet was the same as that used in the treatment of Crohn's disease. Genes other than class II MHC, with common promoter elements, may also be altered by similar maneuvers, and may be important in regulating

intestinal inflammation.

CONCLUSIONS AND SPECULATION

Certain concepts have been laid down in this chapter that can be used as the basis for examining how dietary factors may affect the expression of genes in various cell types including the intestinal epithelium. The essential components for an effective transition between nutrient and gene expression in cells have been documented. These components (see Figure 19-1) comprise a system of recognizing changes in factors as they interact with a cell (sensing) and a mechanism of converting these external molecule-cellular molecule interactions into changes in gene expression (signal transduction). It is not yet fully understood how these mechanisms are effected in the cell. This chapter has presented evidence that nutritional factors do indeed alter gene expression in cells, but it is likely that different molecular species are sensed by completely different mechanisms. In the case of the epithelial cell, (see Figure 19-5) these sensing mechanisms can be at the cell surface or part of the internal mechanisms of the cell. It is also possible that special receptors exist on the surface of the cell and detect more complex external molecules such as large proteins and oligosaccharides. The apical surface of mucosal epithelial cells expresses a number of molecules belonging to the immunoglobulin super family. Their function is at present unknown, but they have been identified because they form the attachments for invading viruses. One member of the Ig super family (ICAM-1) allows rhinovirus attachment in nasal epithelium. As the primary function of these molecules on the mucosal surface cannot be to serve as a conduit for viral infection, it is tempting to suggest that they and other similar molecules might play a role in the immunosurveillance of macromolecules in the gastrointestinal tract. Signaling through these molecules would require alterations in epithelial cell gene expression for this surveillance to be transmitted to the mucosal immune system. If antigens are recognized by epithelial cells of the GI tract, it is possible that this information is correlated with information garnered from the previous penetration of similar antigens across the epithelium.

Finally, this field of nutrient-gene interaction may have important therapeutic implications; the ability of the epithelium to interact with other immune cells makes it an active member of normal human defense. This physiologic role may allow it to influence inflammatory reactions in pathologic situations. The opportunity exists through dietary means to

manipulate the constituents of the lumen of the intestine by opening up a new vista for treatment of human disease.

ACKNOWLEDGMENT

This work was supported by NIH grants: DK07703; DK40561; DK47753; and a grant from the Crohn's and Colitis Foundation of America.

REFERENCES

1. Milagro FI, Mansego ML, De Miguel C, Martinez JA. Dietary factors, epigenetic modifications and obesity outcomes: progresses and perspectives. *Mol Aspects Med*. 2013;34(4):782–812.
2. Newsholme EA, Start C. Regulation in Metabolism. London, UK: Wiley; 1973;1–349.
3. Dickson R, Abelson J, Barnes W, Reznikoff W. Genetic regulation: the lac control region. *Science*. 1975;187:27–35.
4. Sanderson IR, Walker WA. Mucosal barrier. In: Ogra R, Mestecky J, McGhee J, et al, eds. *Handbook of Mucosal Immunology*. 2nd ed. San Diego:Academic Press; 1999:5–17.
5. Rennie MJ, MacLennan PA, Hundal HS, et al. Skeletal muscle glutamine transport, intramuscular glutamine concentration and muscle protein turnover. *Metabolism*. 1989;38:47–51.
6. Manso HE, Filho HC, de Carvalho LE, et al. Glutamine and glutamate supplementation raise milk glutamine concentrations in lactating gilts. *Anim Sci Biotechnol*. 2012;3(1):2. doi: 10.1186/2049-1891-3–2.
7. Feng B, Shiber SK, Max SR. Glutamine regulates glutamine synthetase expression in skeletal muscle cells in culture. *J Cell Physiol*. 1990;145:376–380.
8. Durschlag RP, Smith RL. Regulation of glutamine production by skeletal muscle cells in culture. *Am J Physiol*. 1985;248:C442–C448.
9. Tadros LB, Willhoft NM, Taylor PM, Rennie MJ. Effects of glutamine deprivation on glutamine transport and synthesis in primary tissue culture of rat skeletal muscle. *Am J Physiol*. 1993;265:E935–E942.
10. Low SY, Rennie MJ, Taylor PM. Sodium-dependent glutamate transport in cultured rat myotubes increases after glutamine deprivation. *FASEB J*. 1994;8:127–131.
11. He Y, Hakvoort TB, Köhler SE, et al. Glutamine synthetase in muscle is required for glutamine production during fasting and extrahepatic ammonia detoxification. *J Biol Chem*. 2010;285(13):9516–9524. doi: 10.1074/jbc.M109.092429.
12. Bode BP. Recent molecular advances in mammalian glutamine transporters. *J Nutr*. 2001;131:2475S–2487S.
13. Chaudhry FA, Reimer RJ, Krizaj D, et al. Molecular analysis of system N suggests novel physiological roles in nitrogen metabolism and synaptic transmission. *Cell*. 1999;99:769–780.
14. Chaudhry FA, Reimer RJ, Edwards RH. The glutamine commute: take the N line and transfer to the A. *J Cell Biol*. 2002;157:349–355.
15. Labow BI, Souba WW, Abcouwer SF. Mechanisms governing the expression of the enzymes of glutamine metabolism-glutaminase and glutamine synthetase. *J*

Nutr. 2001;131:2467S–2474S.
16. Lehmann C, Pavlovic D, Zhou J, et al. Intravenous free and dipeptide-bound glutamine maintains intestinal microcirculation in experimental endotoxemia. *Nutrition*. 2012;28(5):588–593. doi: 10.1016/j.nut.2011.09.021.
17. Niederlechner S, Klawitter J, Baird C, et al. Fibronectin-integrin signaling is required for L-glutamine's protection against gut injury. *PLOS One*. 2012;7(11):e50185. doi: 10.1371/journal.pone.0050185.
18. Wright EM, Loo DD, Hirayama BA. Management of type 2 diabetes: new and future developments in treatment. Biology of human sodium glucose transporters. *Physiol Rev*. 2011;91(2):733–794. doi: 10.1152/physrev.00055.2009.
19. Bhartia M, Tahrani AA, Barnett AH. SGLT-2 inhibitors in the treatment of type 2 diabetes. *Diabet Stud.* 2011;8(3):348–354.
20. Zhao FQ, Keating AF. Functional properties and genomics of glucose transporters. *Curr Genomics*. 2007;8(2):113–128.
21. Douard V, Ferraris RP. The role of fructose transporters in diseases linked to excessive fructose intake. *J Physiol*. 2013;591(2):401–414.
22. Cheeseman C. GLUT7: a new intestinal facilitated hexose transporter. *Am J Physiol Endocrinol Metab*. 2008;295(2):E238–E241. doi: 10.1152/ajpendo.90394.2008.
23. DeBosch BJ, Chi M, Moley KH. Glucose transporter 8 (GLUT8) regulates enterocyte fructose transport and global mammalian fructose utilization. *Endocrinology*. 2012;153(9):4181–4191. doi: 10.1210/en.2012–1541.
24. Barker A, Langenberg C, Wareham NJ. Genetic determinants of glucose homeostasis. *Best Pract Res Clin Endocrinol Metab*. 2012;26(2): 159–170. doi: 10.1016/j.beem.2011.12.002.
25. Shoelson SE, Herrero L, Naz A. Obesity, inflammation and insulin resistance. *Gastroenterology*. 2007;132:2169–2180.
26. Nettleton JA, Hivert MF, Lemaitre RN, et al. Meta-analysis investigating associations between healthy diet and fasting glucose and insulin levels and modification by loci associated with glucose homeostasis in data from 15 cohorts. *Am J Epidemiol*. 2013;177(2):103–115.
27. Chaneton B, Hillmann P, Zheng L, et al. Serine is a natural ligand and allosteric activator of pyruvate kinase M2. *Nature*. 2012; 491(7424):458–462. doi: 10.1038/nature11540. Epub 2012 Oct 14
28. Uyeda K, Yamashita H, Kawaguchi T. Carbohydrate responsive element-binding protein (ChREBP): a key regulator of glucose metabolism and fat storage. *Biochem Pharmacol.* 2002; 63:2075–7080.
29. Stoeckman AK, Towle HC. The role of SREBP-1c in nutritional regulation of lipogenic enzyme gene expression. *J Biol Chem*. 2002;277:27029–27035.
30. Shao W, Espenshade PJ. Expanding roles for SREBP in metabolism. *Cell Metab*. 2012;16(4):414–419. doi: 10.1016/j.cmet.2012.09.002.
31. Wong RH, Sul HS. Insulin signaling in fatty acid and fat synthesis: a transcriptional perspective. *Curr Opin Pharmacol*. 2010;10(6): 684–691.
32. Roth U, Curth K, Unterman TG, Kietzmann T. The transcription factors HIF-1 and HNF-4 and the coactivator p300 are involved in insulin- regulated glucokinase gene expression via the phosphatidylinositol 3-kinase/protein kinase B pathway. *J Biol Chem*. 2004;279(4):2623–2631.
33. Carrillo JJ, Ibares B, Esteban-Gamboa A, Feliu JE. Involvement of both phosphatidyl 3-kinase and p44/p42 mitogen-activated protein kinase pathways in the short-term regulation of pyruvate kinase L by insulin. *Endocrinology*. 2001;142:1057–1064.

34. Vaulont S, Vassaur-Cognet M, Kahn A. Glucose regulation of gene transcription. *J Biol Chem*. 2000;275:31555–31558.
35. Decaux JF, Marcillat O, Pichard AL, et al. Glucose-dependent and -independent effect of insulin on gene expression. *J Biol Chem*. 1991;266:3432–3438.
36. Lefrancois-Martinez A-M, Diaz-Guerra M-JM, Vallet V, et al. Glucose-dependent regulation of the L-pyruvate kinase gene in a hepatoma cell line is independent of insulin and cyclic AMP. *FASEB J*. 1994;8:89–96.
37. Jeong JY, Jeoung NH, Park KG, Lee IK. Transcriptional regulation of pyruvate dehydrogenase kinase. *Diabetes Metab J*. 2012;36(5):328–335.
38. Yamada K, Tanaka T, Miyamoto K, Noguchi T. Sp family members and nuclear factor-Y cooperatively stimulate transcription from the rat pyruvate kinase gene distal promoter region via their direct interactions. *J Biol Chem*. 2000;275:18129–18133.
39. Wiederkehr A, Wollheim CB. Mitochondrial signals drive insulin secretion in the pancreatic β-cell. *Mol Cell Endocrinol*. 2012;353(1–2):128–1237.
40. Chandra R, Liddle RA. Recent advances in pancreatic endocrine and exocrine secretion. *Curr Opin Gastrenterol*. 2011;27 (5):439–443.
41. Rorsman P, Braun M. Regulation of insulin secretion in human pancreatic islets. *Annu Rev Physiol*. 2013;75:155–179.
42. Welsh M, Schrerberg N, Gilmore R, Steiner DF. Translational control of insulin biosynthesis. Evidence for regulation of elongation, initiation and signal-recognition-particle-mediated translational arrest by glucose. *Biochem J*. 1986;235:13590–13594.
43. Guest PC, Baileys EM, Rutherford NG, Hutton JC. Insulin secretory granule biogensis: co-ordinate regulation of the biosynthesis of the majority of constituent protein. *Biochem J*. 1991;274:431–437.
44. Nishio Y, Aiello LP, King GL. Glucose induced genes in bovine aortic smooth muscle cells identified by mRNA differential display. *FASEB J*. 1994;8:103–106.
45. Giddings SJ, Chirgwin J, Permutt MA. Effects of fasting and refeeding on prepro insulin messenger RNA. *J Clin Invest*. 1981;67:952–960.
46. Giddings SJ, Chirgwin J, Permutt MA. Effects of glucose on pro insulin messenger RNA in rats in vivo. *Diabetes*. 1982;31:624–629.
47. Gosmain Y, Katz LS, Masson MH, et al. Pax6 is crucial for β-cell function, insulin biosynthesis, and glucose-induced insulin secretion. *Mol Endocrinol*. 2012;26(4):696–709.
48. Ahlqvist E, Turrini F, Lang ST, et al. A common variant upstream of the PAX6 gene influences islet function in man. *Diabetologia*. 2012;55(1):94–104.
49. Kanof ME, Lake AM, Bayless TM. Decreased height velocity in children and adolescents before the diagnosis of Crohn's disease. *Gastroenterology*. 1988;95:1523–1527.
50. Sawczenko A, Azooz O, Paraszczuk J, et al. Intestinal inflammation-induced growth retardation acts through IL-6 in rats and depends on the -174 IL-6 G/C polymorphism in children. *Proc Natl Acad Sci U S A*. 2005;102:13260–13265.
51. Ezri J, Marques-Vidal P, Nydegger A. Impact of disease and treatments on growth and puberty of pediatric patients with inflammatory bowel disease. *Digestion*. 2012;85(4):308–319.
52. Randhawa RS. The insulin-like growth factor system and fetal growth restriction. *Pediatr Endocrinol Rev*. 2008;6(2):235–240.
53. Clemmons DR. Metabolic actions of insulin-like growth factor-I in normal physiology and diabetes. *Endocrinol Metab Clin North Am*. 2012;41(2):425–443.
54. Cianfarani S, Liguori A, Germani D. IGF-1 and IGFBP-3 assessment in the

management of childhood onset of growth hormone deficiency. *Endocr Rev*. 2005;9:66–75.
55. Ekstrom K, Salemyr J, Zachrisson I, et al. Normalization of IGF1/IGFBP axis by sustained nightly insulinization in type 1 diabetes. *Diabetes Care*. 2007;30:1357–1363.
56. Backeljauw PF, Chernausek SD. The insulin-like growth factors and growth disorders of childhood. *Endocrinol Metab Clin North Am*. 2012;41(2):265–282.
57. D'Mello S, Trauernicht A, Ryan A, et al. Innate dysfunction promotes linear growth failure in pediatric Crohn's disease and growth hormone resistance in murine ileitis. *Inflamm Bowel Dis*. 2012:18(2):236–245.
58. Katsumata M, Cattaneo D, White P, et al. Growth hormone receptor gene expression in porcine skeletal and cardiac muscles is selectively upregulated by postnatal undernutrition. *J Nutr*. 2000;130:2482–2488.
59. Wei Y, Puzhko S, Wabitsch M, Goodyer. Transcriptional regulation of the human growth hormone receptor (hGHR) gene V2 promoter by transcriptional activators and repressor. *Mol Endocrinol*. 2009;23(3):373–387
60. Courtland HW, Sun H, Beth-On M, et al. Growth hormone mediates pubertal skeletal development independent of hepatic IGF-1 production. *J Bone Miner Res*. 2011;26(4):761–768.
61. Ohlsson C, Mohan S, Sjögren K, et al. The role of liver-derived insulin-like growth factor-I. *Endocr Rev*. 2009;30(5):494–535.
62. Thissen JP, Triest S, Maes M, et al. The decreased plasma concentration of insulin-like growth factor-1 in protein-restricted rats is not due to decreased numbers of growth hormone receptors on isolated hepatocytes. *J Endocrinol*. 1990;124:159–165.
63. Peterson CA, Gillingham MB, Mohapatra NK, et al. Enterotrophic effect of insulin-like growth factor 1 but not growth hormone and localised expression of insulin-like growth factor 1, insulin-like growth factor binding protein-3 and -5 mRNAs in jejunum of parenterally fed rats. *J Parenter Enteral Nutr*. 2000;5:288–295.
64. Naranjo WM, Yakar S, Sanchez-Gomez M, et al. Protein calorie restriction affects non-hepatic IGF-1 production in lymphoid system: studies using the liver specific IGF-1 gene knock-out mouse model. *Endocrinology*. 2002;143:2233–2241.
65. Alfieri CM, Evans-Anderson HJ, Yutzey RE. Developmental regulation of the mouse IGF-1 exon 1 promoter region by calcineurin activation of NFAT in skeletal muscle. *Am J Physiol Cell Physiol*. 2007;292:C1887–C1894.
66. Zhang J, Whitehead RE, Underwood LE. Effect of fasting on insulin-like growth factor (IGF)-IA and IGF-IB messenger ribonucleic acids and prohormones in rat liver. *Endocrinology*. 1997;138:3112–3118.
67. Bucuvalas JC, Chernausek SD, Alfaro MP, et al. Effect of insulin-like growth factor-1 treatment in children with cystic fibrosis. *J Ped Gastroenterol Nutr*. 2001;33:576–558.
68. Martin JI, Baxter RC. Insulin-like growth factor binding protein-3: biochemistry and physiology. *Growth Regul*. 1992;2:88–99.
69. Conover CA. Potentiation of insulin-like growth factor (IGF) action by IGF-binding protein-3:studies of underlying mechanism. *Endocrinology*. 1992;130:3191–3199.
70. Forbes BE, McCarthy P, Norton RS. Insulin-like growth factor binding proteins: a structural perspective. *Front Endocrinol (Lausanne)*. 2012;3:38.
71. Regan FM., Williams RM, McDonald A, et al. Treatment with recombinant human insulin-like growth factor (rhIGF)-I/rhIGF binding protein-3 complex

improves metabolic control in subjects with severe insulin resistance. *J Clin Endocrinol Metab*. 2010;95:2113–2122.
72. Chang EB, Rao MC. Intestinal water and electrolyte transport. In: Johnson LR, ed. *Physiology of the Gastrointestinal Tract*. 3rd ed. New York, NY: Raven Press; 1994:2027–2081.
73. Thomson AB, Chopra A, Clandinin MT, Freeman H. Recent advances in small bowel diseases: Part I. *World J Gastroenterol*. 2012;18(26):3336–3352.
74. Thomson AB, Chopra A, Clandinin MT, Freeman H. Recent advances in small bowel diseases: Part II. *World J Gastroenterol*. 2012;18(26):3353–3374.
75. Sanderson IR, Parsons DS. Influence of vascular flow on amino acid transport across the frog small intestine. *J Physiol*. 1980;309:447–460.
76. Sanderson IR, Walker WA. Uptake and transport of macromolecules by the intestine: possible role in clinical disorders (an update). *Gastroenterology*. 1993;104:622–639.
77. Sanderson IR, Croft NM. The anti-inflammatory effects of enteral nutrition. *J Parenter Enteral Nutr*. 2005;29:S134–S138.
78. Takahashi K, Sugi Y, Nakano K, et al. Epigenetic control of the host gene by commensal bacteria in large intestinal epithelial cells. *J Biol Chem*. 2011;286(41):35755–35762.
79. Kapoor A, Li HJ, Leiter AB. Intestinal development: the many faces of Wnt signalling. *Gastroenterology*. 2007;133:710–712.
80. Stephen A, Alles M, de Graaf C, et al. The role and requirements of digestible dietary carbohydrates in infants and toddlers. *Eur J Clin Nutr*. 2012; 66(7):765–779.
81. Le Gall M, Tobin V, Stolarczyk E, et al. Sugar sensing by enterocytes combines polarity, membrane-bound detectors and sugar metabolism. *J Cell Physiol*. 2007;213:834–843.
82. Robinson J, Heineke EW, Belgovic ME. Quantitative relationship between intestinal sucrase inhibition and reduction of the glycemic response to sucrose in rats. *J Nutr*. 1990;120:105–111.
83. Goda T, Yamada K, Bustamante S, et al. Precocious increase of sucrase activity by carbohydrates in the small intestine of suckling rats. I. Significance of the stress effect of sugar-induced diarrhea. *J P ediatr Gastroenterol Nutr*. 1985;4:468–475.
84. Goda T, Yasutake H, Tanaka T, Takase S. Lactase-phlorizin hydrolase and sucrase-isomaltase genes are expressed differently along the villus-crypt axis of rat jejunum. *J Nutr*. 1999;129:1107–1113.
85. Hunziker W, Spiess M, Semenza G, Lodish HF. The sucrase-isomaltase- complex: primary structure, membrane-orientation, and evolution of a stalked, intrinsic brush border protein. *Cell*. 1986;46:227–232.
86. Yeh KY, Holt PR. Ontogenic timing mechanism initiates the expression of rat intestinal sucrase activity. *Gastroenterology*. 1986;90:520–526.
87. Rosamond WD, Savaiano DA. Maintenance of sucrase activity in rat small intestine: influence of diet and age. *Dig Dis Sci*. 1988;33:1397–1402.
88. Alpers DH, Gerber JE. Monosaccharide inhibition of human intestinal lactase. *J Lab Clin Med*. 1971;78:265–274.
89. Castillo RO, Feng JJ, Stevenson DK, Kwong LK. Altered maturation of small intestinal function in the absence of intraluminal nutrients: rapid normalization with refeeding. *Am J Clin Nutr*. 1991;53:558–561.
90. Ferraris RP, Villenas SA, Diamond J. Regulation of brush border enzyme activities and enterocyte migration rates in mouse small intestine. *Am J Physiol*.

1992;262:G1047–G1059.
91. Markowitz A, Birkenmeier E, Traber PG. The 5'-flank of the human sucrase isomaltase (SI) gene directs intestine-specific transcription in transgenic mice. *Gastroenterology*. 1991;102:A285.
92. Traber PG, Wu GD, Want W. Novel DNA-binding proteins regulate intestine-specific transcription of the sucrase-isomaltase gene. *Mol Cell Biol*. 1992;12:3614–3627.
93. Bourdeau F, Zhu Y, Traber PG. Sucrase-isomaltase gene transcription requires the hepatocyte nuclear factor-1 (HNF-1) regulatory element and is regulated by the ratio of HNF-1 alpha/HNF-1 beta. *J Biol Chem*. 2001;276:32122–32128.
94. Boudreau FB, Rings EH, Van Wering HM, et al. Hepatocyte-nuclear-factor-1 (HNF-1 alpha), GATA-4 and caudal-related homeodomain protein Cdx2 functionally interact to modulate intestinal gene transcription: implication for the developmental regulation of the sucrase-isomaltase gene. *J Biol Chem*. 2002;277:31909–31917.
95. Bosse T, Fialkovich JJ, Piaseckyj CM, et al. GATA-4 and HNF-1 alpha are partially required for the expression of specific intestinal genes during development. *Am J Physiol Gastrointest Liver Physiol*. 2007;292:G1302–G1304.
96. Freund JN, Deluc I, Raul F. Lactase expression is controlled differently in the jejunum and ileum during development in rat. *Gastroenterology*. 1990;100:388–394.
97. Wright EM, Hirayama BA, Loo DF. Active sugar transport in health and disease. *J Intern Med*. 2007;261:32–43.
98. Kotler DP, Levine GM, Shiau YF. Effects of nutrients, endogenous secretions, and fasting on in vitro glucose uptake. *Am J Physiol*. 1980;238:G219–G227.
99. Hosseinzadeh Z, Bhavsar SK, Shojaiefard M, et al. Stimulation of the glucose carrier SGLT1 by JAK2. *Biochem Biophys Res Commun*. 2011;408(2):208–213.
100. Sala-Rabanal M, Hirayama BA, Loo DD, et al. Bridging the gap between structure and kinetics of human SGLT1. *Am J Physiol Cell Physiol*. 2012;302(9):C1293–C1305.
101. Ferraris RP. Dietary and developmental regulation of intestinal sugar transport. *Biochem J*. 2001;360:265–276.
102. Sharp PA, Debnam ES, Srai SK. Rapid enhancement of brush border glucose uptake after exposure of rat jejunal mucosa to glucose. *Gut*. 1996;39:545–550.
103. Ferraris RP, Diamond JM. Crypt/villus site of substrate-dependent regulation of mouse intestinal glucose transporters. *Proc Natl Acad Sci U S A*. 1993;90:5868–5872.
104. Ferraris RP, Diamond JM. Specific regulation of intestinal nutrient transporters by their dietary substrates. *Ann Rev Physiol*. 1989;51:125–141.
105. Shirazi-Beechey SP. Intestinal sodium-dependent D-glucose co-transporter: dietary regulation. *Proc Nutr Soc*. 1996;55:167–178.
106. Ferraris RP, Diamond J. Crypt-villus site of glucose transporter induction by dietary carbohydrate in mouse intestine. *Am J Physiol*. 1992;262:G1069–G1073.
107. Hu Z, Yang Y, Graham WV, et al. MAPAPK-2 is a critical signalling intermediate in NH3 activation followign Na+ glucose transport. *J Biol Chem*. 2006;281:24247–24253.
108. Palazzo M, Gariboldi S, Zanobbio L, et al. Sodium-dependent glucose transporter-1 as a novel immunological player in the intestinal mucosa. *J Immunol*. 2008;181(5):3126–3136.
109. Perissé J, Sizaret F, François P. FAO nutrition. *Newsletter*. 1969;7:1–9.
110. Wosje K, Khoury PR, Claytor RP, et al. Dietary patterns associated with fat and

bone mass in young children. *Am J Clin Nutr*. 2010; 92(2): 294–303.
111. Morandi A, Meyre D, Lobbens S, et al. Estimation of newborn risk for child or adolescent obesity: lessons from longitudinal birth cohorts. *PLoS One*. 2012;7(11):e49919.
112. Sheppard RW, Holt TL, Thomas BJ, et al. Nutritional rehabilitation in cystic fibrosis, controlled studies of effect on nutritional growth retardation, body protein turnover and course of pulmonary disease. *J Pediatr*. 1986;109:788–794.
113. Kalnins D, Wilschanski M. Maintenance of nutritional status in patients with cystic fibrosis: new and emerging therapies. *Drug Des Devel Ther*. 2012;6:151–161.
114. Fredickson B, Olivecrona T. Decrease of lipase and esterase activities in intestinal contents of newborn infants during test meals. *Pediatr Res*. 1978;12:631–634.
115. Dipalma J, Kirk CL, Hamosh M, et al. Lipase and pepsin activity in the gastric mucosa of infants, children and adults. *Gastroenterology*. 1991;101:116–121.
116. Abrahamse E, Minekus M, van Aken GA, et al. Development of the digestive system—experimental challenges and approaches of infant lipid digestion. *Food Dig*. 2012;3(1–3):63–77.
117. Bernback S, Blackberg L, Hernell O. Fatty acids generated by gastric lipase promote human milk triacylglycerol digestion by pancreatic colipase-dependent lipase. *Biochim Biophys Acta*. 1989;1001:286–293.
118. Hamosh M, Scanlon JW, Ganot D, et al. Fat digestion in the newborn: characterization of lipase in gastric aspirates of premature and term infants. *J Clin Invest*. 1981;67:838–846.
119. Lee PC, Borysewicz R, Struve M, et al. Development of lipolytic activity in gastric aspirates from premature infants. *J Pediatr Gastroenterol Nutr*. 1993;17:291–297.
120. Miled N, Canaan S, Dupuis L, et al. Digestive lipases: from three-dimensional structure to physiology. *Biochimie*. 2000;82:973–986.
121. Armand M. Lipases and lipolysis in the human digestive tract: where do we stand? *Curr Opin Nutr Metab Care*. 2007;10:156–164.
122. Moreau H, Luagier R, Gargouri Y, et al. Human preduodenal lipase is entirely of gastric fundic origin. *Gastroenterology*. 1988;95:1221–1226.
123. Abrams CK, Hamosh M, Lee TC, et al. Gastric lipase: localization in the human stomach. *Gastroenterology*. 1988;95:1460–1468.
124. Borel P, Armand M, Senft M, et al. Gastric lipase: evidence of an adaptive response to dietary fat in the rabbit. *Gastroenterology*. 1991;100:1582–1589.
125. Armand M, Hamosh M, DiPalma JS, et al. Human gastric lipase expression is affected by dietary fat. *Pediatr Res*. 1995;37:119A.
126. Masson CJ, Plat J, Mensink RP, et al. Fatty acid- and cholesterol transporter protein expression along the human intestinal tract. *PLoS One*. 2010;5(4):e10380.
127. Tso P, Nauli A, Lo C-M. Enterocyte fatty acid uptake and intestinal fatty acid-binding protein. *Biochem Soc Trans*. 2004;32:75–78.
128. Milger K, Harrmann T, Becker C, et al. Cellular uptake of fatty acids driven by ER-localised acyl-CoA synthetase FATP4. *J Cell Sci*. 2006;119:4687–4688.
129. Levy E, Menard D, Delvin E, et al. The polymorphism at codon 54 of the FABP2 gene increases fat absorption in human intestinal explants. *J Biol Chem*. 2001;276:39679–39684.
130. Rubin D, Helwig U, Pfeuffer M, et al. The effect of FABP2 promoter haplotype on response to a diet with medium-chain triacylglycerols. *Genes Nutr*. 2012;7(3):437–445.
131. Hajri T, Xia Han X, Bonen A, Abumrad NA. Defective fatty acid uptake

modulates insulin responsiveness and metabolic responses to diet in CD36-null mice. *J Clin Invest*. 2002;109:1381–1389.
132. Goudriaan JR, Dahlmans VE, Febbraio M, et al. intestinal lipid absorption is not affected in CD36 deficient mice. *Mol Cell Biochem*. 2002;239:199–202.
133. Geloen A, Helin L, Geeraert B, et al. CD36 inhibitors reduce postprandial hypertriglyceridemia and protect against diabetic dyslipidemia and atherosclerosis. *PLoS One*. 2012;7(5):e37633.
134. Playford RJ, MacDonald CE, Johnson WS. Colostrum and milk-derived peptide growth factors for the treatment of gastrointestinal disorders. *Am J Clin Nutr*. 2000;72:5–14.
135. Bu HF, Zuo XL, Wang X, et al. Milk fat globule–EGF factor 8/lactadherin plays a crucial role in maintenance and repair of murine intestinal epithelium. *J Clin Invest*. 2007;117(12):3673–3683.
136. Siafakas CG, Anatolitou F, Fusunyan RD, et al. Vascular endothelial growth factor (VEGF) is present in human breast milk and its receptor is present on intestinal epithelial cells. *Paed Res*. 1999;45:652–657.
137. Rautava S, Lu L, Nanthakumar NN, et al. Breast milk-transforming growth factor-β2 specifically attenuates IL-1β-induced inflammatory responses in the immature human intestine via an SMAD6- and ERK-dependent mechanism. *Neonatology*. 2011;99(3):192–201.
138. He Y, Chu SH, Walker WA. Nucleotide supplements alter proliferation and differentiation of cultured human (Caco-2) and rat (IEC-6) intestinal epithelial cells. *J Nutr*. 1993;123:1017–1027.
139. Sanderson IR, He Y. Nucleotide uptake and metabolism by intestinal epithelial cells in tissue culture. *J Nutr*. 1994;124:131S–137S.
140. Schwartz S, Friedberg I, Ivanov IV, et al. A metagenomic study of diet-dependent interaction between gut microbiota and host in infants reveals differences in immune response. *Genome Biol*. 2012;13(4): r32. doi: 10.1186/gb-2012-13-4-r32.
141. Hettinga K, van Valenberg H, de Vries S, et al. The host defense proteome of human and bovine milk. *PLoS One*. 2011;6(4):e19433. doi: 10.1371/journal.pone.0019433.
142. Campanella L, Martini E, Tomassetti M. New immunosensor for lactoferrin determination in human milk and several pharmaceutical dairy milk products recommended for the unweaned diet. *J Pharm Biomed Anal*. 2008;48(2):278–287.
143. Ward PP, Mendoza-Meneses M, Park PW, Conneely OM. Stimulus-dependent impairment of the neutrophil oxidative burst response in lactoferrin-deficient mice. *Am J Pathol*. 2008;172(4):1019–1029.
144. Britton JR, Koldovsky O. Gastric luminal digestion of lactoferrin and transferrin by preterm infants. *Early Hum Dev*. 1989;19:127–135.
145. Levay PF, Viljoen M. Lactoferrin: a general review. *Haematologica*. 1995;80(3):252–267.142.
146. Yang D, de la Rosa G, Tewary P, Oppenheim JJ. Alarmins Link Neutrophils and Dendritic Cells. *Trends Immunol*. 2009;30(11):531–537.
147. Suzuki YA, Lopez V, Lönnerdal B. Mammalian lactoferrin receptors: structure and function. *Cell Mol Life Sci*. 2005;62:2560–2575.
148. Domeneghini C, Di Giancamillo A, Arrighi S, Bosi G. Gut-trophic feed additives and their effects upon the gut structure and intestinal metabolism. State of the art in the pig, and perspectives towards humans. *Histol Histopathol*. 2006;21:273–283.
149. Liao Y, Lopez V, Shafizadeh TB, et al. Cloning of a pig homologue of the human lactoferrin receptor: expression and localization during intestinal maturation in

piglets. *Comp Biochem Physiol A Mol Integr Physiol*. 2007;148(3):584–590.
150. Lipiski P, Sty A, Starzyski RR. Molecular insights into the regulation of iron metabolism during the prenatal and early postnatal periods. *Cell Mol Life Sci*. 2013;70(1):23–38.
151. Britton JR, Koldovsky O. Gastric luminal digestion of lactoferrin and transferrin by preterm infants. *Early Hum Dev*. 1989;19:127–135.
152. Jiang R, Lopez V, Kelleher SL, Lönnerdal B. Apo and Holo-lactoferrin are both internalised by the lactoferrin receptor via clathrin-mediated endocytosis but differentially affect ERK-signaling and cell proliferation in Caco-2 cells. *J Cell Physiol*. 2011;226(11):3022–3031.
153. Ochoa TJ, Cleary TG. Effect of lactoferrin on enteric pathogens. *Biochimie*. 2009;91(1):30–34.
154. Zweytick D, Deutsch G, Andrä J, et al. Studies on lactoferricin-derived *Escherichia coli* membrane-active peptides reveal differences in the mechanism of *N*-acylated *versus* nonacylated peptides. *J Biol Chem*. 2011;286(24):21266–21276.
155. Kruzel ML, Actor JK, Radak Z, et al. Lactoferrin decreases LPS-induced mitochondrial dysfunction in cultured cells and in animal endotoxemia model. *Innate Immun*. 2010;16(2):67–79.
156. Gifford JL, Ishida H, Vogel HJ. Structural characterization of the interaction of human lactoferrin with calmodulin. *PLoS One*. 2012;7(12):e51026.
157. Zimecki M, Stepnaik D, Szynol A, Kruzel ML. Lactoferrin regulates proliferative response of human peripheral blood mononuclear cells to phytohemagglutinin and mixed lymphocyte reaction. *Arch Immunol Ther Exp*. 2001;49:147–154.
158. Nichols BL, McKee KS, Henry JF, Putman M. Human lactoferrin stimulates thymidine incorporation into DNA of rat crypt cells. *Pediatr Res*. 1987;21:563–567.
159. Oguchi S, Walker WA, Sanderson IR. Iron saturation alters the effect of lactoferrin on the proliferation and differentiation of human enterocytes (Caco-2) cells. *Biol Neonate*. 1995;67:330–339.
160. He J, Furmanski P. Sequence specificity and transcriptional activation in the binding of lactoferrin to DNA. *Nature*. 1995;373:721–724.
161. Penco S, Pastorino S, Bianchi-Scarra G, Garre C. Lactoferrin down- modulates the activity of the granulocyte macrophage colony- stimulating factor promoter in interleukin-1 beta-stimulated cells. *J Biol Chem*. 1995;270:12263–12268.
162. Ziegler SF, Artis D. Sensing the outside world: TSLP regulates barrier immunity. *Nat Immunol*. 2010;11:289–293.
163. Sonnenberg GF, Monticelli LA, Elloso MM, et al. $CD4^+$ lymphoid tissue inducer cells promote innate immunity in the gut. *Immunity*. 2012;34(1):122–134.
164. Elinav E, Strowig T, Kau AL, et al. NLRP6 inflammasome is a regulator of colonic microbial ecology and risk for colitis. *Cell*. 2011;145(5):745–757.
165. Naik S, Kelly EJ, Meijer L, et al. Absence of Toll-like receptor 4 explains endotoxin hyporesponsiveness in human intestinal epithelium. *J Pediatr Gastroenterol Nutr*. 2001;32:449.
166. Ey B, Eyking A, Gerken G, et al. TLR2 mediates gap junctional intercellular communication through connexin-43 in intestinal epithelial barrier injury. *J Biol Chem*. 2009;284(33):22332–22343.
167. Franchi L, Muñoz-Planillo R, Núñez G. Sensing and reacting to microbes via the Inflammasomes. *Nat Immunol*. 2012;13(4):325–332.
168. Veldhoen M, Hirota K, Christensen J, et al. Natural agonists for aryl hydrocarbon receptor in culture medium are essential for optimal differentiation of Th17 T

cells. *J Exp Med*. 2009;206(1):43–49.
169. Darwich L, Coma G, Ruth Peña R, et al. Secretion of interferon-γ by human macrophages demonstrated at the single-cell level after costimulation with interleukin (IL)-12 plus IL-18. *Immunology*. 2009;126(3):386–393.
170. Fusunyan RD, Quinn JJ, Ohno Y, et al. Butyrate enhances interleukin (IL)-8 secretion by intestinal epithelial cells in response to IL-1beta and lipopolysaccharide. *Pediatr Res*. 1998;43(1):84–90.
171. Reinecker HC, Loh EY, Ringler DJ, et al. Monocyte-chemoattractant protein 1 gene expression in intestinal epithelial cells and inflammatory bowel disease mucosa. *Gastroenterology*. 1995;108:40–50.
172. Shen A. Clostridium difficile toxins: mediators of inflammation. *J Innate Immun*. 2012;4(2):149–158.
173. Ohtsuka Y, Lee J, Stamm DS, Sanderson IR. MIP-2 secreted by epithelial cells increases neutrophil and lymphocyte recruitement in the mouse intestine. *Gut*. 2001;49:526–533.
174. Canani RB, Di Costanzo M, Leone L, et al. Potential beneficial effects of butyrate in intestinal and extraintestinal diseases. *World J Gastroenterol*. 2011;17(12):1519–1528.
175. Filippo C, Cavalieri D, Di Paola M, et al. Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proc Natl Acad Sci U S A*. 2010;107(33):14691–14696.
176. Jost T, Lacroix C, Braegger CP, Chassard C. New insights in gut microbiota establishment in healthy breast fed neonates. *PLoS One*. 2012;7(8):e44595.
177. Sanderson IR. Dietary regulation of genes expressed in the developing intestinal epithelium. *Am J Clin Nutr*. 1998;68:999–1005.
178. Kalina U, Koyama N, Hosoda T, et al. Enhanced production of IL-18 in butyrate-treated intestinal epithelium by stimulation of the proximal promoter region. *Eur J Immunol*. 2002;32(9):2635–2643.
179. Fusunyan RD, Quinn JJ, Fujimoto M, et al. Butyrate switches the pattern of chemokine secretion by intestinal epithelial cells through histone acetylation. *Mol Med*. 1999;5:631–640.
180. Wehkamp J. Breakdown of antimicrobial host defense barrier as trigger of chronic inflammatory bowel diseases. *J Transl Med*. 2011;9(suppl 2):I14.
181. Cederlund A, Kai-Larsen Y, Printz G, et al. Lactose in human breast milk an inducer of innate immunity with implications for a role in intestinal homeostasis. *PLoS One*. 2013;8(1):e53876.
182. Gutcher I, Becher B. APC-derived cytokines and T cell polaristaion in autoimmune inflammation. *J Clin Invest*. 2007;117:1119–1127.
183. Telega GW, Baumgart DC, Carding SR. Uptake and presentation of antigen to T cells by primary colonic epithelial cells in normal and diseased states. *Gastroenterology*. 2000;119:1548–1559.
184. Campeau JL, Salim SY, Albert EJ, et al. Intestinal epithelial cells modulate antigen-presenting cell responses to bacterial DNA. *Infect Immun*. 2012;80(8):2632–2644.
185. Stockinger B, Pessara U, In RH, et al. A role of Ia-associated invariant chains in antigen processing and presentation. *Cell*. 1989;56:683–689.
186. Sanderson IR, Ouellette AJ, Carter EA, Harmatz PR. Ontogeny of class II MHC mRNA in the mouse small intestinal epithelium. *Mol Immunol*. 1992;29:1257–1263.
187. Sanderson IR, Ouellette AJ, Carter EA, Harmatz PR. Ontogeny of Ia messenger RNA in the mouse intestinal epithelium is modulated by age of weaning and diet.

Gastroenterology. 1993;105:974–980.
188. Smith TJ. Insulin-like growth factor-I regulation of immune function: a potential therapeutic target in autoimmune diseases? *Pharmacol Rev*. 2010; 62(2):199–236.
189. Brocardo MG, Schillaci R, Galeano A, et al. Early effects of insulin-like growth factor-1 in activated human lymphocytes. *J Leukoc Biol*. 2001;70:297–305.
190. Oguchi S, Walker WA, Sanderson IR. Profile of IGF-binding proteins secreted by human intestinal epithelial cells changes with differentiation. *Am J Physiol*. 1994;267:G843–G850.
191. Nishimura A, Fujimoto M, Oguchi S, et al. Short-chain fatty acids regulate IGF-binding protein secretion by intestinal epithelial cells. *Am J Physiol*. 1998;275(1, pt 1):E55–E63.
192. Diaz-Guerra MJ, Bergort MO, Martinez A, et al. Functional characterization of the L-type pyruvate kinase gene glucose response complex. *Mol Cell Biol*. 1993;13:7725–7733.
193. Cuif MH, Porteau A, Kahn A, Vaulot S. Exploration of a liver-specific glucose/insulin-responsive promoter in transgenic mice. *J Biol Chem*. 1993;268:13769–13772.
194. MacFarlane WM, Read ML, Gilligan M, et al. Glucose modulates the binding activity of the beta-cell transcription factor IUF1 in a phosphorylation-dependent manner. *Biochem J*. 1994;303:625–631.
195. Foufelle F, Gouhot B, Pegorier JP, et al. Glucose stimulation of lipogenic enzyme gene expression in cultured white adipose tissue. *J Biol Chem*. 1992;267:20543–20546.
196. Sanderson IR. Short chain fatty acid regulation of signaling genes expressed by the intestinal epithelium. *J Nutr*. 2004;134(9): 2450S– 2454S.
197. Sanderson IR, Walker WA. TLRs in the Gut I. The role of TLRs/Nods in intestinal development and homeostasis. *Am J Physiol Gastrointest Liver Physiol*. 2007;292(1):G6–G10.
198. Hepworth MR, Monticelli LA, Fung TC, et al. Innate lymphoid cells regulate CD4+ T-cell responses to intestinal commensal bacteria. *Nature*. 2013;498(7452):113–117.
199. Kau AL, Ahern PP, Griffin NW, et al. Human nutrition, the gut microbiome and the immune system. *Nature*. 2011;474(7351):327–336.
200. Lozupone CA, Stombaugh JI, Gordon JI, et al. Diversity, stability and resilience of the human gut microbiota. *Nature*. 2012;489(7415):220–230.
201. Arumugam M, Raes J, Pelletier E, et al. Enterotypes of the human gut microbiome. *Nature*. 2011;473(7346):174–180.
202. Wu GD, Chen J, Hoffmann C, et al. Linking long-term dietary patterns with gut microbial enterotypes. *Science*. 2011;334(6052):105–108.
203. Schwartz S, Friedberg I, Ivanov IV, et al. A metagenomic study of diet-dependent interaction between gut microbiota and host in infants reveals differences in immune response. *Genome Biol*. 2012; 13(4):r32
204. Willett WC. Diet and health: what should we eat? *Science*. 1994; 264:532–537.
205. Hostetter TH, Meyer TW, Rennke HG, Brenner BM. Chronic effects of dietary protein on renal structure and function in the rat with intact and reduced renal mass. *Kidney Int*. 1986;30:509–517.
206. Heycock GB. Renal disease. In: McLaren DS, Burman DS, Belton AR, Williams AF, eds. *Textbook of Paediatric Nutrition*. 3rd ed. Edinburgh, UK: Churchill Livingston; 1991:237–256.
207. Moore FA, Moore EE, Jones TN, et al. TEN versus TPN following major abdominal trauma reduced septic morbidity. *J Trauma*. 1989;29:916–922.

208. Feldman EJ, Dowling RH, McNaughton J, Peters TJ. Effects of oral versus intravenous nutrition on intestinal adaptation after small bowel resection in the dog. *Gastroenterology*. 1976;70:712–719.
209. Spector MH, Levine GM, Deren JL. Direct and indirect effects of dextrose and amino acids on gut mass. *Gastroenterology*. 1977;72: 706–710.
210. Baker SS, Baker RD. *Enteric versus Parenteral Feeding and Mucosal Function. Immunophysiology of the Gut*. San Diego, CA: Academic Press; 1993:401–414.
211. Sanderson IR, Udeen S, Davies PSW, et al. Remission induced by an elemental diet in small bowel Crohn's disease. *Arch Dis Child*. 1987;62:123–127.
212. Sanderson IR, Boulton P, Menzies I, Walker-Smith JA. Improvement of abnormal lactulose rhamnose permeability ratio in active small bowel Crohn's disease by an elemental diet. *Gut*. 1987;28:1073–1076.
213. Borrelli O, Cordischi L, Cirulli M, et al., Polymeric diet alone versus corticosteroids in the treatment of active pediatric Crohn's disease: a randomized controlled open-label trial. *Clin Gastroenterol Hepatol*. 2006;4:744–753.

Nutrition and Epigenetics

20

Sang-Woon Choi, MD, PhD
and Simonetta Friso, MD, PhD

INTRODUCTION

Epigenetics is defined as a phenomenon that affects gene expression without changing the underlying base sequence. Epigenetics means super-, over-, or above-genetics. Thus epigenetic marks control the expression of a gene above the level of the genetic sequence by repressing or enhancing the gene transcription.[1]

Epigenetic traits have the peculiar characteristic of being heritable. Epigenetic phenomena can also be reversed or modified by environmental factors, including nutrition. It is known that both single nutrients and our diet as a whole can affect physiologic and pathologic processes in our body from conception to natural death. Because of this, epigenetics is now regarded as one of the most important mechanisms by which a nutrient or diet alters the expression of critical genes, thereby affecting facets of our phenotype, including health traits and disease susceptibility.[2]

DNA methylation, histone modifications, chromatin remodeling, and feasibly microRNA are now considered epigenetic phenomena. DNA methylation, a chemical modification of the cytosine base with a methyl group, is the most extensively studied epigenetic mark. Histone modifications are also common epigenetic marks that take place through the addition of a chemical group to the tail of the histone. Some of these chemical additions include acetylation, methylation, phosphorylation, ubiquitination, and biotinylation. Gene expression is modified by the addition of these small molecules by changing the chromatin structure at the site of the histone modification. Chromatin remodeling is a more dynamic modification of chromatin architecture using an ATP-dependent protein complex or combined modifications of histone tails by multiple

histone modifying enzymes. In recent years, microRNA has also been considered an epigenetic mark because microRNA affects gene expression without altering base pairs. Interestingly microRNA can directly modulate other epigenetic marks, and conversely the expression of microRNA can be controlled by other epigenetic phenomena.[3] In this chapter, we will discuss how nutrients, bioactive food components, and our diet modify these epigenetic marks during our lifetime and influence our health.

DNA METHYLATION

DNA methylation is a major epigenetic mark in eukaryotic cells, which entails the addition of a methyl group to the carbon 5 position of cytosine at cytosine–guanine dinucleotide (CpG) residues, thereby forming 5-methylcytosine (Figure 20-1). 5-Methylcytosine is present in an estimated 4%–6% of the cytosine bases within a human genome, depending on cell type. The human genome contains about 30 million CpG dinucleotides that exist in a methylated or unmethylated state. This dinucleotide can be found in small genomic regions (200 bp to a few kb) known as CpG islands that are located in the proximal promoter regions of 75% of human genes. CpG islands at the promoters of housekeeping or developmental genes are mostly unmethylated, whereas nonregulatory CpGs distributed elsewhere in the genome such as heterochromatin regions are fundamentally methylated. The methylation status of regions within a gene-body such as in exons or introns is also important in regulation of transcription.[4] The DNA methylation reaction is catalyzed by DNA methyltransferases (Dnmts).[5] There are several types of Dnmts, each having a unique function. Dnmt1 is a maintenance Dnmt that copies the methylation pattern of the original DNA strand onto the daughter strands after DNA replication, whereas Dnmt 3a and Dnmt 3b are de novo Dnmts and are responsible for methylation at new cytosine sites. Knockout mutations in any of the three mouse genes that encode Dnmts are lethal.[6] *S*-adenosylmethionine (SAM) is the unique methyl donor to this DNA methylation reaction and after donating a methyl group SAM turns into *S*-adenosylhomocysteine (SAH), which is an inhibitor of DNA methyltransferases.[3]

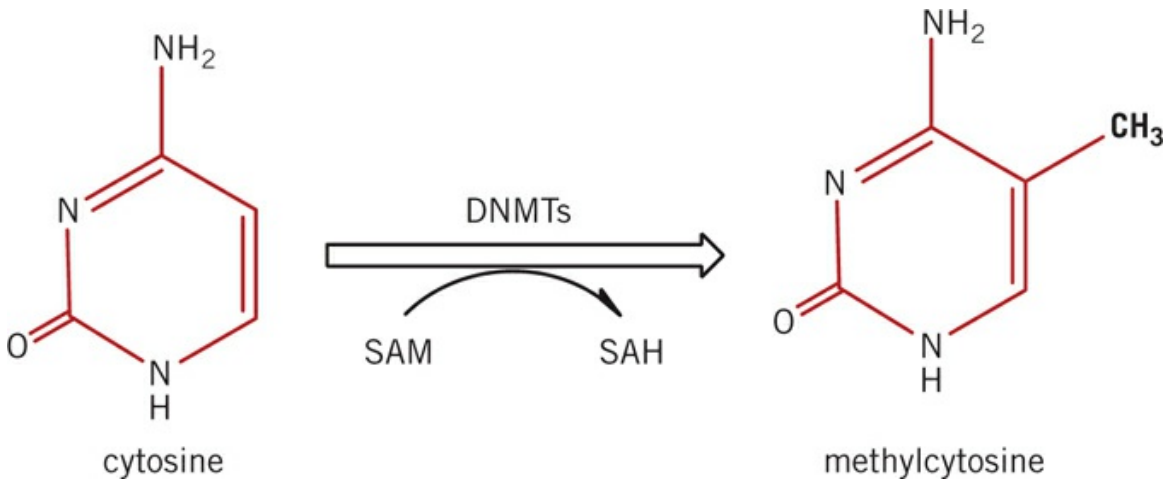


Figure 20-1 DNA methylation: DNA methylation is the addition of a methyl group to the carbon 5 position of cytosine at cytosine–guanine dinucleotide (CpG) residues, thereby forming 5-methylcytosine. The DNA methylation reaction is catalyzed by DNA methyltransferases (DNMTs). *S*-adenosylmethionine (SAM) is the unique methyl donor to this DNA methylation reaction and after donating a methyl group SAM turns to *S*-adenosylhomocysteine (SAH) which is an inhibitor of DNA methyltransferases.

In general, hypermethylation of a specific site of a gene is said to repress the gene by compacting nucleosome structure and recruiting methyl-binding proteins that interfere with the binding of transcription factors and RNA polymerases, whereas hypomethylation activates the gene with the opposite mechanism. However, this is also gene and site specific. Depending on the gene or location of DNA methylation, hypermethylation may increase and hypomethylation may decrease transcription, even though the mechanism is not yet clear.[3]

Compared with DNA methylation reactions, the DNA demethylation process has not been well delineated, even though DNA demethylation is critical during certain cellular processes such as embryonic development and stem cell differentiation. Among several candidate mechanisms, a new epigenetic mark, 5-hydroxymethylcytosine, has been highlighted as a player in the removal of methyl groups from cytosine bases. 5-Hydroxymethylcytosine is as an intermediate in the removal of the methyl group from 5-methylcytosine, returning the cytosine to its unmodified form. The ten-eleven translocation family of proteins are responsible for the hydroxylation of 5-methylcytosine to 5-hydroxymethylcytosine.[7] The

potential regulatory role of 5-hydroxymethylcytosine in gene transcription is somewhat different from that of methylcytosine, in that the affinity of methyl-binding proteins is much lower for 5-hydroxymethylcytosine compared to 5-methylcytosine. Less than 1% of cytosines are hydroxymethylated in mammalian DNA, and the level varies by tissue with the central nervous system having the highest amount. Embryonic stem cells also have high levels of 5-hydroxymethylcytosine, but these levels decrease during differentiation.[8]

DNA METHYLATION DURING OUR LIFETIME

Role of DNA Methylation During Embryonic Development

During embryonic development, each cell secures cell-specific patterns of DNA methylation in a process called DNA methylation reprogramming.[9] On fertilization, methylation patterns from parents are completely erased and newly established through de novo methylation during implantation. These dynamic processes are vital for the fertilization and differentiation of the embryo as well as for establishing the gene expression profiles that characterize each cell. During this reprogramming period, the intrauterine nutritional environment, which is mainly maternally derived, can affect this dramatic epigenetic reprograming, thereby affecting fetal development and even later life health status.[9,10]

Genomic imprinting is a type of DNA methylation by which a gene can be expressed in a parent-of-origin specific manner and plays an essential role in embryonic development and growth. The DNA sequence of the two parental alleles of imprinted genes is identical, whereas the DNA methylation status is different between the two parental alleles. An allele from one parent is repressed by hypermethylation of a specific differentially methylated region (DMR), wheras the other allele from the other parent is active by hypomethylation of the DMR.[11] Currently, approximately 100 imprinted genes have been described in mammals.[12] During the DNA methylation reprogramming period of other regular genes, the parental methylation status in imprinted genes maintain their imprinting status, whereas during germ cell development the parent-specific imprints are erased and new imprints are established. The parental allele-specific DNA methylation pattern is inherited to daughter cells and should be reset in each generation to establish parental specific imprints. The clinical phenotypes of imprinting disorders are diverse but are mainly involved in development and growth, such as Prader–Willi syndrome and

Angelman syndrome.[13] Alteration of imprinting status, as would occur with a loss of imprinting, may cause cancer in the later life.[14] Loss of imprinting of insulin-like growth factor 2 (*IGF2*) is a common molecular event in cancer, especially colorectal cancer, and typically activates the maternally silenced allele.[15]

Impact of Aging on DNA Methylation

During our lifetime, DNA methylation can be changed. Aging is known to affect DNA methylation in a complex fashion. Total 5-methylcytosine in DNA is prone to decrease with aging leading to global hypomethylation in most vertebrate tissues, whereas specific promoter regions tend to undergo a paradoxical hypermethylation in many genes.[10,16,17] We still do not know if these changes are genetically programmed or randomly acquired by environmental factors. Additionally, we do not know whether they are causes or results of the aging process in itself.[2] The most plausible mechanism is that decreased expression of the maintenance *Dnmt1* underlies reduced global hypomethylation, whereas increased expression of de novo *Dnmt3a* and *3b* mediates promoter hypermethylation. Declining levels of sex hormones and diet changes that occur with aging may also contribute to this age-associated DNA methylation changes. In one study, estrogen replacement therapy administered to postmenopausal women increased global DNA methylation in their peripheral mononuclear cells, giving strength to the theory that changes in sex hormones may have an epigenetic effect.[18]

A possible epigenetic connection between aging and cancer has been suggested because global DNA hypomethylation, gene-specific DNA hypermethylation, and altered expression of Dnmts are found in both aging and cancer. This supports the hypothesis that age-associated changes in DNA methylation patterns may provide an epigenetic milieu to develop age- associated diseases such as cancer. Nowadays, the study of DNA methylation has extended to other research fields outside of cancer including neurocognitive diseases,[19] cardiovascular diseases,[20] immune diseases,[21] obesity,[22] and even behavioral diseases[23], due to critical importance of DNA methylationin altering gene expression. It also suggests that we may delay the onset or progress of those diseases by modifying epigenetic mechanisms because epigenetic phenomena can be modulated by environmental factors like nutrition, in contrast to genetic phenomena such as polymorphisms and mutations that are irreversible.

NUTRITIONAL INFLUENCE ON DNA METHYLATION

One-Carbon Metabolism

One-carbon metabolism is a biochemical network that ultimately delivers methyl groups to many biological reactions including DNA methylation (Fig. 20-2). The water-soluble B-vitamins, folate, B_{12}, B_6, and B_2, act as coenzymes in one-carbon metabolism. Folate accepts a methyl group from serine and delivers the methyl group to homocysteine to reform methionine. Methionine, which is derived either from the diet or homocysteine remethylation, turns to SAM, the universal methyl donor to biological methylation reactions including DNA methylation. After donating a methyl group SAM turns to SAH, which is further converted to homocysteine. Importantly, SAH is an inhibitor of methyltransferases including Dnmts. Homocysteine can be remethylated by folate coenzymes as described above or by betaine, which is from the diet or converted from choline. Homocysteine can either be exported directly to blood or further metabolized to cystathionine and cysteine that ultimately form glutathione and taurine.[3] Because so many factors in one-carbon metabolism can be derived from the diet, deficiency or supplementation of those nutrients has a potential to alter DNA methylation by changing the status of SAM and SAH.

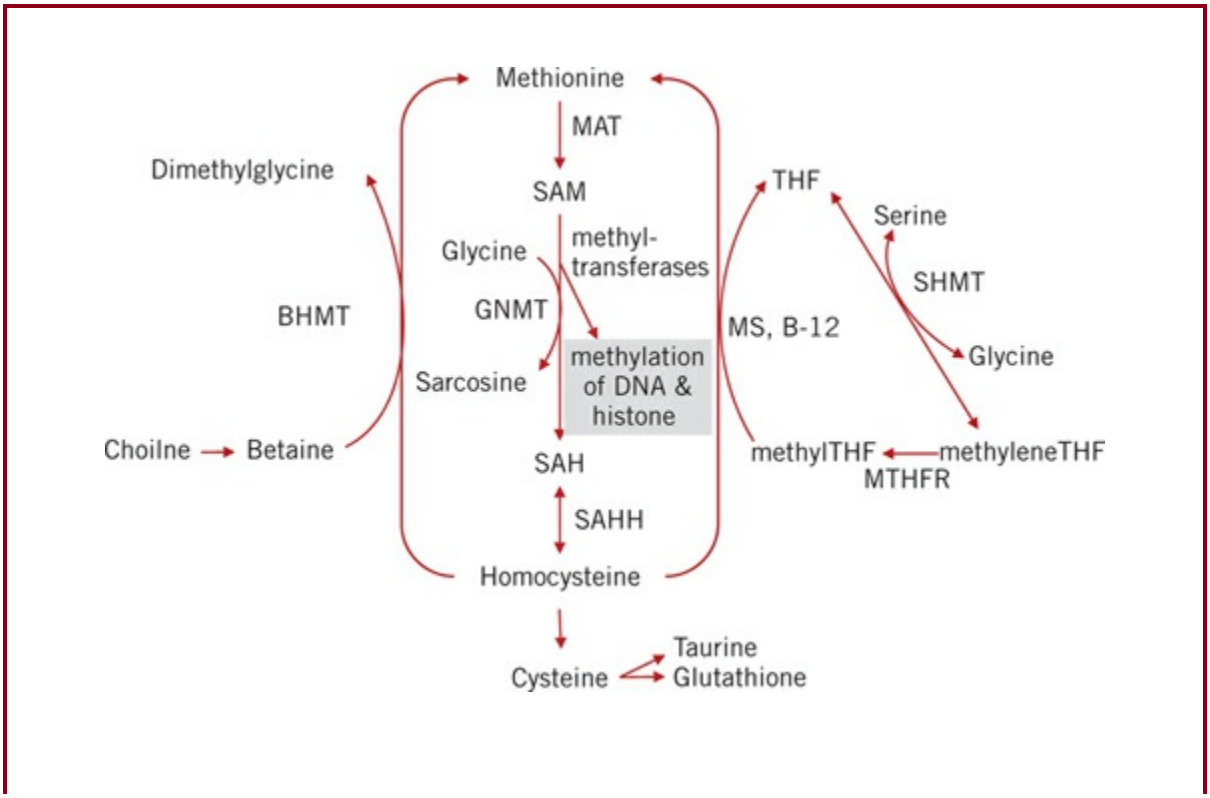


Figure 20-2 One-carbon metabolism: One-carbon metabolism is a biochemical network that ultimately delivers methyl groups to many biological reactions including DNA methylation and histone methylation. B-vitamins act as coenzymes and methionine, choline, betaine, and serine are methyl donors in one-carbon metabolism. BHMT; betaine homocysteine methyltransferase, GNMT; glycine N-methyltransferase, MS; methionine synthase, MTHFR; methylenetetrahydrofolate reductase, SAHH; *S*-adenosylhomocysteine hydrolase, SHMT; serine hydroxymethyltransferase

Folate is the most extensively studied nutrient in regard to DNA methylation. Multiple forms of folate coenzymes are involved in the delivery of methyl groups. Folate depletion decreases and folate supplementation increases global DNA methylation in animal studies, especially when aging is associated with these changes in folate.[24,25] However, the same results have been difficult to reproduce in human studies[26–28] because folate is not the sole determinant of DNA methylation in free living individuals who can get other methyl donor nutrients from the diet. Additionally, humans are exposed to various other environmental factors that may influence DNA methylation compared to animal studies in which animals are under the same environments and same dietary components (except folate) for the study duration.

Maternal folate deficiency has long been known to cause neural tube defects in neonates. Abnormal establishment of DNA methylation patterns from a folate deficiency is one of the most plausible mechanisms for this link between folate and neural tube defects, because low methyl availability induced by a folate deficiency is expected to disturb the DNA methylation reprogramming that is critical for embryonic development.[29] This has been previously shown when assessing the methylation status of particular genes in relation to folic acid intake. In a transgenerational study, periconceptional maternal folic acid supplementation was associated with changes in methylation at a DMR of *IGF2* in children between 12 and 18 months of age.[30] *IGF2* is an imprinting gene in which the methylated allele at the DMR (imprinted allele) is repressed. Children of mothers who took folic acid supplementation had a 4.5% higher methylation at the *IGF2* DMR than control children ($P = 0.014$). This result indicates that periconceptional folic acid supplementation is associated with the imprinting level of *IGF2* in the child, which may affect intrauterine programming of growth and development as well as later life health outcomes.

Deficiency of vitamin B_{12}, a water-soluble B-vitamin, is known to decrease DNA methylation in the rodent colon.[31] Vitamin B_{12} is a coenzyme for methionine synthase that catalyzes the remethylation of homocysteine to form methionine (Figure 20-2). Choline, a water-soluble essential nutrient, is also known to alter DNA methylation after converting to betaine (Figure 20-2). Maternal choline availability is essential for fetal neurogenesis, such as hippocampal development, and memory function throughout the life of the offspring. In a mouse study, dietary choline deficiency altered global and gene-specific DNA methylation in the developing hippocampus of mouse fetal brains, suggesting that maternal choline deficiency during the embryonic period alters fetal brain development through a DNA methylation mechanism.[32] Similarly, prolonged intake of a diet deficient in sources of methyl groups such as methionine, choline, folate, and vitamin B_{12} rapidly induce profound global hepatic DNA hypomethylation in a rodent model, which may contribute to the development of liver cancer.[33,34]

Other nutrients can also modify one-carbon metabolism to alter DNA methylation. Retinoic acid, which controls numerous physiological processes through the retinoid X receptor and retinoic acid receptor (RAR), is also known to alter DNA methylation by increasing abundance and activity of glycine *N*-methyltransferase, which in turn regulates SAM levels by catalyzing the conversion from SAM to SAH[35] (Figure 20-2). Retinoic acid is also known to regulate the expression and enzyme activity of Dnmts.[36] Genome-wide DNA methylation assays demonstrated 166 differentially methylated CpG sites in retinoic acid-treated human embryonic stem cells compared to control cells.[37] Interestingly, another DNA methylation array demonstrated that after retinoic acid treatment 402 gene promoters became demethylated and 88 genes were hypermethylated in neuroblastoma cells.[38] These types of epigenomic studies can demonstrate the effect of nutrients on genome-wide DNA methylation status, but it still needs to be investigated how the nutrient targets-specific genes to be methylated or demethylated and which changes actually have a health effect instead of just occurring as a concomitant phenomenon.

Vitamin D is now considered to play an important role in a wide range of body functions outside of bone health. Vitamin D has taken the forefront in cancer chemoprevention due to the role of the vitamin D receptor, a nuclear receptor that regulates the expression of many genes in cell cycle regulation and differentiation.[39] Recent evidence demonstrates that vitamin D can also modify gene expression through DNA methylation

in cancer cells. Incubation of breast cancer cells with vitamin D_3 reduces the aberrant hypermethylation and restores the expression of *RARβ2*,[40] phosphatase and tensin homologue tumor suppressor gene (*PTEN*),[41] and cadherin-1 gene (*CDH1*) encoding for E-cadherin.[42] These studies suggest that vitamin D may affect gene expression through a DNA methylation mechanism other than the vitamin D receptor that mediates most of biological functions of vitamin D.

DNA methylation may also be affected by periconceptional fasting. Blood DNA methylation status of the *IGF2* gene was assessed in individuals almost 60 years of age who had been exposed to famine periconceptionally during the Nazi embargo of the Western Netherlands near the end of World War II. The methylation status of the same region of the *IGF2* gene was also assessed in participants' same-sex siblings who were not exposed to the famine. Four CpG sites out of five investigated were significantly less methylated among periconceptionally exposed individuals compared with their siblings.[43] This study indicates that maternal undernutrition during the early development period can affect the establishment and maintenance of DNA methylation status in the offspring.

Bioactive Food Components and DNA Methylation

Bioactive food components can be defined as food components that are not essential but helpful for human health by enhancing one or more biological processes that maintain our health. Interestingly, these compounds can alter DNA methylation by directly affecting Dnmts. Evidence indicates that certain bioactive food components, including (–)epigallocatechin-3-gallate (EGCG) in tea, genistein in soybeans, isothiocyanates in cruciferous vegetables, or curcumin in curry spice might inhibit the development of cancer by reducing DNA hypermethylation status in critical genes associated with cancer, such as *p16*, *RARβ*, or *O*-6-methylguanine-DNA methyltransferase (*MGMT*).[44] In a human study, healthy premenopausal women demonstrated that a daily supplementation with isoflavones induced dose- specific changes in methylation of *RARβ2* and cyclin D2 (*CCND2*) measured from the intraductal breast specimens, which were also correlated with serum genistein levels.[45]

Various catechol-containing dietary polyphenols such as bioflavonoids, tea catechins, and coffee polyphenols have also been suggested to indirectly inhibit DNA methylation. The catechol-*O*-methyltransferase-mediated methylation of large amounts of dietary

catechols not only reduces the cellular pools of SAM but also forms an equimolar amount of SAH, an inhibitor of DNA methyltransferase.[46]

HISTONE MODIFICATIONS

DNA is compacted by tightly wrapping approximately 147 base pairs around histone proteins that were previously regarded as an inert packing material for eukaryotic nuclear DNA, forming a nucleosome, the fundamental unit of chromatin. Each nucleosome consists of two pairs of four core histones: H2A, H2B, H3, and H4 (Figure 20-3). Each of the histone proteins consists of a structured core and an unstructured tail domain. Many different types of posttranslational modifications to the N-terminal histone tails can occur, including acetylation, methylation, phosphorylation, ubiquitination, ADP-ribosylation, and biotinylation. These modifications are thought to contribute to the control of gene expression by either altering the ionic charge of the histone tail, modifying chromatin compaction, or by functioning as a binding platform for other proteins. As an essential component of cellular epigenetic machinery, histone modifications are known to influence biological processes including embryonic development and aging.[47] Even though it is not as extensively studied in embryonic development such as DNA methylation, it has been regarded that histone marks are also inherited and the alterations in histone mark control can influence the development process and disease susceptibility in later life. Nutrients and bioactive food components can change histone modifications and thereby alter physiologic and pathologic processes that are mediated by histone marks.

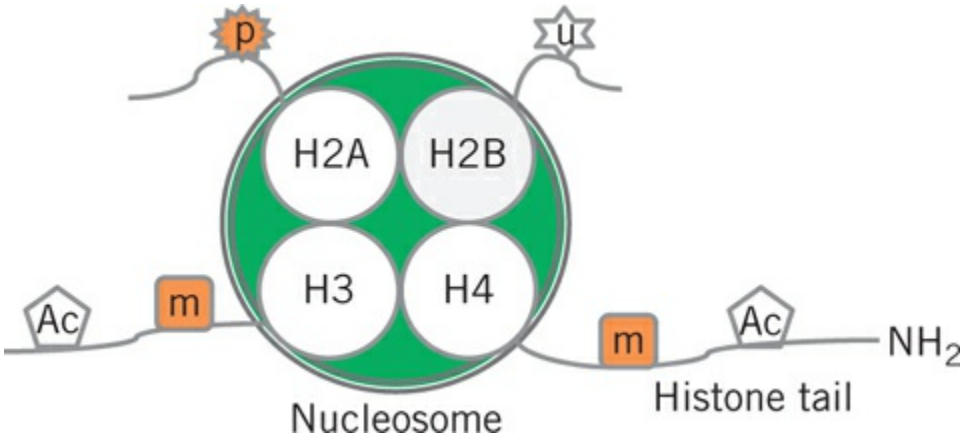


Figure 20-3 Histone modifications: Each nucleosome consists of two

pairs of four core histones (H2A, H2B, H3, and H4) and 147 bp of DNA wrapping around the histone. Each of the histone proteins consists of a core and a tail domain. Many different types of posttranslational modifications to the N-terminal histone tails can occur, such as acetylation (Ac), methylation (m), phosphorylation (p), and ubiquitination (u).

HISTONE ACETYLATION AND BIOACTIVE FOOD COMPONENTS

Histone acetylation is an epigenetic modification caused by the acetylation of lysine residues at the histone tail to alter local DNA compaction, leading to changes in gene expression. The histone acetylation status is maintained by the balance between histone acetyltransferases (HATs) and histone deacetylases (HDACs) (Figure 20-4). HATs are the class of enzymes that transfer acetyl groups onto lysine residues within the histone tail. Through this action the positive charge of the lysine is neutralized. In contrast to HATs, HDACs catalyze the removal of acetyl groups from lysines and restore the positive charge on the histone tail. HATs are generally classified as transcriptional activators, whereas HDACs are repressors.[48] Numerous studies have investigated the effects of bioactive food components on HDAC and HAT. Because the inhibition of HDAC can derepress epigenetically silenced genes in cancer cells, it has been investigated whether certain bioactive food components can be utilized for the prevention and treatment of cancer as HDAC inhibitors.[49]

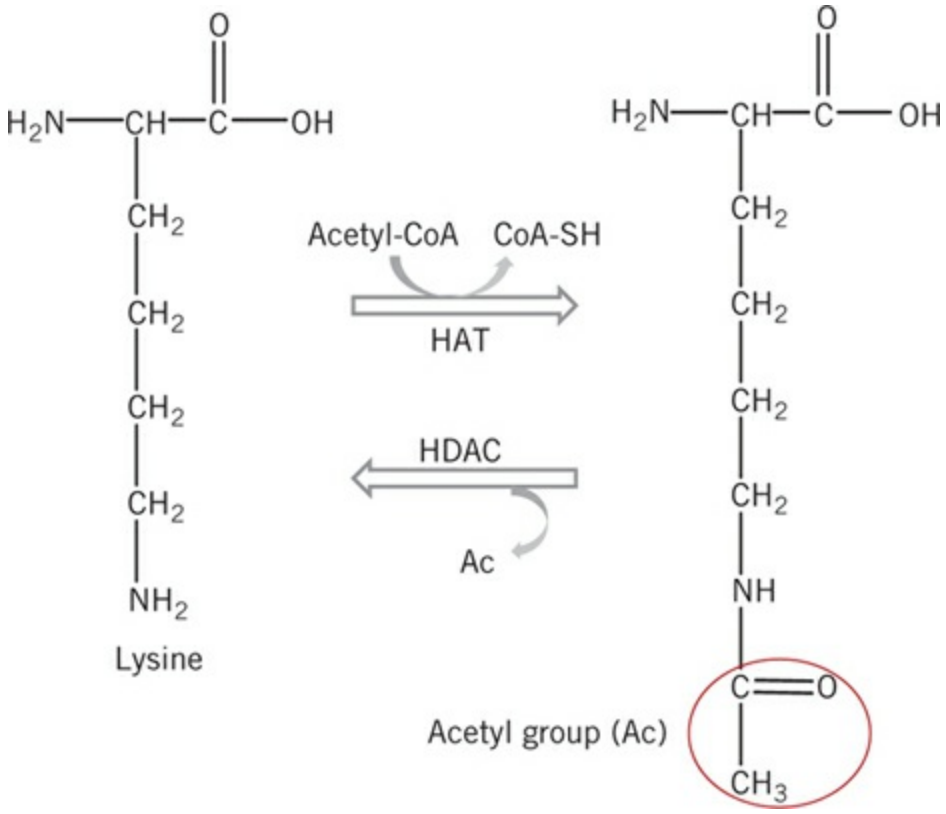


Figure 20-4 Histone acetylation: Histone acetylation is an epigenetic modification that acetylates lysine residues at the histone tail. The histone acetylation status is maintained by the balance between histone acetyltransferases (HAT) and histone deacetylases (HDAC).

Butyrate is a short-chain fatty acid generated during the fermentation of dietary fiber in colon by anaerobic bacteria.[50] Butyrate enters into the active site of HDAC enzymes and inhibits the activity, thereby increasing histone H3 acetylation. Butyrate stimulation may provide an effective method for reprogramming various human adult somatic cells and enhancing derivation of induced pluripotent stem cells.[51] After a report in 1977 that the addition of sodium *n*-butyrate to tissue culture media increases histone acetylation in cultured cancer cells,[52] the health effects of butyrate have been extensively studied as an HDAC inhibitor. This is especially true in cancer research because HDACs have been a promising target in the discovery of new cancer chemotherapeutic agents. Butyrate-induced alterations in histone acetylation have been associated with several processes in both initiation and progression of cancer, including cellular differentiation, cell cycle arrest, apoptosis, and inhibition of

invasion and metastasis in cancer.[53]

Sulforaphane, an isothiocyanate in cruciferous vegetables such as broccoli and broccoli sprouts that has anticancer properties, is also known to have HDAC inhibitory effects. Sulforaphane has been reported to inhibit the growth and proliferation of cancer cells by downregulating HDAC enzymes in different types of human cancer cell lines. In some studies, sulforaphane increased both global and gene-specific histone acetylation around the promoter regions of genes regulating the cell cycle and apoptosis such as *p21* and *Bax*.[54–56] In a human study, a dose of 68 g (1 cup) of broccoli sprouts effectively inhibited the HDAC activity in peripheral blood mononuclear cells 3–6 hours after consumption, with a concomitant induction of histone H3 and H4 acetylation.[57]

Organosulfur compounds in garlic, diallyl sulfide and *S*-allyl mercaptocysteine, also have demonstrated HDAC inhibitory effects. Both compounds are metabolized to allyl mercaptan that is more effective than its precursors at inhibiting HDAC activity.[58] This is due to allyl mercaptan being a good fit for the enzyme's active site. In human colon cancer cells, the HDAC inhibitory effect of allyl mercaptan was accompanied by a rapid, sustained accumulation of acetylated histones. Within 4 hours after the exposure to allyl mercaptan, histone H3 acetylation was increased around the *p21* gene promoter, resulting in elevated p21 protein expression and cell cycle arrest in human colon cancer cells.[58] An animal study demonstrated that diallyl sulfide increases histone H3 and H4 acetylation in colon cells isolated from rats, which was associated with alterations in the expression of a subset of genes.[59]

Resveratrol, a natural polyphenol abundantly found in grape skins and red wine, has diverse biochemical and physiological properties, including antiinflammatory, antioxidation, and antiproliferation effects.[60] Resveratrol is a potent activator of Sirt1, a regulator of cellular lifespan in a yeast model of aging through the reduction of acetylation at histone H4 lysine 16. This change in histone acetylation results in compromised transcriptional silencing at specific loci throughout the genome.[61] In a cultured cell study, the activation of Sirt1 by resveratrol negatively regulated the expression of the antiapoptotic protein, Survivin, by reducing histone H3 lysine 9 acetylation within the promoter of its gene].[62]

Curcumin, a yellow pigment present in the spice turmeric (*Curcuma longa*), has been shown to have anti-inflammatory, antioxidant, and anticancer functions.[63] In cells, curcumin promotes proteasome-dependent degradation of HAT p300 and the closely related cAMP-reponse element

binding protein (CREB). In addition to inducing p300 degradation, curcumin also inhibits the acetyltransferase activity of p300.[64] Curcumin-mediated HAT inhibitory activity was in parallel with decreased histone H3 and H4 acetylation in adult neural-derived stem cells.[65] Because curcumin-mediated epigenetic changes can increase apoptosis in cancer cells and promote neuronal differentiation in stem cells, curcumin might have a therapeutic potential in cancer and neurodegenerative diseases. Additionally, it is also reported that curcumin may prevent diabetes-associated abnormalities in the kidneys by inhibiting p300 and nuclear factor kappa B (NF-κB).[66]

HISTONE METHYLATION AND ONE-CARBON METABOLISM

Histone methylation occurs at the lysine and arginine residues of histone tails. Histone methylation is controlled by histone methyltransferases and histone demethylases. Histone methyltransferases use SAM as the methyl donor and are inhibited by SAH, similar to the Dnmts as discussed above.

Prolonged intake of a diet low in methyl donor nutrients such as methionine, choline, folate, and vitamin B_{12} has been known to induce profound global hepatic DNA hypomethylation and spontaneous liver tumors.[33,34] In this model, liver tumors were significantly decreased in histone H4 lysine 20 trimethylation (44%) and increased in histone H3 lysine 9 trimethylation (40%). Furthermore, the expression of suppressor of variegation 4–20 homolog 2 (Suv4–20h2), a histone methyltransferase that catalyzes histone H4 lysine 20 methylation, was decreased, whereas the expression of suppressor of variegation 3–9 homolog 1 (Suv3–9h1) that catalyzes histone H3 lysine 9 methylation was increased.[67] This study indicates that the development of liver tumors by a methyl-deficient diet may be through the alterations of histone methylation. It also supports a hypothesis that epigenetic reprogramming of the cell caused by the diet is an important prerequisite for the development of cancer.

In a transgenerational animal study using C57BL/6 mice choline deficiency during gestational days 12–17 altered the methylation status of histone H3 in E17 fetal hippocampi.[68] In the ventricular and subventricular zones, monomethylation at histone H3 lysine 9 was significantly decreased by 25%, and in the pyramidal layer dimethylation at histone H3 lysine 9 was also significantly decreased by 37%. This suggests that a maternal deficiency of the methyl donating nutrient choline can influence the

histone methylation status in the offspring's brain.

One of the most interesting observations is that histone methylation is associated with obesity. The histone H3 lysine 9-specific demethylase JmjC domain-containing histone demethylation protein 2A (Jhdm2a, also known as Jmjd1a and Kdm3a) directly regulates genes associated with lipid and energy metabolism such as peroxisome proliferator-activated receptor alpha (*PPARα*) and uncoupling protein 1 (*Ucp1*). The loss of Jhdm2a function in mice results in obesity and hyperlipidemia, along with decreased levels of dimethylation of histone H3 lysine 9 at the PPAR responsive element of the *Ucp1* gene, indicating that histone H3 lysine 9 methylation is important in regulating the expression of metabolic genes.[69] Further studies are needed to understand the role of histone methylation in obesity.

HISTONE BIOTINYLATION

Biotin, an essential water-soluble B vitamin, has been known to modify tails of histone H2A, H3, and H4 through a covalent attachment of biotin to lysine residues catalyzed by biotinidase and holocarboxylase synthase.[70] Even though debiotinylases have not been characterized, histone biotinylation is a reversible process. Although it is rare, biotinylation is a natural modification of human histones. Biotinylated histones are enriched in repeat regions and repressed loci.[71] Biotinylations at histone H4 lysine 8 and lysine 12 have been associated with heterochromatin structures, gene silencing, mitotic condensation of chromatin, and DNA repair.[72] A deficiency in biotin may have profound effects on chromatin structure,[73] but there are many unanswered questions in histone biotinylation, a modification of the histone directly by a nutrient.

CHROMATIN REMODELING

At the chromatin level, which is above the level of DNA and histones, regulation of gene transcription can also occur. By tightly packing the nucleosome to form compact chromatin structure, transcription factors and RNA polymerases can no longer transcribe DNA in those regions and the gene becomes silenced. This compacted chromatin will form not only to prevent transcription of a specific target gene but also to increase integrity of the genome. The mechanism by which chromatin are remodeled to either silence or activate a gene is either through an ATP-dependent

protein complex, or through combined covalent modifications of histone tails by several histone modifying proteins, such as the polycomb repressive complex (PRC).

ATP-DEPENDENT CHROMATIN REMODELING

The SWItch/Sucrose NonFermentable (SWI/SNF) complex is a common ATP-dependent chromatin remodeling protein complex in eukaryotic cells. The ATP-dependent chromatin remodeling mechanism actively alters chromatin structures during gene regulation.[74] This remodeling complex binds to the nucleosome and results in repositioning of that nucleosome.[75] Transcription of targeted genes can be increased or decreased depending on the open or compact chromatin structure induced by the nucleosome repositioning. The SWI/SNF complex plays an essential role in cellular processes, including proliferation, differentiation, and DNA repair and replication[76] as well as in immune responses[77] and carcinogenesis.[78] An animal study demonstrated that the disruption of a novel bromodomain-containing protein 2(Brd2), a transcriptional coactivator/corepressor with SWI/SNF-like functions that regulates chromatin, causes severe obesity, which is accompanied with pancreatic islet expansion, hyperinsulinemia, hepatosteatosis, and increased levels of proinflammatory cytokines.[79] This study suggests that the SWI/SNF chromatin remodeling mechanism may play a critical role in the inhibition of obesity.

POLYCOMB REPRESSIVE COMPLEX

PRC proteins are involved in gene silencing through chromatin remodeling. The formation of the multiple protein complex PRC represses transcription by maintaining the compact chromatin state through particular repressive histone marks and DNA hypermethylation. The composition of the PRC varies between organisms, cell types, developmental stages, and even in some disease states.[80] These complexes are recruited to regions of the DNA, so-called Polycomb response elements, where they regulate gene transcription through histone modifications. In humans, the embryonic ectoderm development enhancer of zeste homolog 2 (EED-EZH2) complex, a component of PRC2, trimethylates histone H3 at lysine 27 and then facilitates binding of PRC1. Recruitment of PRC1 can then lead to transcriptional repression through ubiquitination of histone H2A lysine 119. The addition of the ubiquitin

mark ultimately stops gene transcription by inhibiting mRNA elongation. PRC1 can also be recruited in a PRC2 independent manner.[81] Through this multistep protein recruitment pathway, the gene can be effectively repressed.

PRC is important during embryonic stem cell differentiation. During differentiation, the binding of PRC2, which mediates trimethylation at histone H3 lysine 27, is lost in transcriptionally active developmental genes. Retinoic acid is also involved in differentiation of embryonic stem cells and various types of cultured cancer cells seemingly through histone modifications. Interestingly, a global decrease in histone H3 lysine 27 trimethylation was observed three days after differentiation of mouse embryonic stem cells induced by retinoic acid treatment. The global levels of EZH2, histone H3 lysine 27 methyltransferase, and PRC2 protein also decreased with retinoic acid treatment.[82]

Other research has also focused on the link between dietary components, cancer development, and chromatin remodeling. B lymphoma Mo-MLV insertion region 1 homolog (Bmi-1), a PRC1 protein, is overexpressed in some human cancers including cancers of breast, prostate, lung, and ovary.[83] In a cultured cell study, treatment of SCC-13 cells, a human epidermal squamous carcinoma cell line, with the tea polyphenol EGCG reduced Bmi-1 and EZH2 levels. This was accompanied with diminished cell survival and a global reduction in trimethylation at histone H3 lysine 27.[84] Changes in PRC protein expression were in parallel with reduced expression of proteins that enhance progression through the cell cycle. These findings suggest that EGCG reduces skin cancer cell survival through PRC-mediated epigenetic mechanisms.

MICRORNAS

MicroRNAs are small noncoding RNAs of 20–24 nucleotides in length that play important roles in virtually all cellular processes including development, metabolism, and homeostasis. The primary mechanism by which microRNAs silence target genes is by inhibiting translation of mRNA either by directly binding the target sequence or through the formation of the RNA-induced silencing complex, which is responsible for site-specific cleavage of mRNA and facilitated mRNA degradation.[85] Certain microRNAs are encoded within introns of protein coding genes and are transcribed along with the primary transcript, whereas other microRNAs are transcribed from their own gene, the primary transcript

being called a pri-microRNA, which is then processed into mature microRNA.[86] A single microRNA may have multiple target mRNA and each mRNA may also be regulated by several microRNAs. There is some evidence of feedback between microRNA expression and other epigenetic phenomena.[87] For example, DNA methylation and histone modifications can regulate the transcription of microRNAs, and some microRNAs can alter the expression of Dnmt3A and Dnmt3B, and Polycomb group genes.[88] MicroRNA expression has also been linked to cancer, and aberrant microRNA expression caused by changes in DNA methylation has been discovered in cancer cells.[89] A growing body of evidence indicates that our diet, nutrients, and bioactive components can affect microRNA production. A methyl-deficient diet, which induces aberrant hepatic DNA methylation and histone methylation,[33,67] also changed the expression of microRNA genes, including *miR-34a*, *miR-127*, *miR-200b*, and *miR-16a*, which are involved in the regulation of cell proliferation, apoptosis, cell-to-cell connection, and the epithelial-mesenchymal transition in rat liver.[90] Similar to methyl deficiency, folate deficiency markedly increased *miR-222* expression in human lymphoblastoid cells.[91] In an animal study, Göttingen minipigs fed a high-cholesterol diet showed significantly lower *miR-122* levels as well as an increase in body weight and cholesterol levels compared with those fed the control diet, indicating the association of this microRNA with obesity.[92]

One interesting hypothesis in cancer therapy is that bioactive food components may reduce carcinogenesis through microRNAs.[93] In a cultured cell study using prostate cancer cell line genistein could alter the expression of oncogenic *miR-151* as well as its target genes involved in the progression and metastasis of prostate cancer. Inhibition of *miR-151* in prostate cancer cells by genistein also significantly inhibited cell migration and invasion.[94] In leukemic cells, curcumin upregulates the expression of *miR-15a* and *miR-16-1* and downregulates the expression of Wilms tumor 1 gene (*WT1*), indicating that the antiproliferative effect of curcumin might be through the upregulation of these microRNAs.[95] Additionally, in the human breast adenocarcinoma MCF-7 cell line curcumin reduced the expression of antiapoptotic B cell lymphoma 2 gene (*Bcl-2*) by decreasing the expression of *miR-15a* and *miR-16*.[96]

There is some evidence that exogenous microRNA from food may also have an effect on our cells. *MiR-168a* is abundant in rice, and is one of the most highly enriched exogenous microRNAs in the sera of Chinese subjects. Functional studies have demonstrated that *miR-168a* can bind to

the low-density lipoprotein receptor adapter protein 1 (*LDLRAP1*) mRNA, thereby inhibiting *LDLRAP1* expression in liver.[97] This study provides a new hypothesis that microRNA consumption through diet is a mechanism by which food can affect our phenotype.

CONCLUSION

Chromatin is a quite dynamic structure and has functions outside of being a packaging system for a long string of genomic DNA into the nucleus of each cell. It plays a critical role in controlling the accessibility of transcription factors to DNA. Modifications to the chromatin structure by DNA methylation, histone modifications, and chromatin remodeling can give rise a variety of epigenetic effects on gene expression. Because of its reversible characteristics, epigenetics is now considered an attractive field of nutritional intervention.

During our lifetime, nutrients, bioactive components, or diet can modify many physiologic and pathologic processes through epigenetic mechanisms. However, nutritional epigenetics is a field in its infancy and many questions remain to be answered. Specific time and dose of nutritional exposures and their efficacy on epigenetic regulations need to be examined. Numerous studies have suggested that perinatal nutrition is critical in epigenetic reprogramming of offspring, but few studies exist regarding the epigenetic effects of nutrition on other time periods of the lifecycle. This is particularly critical for the nutritional intervention in aging and age-associated diseases. Investigation into how epigenetic phenotypes acquired by nutrients can be inherited to the next generation independently from the genetic sequence as well as how nutrients select specific genes to be repressed or activated through epigenetic marks needs to be made.

It is not easy to delineate the precise effect of nutrients or bioactive food components on each epigenetic mark and their associations with physiologic and pathologic processes in our body, because the nutrient also interacts with genes, other nutrients, and other lifestyle factors. Furthermore, each epigenetic phenomenon also interacts with each other, adding to the complexity of the system. Nevertheless, we need to evaluate the epigenetic properties of more nutrients or bioactive food compounds to find better ones for our health. Embryonic development and aging are associated with substantial changes in epigenetic patterns and recent work is implicating epigenetic mechanisms in the etiology of pediatric developmental diseases and a majority of age-associated diseases.

Understanding the epigenetic roles of nutrients or bioactive food components in our health and development of these diseases will enable our ability to find a better way to maintain our health through nutritional modulation.

REFERENCES

1. Tammen SA, Friso S, Choi SW. Epigenetics: the link between nature and nurture. *Mol Aspects Med*. 2013;34(4):753–764.
2. Park LK, Friso S, Choi SW. Nutritional influences on epigenetics and age-related disease. *Proc Nutr Soc*. 2011:1–9.
3. Choi SW, Friso, S. Epigenetics: a New Bridge between Nutrition and Health. *Adv Nutr*. 2010;1:8–16.
4. Barrera V, Peinado MA. Evaluation of single CpG sites as proxies of CpG island methylation states at the genome scale. *Nucleic Acids Res*. 2012;40:11490–11498.
5. Zilberman D, Henikoff S. Genome-wide analysis of DNA methylation patterns. *Development*. 2007;134:3959–3965.
6. Goll MG, Bestor TH. Eukaryotic cytosine methyltransferases. *Annu Rev Biochem*. 2005;74:481–514.
7. Ito S, D'Alessio AC, Taranova OV, et al. Role of Tet proteins in 5mC to 5hmC conversion, ES-cell self-renewal and inner cell mass specification. *Nature.* 2010;466:1129–1133.
8. Globisch D, Munzel M, Muller M, et al. Tissue distribution of 5-hydroxymethylcytosine and search for active demethylation intermediates. *PLoS One*. 2010;5:e15367.
9. Reik W, Dean W, Walter J. Epigenetic reprogramming in mammalian development. *Science*. 2001;293:1089–1093.
10. Kim KC, Friso S, Choi SW. DNA methylation, an epigenetic mechanism connecting folate to healthy embryonic development and aging. *J Nutr Biochem*. 2009;20:917–926.
11. Ishida M, Moore GE. The role of imprinted genes in humans. *Mol Aspects Med.* 2013;34(4):826–840.
12. Bartolomei MS, Ferguson-Smith AC. Mammalian genomic imprinting. *Cold Spring Harb Perspect Biol*. 2011;3:a002592.
13. Buiting K. Prader-Willi syndrome and Angelman syndrome. *Am J Med Genet C Semin Med Genet*. 2010;154C:365–376.
14. Tian F, Tang Z, Song G, et al. Loss of imprinting of IGF2 correlates with hypomethylation of the H19 differentially methylated region in the tumor tissue of colorectal cancer patients. *Mol Med Report*. 2012;5:1536–1540.
15. Cheng YW, Idrees K, Shattock R, et al. Loss of imprinting and marked gene elevation are 2 forms of aberrant IGF2 expression in colorectal cancer. *Int J Cancer*. 2010;127:568–577.
16. Richardson B. Impact of aging on DNA methylation. *Ageing Res Rev*. 2003;2:245–261.
17. Thompson RF, Atzmon G, Gheorghe C, et al. Tissue-specific dysregulation of DNA methylation in aging. *Aging Cell*. 2010;9:506–518.
18. Friso S, Lamon-Fava S, Jang H, et al. Oestrogen replacement therapy reduces total plasma homocysteine and enhances genomic DNA methylation in postmenopausal women. *Br J Nutr*. 2007;97:617–621.

19. Qureshi IA, Mehler MF. Epigenetic mechanisms governing the process of neurodegeneration. *Mol Aspects Med*. 2013;34(4):875–882.
20. Udali S, Guarini P, Moruzzi S, et al. Cardiovascular epigenetics: From DNA methylation to microRNAs. *Mol Aspects Med*. 2013;34(4):883–901.
21. Kondilis-Mangum HD, Wade PA. Epigenetics and the adaptive immune response. *Mol Aspects Med*. 2013;34(4):813–825.
22. Milagro FI, Mansego ML, De Miguel C, Martinez JA. Dietary factors, epigenetic modifications and obesity outcomes: Progresses and perspectives. *Mol Aspects Med*. 2013;34(4):782–812.
23. Labonte B, Yerko V, Gross J, et al. Differential glucocorticoid receptor exon 1(B), 1(C), and 1(H) expression and methylation in suicide completers with a history of childhood abuse. *Biol Psychiatry*. 2012;72:41–48.
24. Keyes MK, Jang H, Mason JB, et al. Older age and dietary folate are determinants of genomic and p16-specific DNA methylation in mouse colon. *J Nutr*. 2007;137:1713–1717.
25. Choi SW, Friso S, Keyes MK, Mason JB. Folate supplementation increases genomic DNA methylation in the liver of elder rats. *Br J Nutr*. 2005;93:31–35.
26. Jacob RA, Gretz DM, Taylor PC, et al. Moderate folate depletion increases plasma homocysteine and decreases lymphocyte DNA methylation in postmenopausal women. *J Nutr*. 1998;128:1204–1212.
27. Pizzolo F, Blom HJ, Choi SW, et al. Folic Acid effects on s-adenosylmethionine, s-adenosylhomocysteine, and DNA methylation in patients with intermediate hyperhomocysteinemia. *J Am Coll Nutr*. 2011;30:11–18.
28. Crider KS, Quinlivan EP, Berry RJ, et al. Genomic DNA methylation changes in response to folic acid supplementation in a population-based intervention study among women of reproductive age. *PLoS One*. 2011;6:e28144.
29. Chang H, Zhang T, Zhang Z, et al. Tissue-specific distribution of aberrant DNA methylation associated with maternal low-folate status in human neural tube defects. *J Nutr Biochem*. 2011;22:1172–1177.
30. Steegers-Theunissen RP, Obermann-Borst SA, Kremer D, et al. Periconceptional maternal folic acid use of 400 microg per day is related to increased methylation of the IGF2 gene in the very young child. *PLoS One*. 2009;4:e7845.
31. Choi SW, Friso S, Ghandour H, et al. Vitamin B-12 deficiency induces anomalies of base substitution and methylation in the DNA of rat colonic epithelium. *J Nutr.* 2004;134:750–755.
32. Niculescu MD, Craciunescu CN, Zeisel SH. Dietary choline deficiency alters global and gene-specific DNA methylation in the developing hippocampus of mouse fetal brains. *Faseb J.* 2006;20:43–49.
33. Wainfan E, Dizik M, Stender M, Christman JK. Rapid appearance of hypomethylated DNA in livers of rats fed cancer-promoting, methyl-deficient diets. *Cancer Res.* 1989;49:4094–4097.
34. Pogribny IP, Ross SA, Wise C, et al. Irreversible global DNA hypomethylation as a key step in hepatocarcinogenesis induced by dietary methyl deficiency. *Mutat Res*. 2006;593:80–87.
35. Ozias MK, Schalinske KL. All-trans-retinoic acid rapidly induces glycine N-methyltransferase in a dose-dependent manner and reduces circulating methionine and homocysteine levels in rats. *J Nutr*. 2003;133:4090–4094.
36. Fazi F, Travaglini L, Carotti D, et al. Retinoic acid targets DNA-methyltransferases and histone deacetylases during APL blast differentiation in vitro and in vivo. *Oncogene*. 2005;24:1820–1830.
37. Cheong HS, Lee HC, Park BL, et al. Epigenetic modification of retinoic acid-

treated human embryonic stem cells. *BMB Rep*. 2010;43:830–835.
38. Das S, Foley N, Bryan K, et al. MicroRNA mediates DNA demethylation events triggered by retinoic acid during neuroblastoma cell differentiation. *Cancer Res*. 2010;70:7874–7881.
39. Peterlik M, Grant WB, Cross HS. Calcium, vitamin D and cancer. *Anticancer Res*. 2009;29:3687–3698.
40. Stefanska B, Rudnicka K, Bednarek A, Fabianowska-Majewska K. Hypomethylation and induction of retinoic acid receptor beta 2 by concurrent action of adenosine analogues and natural compounds in breast cancer cells. *Eur J Pharmacol*. 2010;638:47–53.
41. Stefanska B, Salame P, Bednarek A, Fabianowska-Majewska K. Comparative effects of retinoic acid, vitamin D and resveratrol alone and in combination with adenosine analogues on methylation and expression of phosphatase and tensin homologue tumour suppressor gene in breast cancer cells. *Br J Nutr*. 2012;107:781–790.
42. Lopes N, Carvalho J, Duraes C, et al. 1Alpha, 25-dihydroxyvitamin D3 induces de novo E-cadherin expression in triple-negative breast cancer cells by CDH1-promoter demethylation. *Anticancer Res*. 2012;32:249–257.
43. Heijmans BT, Tobi EW, Stein AD, et al. Persistent epigenetic differences associated with prenatal exposure to famine in humans. *Proc Natl Acad Sci U S A*. 2008;105:17046–17049.
44. Fang M, Chen D, Yang CS. Dietary polyphenols may affect DNA methylation. *J Nutr*. 2007;137:223S–228S.
45. Qin W, Zhu W, Shi H, et al. Soy isoflavones have an antiestrogenic effect and alter mammary promoter hypermethylation in healthy premenopausal women. *Nutr Cancer*. 2009;61:238–244.
46. Stefanska B, Karlic H, Varga F, et al. Epigenetic mechanisms in anti-cancer actions of bioactive food components--the implications in cancer prevention. *Br J Pharmacol*. 2012;167:279–297.
47. Greer EL, Shi Y. Histone methylation: a dynamic mark in health, disease and inheritance. *Nat Rev Genet*. 2012;13:343–357.
48. Shahbazian MD, Grunstein M. Functions of site-specific histone acetylation and deacetylation. *Annu Rev Biochem*. 2007;76:75–100.
49. Meeran SM, Ahmed A, Tollefsbol TO. Epigenetic targets of bioactive dietary components for cancer prevention and therapy. *Clin Epigenetics*. 2010;1:101–116.
50. Alvaro A, Sola R, Rosales R, et al. Gene expression analysis of a human enterocyte cell line reveals downregulation of cholesterol biosynthesis in response to short-chain fatty acids. *IUBMB Life*. 2008;60:757–764.
51. Mali P, Chou BK, Yen J, et al. Butyrate greatly enhances derivation of human induced pluripotent stem cells by promoting epigenetic remodeling and the expression of pluripotency-associated genes. *Stem Cells*. 2010;28:713–720.
52. Riggs MG, Whittaker RG, Neumann JR, Ingram VM. n-Butyrate causes histone modification in HeLa and Friend erythroleukaemia cells. *Nature*. 1977;268:462–464.
53. Davie JR. Inhibition of histone deacetylase activity by butyrate. *J Nutr*. 2003;133:2485S–2493S.
54. Myzak MC, Hardin K, Wang R, et al. Sulforaphane inhibits histone deacetylase activity in BPH-1, LnCaP and PC-3 prostate epithelial cells. *Carcinogenesis*. 2006;27:811–819.
55. Myzak MC, Karplus PA, Chung FL, Dashwood RH. A novel mechanism of chemoprotection by sulforaphane: inhibition of histone deacetylase. *Cancer Res*.

2004;64:5767–5774.
56. Pledgie-Tracy A, Sobolewski MD, Davidson NE. Sulforaphane induces cell type-specific apoptosis in human breast cancer cell lines. *Mol Cancer Ther*. 2007;6:1013–1021.
57. Myzak MC, Tong P, Dashwood WM, et al. Sulforaphane retards the growth of human PC-3 xenografts and inhibits HDAC activity in human subjects. *Exp Biol Med (Maywood)*. 2007;232:227–234.
58. Nian H, Delage B, Ho E, Dashwood RH. Modulation of histone deacetylase activity by dietary isothiocyanates and allyl sulfides: studies with sulforaphane and garlic organosulfur compounds. *Environ Mol Mutagen*. 2009;50:213–221.
59. Druesne-Pecollo N, Chaumontet C, Pagniez A, et al. In vivo treatment by diallyl disulfide increases histone acetylation in rat colonocytes. *Biochem Biophys Res Commun*. 2007;354:140–147.
60. Yu W, Fu YC, Wang W. Cellular and molecular effects of resveratrol in health and disease. *J Cell Biochem*. 2012;113:752–759.
61. Dang W, Steffen KK, Perry R, et al. Histone H4 lysine 16 acetylation regulates cellular lifespan. *Nature*. 2009;459:802–807.
62. Wang RH, Zheng Y, Kim HS, et al. Interplay among BRCA1, SIRT1, and Survivin during BRCA1-associated tumorigenesis. *Mol Cell*. 2008;32:11–20.
63. Goel A, Aggarwal BB. Curcumin, the golden spice from Indian saffron, is a chemosensitizer and radiosensitizer for tumors and chemoprotector and radioprotector for normal organs. *Nutr Cancer*. 2010;62:919–930.
64. Marcu MG, Jung YJ, Lee S, et al. Curcumin is an inhibitor of p300 histone acetylatransferase. *Med Chem*. 2006;2:169–174.
65. Kang SK, Cha SH, Jeon HG. Curcumin-induced histone hypoacetylation enhances caspase-3-dependent glioma cell death and neurogenesis of neural progenitor cells. *Stem Cells Dev*. 2006;15:165–174.
66. Chiu J, Khan ZA, Farhangkhoee H, Chakrabarti S. Curcumin prevents diabetes-associated abnormalities in the kidneys by inhibiting p300 and nuclear factor-kappaB. *Nutrition*. 2009;25:964–972.
67. Pogribny IP, Ross SA, Tryndyak VP, et al. Histone H3 lysine 9 and H4 lysine 20 trimethylation and the expression of Suv4-20h2 and Suv-39h1 histone methyltransferases in hepatocarcinogenesis induced by methyl deficiency in rats. *Carcinogenesis*. 2006;27:1180–1186.
68. Mehedint MG, Niculescu MD, Craciunescu CN, Zeisel SH. Choline deficiency alters global histone methylation and epigenetic marking at the Re1 site of the calbindin 1 gene. *FASEB J*. 2010;24:184–195.
69. Tateishi K, Okada Y, Kallin EM, Zhang Y. Role of Jhdm2a in regulating metabolic gene expression and obesity resistance. *Nature*. 2009;458:757–761.
70. Hassan YI, Zempleni J. A novel, enigmatic histone modification: biotinylation of histones by holocarboxylase synthetase. *Nutr Rev*. 2008;66:721–725.
71. Kuroishi T, Rios-Avila L, Pestinger V, et al. Biotinylation is a natural, albeit rare, modification of human histones. *Mol Genet Metab*. 2011;104:537–545.
72. Zempleni J, Chew YC, Bao B, et al. Repression of transposable elements by histone biotinylation. *J Nutr*. 2009;139:2389–2392.
73. Camporeale G, Giordano E, Rendina R, et al. Drosophila melanogaster holocarboxylase synthetase is a chromosomal protein required for normal histone biotinylation, gene transcription patterns, lifespan, and heat tolerance. *J Nutr*. 2006;136:2735–2742.
74. Havas K, Flaus A, Phelan M, et al. Generation of superhelical torsion by ATP-dependent chromatin remodeling activities. *Cell*. 2000;103:1133–1142.

75. Tang L, Nogales E, Ciferri C. Structure and function of SWI/SNF chromatin remodeling complexes and mechanistic implications for transcription. *Prog Biophys Mol Biol*. 2010;102:122–128.
76. Simone C. SWI/SNF: the crossroads where extracellular signaling pathways meet chromatin. *J Cell Physiol*. 2006;207:309–314.
77. Jeong SM, Lee C, Lee SK, et al. The SWI/SNF chromatin-remodeling complex modulates peripheral T cell activation and proliferation by controlling AP-1 expression. *J Biol Chem*. 2010;285:2340–2350.
78. Reisman D, Glaros S, Thompson EA. The SWI/SNF complex and cancer. *Oncogene*. 2009;28:1653–1668.
79. Wang F, Liu H, Blanton WP, et al. Brd2 disruption in mice causes severe obesity without Type 2 diabetes. *Biochem J*. 2010;425:71–83.
80. Lanzuolo C, Orlando V. Memories from the polycomb group proteins. *Annu Rev Genet*. 2012;46:561–589.
81. Schoeftner S, Sengupta AK, Kubicek S, et al. Recruitment of PRC1 function at the initiation of X inactivation independent of PRC2 and silencing. *EMBO J*. 2006;25:3110–3122.
82. Lee ER, Murdoch FE, Fritsch MK. High histone acetylation and decreased polycomb repressive complex 2 member levels regulate gene specific transcriptional changes during early embryonic stem cell differentiation induced by retinoic acid. *Stem Cells*. 2007;25:2191–2199.
83. Cao L, Bombard J, Cintron K, et al. BMI1 as a novel target for drug discovery in cancer. *J Cell Biochem*. 2011;112:2729–2741.
84. Balasubramanian S, Adhikary G, Eckert RL. The Bmi-1 polycomb protein antagonizes the (-)-epigallocatechin-3-gallate-dependent suppression of skin cancer cell survival. *Carcinogenesis*. 2010;31:496–503.
85. Malumbres M. miRNAs and cancer: an epigenetics view. *Mol Aspects Med*. 2013;34(4):863–874.
86. Winter J, Jung S, Keller S, et al. Many roads to maturity: microRNA biogenesis pathways and their regulation. *Nat Cell Biol*. 2009;11:228–234.
87. Sato F, Tsuchiya S, Meltzer SJ, Shimizu K. MicroRNAs and epigenetics. *FEBS J*. 2011;278:1598–1609.
88. Juan AH, Kumar RM, Marx JG, et al. Mir-214-dependent regulation of the polycomb protein Ezh2 in skeletal muscle and embryonic stem cells. *Mol Cell*. 2009;36:61–74.
89. Furuta M, Kozaki KI, Tanaka S, et al. miR-124 and miR-203 are epigenetically silenced tumor-suppressive microRNAs in hepatocellular carcinoma. *Carcinogenesis*. 2010;31:766–776.
90. Tryndyak VP, Ross SA, Beland FA, Pogribny IP. Down-regulation of the microRNAs miR-34a, miR-127, and miR-200b in rat liver during hepatocarcinogenesis induced by a methyl-deficient diet. *Mol Carcinog*. 2009;48:479–487.
91. Marsit CJ, Eddy K, Kelsey KT. MicroRNA responses to cellular stress. *Cancer Res*. 2006;66:10843–10848.
92. Cirera S, Birck M, Busk PK, Fredholm M. Expression profiles of miRNA-122 and its target CAT1 in minipigs (Sus scrofa) fed a high-cholesterol diet. *Comp Med*. 2010;60:136–141.
93. Saini S, Majid S, Dahiya R. Diet, microRNAs and prostate cancer. *Pharm Res*. 2010;27:1014–1026.
94. Chiyomaru T, Yamamura S, Zaman MS, et al. Genistein suppresses prostate cancer growth through inhibition of oncogenic microRNA-151. *PLoS One*.

2012;7:e43812.
95. Gao SM, Yang JJ, Chen CQ, Chen JJ, et al. Pure curcumin decreases the expression of WT1 by upregulation of miR-15a and miR-16-1 in leukemic cells. *J Exp Clin Cancer Res*. 2012;31:27.
96. Yang J, Cao Y, Sun J, Zhang Y. Curcumin reduces the expression of Bcl-2 by upregulating miR-15a and miR-16 in MCF-7 cells. *Med Oncol*. 2010;27(4):1114–1118.
97. Zhang L, Hou D, Chen X, et al. Exogenous plant MIR168a specifically targets mammalian LDLRAP1: evidence of cross-kingdom regulation by microRNA. *Cell Res*. 2012;22:107–126.

Nutrition and the Humoral Regulation of Growth

21

William E. Russell, MD and J. Marc Rhoads, MD

Hormones and nutrients interact to influence growth at all levels of biologic organization. Growth is often loosely defined to include all aspects of somatic enlargement and maturation. The principal impact of malnutrition on the growing child is to diminish or reverse weight gain. When malnutrition is of sufficient duration or intensity, statural growth is retarded, although under some unusual circumstances, malnutrition may selectively impair linear growth without a preceding or concomitant loss in weight.[1] The manifestations of malnutrition effects on growth are often subtle, frequently masquerading as social, ethnic, or racial characteristics. In a study of ethnically homogeneous conscripts in the Polish army, Bielicki and colleagues[2] found that young men from large families, from the countryside, and from lower socioeconomic classes were smaller than their counterparts from smaller, urban, and more privileged families. These authors concluded that the negative impact of these variables on stature could all be attributed to suboptimal nutrition. On a large scale, the effect of malnutrition is often so striking as to produce major population differences in stature that appear to be genetic. For instance, prior to World War II, native Japanese were smaller than Americans of European ancestry and smaller than Japanese-Americans raised in the United States.[3] However, with the significant nutritional improvements that took place in Japan following World War II, these differences vanished.[4]

NUTRIENTS, HORMONES, AND GROWTH

Hormones and Growth Factors

Hormones are generally defined as the secretions of glands that specialize in their production; classic hormones, acting through an endocrine mode of regulation, therefore require conveyance through the circulation to alter the function of distant tissues. Their concentrations and association with binding proteins can be readily measured in the blood and other body fluids. Regulation occurs not only through changes in the concentration of the hormone, but also in the number and cellular distribution of their peripheral receptors and in the systems of second messengers within the target cells that convey these signals to the genome or other cellular compartment. Each of these steps can be regulated by developmental programs and by the local milieu of nutrients and other hormones.

The body-wide endocrine system, however, is also an integrating system that is superimposed on multiple local regulatory systems that employ paracrine modes of regulation.[5] Paracrine control refers to the release by cells of soluble messengers that act on neighboring cells of another type. A circulation is not required because only physical proximity and the presence of receptors are required for effect. In 1980, Sporn and Todaro[6] added the concept of autocrine control to include the substances that cells secrete to regulate their own growth. Although autocrine control was initially thought to be a mode of growth control for neoplastic cells exclusively, autocrine control also characterizes the growth and function of many normal cells. Examples of autocrine control include the release of epidermal growth factor (EGF) by ulcerated gastrointestinal (GI) mucosa and release of transforming growth factor-alpha by normal intestinal epithelial cells.[7] As a consequence of the narrow range of action within the extracellular space of autocrine and paracrine growth factors, their roles in cellular regulation are much more difficult to document in clinical situations. It has recently emerged that proteolytic events at the cell surface are critical regulators of the accessibility of autocrine and paracrine factors to their receptors.[8] Multiple proteases associated with extracellular matrix or cell membranes are known to solubilize and activate growth factors from their cell surface anchored, but biologically inactive, precursors.

It has been proposed that growth factors can exert biologic actions without release into the extracellular space. *Juxtacrine* control refers to the actions of growth factors that remain anchored to the plasma membranes of the cells that produced them; cell-to-cell contact brings them into contact with receptor-bearing target cells.[9] Finally, some growth-

regulatory molecules can activate receptors on intracellular membranes including those in the cytoplasm, the nucleus, nucleolus, or mitochondrion, a mode of action called *intracrine* control.[10] In some instances, intracrine factors originate within their target cell and in other instances they are internalized from the extracellular space. Intracrine action appears to be critical for the regulation of the chicken ovalbumin gene.[11] In conjunction with the neuroendocrine mode of regulation, which makes use of axons to direct regulatory neurotransmitters to their target cells, the multiple systems of cellular regulation provide a diversity of regulatory options, each of which has relevance to an understanding of nutritional influences on growth. The characteristics of endocrine, paracrine, autocrine, juxtacrine, intracrine, and neuroendocrine modes of regulation are summarized in Table 21-1.

TABLE 21-1 Modes of Cellular Regulation

Mode	Agent	Source	Vehicle	Target	Modul
Endocrine	Hormone	Gland	Circulation	Heterologous tissue	Neurons Hormon Growth
Paracrine	Growth factor	Local site	Diffusion	Heterologous cell	Hormon Growth
Autocrine	Growth factor	Local site	Diffusion	Autologous cell	Hormon Growth
Juxtacrine	Growth factor precursor	Local site	Cell contact	Adjacent cell	Proteas
Intracrine	Hormone Growth factor Nutrient	Intracellular	Cytoplasm	Intracellular receptor	?
Neuroendocrine	Neurotransmitter Growth factor	Neuron	Axon	Heterologous tissue	Hormon Growth Neurons

In addition to the protein and steroid molecules that comprise the classic hormones of the endocrine system, locally acting substances, often designated “growth factors” are the principal agents of the paracrine, autocrine, juxtacrine, and intracrine systems.[12] The distinction between hormones and growth factors is generally a functional one. There are instances in which a substance may be released from a gland into the circulation to act as a hormone at a distant site, whereas within a highly specialized tissue the same substance might be locally secreted as a growth factor to influence the function of adjacent cells. In many instances, nature

has retained the basic structure of successful regulatory molecules and, through a divergent evolution, has created new uses for them. The structural homology of insulin and the insulin-like growth factors (IGFs), which are discussed later in this chapter, illustrates how a substance with predominantly nutrient transport-regulating properties, which is found in even primitive unicellular organisms, has evolved to regulate highly differentiated functions.

Nutrients and the Growth of Cells

Holley[13,14] was among the first to propose that the intracellular availability of nutrients is the ultimate regulator of cell growth. He postulated that when cellular nutrition is adequate, mammalian cells require no additional extrinsic growth-promoting substances and that hormones and growth factors regulate tissue growth ultimately by altering the availability of nutrients. In the laboratory, the growth of cells and tissues clearly requires the proper combinations of hormones and growth factors in addition to an adequate supply of nutrients. There is ample evidence that growth factors and hormones influence the transport and utilization of nutrients by cells.[15–19]

Hormones and nutrients interact in a delicate balance: hormones alter the nutritional requirements of cells, and conversely, nutrients alter cellular hormonal requirements. McKeehan and McKeehan[20,21] found that cultured human lung fibroblasts require lower concentrations of various minerals and substrates in the culture medium as the concentration of serum growth factors is increased. Conversely, they found that adequate concentrations of nutrients reduce the need for growth factors, an interaction that has been systematically probed.[22,23]

Although adequate supplies of nutrients are necessary to allow full manifestations of the effects signaled by hormones and growth factors, hormones influence cell growth by mechanisms other than changing their cellular nutritional status. Thyroid and steroid hormones, for example, can change cell function by directly influencing the transcription of target genes in the nucleus, although adequate “nutrition” in the form of high-energy phosphates and other low-molecular-weight substrates is required for full manifestation of the signal. In contrast, most polypeptide hormones and growth factors bind to cell-surface receptors, and their ability to influence the genome requires the initiation of a cascade of reactions that involve lipids, proteins, and ions as second messengers. Specific micronutrients, such as glutamine, allow the amplification of growth factor

signaling, and individually activate intracellular signaling kinases (e.g., mitogen-activated protein kinases) in intestinal and liver cells.[24,25] Furthermore, there are amino acid response elements in the promoter of nutritionally important genes. One gene regulated by amino acid deprivation and the endoplasmic reticulum stress response is asparagine synthetase. Asparagine synthetase is recognized as a target of antileukemic chemotherapy and is essential for maintenance of cell viability.[26]

Nutrients and the Growth of Tissues

The study of cellular proliferation in the laboratory culture dish can provide information on the hormonal and nutritional needs of dividing cells, but the growth and development of the organism as a whole require that the enlargement and maturation of each tissue occur at appropriate rate and in proper sequence.[27] As they can influence nutrient requirements of individual cells, hormones can also affect the nutrient needs of complex tissues as well as the supply of nutrients in the fluid milieu in which they are bathed. The effects of a given hormone on its various target tissues can be quite different: catabolic in one tissue, anabolic in another (Table 21-2). These effects are generally not without coordination, as catabolism in one tissue can serve to increase the supply of nutrients to another tissue that is rendered anabolic by the same hormone. For example, under normal circumstances, anabolic hormones increase the protein content of both viscera and muscles.[28] In the nutrient-deficient state, however, anabolic agents such as growth hormone (GH) and androgens continue to preferentially support carcass protein (muscle) synthesis even at the expense of visceral protein. Androgens, for instance, diminish the rate of muscle breakdown in fasted guinea pigs, while accelerating the loss of protein from the liver.[29] Similarly, the weight loss of food-restricted swine can be reduced by the administration of GH, but the effect is to preferentially retain muscle mass, while depleting body fat.[30] Similar effects are seen in humans (see below) and have led to abuse of GH by competitive athletes. Conversely, an important adaptive response to long-standing malnutrition is a generalized inhibition of anabolic systems, such as those involving GH, estrogens, and androgens, in order to conserve essential body compartments.

TABLE 21-2 Actions of Hormones on Body Compartments

Hormone	Protein	Carbohydrate	Fat

Glucocorticoid	Breakdown in muscle, synthesis in viscera	Permissive for gluconeogenesis, glycogen synthesis	Redistribution
Insulin	Synthesis in muscle and liver	Glycogen synthesis in liver, glycolysis	Lipogenesis, inhibits lipolysis
Growth hormone	Synthesis in muscle	Insulin-like effects (acute), insulin antagonistic (chronic)	Lipolysis, inhibits lipogenesis
Thyroid hormone	Synthesis in liver, breakdown in muscle	Glycogenolysis, gluconeogenesis	Lipolysis
Androgen	Synthesis in muscle and viscera	Increased insulin sensitivity[31,32]	Redistribution[33]

Source: Adapted from Eisenstein and Singh.[34]

Although anabolic hormones preferentially preserve muscle mass when nutrients are in short supply, catabolic hormones serve to protect the viscera. Brief exposure of rats to glucocorticoids or to thyroxine increases the protein and glycogen content of the liver, whereas the protein content of muscle is depleted.[28] These effects are direct ones on hepatic uptake and synthetic processes and not merely the result of increased substrate availability from the carcass.[35] Even a prolonged exposure to glucocorticoids, which depletes both viscera and muscle, reduces visceral mass proportionally less than muscle while increasing adipose mass.

Critical Periods in Tissue Growth

The ability of tissues to recover from a nutritional insult varies greatly at different times during development. The study of normal tissue growth has provided a model of normal tissue development that has been used to analyze the variable effects of malnutrition at different developmental stages. As the organ or tissue approaches its predetermined complement of cells, the organ grows by increasing both its cell number and the size of each cell. When the adult complement of cells is attained, cell division ceases and tissue growth results totally from enlargement of existing cells.[36] Between these two periods, tissue growth appeared to result from both cellular hyperplasia and hypertrophy.

McCance[37] and Widdowson[38,39] performed studies on growing animals that clearly established the existence of critical periods of sensitivity to the effects of malnutrition. They manipulated the litter size and, therefore, the milk supply of rat pups during suckling and observed their subsequent growth. Rats that were nutritionally deprived during this

period were small, and their growth could not be restored by institution of normal food intake.[40] The mechanism for this runting appears to be an irreversible effect of undernutrition on the number of cells. Similar mechanisms are likely to operate in some forms intrauterine growth retardation in children, although the irreversibility of the growth deficit has come into question. About 85% of infants born small for gestational age (SGA) will exhibit catch-up growth in the first 2–3 years of life, but a small group has a high risk of short stature in adult life. It has been demonstrated that irrespective of basal GH secretory status, GH treatment can accelerate growth in SGA children, and lead to normalization of adult stature.[41]

Winick and Noble[42] provided further evidence that the nutritional state of the neonatal period influences cell proliferation and consequently determines the ultimate size of the animal and its organs. Food restriction in the first 21 days of life produced a subnormal complement of cells in most tissues. However, the size of the cells was normal. Adequate nutrition after weaning could not restore normal growth, presumably because of an irreplaceable deficit of cells. Food restriction between 21 and 42 days resulted in a comparable stunting of growth in most tissues, but the brain and lung were able to resume normal growth with the restoration of proper nourishment. In these tissues, the total adult cell number had already been attained, and malnutrition produced a reversible decrease in cell size. When malnutrition was induced between 65 and 86 days, almost all organs underwent a reversible decrease in cell size only.

Growing tissues are as sensitive to an excess of nutrients as to a shortage. When rat litter sizes were reduced to provide supranormal nutrition, the offspring were larger than normal and were found to have hypercellular organs.[43]

As it has evolved from studies such as these, the critical period hypothesis proposes that abnormal nutrition, either excessive or deficient, during the phase of tissue growth by cellular proliferation, can increase or decrease the ultimate size of a tissue by enhancing or impairing cell division. These effects are irreversible because they bring the tissue into the phase of growth by cellular hypertrophy with an abnormal number of cells or abnormal organization. When nutritional surfeit or deficit occurs after establishment of the adult complement of cells, the net effect is a change in cell volume, but not in cell number, and is reversible by normalization of the nutrient supply. Because tissues mature at different rates, the long-term effects of nutritional variations occur most prominently in those tissues that are at a susceptible developmental

period.[44] An important example of this principle of "programming" following an insult at a critical, sensitive period of early life is that mothers who are malnourished during pregnancy often have infants with intrauterine growth retardation, subsequent growth retardation, and ultimately with disproportionately high rates of coronary heart disease, high blood pressure, and high cholesterol, during adult life.[45] Intrauterine growth retardation in girls has recently been linked to the development of hyperandrogenism, insulin resistance, and ovulatory dysfunction in adolescent years.[46–48]

Catch-Up Growth

Parents often want an estimate of their child's ultimate height. Mid-parental height combined with an assessment of skeletal maturity, or "bone age," is widely used to predict adult height and is probably its single best predictor.[49–52] However, in situations where the parents are either unusually short or unusually tall, the child can be relatively less short or tall, respectively—an observation called "regression to the median."[53]

Children have the remarkable capacity to accelerate their growth when recovering from a growth-impairing influence such as malnutrition or disease. At the complex level of organization that is required for statural growth, the ability of nutritionally deprived cells and tissues to be restored to functional health is reflected in the phenomenon of "catch-up growth," a term used by Prader and colleagues.[54] Subsequently, "catch-down" was coined to refer to the complementary process that follows removal of growth-accelerating influences, such as occurs with suppression of precocious puberty, restriction of food in obesity, or cessation of GH therapy in hypopituitary children.

Even under the most favorable circumstances, the accretion of height does not occur at a fixed rate, but is developmentally regulated. Gender-specific, nutritional, and—in some cultures—seasonal influences are also superimposed. The period of most rapid postnatal growth in children occurs in the first months of life, when length increases by 14–22 cm/y. By the second birthday, the mean growth velocity has slowed to about 8 cm/y, and to about 5 cm/y just before puberty. Then, under the influence of sex steroids (but involving the coordinated actions of GH, IGFs, thyroid hormones, and others), growth velocity doubles to 8–10 cm/y before statural growth ceases, as the epiphyses close.

After the second year of life and continuing throughout childhood, growth velocity is modulated to maintain stature at a constant position

relative to the general population. The height percentile attained by about 2 years of age is maintained faithfully throughout childhood and correlates well with the height percentile in adulthood.[55] Statural growth along a predetermined percentile on a growth chart is often called canalization. When growth-retarding or growth-accelerating processes disrupt canalization, growth curves will fall across channels. With amelioration of the abnormal condition, a compensatory acceleration or deceleration occurs to restore the original growth channel (Figure 21-1).

In the first two years of life, catch-up growth can be a normal adaptation in children whose birth size is at variance with their genetic potential. Although statural growth from age 2 to adulthood is primarily controlled by genetic influences, the determinants of body size during fetal life are largely determined by maternal and nutritional influences. The interval between birth and roughly 2 years of age is a preprogrammed period of catch-up or catch-down growth and is characterized by a shift of growth channels as maternal influences give way to genetically determined forces. It is essential for the clinician to be aware of these forces during this period because genetically programmed changes in growth velocity can be misconstrued as manifestations of disease.[56]

From a practical standpoint, for catch-up growth to be complete, the insult must be limited in duration and must not occur during certain vulnerable periods. Williams and coworkers[57,58] noted that the degree of catch-up growth is generally muted if it begins when growth is normally decelerating. On the basis of observations in birds and mammals, Wilson proposed that catch-up growth potential is modified by the nature of the insult, its severity, its duration, and the developmental state of the animal at the onset of the insult. For example, it had been assumed that GH deficiency and hypothyroidism were two conditions with the potential for essentially complete catch-up. However, the catch-up potential of hypothyroid children is diminished in proportion to the duration of the hypothyroidism before treatment.[59] The subjects studied had been hypothyroid for approximately five years and had deficits from their predicted heights of 25 cm at the onset of therapy; however, at the conclusion of therapy an average deficit of 7 cm remained (Figure 21-2). The magnitude of deficit between final height and predicted height was in proportion to the duration of hypothyroidism before treatment began. Similar conclusions resulted from studies of children with GH deficiency of long duration before treatment.[60] Incomplete catch-up growth has also been documented in children with acute nutritional insults, such as diarrhea, when they are superimposed on a background of chronic

malnutrition.[61,62]

In addition to its duration, the specific nature of the insult may be important in determining the extent of catch-up because some growth-arresting conditions appear to cause irreversible changes. Mosier found that cortisone-treated rats failed to regain normal body size after cessation of treatment, whereas rats that were comparably stunted by fasting resumed normal growth when refed.[63] Cartilage from the steroid-treated rats had increased numbers of dead and dying cells, altered cell morphology, and alterations in the matrix. Despite these seemingly irreversible morphologic effects on chondrocytes, GH therapy has been shown to reverse many of the catabolic effects, increase anabolic ones, and promote growth in children with glucocorticoid-induced short stature.[64]

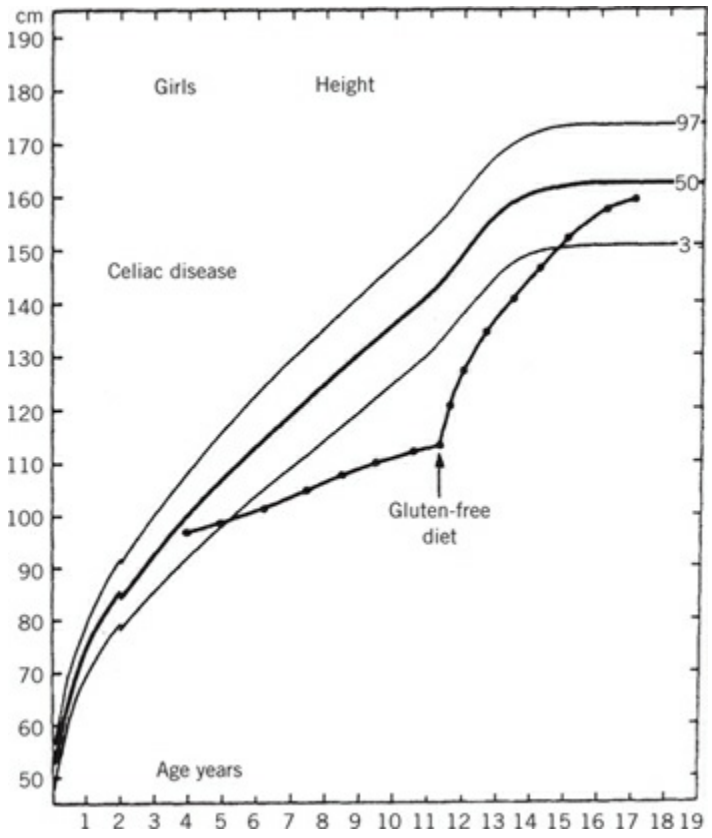


Figure 21-1 Catch-up growth in a girl with celiac disease. After eight years of growth failure, she was placed on a gluten-free diet and exhibited striking catch-up growth. Note return to previous growth percentiles. (Courtesy JM Tanner.)

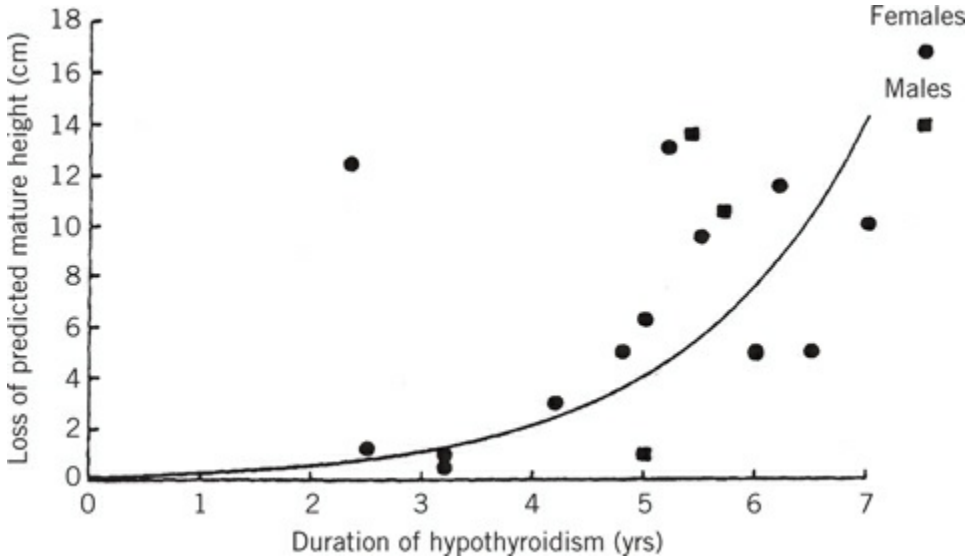


Figure 21-2 Loss of catch-up potential with long-standing hypothyroidism. Relationship between loss in adult height and duration of hypothyroidism before treatment in children. Loss of predicted height is predicted height at time of diagnosis minus actual mature height. Duration of hypothyroidism is chronologic age minus bone age at diagnosis. Adapted from Rivkees et al.[59]

It has been suggested that the catch-up growth of recovery from malnutrition in some experimental conditions is associated with an increase in the efficiency of energy utilization.[65] Fasted rats are capable of a fivefold increase in energy utilization efficiency for weight gain during recovery, perhaps by decreased thermogenesis.[66] The situation may be different in humans: children recovering from malnutrition often have increased appetite and caloric intake, suggesting to others that energy utilization is not efficient.[67] Some marasmic children do not begin to gain weight until 3–5 weeks of refeeding, despite an intake of more than 130 kcal/kg/d.[68] (chapter by Mary Penny in this textbook). In fact, once the infant is resuscitated with respect to fluid balance, infection therapy, glucose control, and temperature control, they usually require between 150 and 220 kcal/kg/d to gain weight.[69]

Implied in the concept of catch-up or catch-down growth is an ability of the organism to constantly monitor its size and determine the extent of deviation from the intended size; to alter its velocity when unrestrained from the growth-perturbing influence; and finally, to again modulate its altered velocity to prevent overshooting or undershooting the channel.[70–73] Williams stressed the theoretical importance of distinguishing between

catch-up growth and compensatory growth.[73] He recognized that the former involved growth to attain a projected (and moving) statural target, whereas the latter involved the growth of a tissue or organ to restore actual lost mass as in the regeneration of liver following a partial hepatectomy.[74]

Two divergent, but not necessarily exclusive, models have been put forward to explain the ability of the organism to monitor body size and initiate catch-up. Tanner[75] postulated the existence of a "sizostat" in the central nervous system (CNS) that maintained communication with the peripheral tissues. Along the same lines, Leibel[76] proposed that the concentration of a circulating hormone, such as insulin (or its ratio to other hormones or metabolites), might reflect the size of a body compartment, such as the adipose mass, by monitoring the number of insulin receptors on adipocytes. With the isolation of leptin, the data that support a neuroendocrine control of weight regulation have been far stronger than that of a neuroendocrine control of statural catch-up. For example, to initiate catch-up growth in conditions of malnutrition, the child must attain a body weight that is 85% of the normal weight for height.[77] The interactions between adipose mass and the CNS are discussed in the section on Reproductive Function in Malnutrition.

In contrast to a neuroendocrine mechanism of catch-up growth, Baron and colleagues[78] focused on the growth plate as the key tissue in catch-up growth. Their data showed that a unilateral growth-suppressing influence, accomplished by transient administration of glucocorticoids to a single growth plate, was accompanied by catch-up growth acceleration at that growth plate only, not systemically.[79] Although their model does not preclude the involvement of systemic hormones and the CNS, they proposed that growth-inhibiting conditions decrease proliferation of growth plate stem cells, thus conserving their proliferative potential. Catch-up growth results from a putative clock, or time tally to use Tanner's term, within the stem cell compartment.

Although it is likely that both systemic (neuroendocrine and endocrine) and local (paracrine, autocrine, and juxtacrine) signals will be implicated as critical regulators of the proliferation that occurs during catch-up or catch-down growth, there is little evidence to date that hormones and growth factors are significant mediators of the catch-up phenomenon. These data are extensively reviewed in reference 77.

GH, IGFs, AND INSULIN

Normal Growth

GH and IGFs in Normal Growth

Growth hormone or somatotropin is essential for normal, balanced somatic growth from childhood to normal adult stature. Growth hormone deficient children are not only short but have profound deficits in lean body mass and excess adipose tissue in addition to abnormalities in glucose homeostasis. The mechanism by which this process is regulated is incompletely understood, especially in the fetus. The actions of GH involve a complex interaction among GH genes, GH receptors, IGFs or somatomedins, IGF receptors, and IGF-binding proteins (IGF-BPs). In addition, the GH–IGF axis is entwined in a regulatory web that is strongly modulated by nutritional signals.

IGFs and the Somatomedin Hypothesis

With the advent of techniques for the culture of tissues and cells in the laboratory almost 50 years ago, it became evident that few of the biochemical events that had been attributed to GH in vivo could be reproduced in isolated tissues. These included effects on chondrocyte proliferation and sulfate incorporation into glycosaminoglycans of growth plate cartilage. These observations led Salmon and Daughaday[80] to propose the “somatomedin hypothesis”: that the growth-promoting effects of GH are not direct ones on its target tissues, but result indirectly from the actions of a “mediator” substance under GH control. Reviewed in reference 81, initially called sulfation factor or thymidine factor, based on the assay system used to detect its presence, it was later determined that these two biological activities resided in the same molecule, which was renamed somatomedin.[82] Other somatomedin species were initially proposed, but somatomedin-C (Sm-C) was determined to be the GH mediator present in serum.

IGFs were purified in parallel, but independent, studies to characterize the source of the insulin-like biological activity of serum, only a small part of which derives from insulin itself. Two molecules were identified, IGF-I and IGF-II, the amino acid sequences of which were found to be remarkably similar to proinsulin.[83,84] Sm-C is identical to IGF-I, and current convention prefers the IGF terminology, although a number of IGF-I assays available from clinical reference laboratories retain the Sm-C designation. The properties of IGF-I and IGF-II are contrasted in Table 21-3. Although the current nomenclature has the disadvantage of dissociating the IGFs from their regulatory link with GH, it does more firmly root these

substances in the realm of nutritional control.

Initial iterations of the somatomedin hypothesis were inspired by endocrine models of growth control: liver was found to be a major producer of somatomedin activity, and the paradigm that evolved had the liver functioning as an endocrine gland under the control of pituitary GH to secrete somatomedins whose ultimate target tissue was the growth plate. To further support an endocrine model, IGFs were found to feed back to the pituitary and inhibit the release of GH.

However, in the 1980s it became clear that multiple tissues express mRNA for IGFs and are involved in IGF production.[85,86] Most compelling was the observation that IGF-I mRNA is expressed in perichondrium and periosteum, suggesting that local production of IGFs in the epiphysis might account for the growth stimulating effects of GH. Indeed, Isaksson[87] had shown that GH elicits an increase in both IGF-I mRNA and protein in rat epiphyses. On the basis of analogy to the work of Greene and colleagues[88] on the role of GH in the differentiation of adipocytes, Isaakson and colleagues proposed that GH stimulates the differentiation of chondrocytes in the germinal zone of the growth plate. Once the stem cells become chondrocytes, their proliferation would result from local (autocrine/paracrine) production of IGF-I.[87] However, inconsistencies remain among the data: GH- or growth hormone receptor (GHR)-deficient mice and humans have no paucity of differentiated chondrocytes, and growth plate chondrocytes can proliferate normally without IGF-I.[81] Nonetheless, the consensus is that locally generated IGFs in the epiphysis are the critical regulators of linear growth.

TABLE 21-3 Insulin-Like Growth Factors

Property	IGF-I	IGF-II
Synonyms	Somatomedin C, basic somatomedin	MSA, neutral somatomedin
Chromosome location	12	11
Molecular weight	7649	7471
Serum concentration	193 ± 58 ng/mL	647 ± 126 ng/mL
GH dependence of serum concentration	+++	+
Insulin-like actions	+	+++
Mitogenicity in cultured cells	+++	+
Developmental role	Postnatal	Fetal
Sensitivity to undernutrition	+++	±

GH, growth hormone; MSA, multiplication stimulating activity.

The weakness of the original somatomedin hypothesis was most clearly illustrated by the creation of a liver-specific knockout of the IGF-I gene. Using Cre/loxP technology, LeRoith and colleagues produced mice with hepatic IGF-I mRNA levels reduced to less than 1% of normal, resulting in a 75% reduction in serum IGF-I.[89,90] Nonetheless, these mice lacking hepatic IGF-I grew normally, and "there were no phenotypic distinctions between the liver-specific IGF-I gene-deleted animals and their wild-type littermates."[81] While these studies do not confirm that bone-derived IGFs are the critical determinants of linear growth, they do support the contention that circulating IGFs are not critical to skeletal growth. As expected based on their markedly reduced serum IGF-I levels, the liver-specific IGF-I knockout animals had elevated concentrations of GH, leading LeRoith to speculate that the major role of circulating IGF-I might be to modulate GH secretion.

IGF-BPs

In contrast to GH, which circulates in association with a fragment of the GH receptor called the GH-binding protein (GH-BP), IGF concentrations in blood are maintained at relatively stable levels by serum binding proteins that markedly delay IGF clearance. IGF binding in serum and other fluids resides in six different molecules, collectively known as the IGF-BPs. The IGF-BPs are summarized in Table 21-4.[91] The major IGF binding complex in serum consists of IGF-BP3 in association with the acid-labile subunit (ALS). IGF-BP5 can also interact with ALS and IGFs in the serum. Circulating IGF-BP3 concentrations, such as those of IGF-I, are GH dependent. It is now clear that many, if not all, of the so-called somatomedin inhibitors identified in early bioassays of somatomedin activity, especially in conditions of malnutrition and disease, were actually IGF-BPs.

All six proteins have higher affinity for both IGF-I and IGF-II than do IGF receptors. IGF-BPs were initially proposed to function as IGF-sequestering agents, thus limiting the concentration of free IGFs. However, IGF-BPs modify IGF actions in a variety of ways. They transport IGFs across vessel walls and capillary membranes, they regulate the concentration of IGFs in the vicinities of specific tissues and cell types, they control IGF interaction with cell-surface IGF receptors, and at least three of these molecules (IGF-BP1, BP3, and BP5) can augment IGF action. Duan has proposed that a binary complex of a single IGF-BP and

IGF-I, which can leave the circulation rapidly, facilitating access of IGFs to their receptors, whereas a ternary complex consisting of IGF, IGF-BP3 (or BP5), and ALS cannot cross the endothelial barrier and remains in the circulation to sequester free IGF-I.[91] The implications of this on nutritional regulation of IGF action are discussed below. As noted in Table 21-4, serum concentrations of the three principal IGF-BPs are profoundly influenced by nutritional signals (mediated by insulin) and by GH. IGF binding and action can also be modulated by posttranslational modifications of the IGF-BPs such as phosphorylation, proteolysis, and matrix association.[92] Three of the IGF-BPs appear to be able to influence cell function independent of their role as an IGF-BP. For example, IGF-BP3 can inhibit proliferation in cells that have no IGF receptors.[93] Specific cell-surface receptors for the IGF-BPs have been proposed, and IGF-BP3 may have direct effects in the nucleus by interacting with the retinoid X receptor α (RXRα).[94]

GH, IGF, and IGF-BP Genes

In humans, GH is encoded within a cluster of five structurally similar genes on the long arm of chromosome 17.[95] These genes include the so-called normal GH gene (GH-N or GH-A), a variant GH gene (GH-V or GH-B), and three genes for placental lactogen (chorionic somatomammotropin). Only GH-N is translated into GH in the pituitary gland, and the other four genes of the family are expressed in placenta. Two distinct forms of GH result from differential splicing of the mRNA that is transcribed from the GH-N gene. The predominant product is a single chain of 191 amino acids with a molecular weight of 22 kDa. The product of alternate splicing of the GH-N gene is 20-kDa GH, which results from a deletion of 15 amino acids and comprises about 10% of circulating GH. Clear evidence for a differential regulation of the secretion of these two GHs is lacking, although they may differ in some of their biologic actions.

TABLE 21-4 Insulin-Like Growth Factor-Binding Proteins

Property	BP-1	BP-2	BP-3	BP-4	BP-5	BP-6
Molecular weight	25,000	33,000	54,000	24,000	31,000	30,000
Chromosome location	7	2	7	17	5	12

Serum concentration (nM)	0.8–2.8	6.1–18.3	60.170	Present		Present
Tissue localization	Serum, amniotic fluid, kidney, reproductive tissues	Serum, CSF	Serum	Serum, CNS	Placenta, kidney, bone, thyroid	Serum, CSFs
Regulation by GH	↓—	↓—	↑↑—			↑—
Regulation by Insulin	↓↓—	↓—				
Proposed function	Transport of IGFs from vascular space; immediate early gene	Transport of IGFs from vascular space	Major component of 150-kDa serum IGF complex intravascular reservoir	Blocks IGF action in cells	Potentiates IGF action in cells	

Each of the IGFs is encoded by a separate gene, although alternative splicing of each of the genes and other posttranscriptional modifications result in multiple IGF mRNA species. The extent to which the modification of IGF gene transcripts is important in the regulation of IGF action is unclear. The IGF-BPs as well arise from a gene superfamily that includes proteins whose function and relationship to IGF physiology are as yet undetermined.[96]

GH–IGF Axis: Physiologic Regulation

Pituitary GH release is regulated by two principal hypothalamic peptides, one stimulatory (GH-releasing factor, GRF or GHRH) and one inhibitory (somatostatin, SRIF) (Figure 21-3). Blood concentrations and patterns of GH secretion result from an interplay between these two substances on the somatotropes of the anterior pituitary. The hypothalamus receives rich innervation from higher centers in the brain, providing a mechanism by which even emotional influences can alter GH secretion (see discussion of “psychosocial dwarfism” later in this chapter). However, other modulating influences on GH release have been identified. Both free fatty acids and the orexigenic (appetite-stimulating) neuropeptide Y are known to directly inhibit GH secretion, whereas the fat-derived hormone, leptin (see below) stimulates GH release, perhaps by inhibiting secretion of neuropeptide Y.[97–101] Very recently, ghrelin has come to the fore as a potent regulator of

GH secretion, food intake, and fat mass. Ghrelin levels surge before every meal, and ghrelin injections rapidly stimulate food intake. Levels are very high in ravenous Prader–Willi patients and are low after gastric bypass.[102] Investigators studying a family of synthetic GH-releasing peptides, the GHRPs,[103] identified a novel CNS receptor for these peptides and then identified ghrelin as its natural ligand. Ghrelin is produced by hypothalamic neurons and by a highly specialized population of endocrine cells in the fundus of the stomach, where its secretion is suppressed by food intake. The model that is emerging ties stomach-produced ghrelin to the nocturnal stimulation of GH secretion, while hypothalamic ghrelin regulates food intake.[104]

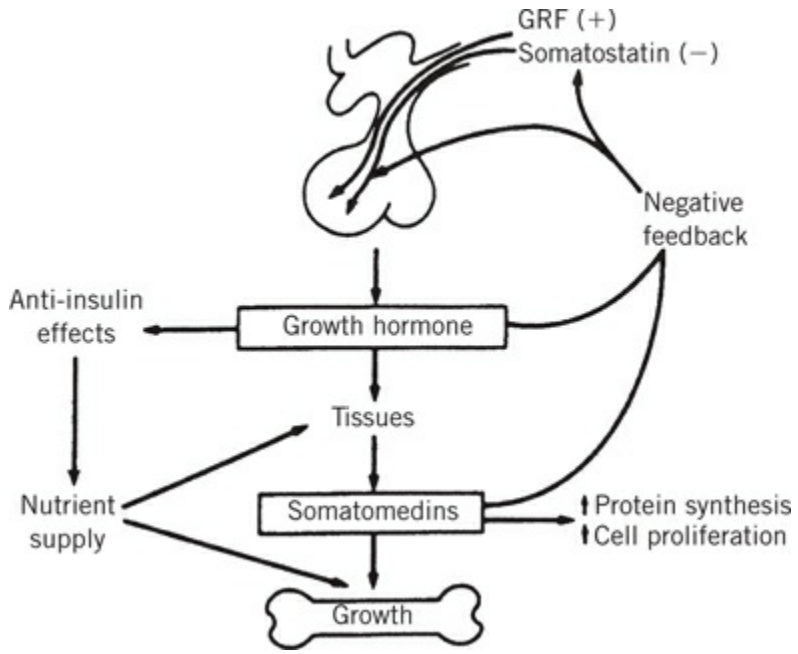


Figure 21-3 The growth hormone–somatomedin axis. (+) stimulatory action; (−) inhibitory action.

Peripherally produced IGFs feedback to inhibit GH secretion by the pituitary. IGF-I can stimulate the secretion of somatostatin by the hypothalamus[105] and may also directly inhibit GH synthesis.[106] It remains to be clarified whether IGFs of peripheral or hypothalamic origin are responsible for the inhibition of GH secretion, but the observations previously cited of enhanced GH secretion in the liver of the IGF-I knockout mouse suggest that circulating IGF-I is a prime regulator of GH

secretion.[89]

In response to GH, but modified by other hormones and most critically by nutritional factors, multiple tissues in the body synthesize and secrete IGFs. Although the liver is the major producer of circulating IGFs, its primacy in the GH–IGF axis has been supplanted. Multiple cell types in virtually every tissue examined from both fetus and adult can synthesize IGFs. Thus a model of IGF action has evolved that is primarily a paracrine one in which IGFs are produced in multiple tissues and act on target cells in the vicinity of the producing cells. Thus the function of the large quantities of circulating IGFs is not clear, but they may be more important for their insulin-like effects on metabolism than on their mitogenic effects.

GH and IGF Concentrations in Blood

Although it was believed that GH circulates in the blood free of binding proteins, it is now recognized that between 25% and 35% of circulating GH is loosely complexed to a 60-kDa glycoprotein referred to as the GH-BP.[107] This protein is identical to the extracellular domain of the GH receptor, but it is unclear whether in humans GH-BP arises from alternative splicing of the GH receptor mRNA, as it does in the rat, or is shed from cell-surface receptors by peptidases. Changes in serum concentrations of GH-BP reflect changes in the number of cell-surface GH receptors.[108] In contrast to the prolonged half-life of IGFs bound to the IGF-BPs (more than 12 hours), the plasma half-life of GH is about 20 minutes. Clinically, IGF-I concentrations in blood have been shown to reliably mirror the total GH secretory activity. Thus, in older children and adults who are well nourished and do not have hypothyroidism, serum IGF-I measurements are more useful than random measurements of GH in diagnosing conditions of GH deficiency (hypopituitarism) or excess (gigantism or acromegaly). The marked influence of nutritional status on IGF production is discussed later in this chapter.

GH secretion in humans is highly episodic. Blood concentrations are generally low or undetectable for much of the day and are interrupted by burst of secretion that results in high concentrations for roughly 90 minutes before returning to the low baseline. This pattern is developmentally regulated.[109] The number and amplitude of secretory episodes are small in young children. Under the influence of gonadal steroids, both the frequency and amplitude increase dramatically and reach their highest levels during puberty, especially during sleep; they then gradually decline over the ensuing decades.

The combined concentration of IGF proteins in the blood is on the order of 750 μg/L or 100 nM. Although GH remains the predominant regulator of IGF production and IGF-BPs, which are the principal regulators of bioavailable IGFs, a number of other hormones influence IGF-I concentrations in blood. IGF-I is low or nonexistent in fetal life, when IGF-II appears to be the predominant somatomedin. During fetal life, placental lactogen or the product of the GH-V gene is likely to be the major regulator of somatomedin production.[110] Prolactin, another GH-like hormone, can also stimulate the production of IGFs in cultured cells and probably in clinical conditions of prolactin excess. Blood levels of the IGFs remain low during infancy and early childhood but rise dramatically at puberty in response to increases in GH secretory activity. In addition, IGF-I concentrations are low in patients with hypothyroidism.[111]

Direct Versus Indirect Actions of GH: A Unifying Hypothesis?

GH initiates a signal by binding to specific receptors on the surface of target cells. A single GH molecule contains two separate receptor-binding domains, which facilitate the formation of receptor homodimers on GH binding.[112] Receptor dimerization is a feature common to a number of growth factor receptors and appears to be a prerequisite for signaling. When activated by ligand, the GH receptor dimerizes and associates with Janus Kinase 2 (JAK2). JAK2 then phosphorylates a family of proteins called signal transducers and activators of transcription (STATs). STAT proteins directly bind to DNA and directly regulate gene transcription. STAT target genes include those that encode a negative regulatory protein called suppressor of cytokine signaling-3 (SOCS-3).

The sequence of GH-initiated signaling events often extends to intracellular pathways that are activated or modulated by other hormones or growth factors, leading to complex intracellular interactions and diverse, but coordinated physiologic events. Among the targets of the activated GH receptors are insulin receptor substrate-1 (IRS-1) and IRS-2, which in turn recruit phosphatidylinositol-3 kinase (PI3-kinase) to the IRS-proteins. Activated PI3-kinase activates protein kinase B (PKB)/Akt, which in turn activates glucose transporters (GLUT4) and increases cellular glucose uptake. PKB/Akt can also activate phosphodiesterase 3B, which lowers cellular concentrations of cAMP, resulting in decreased phosphorylation of the hormone sensitive lipase and inhibition of lipolysis.[81,113]

The actions of GH in vivo have been categorized as either indirect (IGF-mediated) or direct ones. The direct actions vary significantly

depending on the duration of exposure. After short-term administration to GH-deficient individuals or to isolated GH-sensitive tissues in culture, GH has effects very much like those of insulin. They are rapid in onset, transient in duration, and not mediated through the actions of IGFs. They include the stimulation of glucose and amino acid transport and protein synthesis. GH also has inductive effects on a number of enzymes. In patients with GI and pancreatic disease who are taking parenteral nutrition (PN), administration of GH is associated with an increase in protein, potassium, and phosphorus balance.[114]

Chronic GH administration, in contrast, results in biochemical actions that appear antagonistic to insulin; blood sugar rises and fat mobilization and fatty acid oxidation occur. In fact, release of GH is one of the stress responses that the body uses to maintain euglycemia, while individuals with GH-secreting tumors can develop frank diabetes mellitus. About 3% of athletes take GH, often combining it with anabolic steroids. There is no conclusive evidence that GH enhances athletic performance, and an excess can cause acromegaly, myopathy, and hypertension. However, it does produce perceptible fluid shifts and reduced fat mass, increased lean body mass, resulting in a more “toned” appearance.[115] (PMID 23626006)

The origins of these paradoxical “direct” GH actions are not clear but may reside in the structure of the GH molecule itself. The 20-kDa variant has been reported to have only one-fifth the insulin-like activity of the 22-kDa GH molecule, while retaining normal growth-promoting and diabetogenic activity.[116,117] However, analysis of the biologic activities of both the 22-kDa and 20-kDa GH molecules in transgenic mice demonstrated largely overlapping activities with regard to linear growth and carbohydrate metabolism.[118]

The concept that IGFs can mediate the actions of GH in promoting linear growth has been validated by a number of studies that show that IGF-I injections increase tibial width and costal cartilage DNA synthesis in hypophysectomized rats to an extent that is comparable to that in GH-treated controls. In human newborn infants, umbilical cord plasma levels of IGF-I and leptin (see below) show a strong positive correlation with increasing birth weight and birth length.[119] Both IGF-I and leptin, which is secreted by both adipocytes and placenta, are important for fetal growth. The ability of IGF-I to stimulate linear growth in the absence of GH is strongly supported by studies demonstrating overgrowth in transgenic mice overexpressing IGF-I and in IGF-I-treated humans with Laron-type dwarfism, a condition that results from the hereditary absence of GH receptors. A recent study of 55 children with idiopathic short stature

revealed a lower basal level of IGF-I and a lower stimulated level of IGF-I, compared with normal body mass index controls, supporting the principle of an impaired somatotrophic axis. IGF-I receptor levels were unchanged.

IGF-I, IGF-II, and a truncated form of IGF-I, in addition to EGF, are also present in colostrum and in breast milk.[120] Importantly, intestinal IGFs are also locally produced by the mesenchymal cells within the lamina propria. Milk-derived IGFs appear to act on cells in the gut and do not have direct effects on somatic growth. However, by improving bowel function, they may increase nutrient absorption, thereby promoting somatic growth. The trophic effects of IGFs have generally been observed in the distal bowel (ileum) more than jejunum. In the piglet intestine, oral rhIGF-I increased small intestinal weight, protein and DNA content, and jejunal and ileal villus height.[121] In addition, oral rhIGF-I has been shown to increase intestinal sodium absorption and the maximal rate of glucose and alanine absorption in newborn piglets.[122]

When intestinal cells are studied, IGFs are well-known stimulators of cell proliferation[123] but also are potent stimulators of cell migration, a mechanism central to the healing of ulcers, villus damage in viral enteritis, and other lesions. The mechanism by which migration/restitution is stimulated appears to be dependent on protein kinase C.[124] However, other signaling pathways are activated by IGF-I, including the PI3-kinase pathway, c-Jun nuclear kinase, and mitogen-activated protein kinases.[125] Consequently, growth factors have been extensively investigated as therapeutic agents to enhance gut integrity and facilitate repair.

Orally administered growth factors have been found to exert trophic effects to the intestinal mucosa in experimental rotavirus diarrhea and in rats with short bowel syndrome.[126,127] In two studies, the combination of the primary metabolic fuel of the intestine, glutamine, with either systemic GH or IGF-I resulted in a synergistic trophic response.[128,129] A synergistic trophic response of glutamine with transforming growth factor-alpha was seen after ischemia/reperfusion injury of the porcine intestine.[130]

In viral enteritis, exogenous growth factors have enhanced mucosal mass without reducing diarrheal output or increasing specific activities of jejunal disaccharidase enzymes.[126,131] In short bowel syndrome and ischemic bowel disease, growth factors and the trophic amino acid L-glutamine have been shown to have synergistic effects.[132,133] These effects have not been translated into clinical therapy for diarrheal diseases to date, probably because of concerns about adverse events, including tumorigenesis.

Another disease for which enteral growth factor therapy has begun to be studied is short bowel syndrome. In this condition, which is the leading cause of intestinal failure in newborns, the infant's bowel length limits absorption, leading to a prolonged dependence on PN, which, in turn, can result in end-stage liver disease. In several studies, Donovan and colleagues[134] treated malnourished newborn piglets (given 80% of required calories via PN) with enteral IGF-I. They found that enteral IGF-I improved intestinal morphology and markedly increased disaccharidase activity in parenterally fed piglets when compared to those given partial enteral nutrition alone.[135]

Some notable differences between GH and IGF-I responses in the gut have further strengthened the concept that GH has both direct and IGF-mediated actions.[136] Transgenic mice overexpressing GH and IGF-I have increased body weight, as well as increased small bowel length and mass. However, GH had more profound effects on body mass compared to IGF-I (60% vs. 23% increase), while IGF-I had much greater effects on bowel length (44% vs. 20% increase). IGF-I but not GH overexpression increases intestinal crypt cell proliferation while reducing crypt cell apoptosis. In further support, murine knockout models of IGF-I, the GH receptor, or both show that both factors have major independent effects on somatic growth (30%–50%), while lack of functional expression of both growth factors profoundly affects growth. *IGF-I/ghr* double knockouts reach only 17% of the expected adult body weight.[137]

IGF-I and GH have not been to be beneficial in limited studies of humans with short bowel syndrome. On the other hand, a growth factor that has received considerable attention and that is consistently trophic in animal models is glucagon-like peptide-2 (GLP-2). Phase 3 trials have shown that a long-acting GLP-2 analog teduglutide stimulates mucosal growth, reduces malabsorption, and reduces PN dependence in adults with short bowel syndrome (23587733). Mechanisms of action include increasing blood flow and paracrine actions (because there are no GLP-2 receptors on enterocytes); teduglutide increases mucosal levels of IGF-I, epidermal growth factor, and keratinocyte growth factor.

Insulin in Normal Growth

The prominent influence of insulin on normal growth is best illustrated in disorders at the extremes of altered insulin secretion. Especially in the fetus, hyperinsulinemia produces somatic overgrowth. Hyperinsulinemic infants born to poorly controlled diabetic mothers or infants with hyperinsulinism of infancy[138] exhibit visceromegaly, increased body fat,

and increased length. Conversely, the hypoinsulinemic infant born with severe insulin resistance or pancreatic agenesis is small and depleted in body fat.[139] In postnatal life, poorly controlled diabetes mellitus can be associated with a striking growth failure, the so-called Mauriac syndrome.[140] The growth-promoting actions of insulin both in prenatal and postnatal life are complex and likely to result from both direct, largely metabolic, effects mediated through the insulin receptor and indirect, predominantly mitogenic effects mediated through the structurally homologous IGF-I receptor. Insulin mediates the feeding-stimulated enhancement of muscle and skin protein synthesis, but insulin is not related to feeding-induced stimulation of protein synthesis in the viscera.[141]

Insulin Receptor-Mediated Actions of Insulin

High-affinity insulin receptors mediate the rapid biologic actions of insulin in stimulating the cellular transport of substrates and in the induction of enzymes that promote glycogen and fat synthesis and that inhibit gluconeogenesis, lipolysis, and ketogenesis.

An indirect consequence of insulin action appears to be the stimulation of IGF production. A number of clinical studies have shown that the depressed IGF concentration associated with poorly controlled diabetes mellitus can be returned to normal with insulin therapy.[142] Insulin appears to directly stimulate IGF production in whole liver, liver slices, or cultured hepatocytes.[143] In addition, insulin may regulate IGF production by influencing the number of GH receptors on important IGF-producing tissues. In poorly controlled diabetes mellitus, there is a dramatic reduction in the number of somatogenic GH binding sites in liver. These can be restored to normal with insulin therapy.[144]

IGF Receptor–Mediated Actions of Insulin

In a few cell types, such as hepatocytes, certain teratocarcinoma cells, and mammary tumor cells, insulin appears to be directly mitogenic by binding to its own receptor at physiologic concentrations. However, some of the growth-promoting actions of insulin can be attributed to activation of IGF receptors. There are two principal IGF receptors, type 1 and type 2. The insulin receptor and the IGF-I receptor are homologous “heterotetramers,” consisting of two α and β subunits. The binding of insulin or IGFs to their respective α subunits activates a tyrosine kinase on the cytoplasmic portion of their respective β subunits. This kinase, in turn, transphosphorylates

tyrosines on the adjacent β subunit and on other cytoplasmic substrates that are essential for the actions of both insulin and IGF-I.[145] Both IGF-I and IGF-II can bind to and activate the insulin receptor, although with much lower affinity than insulin.

Crossover of IGF and insulin actions also occurs when type 1 IGF receptors are activated by insulin. Although type 1 receptors preferentially bind IGF-I, they can be activated by 10- to 100-fold excess concentrations of insulin, conditions that are achieved in some cases of hyperinsulinism, such as insulin-resistant states. Another level of overlap between IGF-I and insulin action is suggested by the finding that a single α, β subunit of the insulin receptor can associate with an α, β subunit of the type 1 IGF receptor, forming a hybrid tetramer.[146]

The IGF-II receptor is more enigmatic.[147] It preferentially binds IGF-II over IGF-I, but it is also the receptor that (at a different binding site) shuttles mannose-6-phosphate-containing extracellular proteins into lysosomes. The type 2 receptor does not contain an intracellular tyrosine kinase domain, and its role (if any) in intracellular signaling is unclear. Most of the actions of IGF-II are thought to be mediated through IGF-I or insulin receptors. IGF-II receptors do not bind insulin, but physiologic concentrations of insulin dramatically upregulate the number of these receptors on adipocytes.[148]

Insulin Synergy with Other Growth Factors

One further mechanism by which insulin appears to influence cellular growth is to alter the cellular requirements for mitogenic signals from other growth factors. As discussed previously regarding the interactions of nutrients and hormones, growth factor signals frequently converge on common pathways within a cell so that low concentrations of two growth factors can amplify the actions of one another. In cultured hepatocytes, insulin can potentiate the mitogenic signal of EGF or of vasopressin,[149] and in the well-studied BALB/c 3T3 fibroblast, the presence of insulin or IGF-I strongly modulates the mitogenic actions of EGF and platelet-derived growth factor.

Malnutrition

GH in Malnutrition

In humans, experimental fasting and malnutrition generally result in elevated serum concentrations of GH. Studies of malnourished populations in different parts of the world have often led to conflicting conclusions, in

part from differing criteria for malnutrition, from the inaccuracies inherent in characterizing complex GH secretory patterns from a single blood sample, and perhaps to some extent from confounding features such as infection.[150] The normal physiology of nutrient regulation of GH secretion is also complex; amino acids such as arginine stimulate GH release, whereas carbohydrates and fatty acids inhibit GH secretion. Clinical malnutrition results from a combination of dietary deficiencies that could be expected to have variable effects on GH release.

Recently, attention has turned to the effects of fasting on the pattern of GH release from the pituitary. As previously described, GH secretion in humans is episodic, and measurement of GH concentrations on random blood samples, as is typical of field studies, might be expected to give inconsistent results. Ho and coworkers performed frequent blood sampling (every 20 minutes) of fasted volunteers and uncovered an inherent pulsatility in GH secretion in humans that appears to be suppressed in the fed state, perhaps by the negative feedback of IGFs.[151] Over the course of a five-day fast, those authors measured significant increases in GH pulse frequency, maximal pulse amplitude, and 24-hour integrated concentration (Table 21-5). These changes coincided with a decline in concentrations of IGF-I and glucose and a rise in those of free fatty acids and acetoacetate. Subsequent studies led those authors to conclude that fasting-induced enhancement of GH secretion is mediated by an increase in GH-releasing hormone pulsatility and a decrease in somatostatin release.[152]

TABLE 21-5 Enhanced Pulsatility of Growth Hormone Release in Fasting

Measure of Pulsatility	Day 0[a]	Day 1[a]	Day 5[a]
GH peaks (number/24 h)	5.8 ± 0.7	7.3 ± 0.6**	9.9 ± 0.7*
GH peak amplitude (ng/mL)	5.9 ± 1.1	13.1 ± 1.2*	12.3 ± 1.6*
24-hour IGHC (μg/mL/min)	2.8 ± 0.5	7.8 ± 1.1*	8.8 ± 0.8**

GH, growth hormone; IGHC, 24-hour integrated GH concentration.

[a]Frequent venous sampling of six adult male subjects was performed during a control fed day (day 0) and during the first (day 1) and fifth (day 5) days of a fast.

*$P < .005$, compared with day 0; **$P > .0009$.

Serum GH concentrations are elevated in children with both kwashiorkor and marasmus[153–157] but do not allow distinction between the two. Becker and associates[153] observed the magnitude of GH elevation to be inversely related to the severity of protein malnutrition, as judged by

the serum albumin concentration. In their subjects, GH concentrations fell after feeding a protein-rich diet for 36–72 hours. The critical role of protein in the refeeding diet was also demonstrated by Pimstone and colleagues,[157] who found that refeeding with a high-carbohydrate, protein-free diet does not normalize GH concentrations. When caloric deprivation predominates, the GH response appears to be variable; some groups have reported depressed GH concentrations and others have reported GH elevations.[158,159] In short-term malnutrition, the adaptive benefits of elevated GH concentrations are suggested by the known direct effects of GH to increase gluconeogenesis, lipolysis, and fatty acid oxidation. Elevated GH concentrations might be expected to increase glucose production initially, while shifting the organism to a lipolytic, protein-sparing state.

Although short-term protein-calorie malnutrition is associated with elevated GH concentrations, long-standing malnutrition results in depressed GH secretion. Beas and coworkers[160] described a group of infants with low concentrations not only of GH but also of adrenal and thyroid hormones, suggesting generalized pituitary hypofunction. An adaptive function of a hypopituitary state in reducing metabolic activity had previously been proposed by Mulinos and Pomerantz,[161] who likened the malnourished state to a "pseudohypophysectomy."

Clinical Uses of GH in Disease States

GH signaling (as measured by IGF-I concentrations) is reduced in many conditions, including malnutrition, Prader–Willi syndrome,[162] thalassemia major,[163] type 1 diabetes,[164] leukemia,[165] and premature osteoporosis.[166] Also, Laron dwarfism, which results from inactivating mutations in the GH receptor, is characterized by dramatically reduced IGF-I concentrations.[167] The adaptive changes in GH release that accompany malnutrition have prompted a number of studies on the possible uses of GH in promoting anabolism during recovery from operation or injury. Clemmons and coworkers[168] determined the effects of GH injections on obese individuals subjected to hypocaloric diets. Although dietary restriction produced comparable weight loss in subjects receiving GH and those receiving placebo, those treated with GH (0.1 mg/kg every 48 hours) had a significantly lower daily nitrogen deficit than controls. Similar findings were reported by Manson and Wilmore,[169] who administered GH to volunteers receiving hypocaloric PN (Figure 21-4). GH therapy decreased the rate of weight loss and increased nitrogen retention. In

parenterally nourished patients recovering from GI surgery, GH administration has resulted in sustained nitrogen retention,[170,171] suggesting a possible role for this agent in promoting recovery from a variety of catabolic states.

Unfortunately, a series of investigations in critically ill adults with multiorgan system failure in several placebo-controlled, masked trials found that high-dose recombinant human GH treatment produced a twofold increase in mortality, increased the length of stay in hospital, and prolonged the duration of mechanical ventilation.[172] However, similar doses of GH administered to critically ill children with extensive body burns did not increase morbidity or mortality and reduced intravenous albumin and calcium requirements, compared to placebo-treated children. There were increased hyperglycemic episodes and insulin requirements in the treated group, although these were not considered dangerous.[173]

In an animal model of colitis (interleukin-10 knockout mouse), GH injections decreased disease activity, increased weight gain, and reduced apoptosis in the colon. The improvement was mediated by upregulation of a protein called SHP-2, which in turn reduced STAT-3 (see above) activation, while increasing the antiapoptotic protein bcl-2.[174] Similarly, administration or overexpression of IGF-I reduced inflammation and fibrosis in experimental ileocolitis[175] and in toxic hepatitis.[176]

In humans with several gastroenterological conditions, exogenous GH administration has been shown to have beneficial effects. In cystic fibrosis, GH treatment improved growth rate (linear and ponderal) and—by as yet undetermined mechanisms—reduced pulmonary exacerbations.[177,178]

Growth failure in the inflammatory bowel disease Crohn's disease is associated with small bowel involvement, Card-15 mutations, and mutations in granulocyte-macrophage colony stimulating factor (GmCSF). In addition, the liver cell-surface GH receptor is cleaved in the face of endotoxemia, a result of metalloprotease activation by LPS (18323468). In adults with Crohn's disease, a preliminary placebo-controlled trial of GH therapy for four months significantly improved the disease activity index score.[179] In children, inflammatory bowel disease activity was not influenced, but a four-month treatment with GH enhanced the lean body mass, decreased the fat mass, and enhanced linear growth velocity.[180] Denson's group has shown that in children with growth failure secondary to Crohn's disease, treatment with GH plus corticosteroid (compared to corticosteroid controls) was significantly more likely to produce a clinical remission and increased height velocity (PMID 20453679).

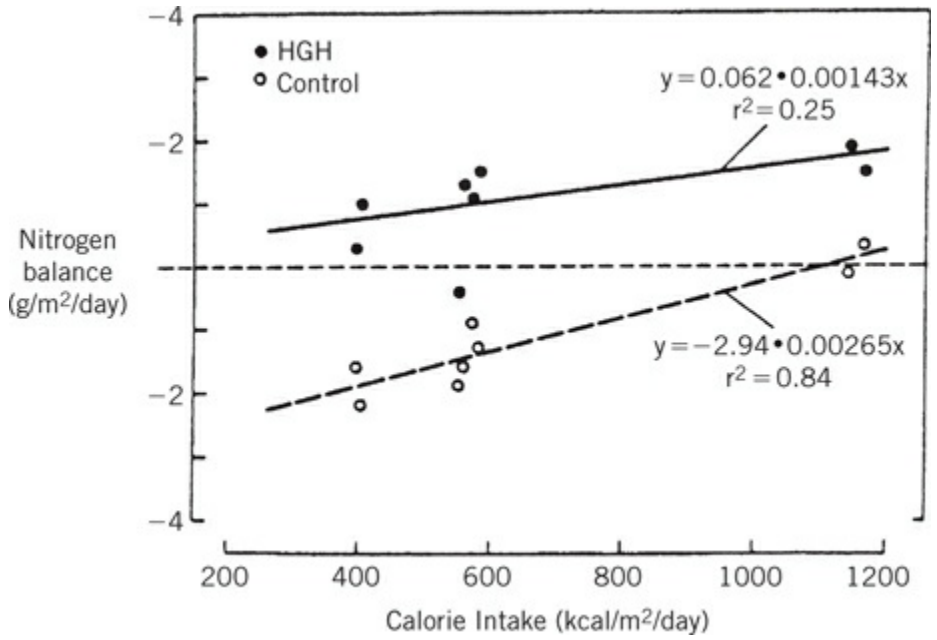


Figure 21-4 Positive nitrogen balance in growth hormone-treated subjects at all levels of hypocaloric calorie provision. Eight male subjects were exposed to various levels of hypocaloric intravenous feeding. Only adequate energy intake resulted in positive nitrogen balance in the controls (*open circles*), whereas growth hormone-treated subjects (*closed circles*) were in positive nitrogen balance at all levels of energy intake. Adapted from Manson and Wilmore.[169]

Substantial worldwide experimental support has emerged for GH treatment of infants born SGA, a condition also called intrauterine growth retardation (IUGR). This condition, highly prevalent in developing countries, is associated prevalent in developing countries, is associated with low body fat at birth, low IGF-I levels up to age 2, and subsequently abnormal catch-up growth characterized by increased adiposity, insulin resistance, and low-grade inflammation.[181] IUGR eventually tends to be associated with subsequently high rates of coronary heart disease, high blood pressure, diabetes, and high cholesterol.[182] Treatment of children manifesting IUGR with daily GH shots increased linear growth velocity in many studies, while delaying the onset of puberty and exhibiting few if any side effects.[183–187] However, treatment for years was required, higher doses were more effective, and younger children (3–5 years old) were more responsive to treatment.[188] Concomitant treatment with a gonadotropin-releasing hormone analog (to postpone puberty) and GH

resulted in increased height, lean body mass, and bone mineral density (PMID 23125290). Of major concern is the global scope of this problem with IUGR, however, because IUGR affects 20% of children in Malawi[189] and 25% of children in Pakistan.[190] Clearly, ethical problems will arise as to which children can be treated with this expensive and long-term therapy.

IGFs in Malnutrition

IGF Bioactivity, IGF Inhibitors, and IGF-BPs

Despite elevated GH levels in fasting or short-term malnutrition, the production of IGF-I is dramatically reduced in malnutrition. Studies using somatomedin bioassays were the first to indicate that serum IGF bioactivity falls with malnutrition. More recent data using highly specific radioimmunoassays indicate that the depressed bioactivity of serum from fasted or malnourished individuals results from a dramatic fall in IGF-I concentrations, minimal if any change in IGF-II concentrations,[191] and changes in IGF-BPs (Table 21-6).

Measuring serum somatomedin bioactivity in a sulfation factor assay, Grant and associates were among the first to show a nutritional influence on somatomedin activity[192]; they found low levels in the serum of malnourished South African children despite elevated GH concentrations. Those authors found both hormones to return toward normal by the ninth day of refeeding. Similar results using somatomedin bioassays were forthcoming from other malnourished populations,[193] including malnourished Thai[194] and Nigerian[195] infants and children and patients with celiac disease[196] and anorexia nervosa.[197]

Bioassays of IGF activity in blood, although relatively nonspecific as to which of the two IGFs is being measured, allow the detection of physiologically important IGF inhibitors in serum. Much of the older data on somatomedin inhibitors can be attributed to changes in the concentration of known IGF-BPs. Increased concentrations of inhibitors have been found in the serum of malnourished children and hypopituitary adults[193,198,199] and in serum from starved, hypophysectomized, or diabetic rats.[200,201] Salmon and colleagues[202] partially purified a 27- to 40-kDa, heat-labile inhibitor from fasted rat serum. Recent studies have suggested that this inhibitor may be the serum-binding protein, IGF-BP-1[203] (see Table 21-4). Serum concentrations of BP-1 almost double with fasting and decrease after meals.[204] In addition, they are elevated in diabetes,[205] consonant with the proposed role of insulin in the regulation

of BP-1 production. Phillips and associates have shown that concurrent with the fall in IGF activity of rat serum during fasting, total IGF inhibitor activity rises.[206] The IGF inhibitory activity of serum appears to be more nutritionally sensitive than the IGF activity itself. After six hours of refeeding, the IGF inhibitor activity had fallen significantly, but there was little change in IGF activity; both returned to normal with continued refeeding.

TABLE 21-6 Effects of Undernutrition on the GH–IGF Axis

Increased GH secretion
GH resistance in peripheral tissues
Decreased GH receptor mRNA
Decreased GH binding
Diminished postreceptor signaling (P)
Decreased somatomedin bioactivity
Decreased IGF-I production
Decreased IGF-I gene transcription
Diminished IGF-I mRNA stability (P)
Diminished IGF-I mRNA translation (P)
Normal IGF-II production
Increased IGF-BP-1 production
Decreased IGF-BP-3 production (P)
Increased IGF-I clearance
Increased IGF inhibitor production
Decreased tissue sensitivity to IGF-I

GH, growth hormone; IGF, insulin-like growth factor; IGF-BP, insulin-like growth factor binding protein; mRNA, messenger ribonucleic acid.

IGF-I and IGF-II in Malnutrition

As measured by specific radioimmunoassays, IGF-I concentrations drop dramatically with fasting in adult volunteers,[207] while those of IGF-II do not.[191] Clemmons found that fasting of overweight adult volunteers for 10 days produced a drop of IGF-I concentrations to hypopituitary levels; refeeding produced a return toward normal[207] (Figure 21-5). The decline in serum IGF-I correlated with nitrogen balance. In some physiologic circumstances, however, the fall in serum IGF-I concentrations becomes dissociated from the changes in nitrogen balance. Smith and coworkers[208] studied six conditioned athletes and concluded that strenuous exercise, which produces negative caloric balance, causes as much of a fall in serum IGF-I as does comparable caloric restriction without exercise, but a much lower nitrogen loss.

In contrast to the apparent insensitivity of serum IGF-II to fasting in adults,[191] Soliman and associates[209] have documented depressed IGF-II concentrations in children with severe marasmus and marasmic kwashiorkor. These levels returned to normal with refeeding. The magnitude of IGF-II depression was clearly not as great as that of IGF-I, and the IGF-II concentrations in sera of children who were underweight or had only kwashiorkor were not depressed. Thus, severe or long-lasting malnutrition in children may eventually depress IGF-II production, although IGF-I remains the more acutely sensitive to malnutrition. In addition, IGF-I levels may reflect imbalanced or abnormal nutrition. For example, infants with short bowel syndrome maintained on PN who developed liver disease had low levels of IGF-I.[210]

Serum IGF-I as an Index of Nutritional Status

The striking correlation between (as yet unidentified) aspects of nutritional status and serum IGF-I concentrations has led some investigators to suggest that the latter might provide a highly sensitive and quantitative way of following nutritional status. Clemmons and colleagues found that IGF-I increased by 170% over 10 days in six malnourished patients with GI disorders that necessitated either total parenteral or tube feedings.[211] All patients were in positive nitrogen balance when first measured on the second day of refeeding. Other commonly used indices of nutritional status, such as prealbumin, retinol-binding protein, and transferrin, fell much less dramatically and normalized much more slowly. Unterman and coworkers came to similar conclusions when they compared IGF-I concentrations with other indices in 37 generally elderly hospitalized patients with a variety of neoplastic, GI, and neurologic diseases.[212] The

IGF-I was more depressed than albumin or transferrin in patients who had no reduction in either triceps skinfold thickness or mid-arm muscle circumference. In six patients given nutritional support, IGF-I concentrations rose by more than 70% in each patient, while the lymphocyte count and transferrin concentration rose in only four patients and albumin concentration in only one. Serum IGF-I measurements have also been suggested as a means to follow the progress of children and adolescents with chronic inflammatory bowel disease.[213] In a study of critically ill patients, serum concentrations of prealbumin, retinol binding protein, and transferrin levels were strongly correlated with levels of IGF-I, IGF-II, IGF-BP3, and ALS.[214]

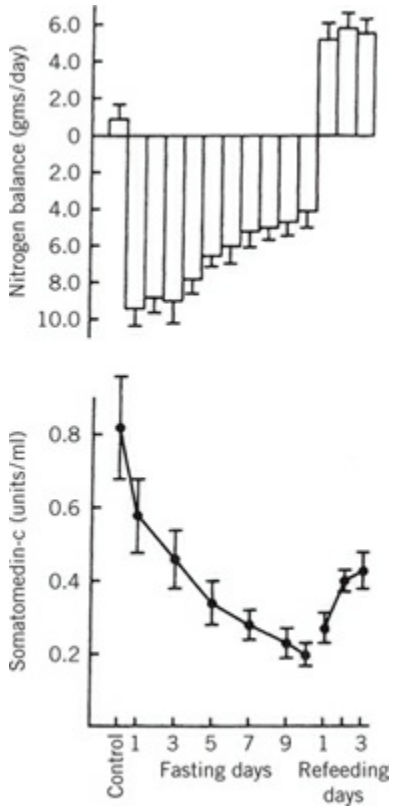


Figure 21-5 The effects of fasting on plasma somatomedin-C (IGF-I) and nitrogen balance. The subjects were seven males 20%–70% above ideal body weight. During the control period, they were fed a 1500 kcal diet for three days. Between days 4 and 13 they fasted, receiving only water, vitamins, and potassium chloride. Refeeding consisted of resumption of the control diet. Each point is the mean ± SEM. Adapted from Clemmons et al.[207]

Mechanisms of IGF Regulation by Nutrients

A number of studies have attempted to determine the specific components of the refeeding diet that regulate IGF-I production in recovery from an acute fast. When normal-weight human volunteers were fasted for five days and then refed with a normal diet, a diet that was isocaloric but protein deficient, or a diet deficient in both protein and calories, Isley and associates[215] concluded that intake of adequate quantities of both protein and calories is critical for the restoration of IGF-I after a fast. Animal studies have indicated that the roles played by protein and calorie intake in regulating serum IGF-I concentrations may be developmentally influenced. Prewitt and coworkers[216] placed 3-week-old rats on diets in which the protein content was varied (5%, 10%, or 15% lactalbumin) at one of three levels of energy intake (100%, 75%, or 50% ad libitum). After two weeks, protein intake played the major role in regulating IGF-I concentrations, whereas after nine weeks, both protein and caloric intake were important.

The malnourished state is one of resistance to the effects of GH. As noted previously, GH secretion increases in the fasted state, while serum concentrations of IGF-I fall. Merimee and colleagues found that GH administered to fasted, GH-deficient subjects induced a minimal increase in serum IGF-I concentrations in comparison with the 10-fold increase GH induced in normally fed, GH-deficient subjects. Studies in fasted[217–219] and diabetic[219] rats have correlated the fall in serum IGF-I concentrations with a decline in the number of hepatic GH receptors, suggesting that a primary effect of malnutrition, perhaps mediated by decreased insulin concentrations, is to decrease GH receptors and consequently IGF-I production. Indeed, fasting results in decreased GH receptor mRNA in the liver.[220,221] The situation may differ in protein-deprived rats, in whom a postreceptor defect in GH action has been suggested.[148] The net result of fasting is a decrease in the concentrations of IGF-I mRNA in the liver and other tissues.[220] This results from decreased transcription of the IGF-I gene, although the effects of protein deprivation may further influence IGF-I mRNA stability and also decrease the translation of IGF-I mRNA into peptide.[222]

The fasted or malnourished state is also one of resistance to the action of IGF-I. Thissen and associates[223] showed that, in contrast with normally fed, hypophysectomized rats, an infusion of recombinant IGF-I into protein-restricted rats did not stimulate an increase in weight gain or linear growth, despite restoring normal serum concentrations. Somewhat

surprisingly, this nutritionally induced IGF-I resistance does not result from a decrease in IGF-I receptors at target tissues; instead, most tissues show increased IGF-I binding.[224]

IGF-BPs in Malnutrition

Among the IGF-BPs, BP3 is unique in its diurnal rhythm that appears to be regulated by food intake. IGF-BP1 is rapidly suppressed following a meal, and it acutely rises in response to hyperglycemia. In this context, IGF-BP1 may serve as a glucose counter-regulatory hormone by further depressing free IGF concentrations in the hypoglycemic state. In addition to its regulation by nutrients, IGF-BP1 is stimulated by glucocorticoids and by cytokines such as TNFα and various interleukins. Nutritional status appears to also influence the phosphorylation status of IGF-BP1. Baxter has postulated that increases in IGF-BP-2, BP-4, and BP-6 during acute illness could serve to redistribute IGFs from ternary complexes in the serum to binary complexes in the extracellular space and make them more available to the tissues.[91]

Insulin in Malnutrition

With fasting or protein-calorie malnutrition, plasma insulin concentrations are reduced, although in some instances high and normal levels have been reported.[209] Some of these discrepancies might be explained by the observations of Lunn and associates[155] that in kwashiorkor there is an early phase of hyperinsulinism that is followed by insulinopenia. Diminished insulin release results in impaired tolerance to glucose or an amino acid load.[225] Pimstone and coworkers[226] noted that the insulin response to oral glucose is abnormal in malnourished children but that intravenous glucose elicits a normal response, suggesting that deficiencies of the so-called “incretins” may be important in determining the response to malnutrition. Those authors also demonstrated that the impaired insulin response to a glucose load in kwashiorkor could be ameliorated by the addition of potassium to the diet. The relationship of malnutrition and diabetes mellitus has been reviewed by Rao,[227] who concluded that a malnourished condition can influence the diabetogenicity of other agents, including dietary toxins, drugs, and viruses, but does not directly cause diabetes. In clinical practice, an initial presentation with malnutrition, dehydration, and hypoglycemia (<40 mg/dL) has been associated with a high mortality (>40%) for children with diarrheal dehydration.[228]

Psychosocial Growth Failure

The interactions of nutritional and hormonal influences have been extensively studied in cases of "psychosocial growth failure" or "deprivation dwarfism," terms used to describe the growth failure that can accompany severe emotional deprivation. Early in this century, it was recognized that emotional deprivation could be associated with growth retardation in institutionalized children.[229] Subsequent studies revealed that such children need not be in an institutional setting and that an impoverished emotional environment in the home is sufficient. Affected children have a voracious appetite with bizarre eating and drinking behavior such as foraging, drinking from the toilet bowl, and gorging. In addition, delayed speech, foul-smelling stools, a protuberant abdomen, and characteristic neurologic postures have been described.[229,230]

Patton and Gardner[231] speculated that emotional deprivation could produce growth retardation in a variety of ways, including failure of the care giver to provide adequate nutrition, depression-induced anorexia in response to the emotional environment, altered GI function, or changes in metabolism. Considerable attention has been paid to whether the growth failure and endocrine-metabolic changes of psychosocial dwarfism are the result of emotional deprivation alone or are secondary to impaired caloric intake.

A large body of evidence suggests that a hypopituitary state can develop in some children with emotional deprivation. Talbot found significant malnutrition in a group of 51 children with nutritional growth failure of no identifiable etiology and postulated that they might have "functional hypopituitarism"as a feature of "anorexia secondary to emotional disturbance.[132] Twenty years later, Powell and colleagues[233,234] studied emotionally deprived children and demonstrated diminished reserves of adrenocorticotropic hormone (ACTH) and GH, which were normalized when the affected children were removed from their adverse surroundings. The authors found little evidence of caloric deprivation in their patients and suggested that psychic factors caused their hypopituitarism. Miller and colleagues[235] demonstrated a loss of GH secretory episodes during a six-hour period of blood sampling in four children with psychosocial deprivation. In one child, GH secretion normalized after hospitalization and rehabilitation. Despite the apparent state of GH deficiency, attempts to induce growth in such children by administration of GH have been unsuccessful.[236,237]

Psychological factors were also invoked by Widdowson to explain her

observations on the growth of children in two German orphanages after World War II.[238] Orphans under the care of a stern and forbidding supervisor who chose meal times to rebuke the children gained weight at about one-third the rate of children at another, more hospitable orphanage despite comparable food intake. When the supervisor of the first orphanage was transferred to the second, the pattern reversed, and the new children under her care gained weight poorly.

A counter theme to the hypothesis of a strictly emotional etiology of psychosocial growth failure has been the suggestion that underfeeding by care givers is the primary cause of growth failure in these children. From their studies on emotionally deprived infants between 3 and 24 months of age, Whitten and colleagues observed dramatic weight gain in 11 of 13 children when the infants were given adequate nutrition in a hospital setting designed to reproduce the impoverished emotional ambience of their homes.[239] During a control period in which these children were fed equal amounts of food but also given a high level of mothering care, their weight gain was no greater. Other studies have found elevated fasting levels of GH in a number of infants and children with emotional deprivation, leading to the speculation that these infants suffered from food deprivation as well as emotional deprivation.[240]

Contemporary dietary and health misconceptions have created a modern-day version of the growth-retarded child from a turn-of-the-century foundling house or war orphanage. Pugliese and colleagues described a group of 14 adolescents who presented with short stature and malnutrition from self-imposed caloric restriction.[241] These children showed no evidence of overt psychiatric disease or anorexia nervosa but had restricted their food intake from a fear of obesity. Similar findings were reported in a group of younger children, 7–22 months of age, who presented with nutritional failure to thrive secondary to parental restriction of food arising from a fear of obesity, cardiovascular disease, or unhealthy eating habits.[242]

It is reasonable to propose from these somewhat disparate observations that the mechanisms involved in the growth failure associated with emotional deprivation form a spectrum (Figure 21-6). At one extreme are children in whom growth failure is nutritional in origin, whether from limited access to food, from emotionally induced anorexia, or from altered GI function. In these children, endocrine alterations are adaptive and occur in response to malnutrition. At the other extreme of the spectrum are those children whose hormonal and metabolic alterations are emotionally induced. The growth failure of these children is probably mediated by

hormone deficiencies rather than by inadequate nutrition. It would appear that many of the children with clinical features of psychosocial growth failure combine features of both extremes of the spectrum.

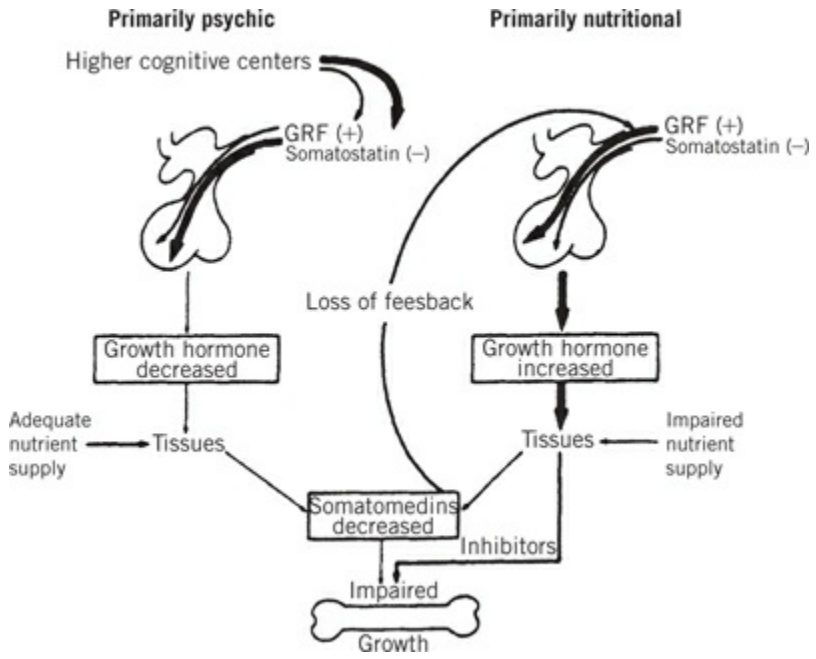


Figure 21-6 Psychosocial growth failure. Two possible mechanisms of psychosocial growth failure are proposed: primarily psychic (neuroendocrine) and nutritional. Under largely psychologic influences, pituitary growth hormone secretion is diminished in the face of normal nutrition (left). When undernutrition predominates, the endocrine changes observed are adaptive ones (right). Somatomedin production is reduced, IGF-binding proteins are increased, and growth hormone secretion is increased.

THYROID HORMONES

Thyroid Hormones and Normal Growth

The actions of the thyroid hormones, like those of insulin, can be broadly separated into directly metabolic ones and those that result from thyroid induction of other genes that regulate cell growth and function. Increased oxygen consumption, heat production, and accelerated metabolism constitute the allergenic effects of the thyroid hormones.[243] The mechanism by which thyroid hormones affect these actions is not clearly understood. Thyroid hormone signaling increases basal thermogenesis by

increasing ATP turnover and decreasing the efficiency of ATP synthesis. Decreased efficiency of ATP utilization has been linked to the generation of a proton leak across the inner mitochondrial membrane.[244] Unlike peptide hormones, which bind to cell membrane receptors and regulate cell function and gene transcription via second messengers, thyroid hormone receptors exist in the cytoplasm and directly regulate the transcription of other genes by binding to regulatory regions of DNA in the genome.[245] Many of the developmental effects of the thyroid hormones may result from induction of the genes for other hormones and growth factors, such as epidermal and nerve growth factors.[246]

Thyroxine (T4) is the major hormone secreted by the thyroid, but it is deiodinated at its outer ring to produce triiodothyronine (T3), which is the metabolically active hormone (Figure 21-7). Deiodination occurs both in the thyroid gland and at peripheral sites such as liver, kidney, and brain. Thyroxine may also be deiodinated at its inner ring to form reverse T3 (rT3), which is metabolically inactive. Regulation of the relative activity of the inner and outer ring deiodinases to vary the concentrations of T3 and rT3 is a major control point in the thyroid economy. Thyroxine release from the thyroid is stimulated by thyroid-stimulating hormone (TSH, thyrotropin), which in turn is released from the anterior pituitary under the influence of the hypothalamic modulator, thyrotropin-releasing hormone (TRH). Perhaps in conjunction with T4, T3 feeds back to inhibit TSH secretion.

The influence of thyroid hormones on growth occurs at many levels. Within the cell, they have profound effects on metabolic processes, including the induction of mitochondrial enzymes and an increase in Na+, K+-ATPase activity. Thyroid hormones influence the GH–IGF axis, as evidenced by the fact that hypothyroid rats have a diminished pituitary content of GH, low circulating GH levels, and depressed IGF levels.[246–249] T3 has been shown to stimulate transcription of the GH gene.[250] In contrast to animal models, hypothyroid children do not consistently demonstrate subnormal GH release.[111,251,252] Furthermore, the growth-retarding effects of hypothyroidism in rats are not reversed by GH treatment. In addition to a GH-mediated influence on IGF secretion, thyroid hormones augment IGF-I actions on cartilage growth,[252] and their absence is evident at the growth plate cartilage by altered morphology.[253] In addition, some thyroid hormone actions on growth plate chondrocytes appear to be IGF-mediated, as the mitogenic actions of T3 on cartilage growth can be blocked with an antibody to IGF-I. In contrast, the effects of

T3 on cartilage morphology and functional maturation do not appear to be IGF-mediated, direct ones.[254]

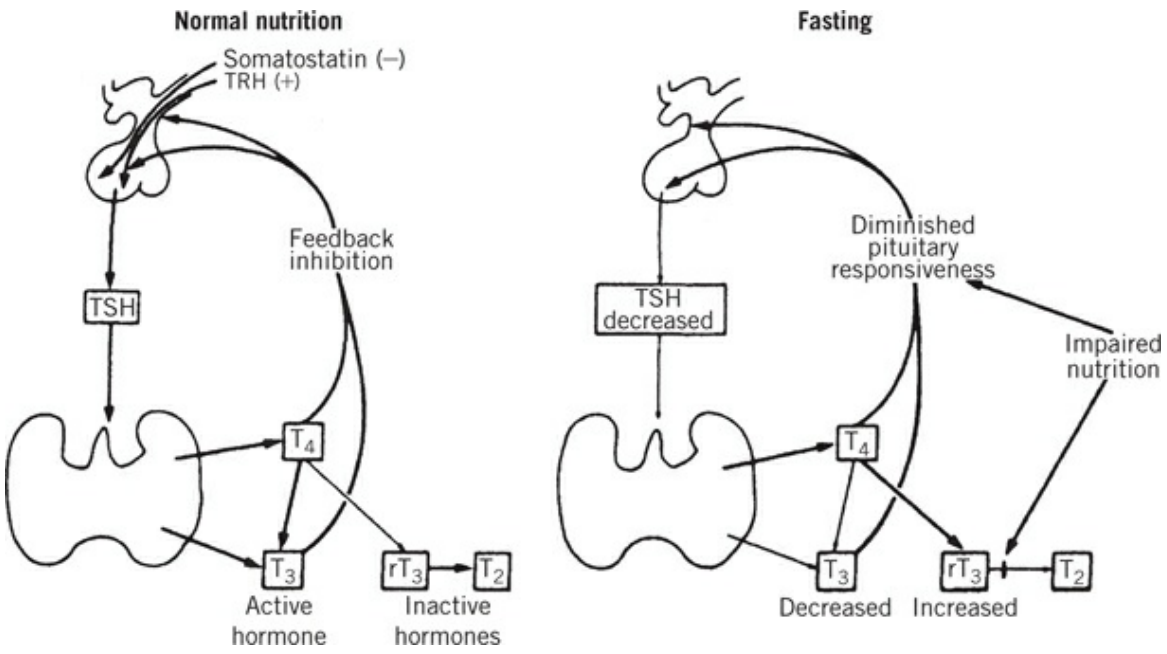


Figure 21-7 The pituitary–thyroid axis. A, When nutrition is normal, thyroxine (T4) is converted primarily to the calorigenic hormone triiodothyronine (T3) in the thyroid and peripheral tissues. Levels of the inactive hormones reverse T3 (rT3) and T2 are relatively low. B, After fasting, reverse T3 concentrations rise primarily because of decreased degradation to T2, and to some extent through preferential conversion of T4 to rT3. Fasting also diminishes the pituitary responsiveness to hypothalamic factors. Thyroid stimulating hormone (TSH) levels do not rise as in other forms of hypothyroidism. (+) stimulatory action; (−) inhibitory action.

Thyroid Hormones and Malnutrition

Fasting and malnutrition induce a hypothyroid-like state characterized by a diminished rate of T3 production and, consequently, a depressed heart rate and basal metabolic rate, as well as a diminished loss of nitrogen.[160,255,256] Similarly, bradycardia is a feature of anorexia nervosa. Concomitant with the fall in T3 in malnutrition is a depression of sympathetic nervous system activity, as measured by norepinephrine secretion.[257] Within 48 hours of a fast, T3 concentrations fall into the hypothyroid range while concentrations of T4 change little, and those of rT3, the inactive

metabolite of T4, rise (Figure 21-7). These changes result from a starvation-induced inhibition of the deiodination of T4 to T3, while the serum half-life of rT3 is prolonged by preventing its deiodination to T2. In addition, malnutrition also results in a diminished synthesis of the plasma thyroid-binding proteins, also leading to depressed thyroid hormone levels.

The low levels of thyroid hormones in the undernourished state are likely to be an adaptive mechanism meant to diminish the extent of catabolism. Silva have shown that when T3 is administered to fasting subjects sufficient to maintain T3 levels in the normal range, nitrogen losses are accentuated.[244] Despite a low serum T3, TSH secretion is not increased in fasting, and the TSH response to TRH is either normal or somewhat blunted.[256,258] The condition of depressed levels of metabolically active thyroid hormones without elevation in TSH in malnutrition or illness has often been called the "euthyroid sick syndrome" or "nonthyroidal illness syndrome," and recent developments in the field have cautiously supported the administration of thyroid hormones to individuals in this physiologic state.[259,260]

The depressed T3 levels of the fasted state can be corrected by refeeding as little as 100 kcal of carbohydrate, whereas the protein or fat content of the diet does not significantly affect the recovery of T3.[261] As little as 50 g of carbohydrate will prevent the effect of dietary restriction on serum T3 levels.[87] As in those subjected to acute starvation, chronically malnourished children and adults have depressed serum T3 and elevated rT3 concentrations. However, the TSH concentrations are either normal or elevated in chronic malnutrition, and the TSH response to TRH is exaggerated.[156,215,262]

CORTISOL AND SEX STEROIDS

Cortisol in Intestinal Maturation

Development of critical intestinal absorptive transporters and digestive enzymes is a prelude to somatic growth. There is a striking maturation of absorptive function of the intestine following birth, with ample evidence to suggest that glucocorticoids regulate the process. Most of the data on the regulation of gut maturation have been derived from studies in rats and other small mammals. Coincident with the period of weaning and, consequently, a decreasing reliance on lactose and other milk sugars for nourishment, there is a fall in the high activity of lactase in the small intestine and a rise in the activity of intestinal sucrase, maltase, and

pancreatic amylase.[263] In addition, there is a loss of the mechanisms by which the ileal and jejunal mucosa take up macromolecules and transport immunoglobulins. Delay of weaning has no significant effect on the timing of these changes. In contrast, deprivation of pituitary hormones by hypophysectomy in the newborn period elicits a marked delay.[264,265] These observations have led to studies of the role of pituitary-derived or pituitary-regulated hormones in the maturation of gut function.

The experimental data support roles for both glucocorticoids and thyroid hormones in intestinal maturation. Coincident with the enzymatic changes in the rat gut at the end of the second week of life, there is a prominent rise in circulating corticosterone. Breast milk might provide an additional source of glucocorticoids.[266] Glucocorticoid administration to preweanlings results in early maturation of intestinal enzymes such as sucrase[263] and of pancreatic amylase, bile acid synthesis, and transport phenomena such as bile salt absorption.[267] The dependence on glucocorticoids is not absolute; however, as adrenalectomy on day 9 of life does not prevent the appearance of sucrase or delay the maturation of sucrase to adult levels of activity.[263] A comparable role for thyroid hormones has also been suggested, although it appears that the ability of thyroxine to induce maturation of the intestinal enzymes requires the presence of intact adrenal glands. Administration of thyroxine to rats results in increased blood concentrations of corticosterone. Thus, the observed effects of thyroxine may be through induction of glucocorticoids.[263]

Cortisol in Malnutrition

Reflecting the metabolic stress, malnutrition in humans and laboratory animals results in elevated cortisol concentrations, with an abolition of the normal diurnal rhythm and a slowed clearance of exogenous cortisol.[268] Fasting also depresses production of cortisol-binding proteins, resulting in a further elevation of free glucocorticoids.[269] Despite the glucocorticoid excess, the adrenal glands remain suppressible with dexamethasone and responsive to ACTH in the malnourished state.

A prominent metabolic effect of cortisol is the diversion of substrates to the liver for protein synthesis. Rao and coworkers[270] have demonstrated that infants with marasmus have higher cortisol levels than do infants with kwashiorkor and that the response of cortisol to ACTH is better preserved in marasmus. Those authors postulated that the development of kwashiorkor is coincident with failure of the adrenal gland to secrete

adequate cortisol to maintain normal protein levels. In rats, the biochemical manifestations of kwashiorkor resulting from a low-protein diet can be reversed by glucocorticoids.[271] As previously indicated, glucocorticoids have a deleterious effect on cartilage growth and act as IGF antagonists in somatomedin bioassays. In his studies on malnourished Nigerian children, Smith[195] postulated that elevated cortisol, acting as an inhibitor of IGF action at the epiphyses was a major contributor to the poor growth of his subjects.

Reproductive Function in Malnutrition

The potent gonadal steroids testosterone and estradiol are primarily responsible for the establishment of reproductive fitness. Other, weaker androgenic hormones, however, which are produced by the gonads and adrenal glands of both sexes, are important in the maintenance of normal anabolism. These include androstenedione, dehydroepiandrosterone, and its metabolite dehydroepiandrosterone sulfate. Whereas reproductive functions are expendable and potentially damaging during times of nutrient deficit, considerable attention has been paid to the importance of weaker androgenic hormones, or anabolic steroids, as possible therapeutic agents for the recovery from catabolic states.

In malnourished adults, hypogonadism often develops, and the secretion of sex steroids by the gonads decreases. In malnourished children, the onset of puberty is delayed.[272] Men suffering from chronic protein-calorie malnutrition often experience gonadal failure. In adults, nutrients appear to influence reproduction at both the hypothalamic and gonadal levels. In a group of Indian men suffering from malnutrition, Smith and colleagues[273] found low-plasma testosterone and elevated gonadotropins, indicating impaired testicular function. During recovery, these men continued to have elevated levels of luteinizing hormone and a subnormal release of testosterone to chorionic gonadotropin (hCG). In contrast, Klibanski[274] found evidence for hypothalamic hypofunction when mildly obese men were fasted for 10 days.

In addition to the effects of malnutrition in delaying puberty, a number of studies on young athletes have suggested a connection between body composition and reproductive fitness. In young female athletes and dancers who train extensively before puberty, menarche can be delayed by several years.[275] In some instances, the delay in menarche can be related directly to the duration of training prior to menarche: five months for every year of training.[276] These observations led Frisch and McArthur[277] to

propose that normal menstrual function depends on the attainment of at least 22% body fat. This model assumed that some aspect of body fat content signals permission for the secretion of gonadotropic hormones to trigger menstruation. The identification of leptin as the adipocyte-derived product of the ob gene, has shed some light on the relationship between body fat and puberty. Leptin levels in the blood increase with adiposity. Leptin receptors have been identified in the anterior pituitary and gonads, and leptin accelerates the pulsatility of gonadotropin releasing hormone as well as being critical for the synthesis of adrenal androgens.[278]

Anabolic Steroids as Therapeutic Agents in Catabolic Disease

Despite all attempts, it has been impossible to synthesize steroids with anabolic actions that are divorced from their androgenic (masculinizing) ones. Coupled with their anabolic effects, these hormones also have behavioral effects such as increased aggressiveness and an enhanced appetite. Like all androgens, synthetic anabolic steroids suppress gonadotropin production and result in a lipogenic profile of cholesterol metabolism. High doses can alter liver function and result in hepatocellular carcinoma. Anabolic agents have been shown to decrease nitrogen loss after minor surgery,[279] but have had no consistent effect on weight gain in geriatric patients[280] or in patients with chronic renal failure.[281]

There has been considerable popular attention in recent years to the use of anabolic steroids to increase performance in bodybuilding and other sports. Well-controlled studies in adult male athletes are few, and the results are conflicting.[282] Those whose performance stands to benefit the most from increased androgenic activity are those who produce relatively low levels of androgens endogenously: females, prepubertal children, and adolescents. These are the groups in whom additional adverse effects such as virilization, balding, sexual precocity, and premature closure of the epiphyses would be the most damaging. At this time, there appears to be no clear role for anabolic agents in the treatment of catabolic states. Their use in augmenting athletic performance entails significant medical risk, including hyperlipidemia, glucose intolerance, liver dysfunction, thromboembolic events, and mood swings.

SUMMARY AND CONCLUSIONS

The humoral controls on growth are highly sensitive to the nutritional

milieu and are programmed to alter and even abort the body's developmental scheme as required during conditions of nutritional want or excess. The specific components of the diet that are monitored by the various nutritionally sensitive endocrine systems are still not known. However, our understanding is growing as to how nutrients can alter the expression of specific genes that encode key regulatory hormones, growth factors, and enzymes. A careful assessment of physical growth and maturation can serve as a sensitive and accurate index of nutritional status and disease, both for individuals and for whole populations.

The capacity of the growing organism to accommodate changes in the availability of nutrients by slowing or halting growth is a generally reversible process, designed to permit adaptation to a changing nutrient supply. However, the ability to resume normal growth after the correction of metabolic insults is dependent on the nature of the insult, its duration, and the developmental phase of the organism during which it occurs. The damage incurred during certain susceptible periods of growth may be irreversible, perhaps owing to a reduction in the number or critical organization of the cells that make up key tissues. The mechanisms by which a tissue or organism can monitor its own size and correct discrepancies with its "intended" size are unknown. They probably involve coordination between mechanisms for catch-up that are extant in individual tissues and regulatory centers in the CNS; classic hormones probably do not play a key regulatory role.

Recent years have brought the discovery of new hormone systems with close ties to the nutritional status of the organism. The discoveries of leptin, an appetite suppressant produced by adipose tissue, and ghrelin, an appetite stimulant produced in the alimentary tract, shed new light on how the body monitors its nutritional status. Other adipocyte-secreted substances such as Acrp30 and resistin may contribute to the insulin-resistant state of obesity. We have an abundance of candidate molecules to begin to elucidate the mechanisms involved in obesity and diabetes and to suggest therapeutic strategies against these disorders.

From our current vantage point, the metabolic changes of undernutrition are adaptive ones, designed to ensure survival until adequate reserves exist to permit growth and reproduction again. In some instances, however, morbidity may actually result from maladaptive endocrine changes. The development of kwashiorkor, for example, may result when the adrenal gland is no longer able to maintain adequate production of glucocorticoid hormones to facilitate withdrawal of proteins from structural stores, such as muscle.

A better understanding of the normal regulation of hormones and growth factors in conditions of altered nutrition has suggested new diagnostic and therapeutic approaches. Administration of GH to individuals in catabolic states may hasten their recovery. Studies of anabolic steroid hormones in catabolic states have generally not been promising.

The future challenge is to better delineate the full spectrum of humoral and neuroendocrine changes that accompany nutritional alterations and to define those components of "nutrition" that are the actual signals that can regulate the production and actions of hormones and growth factors. The accelerated rate at which "new" hormone systems have been discovered illustrates the fact that we have not yet identified all of the relevant humoral control systems, let alone understand them. The characterization of genes identified by the Human Genome Project and the advent of new technologies such as proteomic analysis may allow us to finally identify all of the critical components of the interface between nutrition and cellular control. At this time, the measurement of growth factors, their receptors, and their downstream signaling events within tissues is an invasive proposition and of little clinical practicality. When the physiology of these substances is better understood, however, the promise for therapeutic intervention will be great.

REFERENCES

1. Trowbridge FL, Marks JS, deRomana GL, et al. Body composition of Peruvian children with short stature and high weight-for-height. II. Implications for the interpretation of weight-for-height as an indicator of nutritional status. *Am J Clin Nutr*. 1987;46:411–418.
2. Bielicki T, Szczotka H, Charzewski J. The influence of three socio-economic factors on body height in Polish military conscripts. *Hum Biol*. 1981;53:543–555.
3. Greulich WW. A comparison of the physical growth and development of American-born and native Japanese children. *Am J Phys Anthropol*. 1957;15:489–515.
4. Greulich WW. Some secular changes in the growth of American-born and native Japanese children. *Am J Phys Anthropol*. 1976;45:553–568.
5. Feyrter F. Uber *die Peripheren Endokrinen (Parakrinen) Drusen des Menschen* (The peripheral endocrine [paracrine] glands of the human body). Wein: W Maudrich; 1953.
6. Sporn MB, Todaro GJ. Autocrine secretion and malignant transformation of cells. *N Engl J Med*. 1980;303:878–880.
7. Barnard JA, Beauchamp RD, Russell WE, et al. Epidermal growth factor-related peptides and their relevance to gastrointestinal pathophysiology. *Gastroenterology*. 1995;108:564–580.
8. Werb Z. ECM and cell surface proteolysis: regulating cellular ecology. *Cell*. 1997;91:439–442.

9. Anklessaria P, Teixido J, Laiho M, et al. Cell–cell adhesion mediated by binding of membrane anchored transforming growth factor alpha to epidermal growth factor receptors promotes cell proliferation. *Proc Natl Acad Sci U S A*. 1990;87:3289–3293.
10. Re RN. The origins of intracrine hormone action. *Am J Med Sci*. 2002;323:43–48.
11. O'Malley BW. Did eucaryotic steroid receptors evolve from intracrine gene regulators? *Endocrinology*. 1989;125:1119–1120.
12. Russell WE, Van Wyk JJ. Peptide growth factors. In: DeGroot LJ, ed. *Endocrinology*, 3rd ed. Philadelphia, PA: WB Saunders; 1995:2590–2623.
13. Holley RW. A unifying hypothesis concerning the nature of malignant growth. *Proc Natl Acad Sci USA*. 1972;69:2840–2841.
14. Holley RW. Control of growth of mammalian cells in culture. *Nature*. 1975;258:487–490.
15. Barnes D, Sato G. Serum free cell culture: a unifying approach. *Cell*. 1980;22:649–655.
16. Barsh GS, Cunningham DD. Nutrient uptake and control of animal cell proliferation. *J Supramol Struct*. 1977;7:61–77.
17. Eagle H. Metabolic controls in cultured mammalian cells. *Science*. 1965;148:42–51.
18. Gospodarowicz D, Moran JS. Growth factors in mammalian cell culture. *Ann Rev Biochem*. 1976;45:531–558.
19. Temin HM, Pierson HM, Dulak NC. The role of serum in the control of multiplication of avian and mammalian cells in culture. In: Rothblat GH, Cristofalo JJ, ed. *Growth, Nutrition and Metbolism of Cells in Culture*. New York: Academic; 1972:1.
20. McKeehan WL, McKeehan KA. Serum factors modify the cellular requirement for Ca2+, K+, Mg2+, phosphate ions, and 2-oxycarboxylic acids for multiplication of normal human fibroblasts. *Proc Natl Acad Sci U S A*. 1980;77:3417–3421.
21. McKeehan WL, McKeehan KA. Extracellular regulation of fibroblast multiplication: a direct kinetic approach to analysis of role of low-molecular-weight nutrients and serum growth factors. *J Supramol Struct*. 1981;15:83–110.
22. Rizzino A, Rizzino H, Sato G. Defined media and the determination of nutritional and hormonal requirements of mammalian cells in culture. *Nutr Rev*. 1979;37:369–378.
23. Wu R, Sato GH. Replacement of serum in cell culture by hormones: a study of hormonal regulation of cell growth and specific gene expression. *J Toxicol Environ Health*. 1978;4:427–448.
24. Rhoads M. Glutamine signaling in intestinal cells. *JPEN J Parenter Enteral Nutr*. 1999;23:S38–40.
25. Haussinger D, Graf D, Weiergraber OH. Glutamine and cell signaling in liver. *J Nutr*. 2001;131:2509S–2514S.
26. Barbosa-Tessman IP, Chen C, Zhong C, et al. Activation of the human asparagine synthetase gene by the amino acid response and the endoplasmic reticulum stress response pathways occurs by common genomic elements. *J Biol Chem*. 2000;275:26976–26985.
27. Palsson H. Conformation and body composition. In: Hammond J, ed. *Progress in the Physiology of Farm Animals*. London: Butterworths; 1955:430–542.
28. Munro HN. General aspects of the regulation of protein metabolism by diet and by hormones. In: Munro HN, Allison JB, eds. *Mammalian Protein Metabolism*. New York: Academic; 1964:381–481.
29. Kochakian CD, Tillotson C, Austin J. A comparison of the effect of inanition,

castration and testosterone on the muscles of the male guinea pig. *Endocrinology*. 1957;60:144.
30. Machlin LJ. Effect of porcine growth hormone on growth and carcass composition of the pig. *J Anim Sci*. 1972;35:794–800.
31. Marin P, Krotkiewski M, Bjorntorp P. Androgen treatment of middle-aged, obese men: effects on metabolism, muscle and adipose tissues. *Eur J Med*. 1992;1:329–336.
32. Marin P, Holmang S, Jonsson L, et al. The effects of testosterone treatment on body composition and metabolism in middle-aged obese men. *Int J Obes Relat Metab Disord*. 1992;16:991–997.
33. Jensen MD. Androgen effect on body composition and fat metabolism. *Mayo Clin Proc*. 2000;75:S65–68.
34. Eisenstein AP, Singh SP. Hormonal control of nutrient metabolism. In: Goodheart RS, Shils ME, eds. *Modern Nutrition in Health and Disease*, 6th ed. Philadelphia, PA: Lea & Febiger; 1980.
35. Sokoloff L, Kauffman S. Thyroxine stimulation of amino acid incorporation into protein. *J Biol Chem*. 1961;236:795–803.
36. Enesco M, Leblond CP. Increase in cell number as a factor in the growth of the organs and tissues of the young male rat. *J Embryol Exp Morhpol*. 1962;10:530–564.
37. McCance RA. Critical periods of growth. *Proc Nutr Soc*. 1976;35:309–313.
38. Widdowson EM, McCance RA. Some effects of accelerating growth: I. General somatic development. *Proc R Soc Lond (Biol)*. 1960;152:188–206.
39. Widdowson EM, McCance RA. A review: new thoughts on growth. *Pediatr Res*. 1975;9:154–156.
40. Widdowson EM, McCance RA. The effect of finite periods of undernutrition at different ages on the composition and subsequent development of the rat. *Proc R Soc Lond (Biol)*. 1963;158:329–342.
41. van Pareren Y, Mulder P, Houdijk M, et al. Adult Height after long-term, continuous growth hormone (GH) treatment in short children born small for gestational age: results of a randomized, double-blind, dose-response GH trial. *J Clin Endocrinol Metab*. 2003;88:3584–3590.
42. Winick M, Noble A. Cellular response in rats during malnutrition at various ages. *J Nutr*. 1966;89:300–306.
43. Winick M, Noble A. Cellular response with increased feeding in neonatal rats. *J Nutr*. 1967;91:179–182.
44. Winick M, Brasel JA. Nutrition and cell growth. In: Goodhart RS, Shils ME, eds. *Modern Nutrition in Health and Disease*. Philadelphia, PA: Lea & Febiger; 1980:592–607.
45. Godfrey KM, Barker DJ. Fetal programming and adult health. *Public Health Nutr*. 2001;4:611–624.
46. Ibanez L, Potau N, Marcos MV, et al. Exaggerated adrenarche and hyperinsulinism in adolescent girls born small for gestational age. *J Clin Endocrinol Metab*. 1999;84:4739–4741.
47. Ibanez L, Potau N, Enriquez G, et al. Reduced uterine and ovarian size in adolescent girls born small for gestational age. *Pediatr Res*. 2000;47:575–577.
48. Ibanez L, Potau N, Ferrer A, et al. Reduced ovulation rate in adolescent girls born small for gestational age. *J Clin Endocrinol Metab*. 2002;87:3391–3393.
49. Abitbol C, Chan JC, Trachtman H, et al. Growth in children with moderate renal insufficiency: measurement, evaluation, and treatment. *J Pediatr*. 1996;129:s3–8.
50. Posada ME, Ruben M, Esquivel M. On growth of children: effect of some

socioeconomic and genetic factors. *Acta Paediatr Hung*. 1987;28:37–43.
51. Karlberg J, Lawrence C, bertsson-Wikland K. Prediction of final height in short, normal and tall children. *Acta Paediatr Suppl*. 1994;406:3–9.
52. Gohlke BC, Stanhope R. Final height in psychosocial short stature: is there complete catch-up? *Acta Paediatr*. 2002;91:961–965.
53. Wright CM, Cheetham TD. The strengths and limitations of parental heights as a predictor of attained height. *Arch Dis Child*. 1999;81:257–260.
54. Prader A, Tanner JM, von Harnack GA. Catch-up growth following illness or starvation: an example of developmental canalization in man. *J Pediatr*. 1963;62:646–659.
55. Tanner JM. *Growth at Adolescence*, 2nd ed. Oxford, UK: Blackwell Scientific Publishers; 1962.
56. Smith DW, Truog W, Rogers JE, et al. Shifting linear growth during infancy: illustrations of genetic factors in growth from fetal life through infancy. *J Pediatr*. 1976;89:225–230.
57. Williams JPG, Hughes PCR. Catch-up growth in rats undernourished for different periods during the suckling period. *Growth*. 1975;39:179–193.
58. Williams JPG, Tanner JM, Hughes PCR. Catch-up growth in male rats after growth retardation during the suckling period. *Pediatr Res*. 1974;8:149–156.
59. Rivkees SA, Bode HH, Crawford JD. Long-term growth in juvenile acquired hypothyroidism. *N Engl J Med*. 1988;318:599–602.
60. Burns EC, Tanner JM, Preece MA, et al. Final height and pubertal development in 155 children with idiopathic growth hormone deficiency treated for between 2 and 15 years with growth hormone. *Eur J Pediatr*. 1981;137:155–164.
61. Guerrant RL, Schorling JB, McAuliffe JF, et al. Diarrhea as a cause and an effect of malnutrition; diarrhea prevents catch-up growth and malnutrition increases diarrhea frequency and duration. *Am J Trop Med Hyg*. 1992;47:28–35.
62. Golden MH. Is complete catch-up possible for stunted malnourished children? *Eur J Clin Nutr*. 1994;48:58–70.
63. Mosier HD. Failure of compensatory (catch-up) growth in the rat. *Pediatr Res*. 1971;5:59–63.
64. Allen DB, Julius JR, Breen TJ, et al. Treatment of glucocorticoid-induced growth suppression with growth hormone. *J Clin Endocrinol Metab*. 1998;83 2824–2828.
65. Ashworth A. Growth rates in children recovering from protein-calorie malnutrition. *Br J Nutr*. 1969;23:835–845.
66. Bjorntorp P, Yang MU. Refeeding after fasting in the rat: effects on body composition and food efficiency. *Am J Clin Nutr*. 1982;36:444–449.
67. Krieger I, Whitten CF. Energy metabolism in infants with growth failure due to maternal deprivation, undernutrition or causes unknown. *J Pediatr*. 1969;75:374–379.
68. Talbot N. Correlation between weight increase and caloric intake in infants with severe marasmic malnutrition. 26th annual meeting of the Society for Pediatric Research; Atlantic City, NJ:1966.
69. WHO. *Management of severe malnutrition: A manual for physicians and other senior health workers*. Geneva: WHO; 2006.
70. Forbes GB. A note on the mathematics of “catch-up” growth. *Pediatr Res*. 1974;8:929–931.
71. Mosier HD. Catch-up and proportionate growth. *Med Clin North Am*. 1978;62:337–350.
72. Tanner JM. Catch-up growth in man. *Br Med Bull*. 1981;37:233–238.
73. Williams JPG. Catch-up growth. *J Embryol Exp Morhpol*. 1981;65:89–101.

74. Fausto N, Campbell JS, Riehle KJ. Liver regeneration. *Hepatology*. 2006;43:S45–53.
75. Tanner JM. The regulation of human growth. *Child Dev*. 1963;34:817–847.
76. Leibel RL. A biologic radar system for the assessment of body mass: the model of a geometry sensitive endocrine system is presented. *J Theor Biol*. 1977;66:297–306.
77. Gafni RI, Baron J. Catch-up growth: possible mechanisms. *Pediatr Nephrol*. 2000;14:616–619.
78. Baron J, Klein KO, Colli MJ, et al. Catch-up growth after glucocorticoid excess: a mechanism intrinsic to the growth plate. *Endocrinology*. 1994;135:1367–1371.
79. Kay's SK, Hindmarsh PC. Catch-up growth: an overview. *Ped Endocrinol Rev*. 2006;3:365–378.
80. Salmon WD, Jr, Daughaday WH. A hormonally controlled serum factor which stimulates sulfate incorporation by cartilage in vitro. *J Lab Clin Med*. 1957;49:825–826.
81. Le Roith D, Bondy C, Yakar S, et al. The somatomedin hypothesis: 2001. *Endocr Rev*. 2001;22:53–74.
82. Daughaday WH, Hall K, Raben MS, et al. Somatomedin: proposed designation for sulphation factor. *Nature*. 1972;235:107.
83. Rinderknecht E, Humbel RE. The amino acid sequence of human insulin-like growth factor I and its structural homology with proinsulin. *J Biol Chem*. 1978;253:2769–2776.
84. Zapf J, Schmid CH, Froesch ER. Biological and immunological properties of insulin-like growth factors (IGF) I and II. *Clin Endocrinol Metab*. 1984;13:3–30.
85. D'Ercole AJ, Applewhite GT, Underwood LE. Evidence that somatomedin is synthesized by multiple tissues in the fetus. *Dev Biol*. 1980;75:315–328.
86. Lowe WT, Lasky SR, LeRoith D, et al. Distribution and regulation of rat insulin-like growth factor I messenger ribonucleic acids encoding alternative carboxy-terminal E-peptides: evidence for differential processing and regulation in liver. *Mol Endocrinol*. 1988;2:528–535.
87. Isaksson O, Lindahl A, Nilsson A, et al. Mechanism of the stimulatory effect of growth hormone on longitudinal bone growth. *Endocr Rev*. 1987;8:426–438.
88. Green H, Morikawa M, Nixon T. A dual effector theory of growth hormone action. *Differentiation*. 1985;29:195–198.
89. Yakar S, Liu JL, Stannard B, et al. Normal growth and development in the absence of hepatic insulin-like growth factor I. *Proc Natl Acad Sci U S A*. 1999;96:7324–7329.
90. Sjogren K, Liu JL, Blad K, et al. Liver-derived insulin-like growth factor I (IGF-I) is the principal source of IGF-I in blood but is not required for postnatal body growth in mice. Proc Natl Acad Sci U S A. 1999;96:7088–7092.
91. Duan C, Xu Q. Roles of insulin-like growth factor (IGF) binding proteins in regulating IGF activity. *Gen and Comp Endocrinol*. 2005;142:44–52.
92. Jones JI, Gockerman A, Busby WJ, et al. Insulin-like growth factor binding protein 1 stimulates cell migration and binds to the alpha 5 beta 1 integrin by means of its Arg-Gly-Asp sequence. *Proc Natl Acad Sci U S A*. 1993;90:10553–10557.
93. Valentinis B, Bhala A, DeAngelis T, et al. The human insulin-like growth factor (IGF) binding protein-3 inhibits the growth of fibroblasts with a targeted disruption of the IGF-I receptor gene. *Mol Endocrinol*. 1995;9:361–367.
94. Lee KW, Cohen P. Nuclear effects: unexpected intracellular actions of insulin-like growth factor binding protein-3. *J Endocrinol*. 2002;175:33–40.

95. Miller WL, Eberhardt NL. Structure and evolution of the growth hormone gene family. *Endocr Rev*. 1983;4:97–130.
96. Hwa V, Oh Y, Rosenfeld RG. The insulin-like growth factor binding protein (IGFBP) superfamily. *Endocr Rev*. 1999;20:761–787.
97. Muggeo M, Tiengo A, Fedele D, et al. The influence of plasma triglycerides on human growth hormone response to arginine and insulin: a study in hyperlipidemics and normal subjects. *Horm Metab Res*. 1975;7:367–374.
98. Carro E, Senaris R, Considine RV, et al. Regulation of in vivo growth hormone secretion by leptin. *Endocrinology*. 1997;138:2203–2206.
99. Tannenbaum GS, Gurd W, Lapointe M. Leptin is a potent stimulator of spontaneous pulsatile growth hormone (GH) secretion and the GH response to GH-releasing hormone. *Endocrinology*. 1998;139:3871–3875.
100. Vuagnat BA, Pierroz DD, Laloui M, et al. Evidence for a leptin- neuropeptide Y axis for the regulation of growth hormone secretion in the rat. *Neuroendocrinology*. 1998;67:291–300.
101. Chan YY, Steiner RA, Clifton DK. Regulation of hypothalamic neuropeptide-Y neurons by growth hormone in the rat. *Endocrinology*. 1996;137:2109–2112.
102. Cummings DE. Ghrelin and the short- and long-term regulation of appetite and body weight. *Physiol Behav*. 2006;89:71–84.
103. Ghigo E, Arvat E, Gianotti L, et al. Growth hormone-releasing activity of hexarelin, a new synthetic hexapeptide, after intravenous, subcutaneous, intranasal, and oral administration in man. *J Clin Endocrinol Metab*. 1994;78:693–698.
104. Bowers CY. Unnatural growth hormone-releasing peptide begets natural ghrelin. *J Clin Endocrinol Metab*. 2001;86:1464–1469.
105. Berelowitz M, Szabo M, Frohman LA, et al. Somatomedin-C mediates growth hormone negative feedback by effects on both the hypothalamus and the pituitary. *Science*. 1981;212:1279–1281.
106. Brazeau P, Guillemin R, Ling N, et al. Inhibition by somatomedin of growth hormone secretion stimulated by hypothalamic growth hormone releasing factor (somatocrinin, GRF), or the synthetic peptide hpGRF. *CR Acad Sci III*. 1982;295:651–654.
107. Herington AC, Ymer S, Stevenson J. Identification and characterization of specific binding proteins for growth hormone in normal human sera. *J Clin Invest*. 1986;77:1817–1823.
108. Daughaday WH, Trivedi B, Andrews BA. The ontogeny of serum GH binding protein in man: a possible indicator of hepatic GH receptor development. *J Clin Endocrinol Metab*. 1987;65:1072–1074.
109. Finklestein JW, Roffwarg HP, Boyar RM, et al. Age-related changes in the twenty-four hour spontaneous secretion of growth hormone. *J Clin Endocrinol Metab*. 1972;35:665–670.
110. Frankenne F, Rentier-Delrue F, Scippo ML, et al. Expression of the growth hormone variant gene in human placenta. *J Clin Endocrinol Metab*. 1987;64:635–637.
111. Chernausek SD, Underwood LE, Utiger RD, et al. Growth hormone secretion and plasma somatomedin-C in primary hypothyroidism. *Clin Endocrinol*. 1983;19:337–344.
112. Cunningham BC, Ultsch M, DeVos AM, et al. Dimerization of the extracellular domain of the human growth hormone receptor: crystal structure of the complex. *Science*. 1991;254:821–825.
113. Ridderstrale M. Signaling mechanism for the insulin-like effects of growth

hormone-another example of a classical hormonal negative feedback loop. *Curr Drug Targets Immune Endocr Metab Disord*. 2005;5:79–92.
114. Ziegler TR, Rombeau JL, Young LS, et al. Recombinant human growth hormone enhances the metabolic efficacy of parenteral nutrition: a double-blind, randomized controlled study. *J Clin Endocrinol Metab*. 1992;74:865–873.
115. Simon D, Alberti C, Alison M, Le Henaff L, Chevenne D et al. Effects of recombinant human growth hormone for 1 year on body composition and muscle strength in children on long-term steroid therapy: randomized controlled, delayed-start study. *J Clin Endocrinol Metab*. 2013 Jul;98(7):2746–54.
116. Smal J, Closset J, Hennen G, et al. The receptor binding properties of the 20K variant of human growth hormone explain its discrepant insulin-like and growth promoting activities. *Biochem Biophys Res Comm*. 1986;134:159–165.
117. Kostyo JL, Cameron DM, Olson KC, et al. Biosynthetic 20-kilodalton methionyl-human growth hormone has diabetogenic and insulin-like activities. *Proc Natl Acad Sci U S A*. 1985;82:4250–4253.
118. Stewart TA, Clift S, Pitts-Meek S, et al. An evaluation of the functions of the 22-kilodalton (kDa), the 20-kDa, and the N-terminal polypeptide forms of human growth hormone using transgenic mice. *Endocrinology*. 1992;130:405–414.
119. Vatten LJ, Nilsen ST, Odegard RA, et al. Insulin-like growth factor I and leptin in umbilical cord plasma and infant birth size at term. *Pediatrics*. 2002;109:1131–1135.
120. Francis GL, Upton FM, Ballard FJ, et al. Insulin-like growth factors 1 and 2 in bovine colostrum. Sequences and biological activities compared with those of a potent truncated form. *Biochem J*. 1988;251:95–103.
121. Burrin DG, Wester TJ, Davis TA, et al. Orally administered IGF-I increases intestinal mucosal growth in formula-fed neonatal pigs. *Am J Physiol*. 1996;270:R1085–1091.
122. Alexander AN, Carey HV. Insulin-like growth factor-I stimulates Na+-dependent glutamine absorption in piglet enterocytes. *Dig Dis Sci*. 2002;47:1129–1134.
123. Simmons JG, Pucilowska JB, Lund PK. Autocrine and paracrine actions of intestinal. broblast-derived insulin-like growth factors. *Am J Physiol*. 1999;276:G817–827.
124. Andre F, Rigot V, Remacle-Bonnet M, et al. Protein kinases C-gamma and -delta are involved in insulin-like growth factor I-induced migration of colonic epithelial cells. *Gastroenterology*. 1999;116:64–77.
125. Kuemmerle JF, Bushman TL. IGF-I stimulates intestinal muscle cell growth by activating distinct PI 3-kinase and MAP kinase pathways. *Am J Physiol*. 1998;275:G151–158.
126. Zijlstra RT, Odle J, Hall WF, et al. Effect of orally administered epidermal growth factor on intestinal recovery of neonatal pigs infected with rotavirus. *J Pediatr Gastroenterol Nutr*. 1994;19:382–390.
127. Gillingham MB, Dahly EM, Carey HV, et al. Differential jejunal and colonic adaptation due to resection and IGF-I in parenterally fed rats. *Am J Physiol Gastrointest Liver Physiol*. 2000;278:G700–709.
128. Gu Y, Wu ZH, Xie JX, et al. Effects of growth hormone (rhGH) and glutamine supplemented parenteral nutrition on intestinal adaptation in short bowel rats. *Clin Nutr*. 2001;20:159–166.
129. Ziegler TR, Mantell MP, Chow JC, et al. Gut adaptation and the insulin-like growth factor system: regulation by glutamine and IGF-I administration. *Am J Physiol*. 1996;271: G866–875.
130. Blikslager AT, Rhoads JM, Bristol DG, et al. Glutamine and transforming growth

factor-alpha stimulate extracellular regulated kinases and enhance recovery of villous surface area in porcine ischemic-injured intestine. *Surgery*. 1999;125:186–194.
131. Rhoads JM, Ulshen MH, Keku EO, et al. Oral transforming growth factor-alpha enhances jejunal mucosal recovery and electrical resistance in piglet rotavirus enteritis. *Pediatr Res*. 1995;38:173–181.
132. Talbot NB, Sobel EH, et al. Dwarfism in healthy children; its possible relation to emotional nutritional and endocrine disturbances. *N Engl J Med*. 1947;236:783–793.
133. Ahdieh N, Blikslager AT, Bhat BG, et al. L-glutamine and transforming growth factor-alpha enhance recovery of monoacylglycerol acyltransferase and diacylglycerol acyltransferase activity in porcine postischemic ileum. *Pediatr Res*. 1998;43:227–323.
134. Park YK, Monaco MH, Donovan SM. Enteral insulin-like growth factor-I augments intestinal disaccharidase activity in piglets receiving total parenteral nutrition. *J Pediatr Gastroenterol Nutr*. 1999;29:198–206.
135. Park YK, Dudley MA, Burrin DG, et al. Intestinal protein and LPH synthesis in parenterally fed piglets receiving partial enteral nutrition and enteral insulinlike growth factor 1. *J Pediatr Gastroenterol Nutr*. 2001;33:189–195.
136. Lund PK. Molecular basis of intestinal adaptation: the role of the insulin-like growth factor system. *Ann N Y Acad Sci*. 1998;859:18–36.
137. Butler AA, Yakar S, LeRoith D. Insulin-like growth factor-I: compartmentalization within the somatotropic axis? *News Physiol Sci*. 2002;17:82–85.
138. Glaser B, Phillip M, Carmi R, et al. Persistent hyperinsulinemic hypoglycemia of infancy ("nesidioblastosis"): autosomal recessive inheritance in 7 pedigrees. *Am J Med Genet*. 1990;37:511–515.
139. Hill DJ, Milner RDG. Insulin as a growth factor. *Pediatr Res*. 1985;19:879–886.
140. Lee RGL, Bode HH. Stunted growth and hepatomegaly in diabetes mellitus. *J Pediatr*. 1977;91:82–84.
141. Davis TA, Fiorotto ML, Beckett PR, et al. Differential effects of insulin on peripheral and visceral tissue protein synthesis in neonatal pigs. *Am J Physiol Endocrinol Metab*. 2001;280:E770–779.
142. Amiel SA, Sherwin RS, Hintz RL, et al. Effect of diabetes and its control on insulin-like growth factors in the young subject with type I diabetes. *Diabetes*. 1984;33:1175–1179.
143. Binoux M, Lassarre C, Jardoudin N. Somatomedin production by rat liver in organ culture: III. Studies on the release of insulin-like growth factor and its carrier protein measured by radioligand assays. *Acta Endocr (Copenh)*. 1982;99:422–430.
144. Maes M, Ketelslegers JM, Underwood LE. Low plasma somatomedin-C in streptozotocin-induced diabetes mellitus. *Diabetes*. 1983;32:1060–1069.
145. Myers MJ, Wang LM, Sun XJ, et al. Role of IRS-1-GRB-2 complexes in insulin signaling. *Mol Cell Biol*. 1994;14:3577–3587.
146. Moxham CP, Duronio V, Jacobs S. Insulin-like growth factor-1 receptor beta-subunit heterogeneity: evidence for hybrid tetramers composed of insulin-like growth factor 1 and insulin heterodimers. *J Biol Chem*. 1989;264:13228–3244.
147. Roth RA. Structure of the receptor for insulin-like growth factor II: the puzzle amplified. *Science*. 1988;239:1269–1271.
148. Rechler MM, Nissley SP. The nature and regulation of the receptors for insulin-like growth factors. *Ann Rev Physiol*. 1985;47:425–442.
149. McGowan JA, Strain AJ, Bucher NLR. DNA synthesis in primary cultures of

adult rat hepatocytes in a defined medium: effects of epidermal growth factor, insulin, glucagon and cyclic-AMP. *J Cell Physiol*. 1981;108:353–363.
150. Whitehead RG, Rowland MGM, Cole TJ. Infection, nutrition and growth in a rural African environment. *Proc Nutr Soc*. 1976;35:369–375.
151. Ho KY, Veldhuis JD, Jphnson ML, et al. Fasting enhances growth hormone secretion and ampli.es the complex rhythms of growth hormone secretion in man. *J Clin Invest*. 1988;81:968–975.
152. Hartman ML, Veldhuis JD, Johnson ML, et al. Augmented growth hormone (GH) secretory burst frequency and amplitude mediate enhanced GH secretion during a two-day fast in normal men. *J Clin Endocrinol Metab*. 1992;74:757–765.
153. Becker DJ, Pimstone BL, Hansen JDL, et al. Serum albumin and growth hormone relationships in kwashiorkor and the nephrotic syndrome. *J Lab Clin Med*. 1971;78:865–871.
154. Coward WA, Lunn PG. The biochemistry and physiology of kwashiorkor and marasmus. *Br Med Bull*. 1981;37:19–24.
155. Lunn PG, Whitehead RG, May RW, et al. Progressive changes in serum cortisol, insulin and growth hormone concentrations and their relationship to the distorted amino acid pattern during the development of kwashiorkor. *Br J Nutr*. 1973;29:399–422.
156. Parra A. Insulin-growth hormone adaptations in marasmus and kwashiorkor as seen in Mexico. In: Gardner LI, Amacher P, eds. *Endocrine Aspects of Malnutrition: Marasmus, Kwashiorkor and Psychosocial Deprivation*. Santa Ynez, CA: The Kroc Foundation; 1973:31–43.
157. Pimstone BL, Barbezat G, Jansen JDL, et al. Studies on growth hormone secretion in protein-calorie malnutrition. *Clin Nutr*. 1973;21:482–487.
158. Beas F, Muzzo S. *Growth Hormone and Malnutrition: The Chilean Experience*. Santa Ynez, CA: The Kroc Foundation; 1973.
159. Robinson H, Cocks T, Kerr D, et al. Fasting and post-prandial levels of plasma insulin and growth hormone in malnourished Jamaican children, during catch-up growth and after complete recovery. In: Gardner LI, Amacher P, eds. *Endocrine Aspects of Malnutrition: Marasmus, Kwashiorkor and Psychosocial Deprivation*. Santa Ynez, CA: The Kroc Foundation; 1973:45–72.
160. Beas F, Monckeberg F, Horwitz I, et al. The responses of the thyroid gland to thyroid-stimulating hormone (TSH) in infants with malnutrition. *Pediatrics*. 1966;38:1003–1008.
161. Mulinos MG, Pomerantz L. Pseudo-hypophysectomy: a condition resembling hypophysectomy produced by malnutrition. *J Nutr*. 1940;19:493–504.
162. Miller J, Silverstein J, Shuster J, et al. Short-term effects of growth hormone on sleep abnormalities in Prader-Willi syndrome. *J Clin Endocrinol Metab*. 2006;91:413–417.
163. Low LC. Growth of children with beta-thalassemia major. *Indian J Pediatr*. 2005;72:159–164.
164. Bideci A, Camurdan MO, Cinaz P, et al. Ghrelin, IGF-I and IGFBP-3 levels in children with type 1 diabetes mellitus. *J Pediatr Endocrinol Metab*. 2005;18:1433–1439.
165. Krawczuk-Rybak M, Muszynska-Roslan K, Kitszel A, et al. Relationship between insulin-like growth factors (IGF-I and IGF-II), IGF-binding proteins (IGFBP-3, IGFBP-2), leptin and anthropometric parameters (height, body mass index) during antileukaemic treatment in children. *Rocz Akad Med Bialymst*. 2005;50:208–211.
166. Rusinska A, Chlebna-Sokol D. Insulin-like growth factor-I and mineral metabolism markers in children with idiopathic decrease in bone mass. *Clin Chim*

Acta. 2006;366:257–263.
167. Laron Z, Ginsberg S, Lilos P, et al. Long-term IGF-I treatment of children with Laron syndrome increases adiposity. *Growth Horm IGF Res*. 2006;16:61–64.
168. Clemmons DR, Snyder DK, Williams R, et al. Growth hormone administration conserves lean body mass during dietary restriction in obese subjects. *J Clin Endocrinol Metab*. 1987;64:878–883.
169. Manson JM, Wilmore DW. Positive nitrogen balance with human growth hormone and hypocaloric intravenous feeding. *Surgery*. 1986;100:188–197.
170. Mode A, Norstedt G, Simic B, et al. Continuous infusion of growth hormone feminizes hepatic steroid metabolism in the rat. *Endocrinology*. 1981;108:2103–2108.
171. Ziegler TR, Young LS, Manson JM, et al. Metabolic effects of recombinant human growth hormone in patients receiving parenteral nutrition. *Ann Surg*. 1988;208:6–16.
172. Ruokonen E, Takala J. Dangers of growth hormone therapy in critically ill patients. *Ann Med*. 2000;32:317–322.
173. Ramirez RJ, Wolf SE, Barrow RE, et al. Growth hormone treatment in pediatric burns: a safe therapeutic approach. *Ann Surg*. 1998;228:439–448.
174. Han X, Sosnowska D, Bonkowski EL, et al. Growth hormone inhibits signal transducer and activator of transcription 3 activation and reduces disease activity in murine colitis. *Gastroenterology*. 2005;129:185–203.
175. Theiss AL, Fuller CR, Simmons JG, et al. Growth hormone reduces the severity of fibrosis associated with chronic intestinal inflammation. *Gastroenterology*. 2005;129:204–219.
176. Sanz S, Pucilowska JB, Liu S, et al. Expression of insulin-like growth factor I by activated hepatic stellate cells reduces fibrogenesis and enhances regeneration after liver injury. *Gut*. 2005;54:134–141.
177. Hardin DS, Ellis KJ, Dyson M, et al. Growth hormone improves clinical status in prepubertal children with cystic fibrosis: results of a randomized controlled trial. *J Pediatr*. 2001;139:636–642.
178. Taylor AM, Thomson A, Bruce-Morgan C, et al. The relationship between insulin, IGF-I and weight gain in cystic fibrosis. *Clin Endocrinol*. 1999;51:659–665.
179. Slonim AE, Bulone L, Damore MB, et al. A preliminary study of growth hormone therapy for Crohn's disease. *N Engl J Med*. 2000;342:1633–1637.
180. Mauras N, George D, Evans J, et al. Growth hormone has anabolic effects in glucocorticosteroid-dependent children with inflammatory bowel disease: a pilot study. *Metabolism*. 2002;51:127–135.
181. Ibanez L, Ong K, Dunger DB, et al. Early development of adiposity and insulin resistance after catch-up weight gain in small-for-gestational-age children. *J Clin Endocrinol Metab*. 2006;91:2153–2158.
182. Godfrey KM, Barker DJ. Fetal nutrition and adult disease. *Am J Clin Nutr*. 2000;71:1344S–1352S.
183. de Zegher F, bertsson-Wikland K, Wilton P, et al. Growth hormone treatment of short children born small for gestational age: metanalysis of four independent, randomized, controlled, multicentre studies. *Acta Paediatr Suppl*. 1996;417:27–31.
184. de Zegher F, bertsson-Wikland K, Wollmann HA, et al. Growth hormone treatment of short children born small for gestational age: growth responses with continuous and discontinuous regimens over 6 years. *J Clin Endocrinol Metab*. 2000;85:2816–2821.
185. Bundak R, Darendeliler F, Gunoz H, et al. Growth hormone treatment in short

children with intrauterine growth retardation. *J Pediatr Endocrinol*. 2001;14:313–318.
186. Boguszewski M, bertsson-Wikland K, Aronsson S, et al. Growth hormone treatment of short children born small-forgestational-age: the Nordic Multicentre Trial. *Acta Paediatr*. 1998;87:257–263.
187. Rekers-Mombarg LT, Kamp GA, Massa GG, et al. Influence of growth hormone treatment on pubertal timing and pubertal growth in children with idiopathic short stature. Dutch Growth Hormone Working Group. *J Pediatr Endocrinol*. 1999;12:611–622.
188. de Zegher F, Butenandt O, Chatelain P, et al. Growth hormone treatment of short children born small for gestational age: reappraisal of the rate of bone maturation over 2 years and metanalysis of height gain over 4 years. *Acta Paediatr Suppl*. 1997;423:207–212.
189. Verhoeff FH, Brabin BJ, van Buuren S, et al. An analysis of intra-uterine growth retardation in rural Malawi. *Eur J Clin Nutr*. 2001;55:682–689.
190. Shamim A, Khan HO, Rana JS, et al. Intrauterine growth restriction: a perspective for Pakistan. *JPMA J Pak Med Assoc*. 1999;49:82–85.
191. Davenport ML, Svoboda ME, Koerber KL, et al. Serum concentrations of insulin-like growth factor II are not changed by short-term fasting and refeeding. *J Clin Endocrinol Metab*. 1988;67:1231–1236.
192. Grant DB, Hambley J, Becker DJ, et al. Reduced sulphation factor in undernourished children. *Arch Dis Child*. 1973;48:596–600.
193. Van den Brande JL, Du Caju MVL. Plasma somatomedin activity in children with growth disturbance. In: Raiti S, ed. *Advances in Human Growth Hormone Research*. Washington, DC: U.S. Government Printing Office; 1974:98–115.
194. Hintz RL, Suskind R, Amatayakul K, et al. Plasma somatomedin and growth hormone values in children with protein calorie malnutrition. *J Pediatr*. 1978;92:153–156.
195. Smith IF. Blood plasma levels of cortisol, insulin, growth hormone and somatomedin in children with marasmus, kwashiorkor and intermediate forms of protein-energy malnutrition. *Proc Soc Exp Biol Med*. 1981;167:607–611.
196. Lecornu M, David L, Francois R. Low somatomedin activity in celiac disease. *Helv Paediatr Acta*. 1978;33:509–516.
197. Rappaport R, Prevot C, Czernichow P. Somatomedin activity and growth hormone secretion. I. Changes related to body weight in anorexia nervosa. *Acta Paediatr Scand*. 1980;69:37–41.
198. Ashton IK, Franics MJO. An assay for plasma somatomedin: 3H thymidine incorporation by isolated rabbit chondrocytes. *J Endocrinol*. 1977;74:205–212.
199. Van den Brande JL. Further observations on plasma somatomedin activity in children. *Adv Metab*. 1975;8:171–182.
200. Phillips LS, Young HS. Nutrition and somatomedin. II. Serum somatomedin activity and cartilage growth activity in streptozotocin-diabetic rats. *Diabetes*. 1976;25:516–527.
201. Salmon WD. Interaction of somatomedin and a peptide inhibitor of serum of hypophysectomized and starved, pituitary-intact rats. *Adv Metab*. 1975;8:183–199.
202. Salmon WD, Holladay LA, Burkhalter VJ. Partial characterization of somatomedin inhibitor in starved rat serum. *Endocrinology*. 1983;112:360–370.
203. Graubert MD, Goldstein S, Phillips LS. Nutrition and somatomedin. XXVII. Total and free IGF-I and IGF binding proteins in rats with streptozotocin-induced diabetes. *Diabetes*. 1991;40:959–965.
204. Busby WH, Snyder DK, Clemmons DR. Radioimmunoassay of a 26,000-dalton

plasma insulin-like growth factor-binding protein: control by nutritional variables. *J Clin Endocrinol Metab*. 1988;67:1225–1230.
205. Unterman TG, Patel K, Mahathre VK, et al. Regulation of low molecular weight insulin-like growth factor binding proteins in experimental diabetes mellitus. *Endocrinology*. 1990;126:2614–2624.
206. Phillips LS, Goldstein S, Gavin JI. Nutrition and somatomedin. XVI. Somatomedins and somatomedin inhibitors in fasted and refed rats. *Metabolism*. 1988;37:209–216.
207. Clemmons DR, Klibanski A, Ridgeway EC, et al. Reduction of plasma immunoreactive somatomedin-C during fasting in humans. *J Clin Endocrinol Metab*. 1981;53:1247–1250.
208. Smith AT, Clemmons DR, Underwood LE, et al. The effect of exercise on plasma somatomedin-C/insulin-like growth factor I concentrations. *Metabolism*. 1987;36:533–537.
209. Soliman AT, Gassan AEHI, Aref MK, et al. Serum insulin-like growth factors I and II concentrations and growth hormone and insulin responses to arginine infusion in children with protein-energy malnutrition before and after nutritional rehabilitation. *Pediatr Res*. 1986;20:1122–1130.
210. Nucci AM, Reyes J, Yaworski JA, et al. Serum growth factors and growth indices pre- and post-pediatric intestinal transplantation. *J Pediatr Surg*. 2003;38:1043–1047.
211. Clemmons DR, Underwood LE, Dickerson RN, et al. Use of plasma somatomedin-C/insulin-like growth factor I measurements to monitor the response to nutritional repletion in malnourished patients. *Am J Clin Nutr*. 1985;41:191–198.
212. Unterman TG, Vasquez RM, Slas AJ, et al. Nutrition and somatomedin. XIII. Usefulness of somatomedin-C in nutritional assessment. *Am J Med*. 1985;78:228–234.
213. Kirschner BS, Sutton MM. Somatomedin-C levels in growth-impaired children and adolescents with chronic inflammatory bowel disease. *Gastroenterology*. 1986;91:830–836.
214. Baxter RC, Hawker FH, To C, et al. Thirty-day monitoring of insulin-like growth factors and their binding proteins in intensive care unit patients. *Growth Horm IGF Res*. 1998;8:455–463.
215. Isley WL, Underwood LE, Clemmons DR. Dietary components that regulate serum somatomedin-C concentrations in humans. *J Clin Invest*. 1983;71:175–182.
216. Prewitt TE, D'Ercole AJ, Switzer BR, Van Wyk JJ. Relationship of serum immunoreactive somatomedin-C to dietary protein and energy in growing rats. *J Nutr*. 1982;112:144–150.
217. Baxter RC, Bryson JM, Turtle JR. The effect of fasting on liver receptors for prolactin and growth hormone. *Metabolism*. 1981;30:1086–1090.
218. Postel-Vinay MC, Cohen-Tanugi E, Charrier J. Growth hormone receptors in rat liver membranes: effect of fasting and refeeding, and correlation with plasma somatomedin activity. *Mol Cell Endocrinol*. 1982;28:657–669.
219. Maes M, Underwood LE, Ketelslegers JM. Plasma somatomedin-C in fasted and refed rats: close relationship with changes in liver somatogenic but not lactogenic binding sites. *J Endocrinol*. 1983;97:243–252.
220. Bornfeldt KE, Arnqvist HJ, Enberg B, et al. Regulation of insulin-like growth factor-I and growth hormone receptor gene expression by diabetes and nutritional state in rat tissues. *J Endocrinol*. 1986;122:651–656.
221. Straus DS, Takemoto CD. Effect of fasting on insulin-like growth factor-I and

growth hormone receptor mRNA levels and IGF-I gene transcription in rat liver. *Mol Endocrinol*. 1991;4:91–100.
222. Thissen JP, Triest S, Moats-Staats BM, et al. Evidence that pretranslational and translational defects decrease serum IGF-I concentrations during dietary protein energy restriction. *Endocrinology*. 1991;129:429–435.
223. Thissen JP, Ketelslegers JM, Underwood LE. Nutritional regulation of the insulin-like growth factors. *Endocr Rev*. 1994;15:80–101.
224. Lowe WT, Adamo M, Werner H, et al. Regulation by fasting of rat insulin-like growth factor I and its receptor. Effects on gene expression and binding. *J Clin Invest*. 1989;84:619–626.
225. Cahill GF. Hormone fuel interrelationships during fasting. *J Clin Invest*. 1966;45:1751–1769.
226. Pimstone BL, Becker DJ, Weinkove C, et al. Insulin secretion in protein-calorie malnutrition. In: Gardner LI, Amacher P, eds. *Endocrine Aspects of Malnutrition: Marasmus, Kwashiorkor and Psychosocial Deprivation*. Santa Ynez, CA: The Kroc Foundaton; 1973:289–305.
227. Rao RH. Diabetes in the undernourished: coincidence or consequence? *Endocrinology*. 1988;9:67–87.
228. Bennish ML, Azad AK, Rahman O, et al. Hypoglycemia during diarrhea in childhood. Prevalence, pathophysiology, and outcome. *N Engl J Med*. 1990;322:1357–1363.
229. Goldbloom RB. Failure to thrive. *Pediatr Clin North Am*. 1982;29:151–166.
230. Powell GF, Brasel JA, Blizzard RM. Emotional deprivation and growth retardation simulating idiopathic hypopituitarism. I. Clinical evaluation of the syndrome. *N Engl J Med*. 1967;276:1271–1287.
231. Patton RG, Gardner LI. Deprivation dwarfism (psychosocial deprivation): disordered family environment as a cause of so-called idiopathic hypopituitarism. In: Gardner LI, ed. *Endocrine and Genetic Diseases of Childhood and Adolescence*. Philadelphia, PA: WB Saunders; 1975:85–89.
232. Talbot NB. Dwarfism in healthy children: its possible relation to emotional, nutritional and endocrine disturbances. *N Engl J Med*. 1947;236:783–793.
233. Powell GF, Brasel JA, Raiti S, et al. Emotional deprivation and growth retardation simulating idiopathic hypopituitarism: II. Endocrinologic evaluation of the syndrome. *N Engl J Med*. 1967;276:1279–1283.
234. Powell GF, Hopwood NJ, Barratt ES. Growth hormone studies before and during catch-up growth in a child with emotional deprivation and short stature. *J Clin Endocrinol Metab*. 1973;37:674–679.
235. Miller JD, Tannenbaum GS, Colle E, et al. Daytime pulsatile growth hormone secretion during childhood and adolescence. *J Clin Endocrinol Metab*. 1982;55:989–993.
236. Frasier SD, Rallisen ML. Growth retardation and emotional deprivation: relative resistance to treatment with human growth hormone. *J Pediatr*. 1946;80:603–609.
237. Tanner JM, Whitehouse RH, Hughes PCR, et al. The effects of human growth hormone treatment for 1 to 7 years on growth of 100 children with growth hormone deficiency, low birth weight, inherited smallness, Turner's syndrome and other complaints. *Arch Dis Child*. 1971;46:745–782.
238. Widdowson EM. Mental contentment and physical growth. *Lancet*. 1951;1:1316–1318.
239. Whitten CR, Pettit MG, Fishoff J. Evidence that growth failure from maternal deprivation is secondary to undereating. *JAMA*. 1969;209:1675–1682.
240. Krieger I, Mellinger RC. Pituitary function in the deprivation syndrome. *J Pediatr*.

1971;79:216–225.
241. Pugliese MT, Lifshitz F, Grad G, et al. Fear of obesity: a cause of short stature and delayed puberty. *N Engl J Med*. 1983;309:513–518.
242. Pugliese MT, Weyman-Daum M, Moses N, et al. Parental health beliefs as a cause of nonorganic failure to thrive. *Pediatrics*. 1987;80:175–182.
243. Bernal J, Refetoff S. The actions of thyroid hormone. *Clin Endocrinol*. 1977;7:227–249.
244. Silva JE. The thermogenetic effect of thyroid hormone and its clinical implications. *Ann Int Med*. 2003;139:205–13.
245. Evans RM. The steroid and thyroid hormone receptor superfamily. *Science*. 1988;240:889–895.
246. Fisher DA, Hoath S, Lakshmanan J. The thyroid hormone effects on growth and development may be mediated by growth factors. *Endocr Exp*. 1982;16:259–271.
247. Burstein PJ, Draznin B, Johnson DJ, et al. The effect of hypothyroidism on growth, serum growth hormone, the growth hormone-dependent somatomedin, insulin-like growth factor, and its carrier protein in rats. *Endocrinology*. 1979;104:1107–1111.
248. Coiro U. Effect of hypothyroidism and thyroxine replacement on growth hormone in the rat. *Endocrinology*. 1979;105:641–646.
249. Urist MR, DeLang RJ, Finerman GAM. Bone cell differentiation and growth factors. *Science*. 1983;220:680–686.
250. Wegnez M, Schachter BS, Baxter JD, et al. Hormone regulation of growth hormone messenger RNA. *DNA*. 1982;1:145–153.
251. Root AW, Rosenfield RL, Bongiovanni AM, et al. The plasma growth hormone response to insulin-induced hypoglycemia in children with retardation of growth. *Pediatrics*. 1967;39:844.
252. Froesch ER. Nonsuppressible insulin-like activity and thyroid hormones: major pituitary-dependent sulfation factors for chick embryo cartilage. *Proc Natl Acad Sci U S A*. 1976;73:2904–2908.
253. Dearden LC, Espinosa T. Comparison of mineralization of the tibial epiphyseal plate in immature rats following treatment with cortisone, propylthiouracil or after fasting. *Calcif Tissue Res*. 1974;15:93–110.
254. Burch WM, Van Wyk JJ. Triiodothyronine stimulates cartilage growth and maturation by different mechanisms. *Am J Physiol (Endocr Metab 15)*. 1987;252:E176–182.
255. Jung RT, Shetty PS, James WPT. The effect of refeeding after semistarvation on catecholamine and thyroid metabolism. *Int J Obes*. 1980;4:95–100.
256. Vagnati L, Finley RJ, Hagg S, et al. Protein conservation during prolonged fast: a function of triiodothyronine levels. *Trans Assoc Am Physiol*. 1978;91:169–179.
257. O'Dea K, Esler M, Leonard P, et al. Noradrenaline turnover during under- and overeating in normal weight subjects. *Metabolism*. 1982;31:896–899.
258. O'Brian JT. Thyroid hormone homeostasis in states of relative caloric deprivation. *Metabolism*. 1980;29:721–729.
259. Wartofsky L, Burman KD. Alterations in thyroid function in patients with systemic illness: the "euthyroid sick syndrome". *Endocr Rev*. 1982;3:164–217.
260. Nicoloff JT, LoPresti JS. Nonthyroidal illnesses. In: DeGroot LJ, ed. *Endocrinology*, 3rd ed. Philadelphia, PA: WB Saunders; 1995:665–675.
261. DeGroot LJ. Non-thyroidal illness syndrome is a manifestation of hypothalamic-pituitary dysfunction, and in view of current evidence, should be treated with appropriate replacement therapies. *Crit Care Clin*. 2006;22:57–86.
262. Rastogi GK, Sawhney RC, Panda NC, et al. Thyroid hormone levels in adult

protein calorie malnutrition (PCM). *Horm Metab Res*. 1974;6:528–529.
263. Henning SJ. Ontogeny of enzymes in the small intestine. *Ann Rev Physiol*. 1985;47:231–245.
264. Yeh KY, Moog F. Development of the small intestine in the hypophysectomized rat. II. Influence of cortisone, thyroxine, growth hormone, and prolactin. *Dev Biol*. 1975;47:173–184.
265. Moog F, Yeh KY. Pinocytosis persists in the ileum of hypophysectomized rats unless closure is induced by thyroxine or cortisone. *Dev Biol*. 1979;69:159–169.
266. Yeh KY. Corticosterone concentrations in the serum and milk of lactating rats: parallel changes after induced stress. *Endocrinology*. 1984;115:1364–1370.
267. Little JM, Lester R. Ontogenesis of intestinal bile salt absorption in the neonatal rat. *Am J Physiol (Gastrointest Liver Physiol)*. 1983;239:G319–323.
268. Alleyne GAO, Young VH. Adrenocortical function in children with severe protein-calorie malnutrition. *Clin Sci*. 1967;33:189–200.
269. Leonard PJ, MacWilliam KW. The binding of aldosterone in the serum in kwashiorkor. *Am J Clin Nutr*. 1965;16:360–362.
270. Rao KSJ, Srikantia SG, Gopalan C. Plasma cortisol levels in protein-calorie malnutrition. *Arch Dis Child*. 1968;43:365–367.
271. Lunn PG, Whitehead RG, Baker BA, et al. The effect of cortisone acetate on the course of development of experimental protein-energy malnutrition in rats. *Br J Nutr*. 1976;36:537–550.
272. Eveleth PB, Tanner JM. *Worldwide Variation in Human Growth*. Cambridge, UK: Cambridge University Press; 1976.
273. Smith SR. The pituitary-gonadal axis in men with proteincalorie malnutrition. *J Clin Endocr Metab*. 1975;41:60–69.
274. Klibanski A. Reproductive function during fasting in men. *J Clin Endocr Metab*. 1981;53:258–263.
275. Warren MP. The effects of exercise on pubertal progression and reproductive function in girls. *J Clin Endocr Metab*. 1980;51:1150–1157.
276. Frisch RE, Gotz-Welbergen AV, McArthur JW, et al. Delayed menarche and amenorrhea of college athletes in relation to age of onset of training. *JAMA*. 1981;246:1559–1563.
277. Frisch RE, McArthur JW. Menstrual cycles: fatness as a determinant of minimum weight for height necessary for their maintenance or onset. *Science*. 1974;185:949–951.
278. Shalitin S, Phillip M. Role of obesity and leptin in the puberal process and pubertal growth-a review. *Int J Obes Relat Metab Disord*: *J Int Assoc Study Obes*. 2003;27:869–874.
279. Tweedle D, Walton C, Johnston IDA. The effect of an anabolic steroid on postoperative nitrogen balance. *Br J Clin Pract*. 1972;27:130–132.
280. Watson RN, Bradley MH, Callahan R, et al. A six month evaluation of an anabolic drug, norethandrolone, in underweight persons. *Am J Med*. 1959;26:238–242.
281. Thaysen JH. Anabolic steroids in the treatment of renal failure. In: Gross F, ed. *Protein Metabolism*. Berlin, Germany: Springer-Verlag; 1962:450–478.
282. Wilson JD. Androgen abuse by athletes. *Endocr Rev*. 1988;9:181–199.

Gastrointestinal Development: Implications for Infant Feeding

22

Josef Neu, MD, Martha Douglas-Escobar, MD, and Sandra Fucile, OTR, PhD

The field of gastrointestinal (GI) development has been advancing at a rapid pace in the past 30 years.[1] The level of GI maturity of an individual infant is a major determinant of whether the infant will be able to meet nutritional needs by sole use of the GI tract or parenteral means will be necessary. The GI tract is not only an organ for digestion and absorption of nutrients; it also performs major endocrine, neural, and immunologic functions. In this chapter, anatomic, functional, and biochemical development will be described and related to means by which enteral nutrition can be used to optimize health and prevent disease (refer to Table 22-1 for a chapter overview).

ANATOMIC DEVELOPMENT

The intestine undergoes tremendous growth during fetal life. It elongates 1000-fold from 5 to 40 weeks gestation. The length doubles in the last 15 weeks of gestation reaching a mean length at birth of 275 cm. In the small intestine, fingerlike projections, villi, are already formed at 16 weeks gestation. In the large intestine, villi are also present, but these partially regress at around 29 weeks gestation. Microvilli begin to cover the apical surface of the small intestinal epithelium so that by adulthood, the intestinal surface provides the largest interface between the outside

environment and the internal milieu (approximately 200 m^2, which is about the size of a tennis court).

There are numerous cell types in the small intestine. These include the enterocytes (intestinal absorptive epithelium), Paneth cells (involved in secretion of defensins and other peptides implicated in the innate immunity), goblet cells (involved in the secretion of the mucous lining the intestine), and other cell types involved in the intestinal neuroendocrine and immune systems. After intestinal epithelial cells undergo mitotic division in the crypt, they migrate up the villus, where they undergo differentiation and become actively absorbing cells, then after apoptosis, become sloughed into the intestinal lumen. There is only one population of cells that migrate deeper into the crypt and these are the Paneth cells. Specific details regarding these cell types will be discussed later in this chapter.

TABLE 22-1 Developmental Challenges for very low-birth-weight (VLBW) Infant's Enteral Nutrition

Specific Developmental Challenges	
1. Mechanical development	a. Coordination of suck–swallow and breathing b. Low gastroesophageal sphincter tone c. Gastric emptying d. Intestinal motility
2. Absorption development	a. Proteins: only enteral protein are trophic for intestine b. Lipids: differences between medium- and long-chain fatty acids c. Carbohydrates: specially lactase enzyme
3. Innate defenses development	a. Gastrin b. Pancreatic secretions c. Secretion of defensins d. Specialized gut junctions e. Microbiota
4. Other challenges	a. Lack of initial enteral feedings b. Low volume of feeds c. Need for nutrients that decrease inflammatory state d. Need for nutrients for the colonocyte: short fatty acids

DEVELOPMENT OF MECHANICAL FUNCTION

Oral feeding is a major developmental milestone for the premature neonate and is one of the main criteria for hospital discharge. Oral feeding readiness is linked to GI tract functional aspects such as suck–swallow coordination, gastroesophageal sphincter tone, gastric emptying, and intestinal motility. These will be discussed separately albeit they function along as a continuum in the GI tract to propel food from the mouth to digestive absorptive areas.

Suck–Swallow Coordination

Suck and swallow are present as early as 17–18 weeks gestation. However, preterm infants born less than 32 weeks gestation have underdeveloped oral musculature, consequently they have difficulty latching on the nipple, difficulty initiating and sustaining a suck, and an immature and inefficient suck and swallow pattern compared to term infants.[2] In addition, they also present with difficulty coordinating suck-swallow-breathe processes due to their cardiorespiratory immaturity. Although respiration is always inhibited to accommodate swallow, preterm infants have a higher occurrence of swallows during inspiration and prolonged respiratory pauses.[2] These patterns put them at high risk for apnea, bradycardia, oxygen desaturations, and tracheal aspiration during oral feeding. Therefore, the current practice is to provide enteral feedings by tube (naso- or orogastric) until about 33–34 weeks gestation, when the coordination of suck-swallow-respiration begins to resemble that of full term infants. However, this practice remains subject to other immaturities such as, gastroesophageal reflux, poor gastric emptying, and dismotility. Also, feeding tubes may produce hypersensitivity and oral aversion because they are a constant nociceptive stimulus in the oral, pharyngeal, and esophageal areas. Provision of early (before oral feedings are started) sensorimotor therapies, and different infant positions and nipples are various interventions that can be used to prevent or reduce suck and swallow problems.

Gastroesophageal Sphincter Tone

Ganglia are present in the esophagus at five weeks gestation and complete their migration to the rectum by 24 weeks gestation. Despite this, the esophagus of the premature infant demonstrates slower propagation velocities and longer contraction duration than in the older child. At about 28 weeks gestation, the lower esophageal sphincter resting pressure is only

4 mmHg, but it increases to adult values (18 mm Hg) by term.[3] The prevalence of gastroesophageal reflux *disease* (GERD) is a debatable issue. At present, most of the techniques to determine GERD rely on acid measurement, but it is known nonacidic reflux cause reflex apnea. Modest changes in intra-abdominal pressure (such as changing a diaper) dramatically increases gastroesophageal reflux (GER) episodes.[4] Although GER episodes are frequently associated with apnea and bradycardia, there is no clear evidence of cause–effect relationship. Patients with persistent apnea-bradycardia and reflux even after changes in feeding techniques (e.g., changing bolus feeds to feeds over 1–2 hours) often receive empirical antireflux medication trial. Studies employing newer techniques of measuring reflux based on nonacid measurements (e.g., impedance) may yield important new information in this area. The recent suggestion that low-birth-weight infants with GER have an increased risk of esophageal adenocarcinoma in adulthood, makes this area a research priority.[5]

Gastric Emptying

Gastric emptying is slower in premature than in term infants[6,7] and leads to a greater residual volume of gastric contents—thus, it is closely tied to the problem of GER. Infants born between 32 and 39 weeks gestation decrease gastric emptying in response to progressive increments of caloric density.[8] Isocaloric formulas do not change gastric emptying even when the osmolality increases from 279 to 448 mOsm/kg.[8] During the last trimester, the fetus normally swallows up to 450 mL/d of low caloric density amniotic fluid.[9] Preterm infants have difficulty tolerating formula or breast milk with high caloric density. In adults, the rate of gastric emptying is controlled by feedback from the small and large intestine. Stimulation of duodenal receptors by acid, fat, carbohydrates, tryptophan, or increasing osmolality decreases rates of gastric emptying.[8] Very little information is available on the ability of duodenal feedback to control the rate of gastric emptying in infants born between 25 and 32 weeks gestation. One technique to improve feeding tolerance is the use formulas that promote fast gastric emptying (e.g., formulas with low carbohydrate or lipids). However, this technique may not be practical because extreme premature newborns have higher caloric demands to grow. Transpyloric feeding (through nasojejunal tube) is often employed to decrease the likelihood of reflux-related aspiration and apnea, but there is little evidence supporting it decreases premature infants mortality or morbidity.

Several studies suggest that medications used to increase gastric emptying rates in children and adults are also effective in premature infants. Erythromycin improves feeding tolerance without significant side effects in premature infants, although some randomized trial results were inconclusive.[10–12] Metoclopramide increases gastric emptying rate in preterm and term infants,[13] but its known side effects (such as extrapyramidal movement disorders or tardive dyskinesia) are difficult to monitor in this age group. Cisapride is another prokinetic agent, but it was removed from the US market because its association with cardiac arrhythmias.[14,15]

Intestinal Motility

The small bowel motility patterns are not completely developed in preterm infants born before 28 weeks gestation. Gastro-anal transit ranges from 8 to 96 hours in premature infants as compared with 4–12 hours in adults. Infants born between 27 and 30 weeks gestation have disorganized small bowel motility patterns that gradually progress to a more mature pattern and migrating myoelectric complexes are present at 33–34 weeks gestation.[16] Important motility mechanisms such as the motilin receptor and the cyclic release of the motilin are not present until 32 weeks gestation. It is intriguing that erythromycin, a motilin agonist, improved feeding tolerance in some premature infants.[11,12] Whether erythromycin might act through mechanisms not related to motilin remains speculative. Minimal enteral feedings increase GI motility (details will be discussed later in this chapter).

DEVELOPMENT OF DIGESTIVE ABSORPTIVE FUNCTION

Protein Digestion and Absorption

Our knowledge of digestive–absorptive processes has increased significantly in the past three decades. Hydrolysis of protein begins in the acid environment of the stomach and proceeds distally in the intestine through the action of various proteases. Gastric acid secretion is limited in very low-birth-weight (VLBW) infants. In the first 24–48 hours after birth, intragastric pH remains around 5.5–7 (whereas the normal gastric pH in children and adults is 2–3) and is relatively resistant to pentagastrin. Pentagastrin doubles from the first to fourth week of postnatal life in

preterm infants.[17] Gastric acid serves as a barrier to microorganisms, thus the use of H2 blockers to decrease acid production may lead to a higher load of bacteria in the more distal regions of the intestine.[18] Critically ill low-birth-weight infants treated with H2 blockers have a higher incidence of nosocomial sepsis and necrotizing enterocolitis (NEC).[19,20]

The pancreatic protease digestive cascade is catalyzed by food-stimulated secretion of enterokinase from the upper small intestinal epithelium. Enterokinase catalyzes the activation of trypsinogen to trypsin, which in turn activates several other inactive zymogens into proteases. Active luminal enterokinase is detectable at 24 weeks gestation, but its activity level is relatively low and reaches only 25% of adult activity at term.[21] Low enzymatic activity may limit protein digestion and may be responsible for transluminal passage of larger antigens and/or microorganisms without breakdown by luminal enzymes. After luminal digestion, small peptides and amino acids are absorbed and transported to the villus capillaries. The absorptive proteases are present and fully active before 24 weeks gestation and should not be limiting step in the protein absorption.[22] This fact suggests that hydrolyzed protein could be better tolerated in premature infants. Indeed, one study demonstrated that hydrolyzed protein formula led to faster establishment of full enteral feeding than a nonhydrolyzed protein formula.[23] However, the mentioned study did not have a human milk group. Human milk is much better tolerated than formula.[24] Many hydrolyzed protein formulas are not nutritionally formulated for premature infants.

Lipid Digestion and Absorption

Lipid makes up about 50% of the nonprotein energy content of human milk and formulas. However, both preterm and term newborns have a relative pancreatic insufficiency when compared to older children and adults, which may result in mal absorption of certain types of lipids. The digestion of lipid can be split into three phases.[25] The luminal phase of lipid digestion involves lipase de-esterification of triglycerides to 3-monoglyerides and free fatty acids and bile acid-mediated micellar solubilization. Several lipases participate in the fat digestion including lingual, gastric, pancreatic, and epithelial lipases as well as the human milk lipase. The lingual lipase is secreted by glands at the base of the tongue and is involved in gastric lipid hydrolysis. Other lipases are produced in the stomach or secreted from the pancreas. The human milk lipase (absent in cow’s milk and formulas) becomes active in the small intestine lumen

only in the presence of bile acids.[25] Bile acids synthesized in the liver are critical to efficient fat digestion and absorption. VLBW infants have a low concentration of bile acids in the duodenum due to low synthesis and ileal reabsorption of bile.[26,27] Therefore, VLBW have a lower luminal micellar solubilization with the end result of lower lipid absorption than term infants. Long-chain fatty acids required bile acids for solubilization, therefore their absorption is inefficient when compared to medium-chain fatty acids. Nevertheless, meta-analysis of studies comparing medium-chain triglycerides (MCT) to long-chain triglycerides feeding do not show clear weight gain advantage.[28]

Lipid absorption and assimilation phases include permeation of fatty acids and 2-monoglycerides from the lumen into the cell, intracellular reesterification, chylomicron formation, and transport into the circulation. After luminal digestion, fatty acids and monoglycerides approximate the absorptive surface and enter the intracellular milieu by processes that are not yet well understood. MCT that entered to the enterocyte do not need reesterification or chylomicron formation and are taken directly into the portal venous system. In contrast, intracellular long-chain fatty acids undergo reesterification and chylomicron formation before entering into the lymphatic system (Table 22-2).[29] Therefore, MCT predominant formula is recommended when there is obstruction of the lymphatic system (e.g., chylous ascites).

Linoleic or Linolenic fatty acids are considered essential fatty acids (i.e., must be ingested because the human body cannot produce them).[29] However, there is considerable debate about the capability of preterm infants to convert the dietary essential fatty acids (18-carbon linoleic and linolenic fatty acids) into longer chain fatty acids (with 20 or more carbons). Long-chain polyunsaturated fatty acids (LCPUFAs) formed using desaturase and elongase enzymatic pathways are critical in the formation of eicosanoids and structural components of the central nervous system.[30] Preformed LCPUFAs are abundant in human milk, but are not found in commercial intravenous lipid preparations or, until recently, most neonatal formulas. A meta-analysis and systematic review showed advantages of docosahexaenoic acid (an omega-3 LCPUFA) supplemented formulas over unsupplemented formulas in behavioral and electrophysiologic-based measurements of visual acuity in preterm and term infants.[31,32] Although research suggests that LCPUFA supplementation provides visual benefit in VLBW infants, improvement in cognitive neurodevelopment is currently being investigated.[30]

Most of the lipids from the formulas or intravenous lipids are derived from vegetable oil (rich in omega-6 but not omega-3 LCPUFAs). Therefore, the majority of essential fatty acids provided to neonates are derived from the omega-6 family (linoleic acid). Ingestion of products derived from omega-6 fatty acids (2-series prostaglandins) have a greater systemic inflammatory effect than those derived from omega-3 fatty acids (3-series prostaglandins).[33] Although there is a considerable amount of information in adults about the omega-3 fatty acids ameliorating inflammation (e.g., rheumatoid arthritis, coronary artery disease, and other conditions), there is a paucity of information for neonates, particularly preterm infants. In animal studies, using a rat model, omega-3 fatty acid supplementation has been found to decrease the occurrence of NEC.[34] Whether providing omega-3 lipids supplementation will decrease NEC, chronic lung disease, or neurologic problems related to inflammation in human infants warrants further investigation. New designs of lipid solutions are attempting to add MCT and fish oil to the traditional oleic acid solutions.

TABLE 22-2 Comparison of Short, Medium, and Long Chain Fatty Acid Digestion and Assimilation

Overview of Fatty Acid Uptake
Short- and medium-chain fatty acids
Enter portal blood directly from enterocytes
Bound to albumin in blood
Albumin–FFA complex
Oxidized in liver or elongated and used for triglyceride formation
Long-chain fatty acids
Form Chylomicrons in intestinal cell
Drain into the lymphatics via the lacteal
Enter bloodstream at the thoracic duct
Upstream from liver
Slow entry into the blood

Overall, premature infants receive very little, if any, preformed omega-3 LCPUFAs while on minimal enteral feeding and/or parenteral nutrition and this is likely to have detrimental consequences in both nervous tissue formation and inflammatory responses.

Carbohydrate Digestion and Absorption

Neonates ingest carbohydrates either as natural lactose (the primary carbohydrate in human and mammalian milks) or glucose polymers, sucrose, or hydrolyzed starches. Pancreatic amylase hydrolyzes starches by cleaving internal α-1-4 glucose bonds to maltose, maltotriose, limit dextrin, and glucose. Pancreatic secretion of amylase is limited during the first several months of life; therefore starch hydrolysis is a limiting factor and leaves undigested starch in the intestinal lumen. Glucose polymers (18–29 glucose units) can be hydrolyzed by salivary amylases, but this digestion is not enough to compensate for the diminished concentrations of pancreatic amylase.[35] Many term and preterm infant formulas include partially hydrolyzed starches (such as corn syrup solids or tapioca) to overcome problems related with carbohydrate hydrolysis. The more extensively the starch is hydrolyzed, the greater increase in formula osmolality (and risk of hyperosmolar diarrhea and/or intestinal inflammation). Maturation of carbohydrate absorption mechanism has a defined progression during human fetal development.[22,29,36,37] The intestinal epithelial absorptive enzymes, lactase, sucrase, maltase, isomaltase, and glucoamylase, are at mature levels in the term newborn.[21] Preterm infants have sucrase, maltase, and isomaltase almost fully active, but low lactase activity, depending upon gestational (lactase activity increases markedly from 24 to 40 weeks gestational age).[21] Despite this developmental lactase deficiency, clinical lactose intolerance is uncommon. Postnatal adaptive responses to ingested carbohydrates lead to competent carbohydrate utilization.[38] Furthermore, there is a colonic salvage of nonabsorbed carbohydrates. The colonic microbiota ferment the nonabsorbed carbohydrates to hydrogen gas and short-chain fatty acids (SCFAs), which are easily absorbed by the colonocyte.[36]

Feeding route and composition of the milk (or formula) affect the lactase activity in the small bowel.[38] Preterm infants receiving early enteral feedings (before 4 days of age) have greater lactase activity than control group (feedings initiated after 15 days of age) at 10 and 28 days of age by 100% and 60%, respectively. Lactase activity was greater in human milk- versus formula-fed infants at 10 days of age. Lactase activity was inversely related with the time required to achieve full enteral feedings and the total number of abdominal X-ray examinations per infant. These findings show that early enteral feeding increases lactase activity in preterm infants.

VLBW infants' feedings are seldom initiated at levels intended to meet

the infants' entire nutritional requirements and usually feeds are advanced slow, therefore they are unlikely to exceed the lactose hydrolysis capacity and intestinal salvage ability. The use of lactose-free formula instead of human milk or lactose containing formula is not appropriate to feed premature infants and theoretically can be harmful. High lactose concentrations in human milk should not be a contraindication for its use in the VLBW infants. This is especially unlikely when the quantity fed is less than 50% of the caloric requirement provided via the GI tract (unless the infant's bowel has been radically shortened by surgery). Human milk also contains several lactose-derived oligosaccharides and other glycoconjugates that may play an important immunologic role.

Studies examining the crypt to villus gradient of intestinal carbohydrase activities demonstrate that the majority of lactase activity is found at the mid-to-upper villus, whereas sucrase, maltase, and glucoamylase are concentrated at the mid-villus region.[39,40] This is likely to be pertinent to intestinal injury and villus damage. Lactase is usually the first enzyme to be lost and the last to be fully regenerated after injury. Congenital lactase deficiency is extremely rare. A large proportion of the world's population develops lactose intolerance in the first few years of life because of a decline in lactase activity, but further discussion is beyond the scope of this chapter.

INNATE DEFENSE AND BARRIER FUNCTION DEVELOPMENT

The GI tract presents the largest surface area of the body to which antigens and microbes are exposed. The intestine must therefore have intricate mechanisms to allow entry of nutrients and other beneficial molecules while preventing potentially harmful microbes and other agents from gaining entry into the inner milieu. Shortly after birth, the establishment of an intestinal bacterial community is very important for maintenance of normal homeostasis. Several factors lead to a hostile GI environment that predisposes the VLBW infant in the neonatal intensive care unit (NICU) to disease. These include the introduction of feeding tubes into the stomach or more distal intestine, the use of broad-spectrum antibiotics that select for the NICU-resistant pathogens, the restriction of enteral feedings, and the intrinsic functional problems of the preterm infant's GI tract. The combination of total parenteral nutrition (TPN), lack of enteral nutrition, and the introduction of a hostile microbiota into an immature GI tract offers a fertile environment for GI-derived systemic inflammation and

programming of pathologic immune responses.

Intestinal defenses may be broadly divided into innate and adaptive categories. The importance of the innate system is that it constitutes the first line of host defense. Each of the components of the innate system acts in concert to maintain mucosal integrity in the intestine. Several of these components are presented here.

Breast Milk

Breast milk is a protective component of innate defense for the neonate. Breast milk provides immunoglobulins, lactoferrin, lysozyme, glycoconjugates, oligosaccharides, and various types of white blood cells. Antibodies in human milk reflect the antigenic repertoire of the mother's intestine and respiratory tract. Colonization of the lactating mother's intestine with specific strains of *Escherichia coli* results in the appearance of corresponding antibodies in the mother's milk.[41] This effect is achieved by seeding primed plasma cells to the breast tissue. The array of antibacterial, antiviral, antifungal, and antifood antibodies in human milk is considerably vast. This repertoire varies from mother to mother and is modified by the antigens ingested and inhaled by the infant.[42] Mother and babies in close contact have similar bacterial colonization and the maternal breast milk has specific antibodies for the infant protection. This defense is referred to as "enteromammary immune system."[43] Furthermore, mother's milk contains commensal bacteria that may play a protective role and act as a natural "probiotic."[44] Oligosaccharides in mother's milk are "prebiotic" because they provide nutrients for beneficial commensal bacteria. In addition, the oligosaccharides are "postbiotic" because its fermentation produces SCFAs used as a main fuel by the colonocyte. Butyrate, one of the short fatty acids, is an antiapoptotic, pro-proliferative agent that is associated in the strengthening of intercellular tight junctions,[45] and stimulation of the synthesis of the glucagon-like peptide-2 (an intestine trophic hormone).[46] Human milk contains high levels of interleukin-10 (IL-10) and transforming growth factor-beta, anti-inflammatory cytokines that modulate intestinal inflammation.[47]

Gastric Acid and Pancreaticobiliary Secretions

Gastric acid and pancreaticobiliary secretions decrease the load of viable microorganisms and intact dietary protein antigens that reach the small intestine, as previously discussed.[48] The newborn relative pancreatic insufficiency can last through the first year of life.[49] Low gastric and

pancreaticobiliary secretions allow a greater bacterial and/or antigenic load to pass into the distal intestine. In normal conditions, GI tract proteolysis destroy the spatial structure of antigenic proteins, including the antigen epitopes for immunologic recognition.[50] Animal studies demonstrate that GI alkalization increases the passage of intact proteins such as bovine serum albumin in rats.[51] In addition, H2 blockers lead to greater bacterial translocation in newborn rats.[18] These findings are concerning because during the first few days of life, many VLBW infants have no or minimal stimulation of gastric acid and intestinal proteases (due to lack of enteral feeding during first days of life and H2 blocking agents therapy). Therefore, these infants have more intestinal exposure to intact protein molecules and bacteria, increasing the risk for allergies and infections. This could explain why VLBW infants receiving H2 blocking agents have high incidence of hospital-acquired sepsis and NEC.[20,52]

SURFACE EPITHELIUM

Intestinal Absorptive Epithelium

The absorptive lining epithelium of the primitive intestine is initially stratified but becomes a single layer of columnar cells at the end of the first trimester. The intestinal epithelial enterocyte derives from the same undifferentiated stem cell that gives rise to columnar absorptive epithelium, mucus producing goblet cells, Paneth and M cells in the intestine.[53] The function of these cells, beyond nutrient absorption, is well recognized, and some of their major roles include bacterial mucosal cross talk and innate barrier function.

Goblet cells. These are specialized mucus-secretory cells found throughout the intestine. Goblet cells mature from the base of the crypt (proliferative zone) and migrate along the crypt–villus axis. After 5–7 days, they reach the tip of the villi and are exfoliated (by a particular programmed cell death called “anoikis”). Goblet cells secrete mucus/mucin, which are very important in the nonspecific protection of the gut lining. Intestinal mucus is a complex gel that covers the surface of the villous epithelium and contributes significantly to cytoprotection at the same time that it offers many ecologic advantages for the microbiota. It is composed primarily of water and electrolytes but also contains mucins, glycoproteins, immunoglobulins, glycolipids, and albumin.[54] Bioactive factors such as lactoferrin, lysozyme, and glycolipids from breast milk may also be contained within the surface mucus gel.[55] Mucus is secreted

into the lumen of the intestine from storage vacuoles in goblet cells, and these cells are interspersed among enterocytes along the villus to crypt axis. Mucous gels are sufficiently stable to keep foreign dietary antigens and pathogenic bacteria trapped. In response to a threatening insult, the intestine can release rapid outpouring of mucin and resident microbiota can induce an increase in the production of mucins. Mucins may be classified according to their structure as gel forming, soluble, and membrane bound. Mucins develop binding sites for lectins, selectins, adhesion molecules, growth factors, cytokines, and chemokines, suggesting that they are implicated in cellular signaling. Mucins lower the diffusion of large molecules (which limit the antigenic load exposure), but allow the passage of small nutrient molecules. Mucins composition change during development. Mucin from small intestine of newborn rats contains more protein than does the adult rat intestine.[56] Carbohydrate composition also changes; the newborn has less fucose and *N* acetyl-cysteine than does the mucin from adults. Levels of mucin in the small intestine decrease in experimental protein-calorie under nutrition, which reduce the GI protection during the perinatal period.[57] Perinatal malnutrition lowers the amounts of mucin, diminishing infant's protection. Certain nutrients such as threonine have a major effect on mucin production[58] and need to be considered in the nutrition of critically ill infants.

Paneth Cells. Paneth cells represent one of the four major epithelial cell lineages in the small intestine and are the only lineage that migrates downward into the crypt base after originating in the crypt stem cell region. The location of Paneth cells adjacent to crypt stem cells house suggests that they play a critical role in defending epithelial cell renewal. In response to pathogen attack, the Paneth cells secrete a wide spectrum of *antimicrobial peptides* (AMPs) against gram-negative and gram-positive bacteria, fungi, protozoa, and viruses. The AMPs are able to kill the pathogens by different mechanisms. They are able to attach to and insert into microbial membrane bilayers to form pores, reduce the synthesis of cell wall and nucleic acid, and inhibit enzymatic activity. Some of the major enteric pathogens have developed resistances to AMPs as a way to evade innate mucosal defenses.

One of the main group of AMPs secreted by Paneth cells are alpha-defensins, also known as cryptdins.[59] These peptides have hydrophobic positively charged domains that can interact with phospholipids in the cell membranes. These domains allow formation of pores that disrupt bacterial membrane function, leading to bacterial death. Paneth cells are stimulated to secrete defensins when exposed to gram-positive or gram-negative

bacteria or other bacterial products such as lipopolysaccharide (LPS). Alteration of AMPs is detrimental to the neonate's small bowel defense against *E. coli* infection. In addition to defensins, Paneth cells secrete other antimicrobial enzymes such as lysozyme and phospholipase A2. These battery of peptides secreted by Paneth cells are an effective host protection against bacteria, fungi, and even some enveloped viruses.

M-cells. M-cells in the intestine do not have well-developed microvilli and allow macromolecular transport, whereas the absorptive enterocyte has better developed microvilli. M-cells are present only in the follicles that overlie organized mucosa- associated lymphoid tissues.[60] M-cells are specialized in presenting foreign antigens and microorganisms to organized lymphoid tissues within the mucosa of the small and large intestines. Thus, M-cells play a major role in specific immunity to luminal antigens. However, M-cells' role in neonatal diseases is unknown.

Specialized interepithelial junctions. Unique mechanisms are required to allow or prevent passage of substances between intestinal epithelial cells. Special junctions and interdigitations join the intestinal epithelial cells as seen in Figure 22-1. The tight junction (zonula occludens) is the most apical component of the junctional complex and serves as the permeability barrier between the external and internal milieus of the body. The tight junction completely encircles the apical end of absorptive cells as a belt-like band. Several transmembrane proteins on the cytosolic leaflet are part of the tight junction. These include the claudin and occludin protein families. Interactions of some of these proteins with the actin cytoskeleton are a major determinant of tight junction structure and play a role in the regulation of tight junction assembly.[61] Just below the tight junction, the adhesion junction (zonula adherens) is connected to actin filaments in the cytoplasm and anchors each cell to adjacent cells. Below this, the desmosome (macula adherens) functions in a similar manner to the zonula adherens. The relationship between density of zonula occluden strands and epithelial resistance has been demonstrated in cell culture studies.[62] These junctions allow for physiologic passage of fluids, electrolytes, and small macromolecules up to 11 amino acids long,[63] but larger proteins may not pass this route under physiologic conditions. However, pathologic insults may alter these junctions so that they allow abnormal passage of particles. In addition, these intercellular junctions are exquisitely sensitive to modulation by small molecules. SCFAs produced by bacterial fermentation such as butyrate has been shown to enhance restoration of mucosal barrier function after injury.[64] Administration of glutamine (GLN), an amino acid known to be conditionally essential

during critical illness, can also prevent the disruption of tight junctions and subsequent increased paracellular permeability induced by a variety of insults.[65,66] Our ability to manipulate these junctions may be key for prevention of several diseases that are autoimmune in nature and may at least partially relate to "a leaky gut" in its pathogenesis.[67] Figure 22-2 illustrates some of these diseases.

Epithelial turnover and restitution. It is unclear whether the infant can respond to injury to the same extent as the adult. In mammals, the intestinal epithelial turnover is much slower in the infant (4–5 days) than in the adult (two days).[68] If the turnover in human neonates is longer than the adult, as in other mammals, is of significance in that regeneration of injured mucosa in the infant will be much slower than the adult. In addition, environmental factors may contribute to poor restitution in intestinal epithelial cells after injury. In an animal model of NEC, it was determined that intestinal restitution was significantly impaired compared to control animals.[69] The mechanism of this effect was evaluated in vitro with rat crypt cells (IEC-6), which were shown to undergo significant inhibition of restitution after exposure to LPS.[69] GLN has been demonstrated to prevent cytokine-mediated apoptosis in human colonic cells.[70] Another amino acid, arginine, also induced intestinal epithelial migration, but through the mammalian target of rapamycin (mTOR) pathway.[71] The role of these amino acids in counteracting intestinal stresses and insults warrants further investigation.

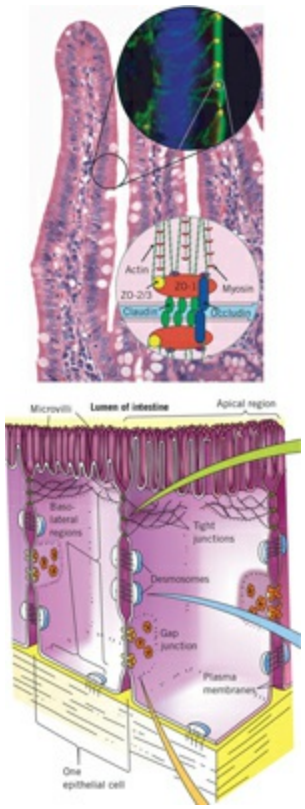


Figure 22-1 Top, Intestinal villi with immunofluorescence of tight junction proteins. Bottom, Relation of tight junctions to the other junctional complexes. (With permission from Clayburgh DR, et al.[100])

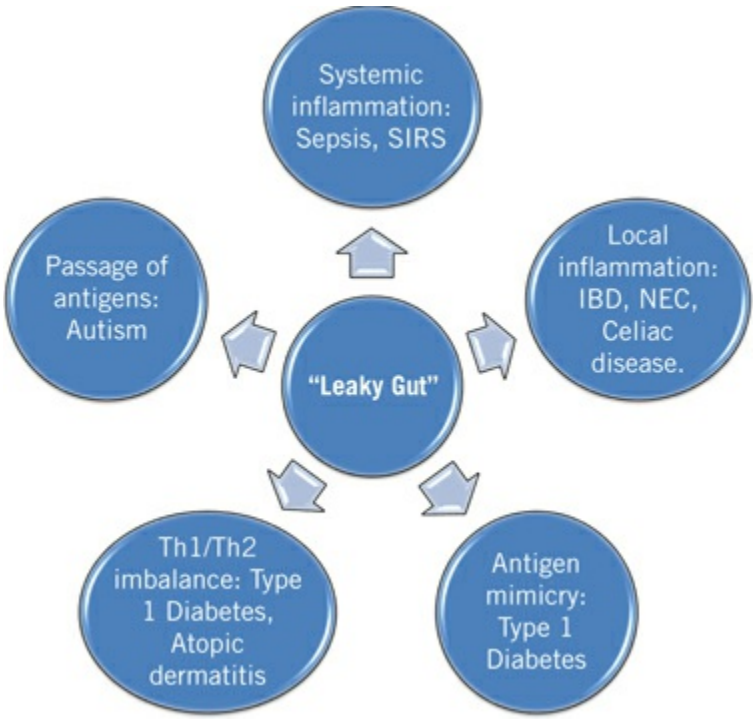


Figure 22-2 Benefits of minimal enteral nutrition for very low-birth-weight (VLBW) infant.

THE INTESTINAL ECOSYSTEM AND MICROBIOTA

It is clear that there is a close interplay between the major cell types lining the intestine, nutrients, and the microbiota, which are all constituents of the intestinal ecosystem. GI infections and antibiotic treatment reduce the normal commensal bacteria in the ecosystem. Reduced commensal bacteria alter normal stimulation of the GI immune response as well as the amount of nutrients available. Similarly, the lack of enteral nutrients negatively affects the host and the microbiota.

We are beginning to appreciate the profound changes that occur in the intestinal ecosystem when young mammals are weaned from their mother's milk. One example is the changes in the angiogenins, potent AMPs released from the Paneth cells of the crypt.[72] Mice rose in a conventional environment have a marked increase in angiogenins at the time of weaning from mother's milk to an adult diet. This change occurs to a much lesser degree in germ-free mice.[73] Commensal bacteria *cross talk*

with the GI tract and therefore the production of angiogenins is critical for normal villus capillary development. For example, mice grown in a germ-free environment have a lag in development of the villus capillaries in the small intestine when compared with mice grown in an environment containing normal mixed flora or only one species of commensal bacterium (Bacteroides thetaiotaomicron).[74]

Most of premature neonates receive broad-spectrum antibiotics at some point in their intensive care hospitalization. The effect of broad-spectrum antibiotics on the resident intestinal microbiota is not known, but there is indirect evidence that may be detrimental. In rodents, when the intestinal tract is depleted of microbes, there is a reduction in the ability of the intestine to tolerate otherwise benign insults.[75,76] Furthermore, genes responsible for intestinal development are altered even after treatment with only one antibiotic.[77] Most recently, there has been an explosion of research on oral supplementation with commensal bacteria (probiotics) for the prevention of diseases, such as NEC.

DEVELOPMENT, INFLAMMATION, AND NUTRITION

The reliance on parenteral nutrition while providing few or no enteral nutrients may be highly detrimental because it promotes intestinal inflammation, atrophy, and sepsis as seen in Figure 22-3. The role of enteral nutrients in the prevention of the intestine-derived inflammatory response syndrome is becoming increasingly recognized.[78] Certain individual nutrients may play an important role in modulating these effects. For example, GLN depletion in LPS-treated Caco-2 (an adult transformed cell line) and, even more dramatically, in H4 (human fetal cells) results in marked upregulation of IL-8 production.[79] Similarly, in infant rats fed a commercial diet by gastrostomy and stimulated with LPS, GLN, decreases chemokine-induced neutrophil chemoattractant (the rodent counterpart of human IL-8) production in the intestine.[80] Several studies have suggested similar effects in vitro for probiotics, but studies in neonatal animals showing regulation of intestinal inflammation by probiotics have only recently been published.[81] Studies in human intestinal epithelial cell cultures have also shown that human milk suppresses IL-1 beta-induced IL-8 production.[82] Many of these nutritional agents appear to be acting through modulation of nuclear factor kappa B (NF-κB) signaling and transcriptional regulation, but precise mechanisms are yet to be

defined.

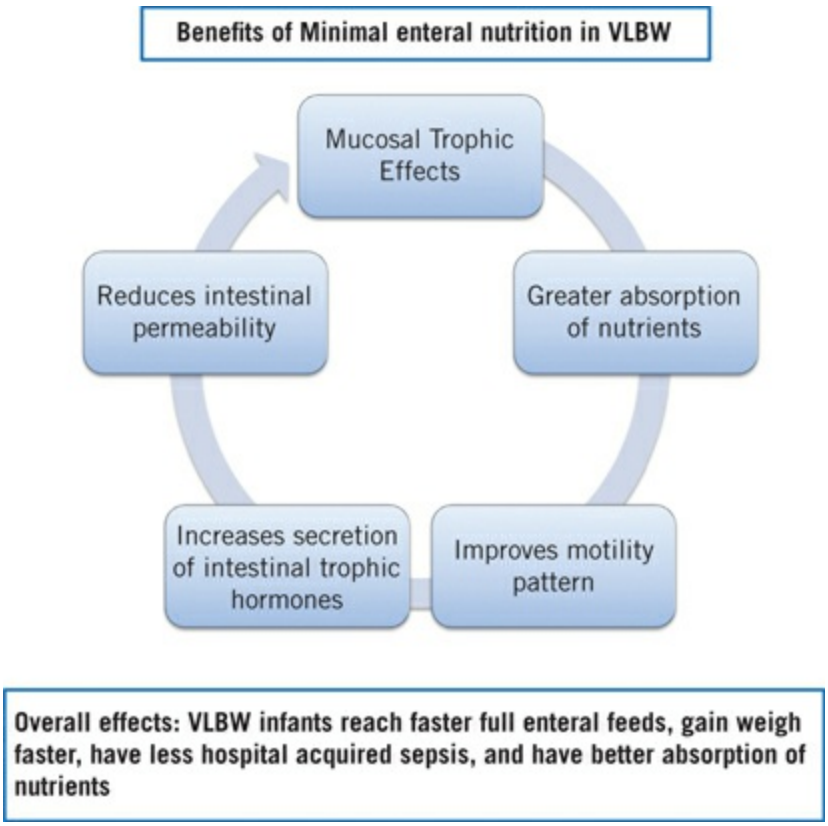


Figure 22-3 An amino acid: an amino (NH_2) group, a carboxyl group (COOH) and a side chain attached to a central α-carbon (R).

INTERVENTIONAL APPROACHES TO NEONATAL ENTERAL FEEDING

Total Parenteral Nutrition, Minimal Enteral Nutrition, and Trophic Feeding

TPN may cover the neonate caloric needs, nevertheless lack of enteral nutrition contributes to breakdown of the intestinal mucosa, bacterial translocation, and systemic inflammation (Figure 22-3).[78] These changes are even more pronounced if the neonate is a premature or VLBW infant. VLBW infants are frequently stressed due to prematurity problems such as respiratory insufficiency, thermoregulation, systemic infections, and others. Because of the high protein requirements in VLBW infants, they are commonly started on TPN to provide more protein intake.[83] Most

VLBW infants receive TPN during the first weeks after birth (during the acute illness), then they start enteral feeds—either human milk or formula. This practice has improved neonatal nutrition, however, it is still suboptimal because of the lack of early enteral nutrients to stimulate the GI ecosystem. During the first weeks of life, some VLBW infants receive "minimal enteral nutrition" (enteral feeds that are <20% of total nutritional requirements to grow for at least five days) (Figure 22-3). Despite often being termed "trophic enteral feeds" it is unlikely that minimal enteral nutrition provides significant GI trophic effect and enhances the barrier function.[84] TPN is associated with hospital-acquired sepsis,[85] probably due to the translocation of intestinal bacteria and intestinal inflammation.[86] In an animal study, enteral nutrition reversed intestinal bacterial translocation and inflammatory cascade triggered by TPN.[87] Neonates, especially VLWB, may benefit from early intravenous amino acid intake because it can decrease catabolism and increase protein synthesis and somatic growth. Early enteral nutrition is very important to maintain GI barrier function and optimal GI tract development as well as a prevention or modulation inflammatory diseases, NEC, hepatic inflammation, chronic lung disease, and progression of periventricular leukomalacia.[88–90] There is a great need for optimization of an enteral formulation that can be safely used in critically ill infants as an adjunct to minimal enteral nutrition shortly after birth that will decrease the need for parenteral nutrition.

How to nourish the VLBW infant during the first 2–3 weeks of life, when they are more critically ill and in catabolic state is one of the main challenges in neonatology. This is a period of extreme vulnerability and high nutrient requirement. A deficiency of essential nutrients during this time may result in significant short-term morbidity as well as lifelong consequences. Hence, "When and how quickly should we start and advance enteral feedings in preterm infants?" is a prominent clinical question. Because of individual characteristics of each infant, one feeding protocol or guideline is not appropriate for all infants. From the available studies, minimal enteral feedings should be instituted within the first days of life. There is no evidence that minimal enteral nutrition increases the risk of NEC in infants with mechanical ventilation, continuous positive airway pressure, indomethacin therapy apnea and bradycardia episodes, or umbilical catheters. Infants on early minimal enteral nutrition have faster maturation of GI motor patterns and release of GI hormones than those without enteral feedings. In addition, infants treated with early minimal enteral nutrition reach full enteral feeds faster and have fewer feeding intolerance, without an increase in the incidence of NEC. Other important

consideration is the quantity and quality of the early enteral nutrition.[91] The volume of feeds does not appear to have a benefit in maturing motor function. Motor responses are similar if feedings are intragastric or transpyloric. Motor responses are unchanged with the formula temperature (chilled, room temperature, or warmed to body temperature). Speed of volume infusion affects the gastric emptying. Infants fed at slow infusion rates (over 120 minutes) display intense feeding response accompanied by brisk gastric emptying. However, when the same feeding volume is given over 15 minutes, this produces a duodenal motor response that is less intense and is associated with delayed gastric emptying. Diluted formulas produce less intense motor responses. Because two-thirds of preterm infants have immature duodenal fed pattern and delayed gastric emptying, many of them are not physiologically prepared to process bolus feedings as well as slow infusion feedings. Taken together, Figure 22-4 shows a strategy for starting early minimal enteral feedings in VLBW infants. However, multicenter randomized studies should test whether this approach is safe and efficient in VLBW infants.

FUTURE DIRECTIONS

The fetus receives about 450 mL/d of amniotic fluid through the GI tract during the last trimester of pregnancy,[9] whereas GI tract of premature infants during the first weeks of life receive only 5%–10% of the volume that would go through the intestine per day in utero. Thus, the GI tract of premature infants might be able to tolerate more volume than what is commonly provided. Osmolality and lipid composition of the feeds are critical factors for tolerance to enteral feeding, because they modulate gastric emptying and intestinal motility. There are several individual nutrients that could potentially enhance intestinal growth and function. Here, we will discuss a few of them; enteral protein, GLN, long-chain omega 3 polyunsaturated fatty acids, and SCFAs (Figure 22-5).

Protein

Protein is the major macronutrient required for growth, but parenteral amino acids do not provide trophic stimulation for intestinal growth. In animal models, trophic effect on the GI mucosa occurs when at least 40% of the caloric intake required for normal growth is supplied by enteral route.[84,92] This trophic effect is most likely derived from enteral protein rather than lipid or carbohydrates. Prevention of epithelial atrophy and mucosal growth is critical for the neonate to have normal GI tract

functions (such as digestion, absorption, barrier function) and to keep microbiota (that could protect from infections and modulate the inflammatory cascade).

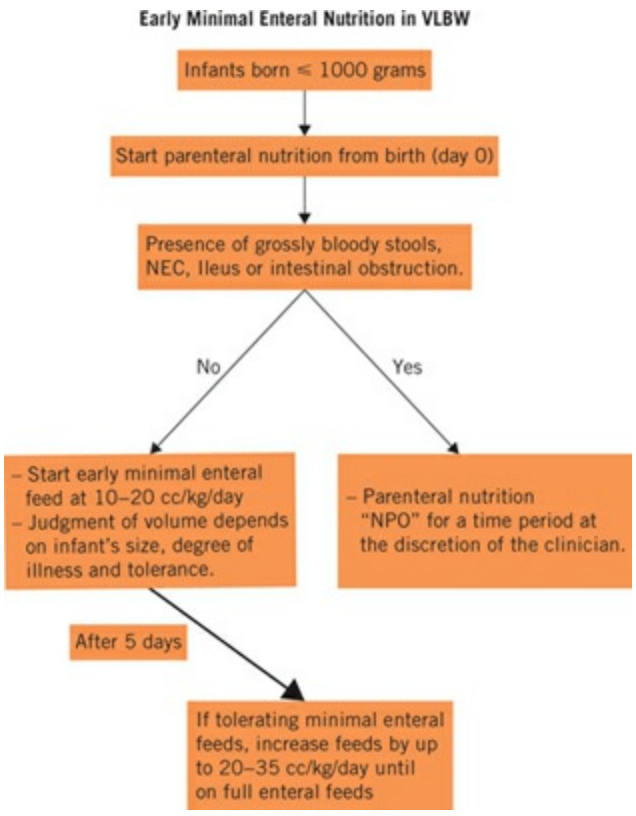


Figure 22-4 Early minimal enteral feedings in very low-birth-weight (VLBW) infants

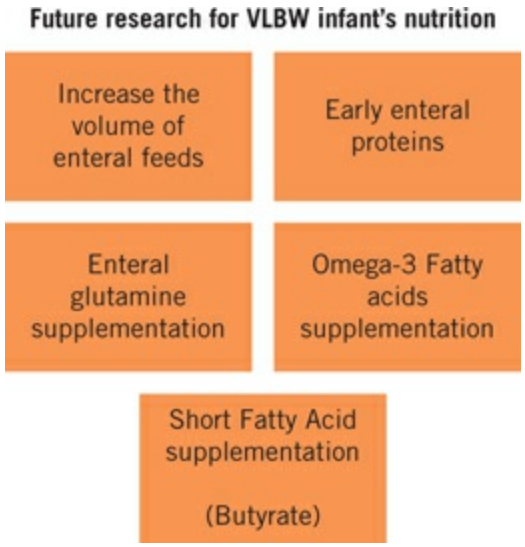


Figure 22-5 Future direction in very low-birth-weight (VLBW) infant's nutritional manipulation.

GLN

Most critically ill premature infants are not supplemented with the amount of GLN they would be receiving had they remained in utero. Lack of intestinal mucosal integrity and exquisite susceptibility to intestinal injury in VLBW infants could be associated to delayed enteral feeding and GLN deprivation. Several studies have demonstrated the efficacy and safety of GLN supplementation in highly stressed critically ill infants.[93–95] However, infants born prematurely receive very little, if any, GLN during the first weeks of life, because they rarely reach full enteral feedings (which contain GLN) for weeks and GLN is absent in routinely used TPN. VLBW infants with enteral GLN supplementation have less hospital-acquired sepsis, along with less CD16 and HLA DR markers on peripheral blood lymphocyte populations, consistent with decreased inflammatory response (probably secondary to decreased translocation of proinflammatory antigens).[93,95] In a recent multicenter trial, 649 VLBW infants with enteral GLN supplementation improved intestinal function and had less grades 3–4 intraventricular hemorrhages and periventricular leukomalacia.[94] However, one large study of intravenous GLN supplementation in neonates did not show any benefit in extremely low-birth-weight infants.[96] Studies on GLN supplementation should target infant subgroups (e.g., infants born to mothers with chorioamnionitis or

other inflammatory conditions), as well as compare dosage response, route, and timing of administration to fully understand and determine the beneficial effects of GLN.[97]

Omega-3 Fatty Acids

An important change to the composition of infant formula in the past several years has been the addition of LCPUFAs. The LCPUFAs added are docosahexaenoic acid (DHA, 22:6*n*–3) and arachidonic acid (AA, 20:4*n*–6). DHA and AA are integral structural components of all cells. Human milk contains DHA and AA. Infants fed with unsupplemented formulas have lower DHA and AA concentrations in their plasma and erythrocytes than do infants who are breastfed or fed supplemented formulas. Critically ill VLBW infants receive only minimal quantities of DHA and AA during the critical illness, through minimal enteral nutrition or TPN. Intravenous lipids are composed of the pro-inflammatory omega-6 fatty acids. Omega-3 LCPUFAs from fish oils and seed extracts decrease the production of inflammatory eicosanoids and cytokines. They inhibit AA metabolism and alter the expression of inflammatory genes through effects on transcription factor activation.[98] Thus, omega-3 LCPUFAs are potential anti-inflammatory agents. Future randomized multicenter studies should determine whether omega-3 LCPUFAs could improve clinical outcomes in VLBW infants.

SCFAs

SCFAs, especially butyrate, play central metabolic roles in maintaining the mucosal barrier in the gut. These SCFAs are a product of bacterial metabolism of carbohydrates, and responsible for several beneficial effects of commensal bacteria or probiotics.[45] Therefore, SCFAs are considered "postbiotics." The use of probiotics for premature infants is very controversial due to safety concerns in providing live bacteria to immunocompromised hosts. Butyrate offers some of the benefits of probiotic bacteria without side effects. Butyrate increases wound healing and reduces inflammation in the small intestine. Butyrate is the main energy source for colonic epithelial cells and affects cellular proliferation and differentiation.[45] Luminal provision of butyrate improves wound healing after intestinal surgery and ameliorates symptoms of inflammatory bowel disease. Furthermore, butyrate stimulates intestinal L-cell to produce glucagon-like peptide-2 (a highly trophic peptide for intestine).[46,99] There is a paucity of information on the effects of butyrate

as a nutrient for the developing intestine.[45]

SUMMARY

We reviewed several aspects of anatomic, physiologic, and biochemical development of the GI tract. We explained the importance of the GI tract beyond the classic paradigm of digestion and absorption of nutrients for optimization of growth. Nutritional needs of premature infants are often difficult to meet due to GI tract functional problems such as coordination of suck and swallow, frequent GER, slow gastric emptying, and slow intestinal motility. We presented the importance of the GI tract for the neonatal immune system as well as for the GI colonization and development of the microbiota. Feeding practices (such as route, volume, quantity, and quality of enteral feeds) can alter the intestinal ecosystem and can have implications for health *beyond the neonatal period*. Enteral feeds are crucial for the normal development of the GI tract; thus we propose to start enteral feeds very early in premature infants. We also summarized some of the new horizon on nutritional manipulation for feeding problems in preterm infants.

REFERENCES

1. Montgomery RK, Mulberg AE, Grand RJ. Development of the human gastrointestinal tract: twenty years of progress. *Gastroenterology* 1999;116(3):702–731.
2. Lau C, Smith EO, Schanler RJ. Coordination of suck-swallow and swallow respiration in preterm infants. *Acta Paediatr Jpn*. 2003;92(6):721–727.
3. Jadcherla SR. Gastroesophageal reflux in the neonate. *Clin Perinatol*. 2002;29(1):135–158.
4. Newell SJ, Booth IW, Morgan ME, et al. Gastro-oesophageal reflux in preterm infants. *Arch Dis Child*. 1989;64(4):780–786.
5. Kaijser M, Akre O, Cnattingius S, Ekbom A. Preterm birth, low birth weight, and risk for esophageal adenocarcinoma. *Gastroenterology*. 2005;128(3):607–609.
6. Cavell B. Gastric emptying in infants fed human milk or infant formula. *Acta Paediatr Scand*. 1981;70(5):639–641.
7. Cavell B. Reservoir and emptying function of the stomach of the premature infant. *Acta Paediatr Scand Suppl*. 1982;296:60–61.
8. Lebenthal E, Siegel M. Understanding gastric emptying: implications for feeding the healthy and compromised infant. *J Pediatr Gastroenterol Nutr*. 1985;4(1):1–3.
9. Pritchard JA. Fetal swallowing and amniotic fluid volume. *Obstet Gynecol*. 1966;28(5):606–610.
10. Patole S, Rao S, Doherty D. Erythromycin as a prokinetic agent in preterm neonates: a systematic review. *Arch Dis Child Fetal Neonatal Ed*. 2005;90(4):301–306.
11. Ng PC, So K, Fung KS, et al. Randomised controlled study of oral erythromycin

for treatment of gastrointestinal dysmotility in preterm infants. *Arch Dis Child Fetal Neonatal Ed*. 2001;84(3):F177–182.
12. Ng SC, Gomez JM, Rajadurai VS, et al. Establishing enteral feeding in preterm infants with feeding intolerance: a randomized controlled study of low-dose erythromycin. *J Pediatr Gastroenterol Nutr*. 2003;37(5):554–558.
13. Hyman PE, Abrams C, Dubois A. Effect of metoclopramide and bethanechol on gastric emptying in infants. *Pediatr Res*. 1985;19(10):1029–1032.
14. Barnett CP, Omari T, Davidson GP, et al. Effect of cisapride on gastric emptying in premature infants with feed intolerance. *J Paediatr Child Health*. 2001;37(6):559–563.
15. Barbey JT, Lazzara R, Zipes DP. Spontaneous adverse event reports of serious ventricular arrhythmias, QT prolongation, syncope, and sudden death in patients treated with cisapride. *J Cardiovasc Pharmacol Ther*. 2002;7(2):65–76.
16. Berseth CL. Gastrointestinal motility in the neonate. *Clin Perinatol*. 1996;23(2):179–190.
17. Hyman PE, Clarke DD, Everett SL, et al. Gastric acid secretory function in preterm infants. *J Pediatr*. 1985;106(3):467–471.
18. Basaran UN, Celayir S, Eray N, et al. The effect of an H2-receptor antagonist on small-bowel colonization and bacterial translocation in newborn rats. *Pediatr Surg Int*. 1998;13(2–3):118–120.
19. Graham PL 3rd, Begg MD, Larson E, et al. Risk factors for late onset gram-negative sepsis in low birth weight infants hospitalized in the neonatal intensive care unit. *Pediatr Infect Dis J*. 2006;25(2):113–117.
20. Guillet R, Stoll BJ, Cotten CM, et al. Association of H2-blocker therapy and higher incidence of necrotizing enterocolitis in very low birth weight infants. *Pediatrics*. 2006;117(2):e137–142.
21. Antonowicz I, Lebenthal E. Developmental pattern of small intestinal enterokinase and disaccharidase activities in the human fetus. Gastroenterology. 1977;72(6):1299–1303.
22. Lebenthal E, Lee PC, Heitlinger LA. Impact of development of the gastrointestinal tract on infant feeding. *J Pediatr*. 1983;102(1):1–9.
23. Mihatsch WA, Franz AR, Hogel J, Pohlandt F. Hydrolyzed protein accelerates feeding advancement in very low birth weight infants. *Pediatrics*. 2002;110(6):1199–1203.
24. Schanler RJ. The use of human milk for premature infants. *Pediatr Clin North Am*. 2001;48(1):207–219.
25. Hamosh M. Digestion in the newborn. *Clin Perinatol*. 1996;23(2):191–209.
26. Balistreri WF. Immaturity of hepatic excretory function and the ontogeny of bile acid metabolism. *J Pediatr Gastroenterol Nutr*. 1983;2 Suppl 1:S207–214.
27. Boehm G, Braun W, Moro G, Minoli I. Bile acid concentrations in serum and duodenal aspirates of healthy preterm infants: effects of gestational and postnatal age. *Biol Neonate*. 1997;71(4):207–214.
28. Klenoff-Brumberg HL, Genen LH. High versus low medium chain triglyceride content of formula for promoting short term growth of preterm infants. *Cochrane Database Syst Rev*. 2003;1:(CD002777).
29. Neu J, Koldovsky O. Nutrient absorption in the preterm neonate. *Clin Perinatol*. 1996;23(2):229–43.
30. Georgieff MK, Innis SM. Controversial nutrients that potentially affect preterm neurodevelopment: essential fatty acids and iron. *Pediatr Res*. 2005;57(5 Pt.2):99R–103R.
31. SanGiovanni JP, Parra-Cabrera S, Colditz GA, et al. Meta-analysis of dietary

essential fatty acids and long-chain polyunsaturated fatty acids as they relate to visual resolution acuity in healthy preterm infants. *Pediatrics.* 2000;105(6):1292–1298.
32. Morale SE HD, Castaneda YS, Wheaton DH, et al. Duration of long-chain polyunsaturated fatty acids availability in the diet and visual acuity. *Early Hum Dev*. 2005;81(2):197–203.
33. Calder PC. Can n-3 polyunsaturated fatty acids be used as immunomodulatory agents? *Biochem. Soc. Trans*. 1996;24(1):211–220.
34. Caplan MS, Russell T, Xiao Y, et al. Effect of polyunsaturated fatty acid (pufa) supplementation on intestinal inflammation and necrotizing enterocolitis (nec) in a neonatal rat model. *Pediatr Res*. 2001;49(5):647–52.
35. Murray RD, Kerzner B, Sloan HR, et al. The contribution of salivary amylase to glucose polymer hydrolysis in premature infants. *Pediatr Res*. 1986;20(2):186–191.
36. Kien CL. Digestion, absorption, and fermentation of carbohydrates in the newborn. *Clin Perinatol*. 1996;23(2):211–228.
37. Neu J. Functional development of the fetal gastrointestinal tract. *Semin. Perinatol*. 1989;13(3):224–235.
38. Shulman RJ, Schanler RJ, Lau C, et al. Early feeding, feeding tolerance, and lactase activity in preterm infants. *J Pediatr*. 1998;133(5):645–649.
39. Kim YS, Perdomo J, Ochoa P, Isaacs RA. Regional and cellular localization of glycosyltransferases in rat small intestine. Changes in enzymes with differentiation of intestinal epithelial cells. *Biochim Biophys Acta*. 1975;391(1):39–50.
40. Boyle JT, Celano P, Koldovsky O. Demonstration of a difference in expression of maximal lactase and sucrase activity along the villus in the adult rat jejunum. *Gastroenterology*. 1980;79(3):503–507.
41. Mellander L, Carlsson B, Jalil F, et al. Secretory IgA antibody response against Escherichia coli antigens in infants in relation to exposure. *J Pediatr.* 1985;107:430–433.
42. Goldblum RM, Ahlstedt S, Carlsson B. Antibody forming cells in human colostrum after oral immunisation. *Nature.* 1975;257:797–799.
43. Kleinman RE, Walker WA. The enteromammary immune system: an important new concept in breast milk host defense. *Dig Dis Sci*. 1979;24(11):876–882.
44. Martin R, Olivares M, Marin ML, et al. Probiotic potential of 3 Lactobacilli strains isolated from breast milk. *J Hum Lact*. 2005;21(1):8–17.
45. Scheppach W, Weiler F. The butyrate story: old wine in new bottles? *Curr Opin Clin Nutr Metab Care*. 2004;7(5):563–567.
46. Bartholome A, Albin D, Baker D, et al. Supplementation of total parenteral nutrition with butyrate acutely increases structural aspects of intestinal adaptation after an 80% jejunoileal resection in neonatal piglets. *JPEN J Parenter Enteral Nutr*. 2004;28(4):210–222.
47. Fituch CC, Palkowetz KH, Goldman AS, Schanler RJ. Concentrations of IL-10 in preterm human milk and in milk from mothers of infants with necrotizing enterocolitis. *Acta Paediatr*. 2004;93(11):1496–1500.
48. Dinsmore JE, Jackson RJ, Smith SD. The protective role of gastric acidity in neonatal bacterial translocation. *J Pediatr Surg*. 1997;32(7):1014–1016.
49. Lebenthal E, Lee PC. Development of functional response in human exocrine pancreas. *Pediatrics*. 1980;66:556–560.
50. Rubinstein E, Mark Z, Haspel J, et al. Antibacterial activity of the pancreatic fluid. *Gastroenterology* 1985;88:927–932.

51. Walker WA, Wu M, Isselbacher KJ, Bloch KJ. Intestinal uptake of macromolecules. IV.--The effect of pancreatic duct ligation on the breakdown of antigen and antigen-antibody complexes on the intestinal surface. *Gastroenterology*. 1975;69(6):1223–1229.
52. Beck-Sague CM, Azimi P, Fonseca SN, et al. Bloodstream infections in neonatal intensive care unit patients: results of a multicenter study. Pediatr *Infect Dis J*. 1994;13(12):1110–1116.
53. Cheng H, Leblond CP. Origin, differentiation and renewal of the four main epithelial cell types in the mouse small intestine. I. Columnar cell. II. Mucus cell. III. Entero-endocrine cells. IV. Paneth cells. *Am J Anat*. 1974;141:461–536.
54. Laboisse CL. Structure of gastrointestinal mucins: searching for the Rosetta stone. *Biochimie*. 1986;68(5):611–617.
55. Clamp JR, Creeth JM. Some non-mucin components of mucus and their possible biological roles. Ciba Found Symp. 1984;109:121–136.
56. Shub MD, Pang KY, Swann DA, Walker WA. Age-related changes in chemical composition and physical properties of mucus glycoproteins from rat small intestine. *Biochem J*. 1983;215(2):405–411.
57. Sherman P, Forstner J, Roomi N, et al. Mucin depletion in the intestine of malnourished rats. *Am J Physiol*. 1985;284(4Pt1):G418–423.
58. Faure M, Moennoz D, Montigon F, et al. Dietary threonine restriction specifically reduces intestinal mucin synthesis in rats. *J. Nutr*. 2005;135(3):486–491.
59. Bevins CL. Paneth cell defensins: key effector molecules of innate immunity. *Biochem Soc Trans*. 2006;34(Pt.2):263–266.
60. Gebert A, Rothkotter HJ, Pabst R. M cells in Peyer/s patches of the intestine. *Int Rev Cytol*. 1996;167:91–159.
61. Denker BM, Nigam SK. Molecular structure and assembly of the tight junction. *Am J Physiol*. 1998;274:F1–F9.
62. Madara JL, Dharmsathaphorn K. Occluding junction structure-function relationships in a cultured epithelial monolayer. *J. Cell Biol*. 1985;101: 2124–2133.
63. Atsook K, Madara JL. An oligopeptide permeates intestinal tight junctions at glucose-elicited dilatations. Implications for oligopeptide absorption. Gastroenterology 1991;100:719–724.
64. Venkatraman A, Ramakrishna BS, Pulimood AB. Butyrate hastens restoration of barrier function after thermal and detergent injury to rat distal colon in vitro. *Scand J Gastroenterol* 1999;34(11):1087–1092.
65. Li N, Lewis P, Samuelson D, et al. Glutamine regulates Caco-2 cell tight junction proteins. *Am J Physiol Gastrointest Liver Physiol*. 2004;287(3):G726–733.
66. Seth A, Basuroy S, Sheth P, Rao RK. L-Glutamine ameliorates acetaldehyde-induced increase in paracellular permeability in Caco-2 cell monolayer. *Am J Physiol Gastrointest Liver Physiol*. 2004;287(3):G510–517.
67. Liu Z, Li N, Neu J. Tight junctions, leaky intestines, and pediatric diseases. *Acta Paediatr*. 2005;94(4):386–393.
68. Koldovsky O, Sunshine P, Kretchmer N. Cellular migration of intestinal epithelia in suckling and weaned rats. *Nature*. 1966;212(68):1389–90.
69. Cetin S, Ford HR, Sysko LR, et al. Endotoxin inhibits intestinal epithelial restitution through activation of Rho-GTPase and increased focal adhesions. *J Biol Chem*. 2004;279(23):24592–24600.
70. Evans ME, Jones DP, Ziegler TR. Glutamine prevents cytokine- induced apoptosis in human colonic epithelial cells. *J Nutr*. 2003;133(10): 3065–3071.
71. Rhoads JM, Niu X, Odle J, Graves LM. Role of mTOR signaling in intestinal cell

migration. *Am J Physiol Gastrointest Liver Physiol*. 2006;291(3):G510–517.
72. Hooper LV, Stappenbeck TS, Hong CV, Gordon J. Angiogenins: a new class of microbicidal proteins involved in innate immunity. *Nat Immunol*. 2003;4(3):269–273.
73. Hooper LV. Bacterial contributions to mammalian gut development. *Trends Microbiol*. 2004;12(3):129–134.
74. Stappenbeck TS, Hooper LV, Gordon JI. Developmental regulation of intestinal angiogenesis by indigenous microbes via Paneth cells. *Proc Natl Acad Sci USA*. 2002;99(24):15451–15455.
75. Rakoff-Nahoum S, Paglino J, Eslami-Varzaneh F, et al. Recognition of commensal microflora by toll-like receptors is required for intestinal homeostasis. *Cell*. 2004;118(2):229–241.
76. Madara J. Building an intestine--architectural contributions of commensal bacteria. *N Engl J Med*. 2004;351(16):1685–1686.
77. Schumann A, Nutten S, Donnicola D, et al. Neonatal antibiotic treatment alters gastrointestinal tract developmental gene expression and intestinal barrier transcriptome. *Physiol. Genomics* 2005;23(2):235–245.
78. Kudsk KA. Current aspects of mucosal immunology and its influence by nutrition. Am J Surg 2002;183(4):390–398.
79. Liboni K, Li N, Neu J. Mechanism of glutamine-mediated amelioration of lipopolysaccharide-induced IL-8 production in Caco-2 cells. *Cytokine*. 2004;26(2):57–65.
80. Li N, Liboni K, Fang MZ, et al. Glutamine decreases lipopolysaccharide-induced intestinal inflammation in infant rats. *Am J Physiol Gastrointest Liver Physiol*. 2004;286(6):G914–921
81. Zhang L, Li N, des Robert C, et al. Lactobacillus rhamnosus GG decreases lipopolysaccharide-induced systemic inflammation in a gastrostomy-fed infant rat model. *J Pediatr Gastroenterol Nutr*. 2006;42(5):545–552.
82. Minekawa R, Takeda T, Sakata M, et al. Human breast milk suppresses the transcriptional regulation of IL-1beta-induced NF-kappaB signaling in human intestinal cells. *Am J Physiol Cell Physiol*. 2004;287(5):C1404–1411.
83. Thureen PJ, Melara D, Fennessey PV, Hay WWJ. Effect of low versus high intravenous amino acid intake on very low birth weight infants in the early neonatal period. *Pediatr Res*. 2003;53(1):24–32.
84. Niinikoski H, Stoll B, Guan X, et al. Onset of small intestinal atrophy is associated with reduced intestinal blood flow in TPN-fed neonatal piglets. *J. Nutr*. 2004;134(6):1467–1474.
85. Stoll BJ, Hansen N, Fanaroff AA, et al. Late-onset sepsis in very low birth weight neonates: the experience of the NICHD Neonatal Research Network. *Pediatrics*. 2002;110(2 Pt.1):285–291.
86. Alverdy JC, Laughlin RS, Wu L. Influence of the critically ill state on host-pathogen interactions within the intestine: gut-derived sepsis redefined. *Crit Care Med*. 2003;31(2):598–607.
87. Wildhaber BE, Yang H, Spencer AU, et al. Lack of enteral nutrition-effects on the intestinal immune system. *J Surg Res*. 2005;123(1):8–16.
88. Harris MC, D'Angio CT, Gallagher PR, et al. Cytokine elaboration in critically ill infants with bacterial sepsis, necrotizing entercolitis, or sepsis syndrome: correlation with clinical parameters of inflammation and mortality. *J Pediatr*. 2005;147(4):462–468.
89. Speer CP. Pre- and postnatal inflammatory mechanisms in chronic lung disease of preterm infants. *Paediatr Respir Rev*. 2004;5(Suppl A):S241–244.

90. Volpe JJ. Neurobiology of periventricular leukomalacia in the premature infant. *Pediatr Res*. 2001;50(5):553–562.
91. Berseth CL, Bisquera JA, Paje VU. Prolonging small feeding volumes early in life decreases the incidence of necrotizing enterocolitis in very low birth weight infants. *Pediatrics*. 2003;111(3):529–534.
92. Owens L, Burrin DG, Berseth CL. Minimal enteral feeding induces maturation of intestinal motor function but not mucosal growth in neonatal dogs. *J Nutr*. 2002;132(9):2717–2722.
93. Neu J, Roig JC, Meetze WH, et al. Enteral glutamine supplementation for very low birth weight infants decreases morbidity. *J. Pediatr*. 1997;131(5):691–699.
94. Vaughn P, Thomas P, Clark R, Neu J. Enteral glutamine supplementation and morbidity in low birth weight infants. *J. Pediatr*. 2003; 142:662–668.
95. van den Berg A, van Elburg RM, Westerbeek EA, et al. Glutamine-enriched enteral nutrition in very-low-birth-weight infants and effects on feeding tolerance and infectious morbidity: a randomized controlled trial. *Am J Clin Nutr*. 2005;81(6):1397–1404.
96. Poindexter BB, Ehrenkranz RA, Stoll BJ, et al. Effect of parenteral glutamine supplementation on plasma amino acid concentrations in extremely low-birth-weight infants. *Am J Clin Nutr*. 2003;77(3):737–743.
97. Neu J. Glutamine supplements in premature infants: why and how. J Pediatr Gastroenterol Nutr. 2003;37(5):533–535.
98. Calder PC. Polyunsaturated fatty acids and inflammation. *Biochem. Soc. Trans*. 2005;33:423–427.
99. Tappenden KA, Albin DM, Bartholome AL, Mangian HF. Glucagon-like peptide-2 and short-chain fatty acids: a new twist to an old story. *J. Nutr*. 2003;133(11):3717–320.
100. Clayburgh DR, Shen L, Turner JR. A porous defense: the leaky epithelial barrier in intestinal disease. *Lab Invest*. 2004;84(3):282–291.

23

Diet, Environment, and Gut Microbiota in the Metagenomic Era

Carlotta De Filippo, BSc, PhD and Paolo Lionetti, MD, PhD

INTRODUCTION

The human gut microbiota is a complex consortium of trillions of microorganisms, whose collective genomes or “metagenome” contains at least 100 times as many genes as our own eukaryote genome.[1] This essential “organ,” the microbiome, provides the host with enhanced metabolic capabilities, energy, nutrients, and bioactive compounds and provides an effective barrier or resistance to invading pathogens, resulting in education of the immune system and modulation of gastrointestinal (GI) development. Furthermore, this microbial community is also involved in detoxification of harmful exogenous and endogenous compounds. The role of the resident microbiota in the human gut and its profound effects on host health and daily well-being is now recognized as crucial. The composition of the intestinal microbiota plays a critical role in mammalian metabolism and, likely, in human health. Twins and mother–daughter pairs have more similar microbiota compositions than unrelated individuals,

suggesting that there could be a genetic influence over the microbiota.[2] However, monozygotic and dizygotic adult twins have equally similar microbiota, suggesting environment rather than genetics may drive familial similarities.[2] The extent at which factors such as environment and diet shape the human gut microbiota is still unclear, partly because these factors are often confounded. However, in terms of microbiota function, metabolic output and interactions with the host, diet, and other exogenous factors like antibiotics or xenobiotic compounds also play a dominant role.

NEW TECHNOLOGIES TO DESCRIBE HUMAN MICROBIOTA—THE METAGENOMICS APPROACH

In the twentieth century, our knowledge of the human microbiota was constrained by the ability to describe and study the biological functions of less than 100 cultivable bacteria. The species we described until year 2000 were also the most abundant ones, and given the special attention of funding agencies toward pathogens, we fundamentally ignored the genome to functions relation for the majority of our commensal, except in the case of a handful of bacterial species used in food production (lactobacilli) and shown to dominate the gut microbiota of breast-fed infants (bifidobacteria).

In the beginning of this century, a limited number of studies on cloning and sequencing libraries of 16S rRNA started opening a window with a view on the astonishing diversity of human microbiota. Despite their importance, these initial, noncultivation-based, diversity studies were limited by the costs and complexity of Sanger sequencing methods. In the past 10 years, the picture of our gut microbiota has rapidly passed from black and white to a surprising explosion of bright colors, thanks to the advent of next-generation sequencing (NGS) technologies.

METAGENOMICS AND THE NEXT-GENERATION SEQUENCING ERA

Metagenomics is the application of modern genomic techniques to the study of communities of microbial organisms directly in their natural environments, bypassing the need for isolation and lab cultivation of individual species, given that less than 1% of the microbes present in nature are cultivable. The studies in the case of human microbiota (the

bacterial community of a body district) can consist of DNA and also transcriptome, proteome, and metabolome analysis to assess the type of organisms and the genes present in the community: the so-called metagenome or microbiome.[3]

The molecular study of both the taxonomic and the functional gene composition of environmental microbial communities is now highly improved by the use of NGS,[4] mainly using Roche/454 and Illumina. Advances in DNA sequencing have dramatically reduced costs and markedly increased capacity, thus allowing culture-independent metagenomic methods to be readily deployed to characterize microbial communities (microbiota) associated with human body habitats at various stages of the human life cycle and in various populations.[5] The 16S rRNA gene is widely considered the major molecular marker in the taxonomic classification of bacteria and is considered a good phylogenetic chronometer. The 16S rRNA gene is universal in bacteria, contains regions that vary in genetic mutation rate, and is long enough to provide meaningful taxonomic data.[6] The 16S rRNA bacterial gene contains nine hypervariable regions V1–V9 (Figure 23-1).[7] The V1, V2, V3, V5, and V6 regions result in having a greater discriminating power in identifying bacterial genus and species in animal commensal microbial communities.[8,9]

The 454 Genome Sequencer FLX Titanium series (Roche) platforms is particularly suited for 16S rRNA-based surveys. This platform can produce in a single run about one million “reads” of high quality with an average size of 400 bp that is now increased to 700 bp, thanks to the last platform upgrade. Basically, using these platform researchers, we will be able to sequence the entire 16S rRNA gene without cloning and without problems in assembling incomplete partial sequences from short reads.

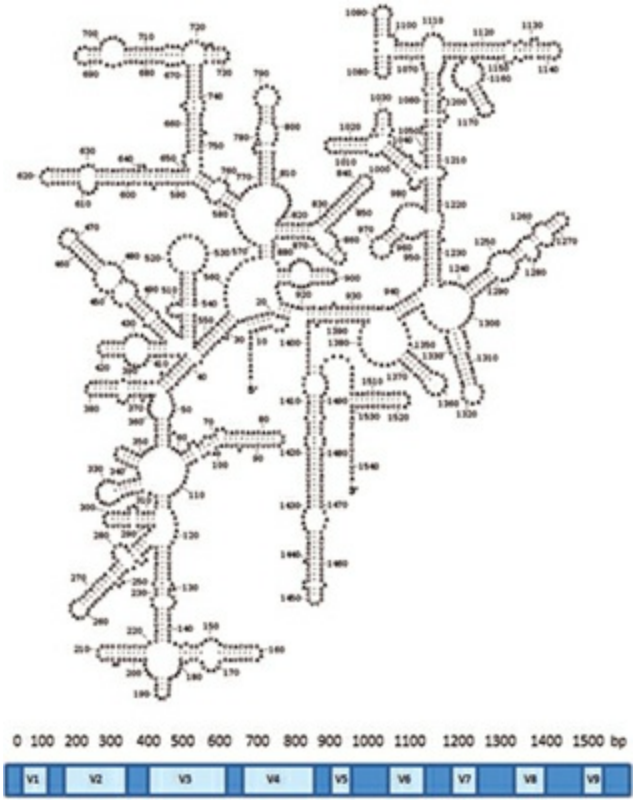


Figure 23-1 RNA secondary structure *Escherichia coli* 16S rRNA. 16S rRNA is a component of the 30S small subunit of prokaryotic ribosomes. It is 1.542 kb (or 1542 nucleotides) in length. The genes coding for it are referred to as 16S rDNA and are used in reconstructing phylogenies, in fact among the well-conserved structures of 16S rRNA (blue boxes), there exist nine hypervariable regions (V1–V9) that vary in their secondary structure from taxon to taxon. New DNA-sequencing technologies (NGS), such as massively parallel pyrosequencing, can provide orders of magnitude more DNA sequences than ever before to identify microbial taxa by comparing short tags from 16S hypervariable regions against a large database of known 16S genes. Using this technique, hypervariable region tags provide equivalent taxonomy and relative abundances of microbial communities as full-length rRNA sequences.

A significant further step has been made possible by approaches enabling the sequencing of every bacterial genome in the gut directly without isolation or cultivation in the laboratory, like Illumina-based NGS techniques. Illumina HiSeq2000 technology is based on a proven chemical technology useful for the sequencing of whole bacterial genomes.[10,11]

High-throughput sequencing technologies now allow for the first time access to the entire genetic repertoire or metagenome of a given environment without prior isolation or knowledge of microbial species make up. In metagenomic approaches, the analysis of the large amount of information contained in sequences generated by microbial sequencing “high throughput” from a complex environmental sample is now a major challenge. Importantly Illumina produces short reads at a significantly reduced cost. The real challenge of this technology is the heavy reliance on hardcore bioinformatics and computation required to assemble short reads from mixed samples by blasting the reads to databases of known sequences. The sequencing and subsequent bioinformatics analysis of DNA belonging to a microbial community is a great way to get information about its content and potential functional genes.[10,11] These new metagenomics surveys not only capture the microbial organism and genetic diversity associated with humans but also make it possible to investigate gene functions.[12] The combination of 16S rRNA and whole metagenome-based approaches holds the key to understanding the functional contributions that our microbes make to our physiologic phenotypes, our health, and our disease predisposition. However, the ability to reconstruct gut active metabolic pathways requires the combination of information on genes and protein sequences with the knowledge of metabolic patterns, which can be inferred from the measurement of the levels of associated metabolites in biological fluids (i.e., feces, urine, and blood).

METABOLOMICS

Direct sequencing metagenomic approaches allow us to access the genetic potential of a microbial community but does not provide in depth, real-time insight into the metabolic flux or kinetic of a microbial community at a given point in time. The human metabolome is the complete set of metabolites, end products of cellular processes, present in human biofluids at a given time. Metabolite profiling approaches represent the key to understanding the functional contribution of the gut microbiota to the host organism and its co-evolution with diet. Metabolomics, defined as a systematic study of the unique chemical fingerprints that specific cellular processes leave behind, allows us to characterize the metabolite profiles within a human biological sample such as stool, blood, urine, and breath, by using high-resolution analytical chemistry methods such as gas chromatography–mass spectrometry (GC-MS) or nuclear magnetic

resonance (NMR). GC-MS analysis represents a robust method for quantification of selected metabolites, with more satisfactory sensitivity and resolution than the conventional NMR approach.[13,14] This analysis is applied in metabolomics research of urinary and serum samples, brain tissue extracts, and fecal samples and allows the analysis of a certain class of metabolites such as fatty acids, phenolic acids, and amino acids in fecal water. The variation in metabolic profiles could depend on host genotype and disease status and also food intake (fruit, vegetables, dietary proteins, and fats) and on the composition of intestinal microbiota. Dietary carbohydrates, specifically resistant starches, and dietary fiber contained in fruits, vegetables, and whole grain cereals are readily fermented by the colonic microbiota to short-chain fatty acids (SCFAs), primarily acetate, propionate, and butyrate. These metabolites and their biologically active compounds modulate human nutrition and health, by decreasing the risk of developing GI and autoimmune disorders, allergy, and inflammatory bowel disease (IBD) through enhanced mucosal integrity and regulation of immune homeostasis, and also through protection from metabolic or obesity-related diseases.[15] SCFAs have been shown to impact on a number of physiological processes related to human energy metabolism including satiety and food intake (via induction of incretins), reducing low-grade systemic inflammation related to type 2 diabetes and nonalcoholic fat liver disease, and regulating lipogenesis and cholesterol metabolism in the liver.[16] Another important set of metabolites are amino acids and the by-products of their metabolism. The proportion of dietary amino acids that are metabolically available in the human body is influenced by digestibility, absorption, and genes of the gut microbiota involved in metabolism of amino acids. The availability of amino acid is important in humans for protein synthesis, modulation of gene expression, and intestinal integrity. Amino acid composition of foods varies greatly. In cereals and legumes, lysine and methionine concentrations, respectively, are significantly lower compared to foods rich in animal protein.[13] Amino acid availability in the colon is dependent on both endogenous sources (sloughed off epithelial cells and host digestive enzymes) and host diet. Food processing, especially heating, can affect the digestibility of proteins, impacting on the amount of protein reaching the colon as can the quantity of protein ingested. The Maillard reaction that occurs when food containing a mixture of carbohydrate/sugars and protein (i.e., most complex foods) is heated leads to the generation of advanced glycation end products, some of which are resistant to digestion by human proteases and reach the colon intact where they become available to the resident gut

microbiota. In terms of gross metabolic activity, there is a functional gradient within the colon. Carbohydrate fermentation is a more energetically favorable process for microorganisms, and available carbohydrates entering the colon are fermented preferentially, compared with other energy sources including protein. As this carbohydrate is used up, however, especially in modern affluent diets with low dietary fiber, microorganisms look for alternative sources of energy and switch to a proteolytic fermentation. Proteolytic fermentation by the gut microbiota, in addition to generating SCFAs and branched-chain fatty acids, results in the production of a range of harmful compounds that can be variably cytotoxic, genotoxic, or carcinogenic.[17] Thus, one important protective mechanism of diets rich in dietary fiber may be to prolong saccharolytic fermentation in the colon and reduce the harmful activities of amino acid fermentation.[18] By uniting metagenomics data with analyses of the products of microbial community metabolism (metabolomics), it is possible to shed light on how microbial communities function in a diverse human population with different dietary habits.[19]

THE CORE MICROBIOME

The reconstruction of the evolution of our dietary habits is crucial for understanding the evolutionary context of our modern diet and diet-associated diseases. The gut microbiota recovers energy and biologically active molecules, such as indigestible plant polysaccharides, from foods that would otherwise remain undigested and excreted by the intestinal tract, thanks to its microbiome enriched with genes for the breakdown of complex sugars. Resident bacteria are also involved in the biosynthesis of essential vitamins, isoprenoids, polyphenols, amino acids and the xenobiotic metabolism.

Turnbaugh et al.[2] performed a microbiota analysis on adult monozygotic and dizygotic female twin pairs and their mothers, similar for their physiological lean or obese state, trying to understand how the genome, environment, and fatness phenotype affect the microbial composition of the gut. The microbiome appears to be shared among relatives, while the composition of microbiota (mainly constituted by Bacteroidetes, Firmicutes, and in a lesser extent Actinobacteria and Proteobacteria) was found to be variable. Among all 154 individuals, 93%–98% of the genes belonged to a core functional group of genes shared among the microbiome.[2] Therefore, such findings suggest that humans may share a functional core microbiome instead of a core

microbiota. Deviations from this core turned out to be associated with different physiological states such as obesity, as well as reduced bacterial diversity and changes in bacterial genes and metabolic pathways present. The short-term temporal variations in the microbial community composition of an individual were found to be less marked than interpersonal differences. Close symbiotic integration of functions from the intestinal cells with bacterial cells suggests that human beings should be considered as “superorganisms” whose metabolism involves integration of metabolic processes encoded by the host genome with those encoded by the microbiome.[20] Initial work showed that the human microbiome composition varies among healthy people[1,20,21] and also between lean and obese individuals, and that it is responsive to dietary modulation for weight reduction.[22] Eckburg et al.[21] showed that the gut microbiota is highly diverse between individuals and comprises about 60% novel bacterial phylotypes and ~80% uncultivated species. This study supports the theory that gut bacteria evolution is a classic case of adaptive radiation, where a few successful colonizers gave rise to a variety of descendants, through diversification of an initial population bottleneck into various strains that likely correspond to ecotypes.

HOW DIET AND ENVIRONMENT COULD SHAPE OUR GUT MICROBIOTA

Despite the increasing number of metagenomic studies published, the complex interaction between diet and gut microbiota in human populations is still wide open to new discoveries. It is well known that human gut microbiota recover energy and biologically active molecules from food that would otherwise be discharged from the intestinal tract. Certain foods or food compounds interact directly with the gut microbiota, specifically those compounds which escape digestion in the stomach and upper gut and reach the colon, which is the home to majority of gut microorganisms. These compounds include nondigestible polysaccharides (prebiotics and oligosaccharides not targeted by human digestive enzymes or polysaccharides resistant to digestion due to chemical structure or physical inaccessibility) and plant polyphenolic secondary metabolites, 90% of which reach the colon.[19,23] These compounds are major components of whole plant foods, fruit, vegetables, and whole grain cereals. The fact that these compounds shape the activities of the gut microbiota is not that surprising when you consider how the gut microbiota must have evolved

alongside its human host. For humans from the earliest times, and our closest primate progenitors, plant-based foods formed the dietary mainstay. For nearly a million years, humans existed as hunter–gatherers, collecting wild fruits, vegetables, grasses, and tubers and supplementing their diet with wild animals and insects when available. Major changes in our diet occurred with the birth of agriculture,[24] reducing the diversity of species consumed, and with the agricultural and industrial revolutions (increasing access to processed and refined foods). However, it has been the last 50 years that has witnessed the most dramatic shift in human diet since we first diverged from our ancestral hunter–gatherer diets. After the Second World War and subsequent "Green Revolution" of the 1960s, intensive farming, dietary patterns in the "developed world" changed dramatically with increased access to processed energy-dense foods with high intake of saturated fat, red and processed meats, salt, sugar, and refined carbohydrates; low consumption of fresh fruit, whole-grain cereals, and vegetables; and consequently, low intake of dietary fiber and plant polyphenolic compounds. This dietary shift toward the "Western style" or modern affluent diet has long been linked to an increased incidence of chronic diet–associated diseases including obesity, type 2 diabetes, nonalcoholic fatty liver disease, cardiovascular disease, and certain cancers.[25,26] More recently, a dietary link to the concomitant increase in autoimmune diseases has also been suggested, but mechanisms remain elusive.[27,28] Similarly, high dietary intake of whole plant foods is considered protective against these same chronic human diseases.[29,30] One obvious implication of this dietary shift is that the type and quantity of food macromolecules reaching the colonic microbiota has changed compared to more traditional diets (e.g., traditional Mediterranean, African, and Japanese) and hunter–gatherer diets with probable implications for microbiota-related functions such as fiber fermentation and release of biologically active derivatives of phytochemicals. This identifies the human gut microbiota and their interactions with diet, as a focal point about which human health swings, with the potential to protect against chronic diseases of modern lifestyles and on the contrary as a contributory factor of autoimmune disease, metabolic diseases, and cancer.[31]

Until very recently, we lacked the tools to study in detail the consequences of modern affluent dietary composition on the makeup and activities of the human gut microbiota. However, the emergence of culture-independent microbiological techniques and most recently the application of postgenomics high-resolution genetic or metabolite profiling

approaches to study the gut microbiota have provided unprecedented insight into the microbial ecology of the human gut microbiota. A new subject area is emerging, which combines host genomics, immunology, metagenomics, metabolomics, food science (food chemistry, genomics, and biotechnology), and nutrition to service the observable interrelatedness of the foods we eat, our resident microbiota, and the human system in all its genetic, immunological, and metabolic diversity. Although usually used to define the interrelationship between plants and their mycorrhizal fungal partners, nutritional symbiosis defines well the relationship among foods ingested, the intestinal microbiota, and the human body.

Recent metagenomic studies have also highlighted important and previously unimagined means by which foods can modulate the composition and functioning of the gut microbiota and its composition species.

Gene transfer between bacteria in the gut has recently been shown to be influenced by the interactions between food and the gut microbiota. Among the Japanese, whose diet includes regular consumption of sushi, Hehemann et al.,[32] working with *Zobellia galactanivorans*, a member of the marine Bacteroidetes, discovered an enzyme (porphyranase) responsible for breaking down porphyran, an abundant polysaccharide in the red algae species *Porphyra*, on which *Z. galactanivorans* is often found. While searching gene-sequence databases, investigators came across predicted porphyranase sequences in metagenomes derived from human feces and in the genome of the resident human gut bacterium *Bacteroides plebeius*, suggesting that *B. plebeius* acquired the genes laterally from marine bacteria. After it turned out that these sequences were present in Japanese individuals but not in residents of the United States, the authors concluded that *Z. galactanivorans* were introduced through *Porphyra* (“nori”), the traditional seaweed used to wrap sushi and a common component of the Japanese diet. The researchers hypothesize that by regularly consuming seaweeds, Japanese communities produced a selective force that led to retaining the beneficial porphyranase genes in their gut microbiomes.[32] Earlier studies with animal models with a “humanized” gut microbiota showed that high-fat, Western-style diet can increase the rate of conjugative transfer of plasmids between bacteria within the GI tract.[33]

Such interplay between food and gut microbiota underlines the importance of food microbial composition in terms of both quality (e.g., presence of probiotic species) and safety (e.g., ratio of probiotics and pathogens) and implicates diet in the co-evolution of the human gut

microbiota in health and disease.

Another prominent role of gut microbiota is energy extraction from fiber and complex carbohydrates. Plant materials that are in fact indigestible by humans are a complex carbohydrate known as dietary fiber. Our bodies need roughage to properly digest food and eliminate waste because the bulky substance cleans the colon as it makes its way through our digestive tract. Dietary fiber comes from vegetables, fruit, grain, and legumes. The plant polysaccharides are rich in xylan-, pectin-, and arabinose- containing carbohydrate structures. The human genome lacks most of the enzymes required for degrading these glycans (see http://www.cazy.org/). However, the distal gut microbiome provides this capacity. At least 81 different glycoside hydrolase families are represented in the microbiome, many of which are not present in the human "glycobiome."[34] Host GI mucus provides a consistent reservoir of glycans for the microbiota and thus, in principle, can serve to mitigate the effects of marked changes in the availability of dietary polysaccharides. Dietary carbohydrates, specifically resistant starches and dietary fiber (e.g., inulin, fructooligosaccharides), contained in fruits, vegetables, and whole grain cereals are readily fermented by the colonic microbiota to SCFAs, primarily acetate, propionate, and butyrate.

Gnotobiotic mouse models of the human gut microbiota have indicated that linked terminal fucose in host glycans is an attractive and accessible source of energy for members of the microbiota such as the Bacteroidetes.[35]

MICROBIOME VARIATION IN HUMAN POPULATIONS

Awareness of the importance of work on human cohorts is stimulating the boost of comparative metagenomics studies in individuals from different countries, different disease states, and different ages. However, addressing comparative metagenomics studies directly in humans is challenging because of numerous uncontrolled variables, such as genetic variation, geographical, environmental, and dietary differences.

The gut microbiota profile reflects people's country of origin. Yet one of the most prominent differences between human populations prior to world globalization or "Westernization" was diet. Examples included the so-called "Mediterranean diet" characterized by the traditional abundance of olive oil and legumes, or the fish and fat diet of Eskimo population.

The geographical diversity of diet could therefore profoundly explain geographically different microbiota as a result of the interplay of the aforementioned variables.

Several studies analyzed variables such as different geographical locations, diets, and ages. A recent cross-sectional study on intestinal microbiota composition was performed on 230 healthy subjects (two age groups: 20–50 years and older than 60 years) living in four European locations: France, Germany, Italy, and Sweden.[36] The analysis used a set of 14 group- and species-specific 16S rRNA-targeted oligonucleotide probes to analyze fecal samples by fluorescence in situ hybridization coupled with flow cytometer. Age-related differences in the microbiota makeup were detected but differed between the study populations from the four countries, each showing a characteristic colonization pattern. Marked country–age interactions were observed for the German and Italian study groups. These interactions were opposite for the predominant bacterial groups such as *Eubacterium rectale, Clostridium coccoides, and Bacteroides–Prevotella*. Differences between European populations were observed for the *Bifidobacterium* group only. Proportions of bifidobacteria were two- to threefold higher in the Italian study population than in any other study group, and this effect was independent of age. Higher proportions of enterobacteria were found in all elderly volunteers independent of the location. Gender effects were observed for the *Bacteroides–Prevotella* group, with higher levels in males than in females.

The utilization of whole-microbiome sequencing moved these types of population studies beyond a description of microbial species or genes present in particular habitats, linking the structure and dynamic operations of microbial communities reciprocally to human biology and pathobiology.

The MetaHIT consortium published a first paper describing metagenomic data set, using Illumina sequencing technology, containing almost 200-fold more gut microbial sequences from fecal samples of 124 European individuals, including healthy, overweight, and obese adults, as well as patients with IBD.[10] The functional analysis of the gene catalogue identified a core set of genes that seem to be essential for bacterial survival in the gut. These include typical housekeeping genes and others that may encode products involved in adhesion to host proteins or in harvesting the sugars that are carried on blood or epithelial cells. Some functions were present in all 124 individuals included in the study cohort but detected in only a small proportion of previously sequenced genomes of gut bacteria. Therefore, the genes encoding these functions may be essential for the

functioning of the gut ecosystem as a whole but may be encoded across different bacterial species. Recently, the MetaHIT consortium[11] described the worldwide microbiome variation according to a series of distinct enterotypes. This study increases the number of individuals and populations from which gut microbiomes have been sequenced. The authors surveyed the gut metagenomes of 22 European individuals and combined the findings with existing data from Japanese and American individuals. They found that the microbiome clusters into distinct phylogenetic groups (enterotypes), which do not reflect country or continent of origin, body mass index (BMI), age, or gender. Each of these three enterotypes is identifiable by the variation in the levels of one of three genera: *Bacteroides* (enterotype 1), *Prevotella* (enterotype 2), and *Ruminococcus* (enterotype 3). The authors affirmed that enterotypes appear complex, are probably not driven by nutritional habits, and cannot simply be explained by host properties such as age or BMI, although there are functional markers such as genes or modules that correlate remarkably well with individual features. Although these studies infer differences related to different diets, they can hardly describe in detail the actual dietary variables.

Children from different human populations provide an attractive model to study the role of geography and diet on microbiota variation. Infants living in developing countries have been shown to be colonized at younger ages with fecal bacteria and have more rapid transfer of enteric microbial strains than infants living in developed countries.[37]

In our recent work,[38] investigating diet–gut microbiota interaction, we discuss how the ability of modern *Homo sapiens* to live in different environments and to follow a wide range of diets has affected our gut microbial ecology. We characterized the fecal microbiota of 14 African children (BF) living in a rural village (Boulpon, Burkina Faso, Africa) and 15 European children (EU) living in an urban area (Florence, Italy), by sequencing the 16S ribosomal RNA gene, with the aim of elucidating the effects of different diets on gut microbiota. The experimental design includes a “natural” control set, breast-fed children from Burkina Faso and Italy. There are five major advancements. The first key finding is that Burkinabè children cluster separately from Italian children and breast-fed toddlers form a third cluster in between (Figure 23-2A). This can be solely the result of diet. Contributions from sanitation could play a role, but this is indeed minor; in fact, if sanitation or hygiene did cause the difference, then one would expect that breast-fed children would also cluster differently. While we clearly observed that mother’s milk significantly

reduces the differences and that is why the breast-fed children cluster separately, the cluster contains both Italian and Burkinabè toddlers, and this is apart from the other Burkinabè and Italian children. The second key finding is that the BF microbiota was significantly enriched in Bacteroidetes and depleted in Firmicutes compared to their EU counterparts, suggesting a co-evolution of intestinal bacteria with their diet, rich in plant polysaccharides (Figure 23-2B). The third key finding is that the fecal samples from children in Burkina Faso especially differed from the Italian subjects because of the presence of *Prevotella and Xylanibacter* (Bacteroidetes), *Treponema* (Spirochaetes), and *Butyrivibrio* (Firmicutes); all appeared in the African but were not found in the Italian samples. This peculiar microbiota is at risk of being lost in urbanized Africans. We hypothesize that these distinctive bacterial genera might help to extract energy from the polysaccharides in the children's heavier fiber diet. Theses bacteria are capable of fermenting cellulose and xylan through a number of carbohydrate-active enzymes, producing anti-inflammatory effects at the same time. The speculation is that there is a link between these microbes that colonize the human gut and termites as part of the Burkinabè diet. The fourth key finding is that SCFA levels are statistically much higher in Burkinabè children compared with the Italian ones, and the four Burkina-specific species (*Prevotella, Xylanibacter, Treponema, and Butyrivibrio*) have the enzymes needed to digest fibers and produce these beneficial molecules. The presence of these species would allow Burkinabè children to maximize the energy intake from indigestible components by producing high levels of SCFAs that supply the host with additional energy. Normal colonic epithelia derive 60%–70% of their energy supply from SCFAs, particularly butyrate.[39] Propionate is largely taken up by the liver and is a good precursor for gluconeogenesis, liponeogenesis, and protein synthesis.[40] Acetate enters the peripheral circulation to be metabolized by peripheral tissues and is a substrate for cholesterol synthesis.[41,42] Together, they account for 10% of calories extracted from a Western diet each day[39] and probably a more abundant amount of calories in the diet of a typical rural village in Burkina Faso. In addition, SCFAs have several functions: they can reduce inflammation in colitis, promoting tissue renewal[43]; increase the absorption of sodium and water in diarrhea[44]; and regulate enteric neurons and control of GI motility.[45] The last key finding is that biodiversity is significantly reduced in Italian children with respect to Burkinobè children. The different bacterial compositions are likely to have profound influences on another

organ as well as the immune system, possibly explaining the absence of IBDs in African children and adults. Exposure to the large variety of environmental microbes associated with a high-fiber diet could increase the potentially beneficial bacterial genomes, enriching the microbiome. Reduction in microbial richness is possibly one of the undesirable effects of globalization and eating generic, nutrient-rich, uncontaminated foods. Both in the Western world and in the developing countries, diets rich in fat, protein, and sugar, together with reduced intake of unabsorbable fibers, are associated with a rapid increase in the incidence of noninfectious intestinal diseases. The potential protective effects of the diet on bowel disorders was first described by Burkitt[46] who, working in Africa in the 1960s, noticed the remarkable absence of noninfectious colonic diseases in Africans consuming a traditional diet rich in fiber. We extend this observation proposing that this health improvement is the combined result of a fiber-rich diet and microbiota. The speculation regarding the influence of a Burkinabè type of diet in reducing IBD and allergies have to be thoroughly tested in the future, but several independent pieces of evidence link Crohn's disease and allergies to alterations in intestinal microbiota and consequently in immune responses, and the increase in SCFAs that we report in rural Burkinabè children could explain why such diseases that are increasing in the Western world are virtually absent in Africa.

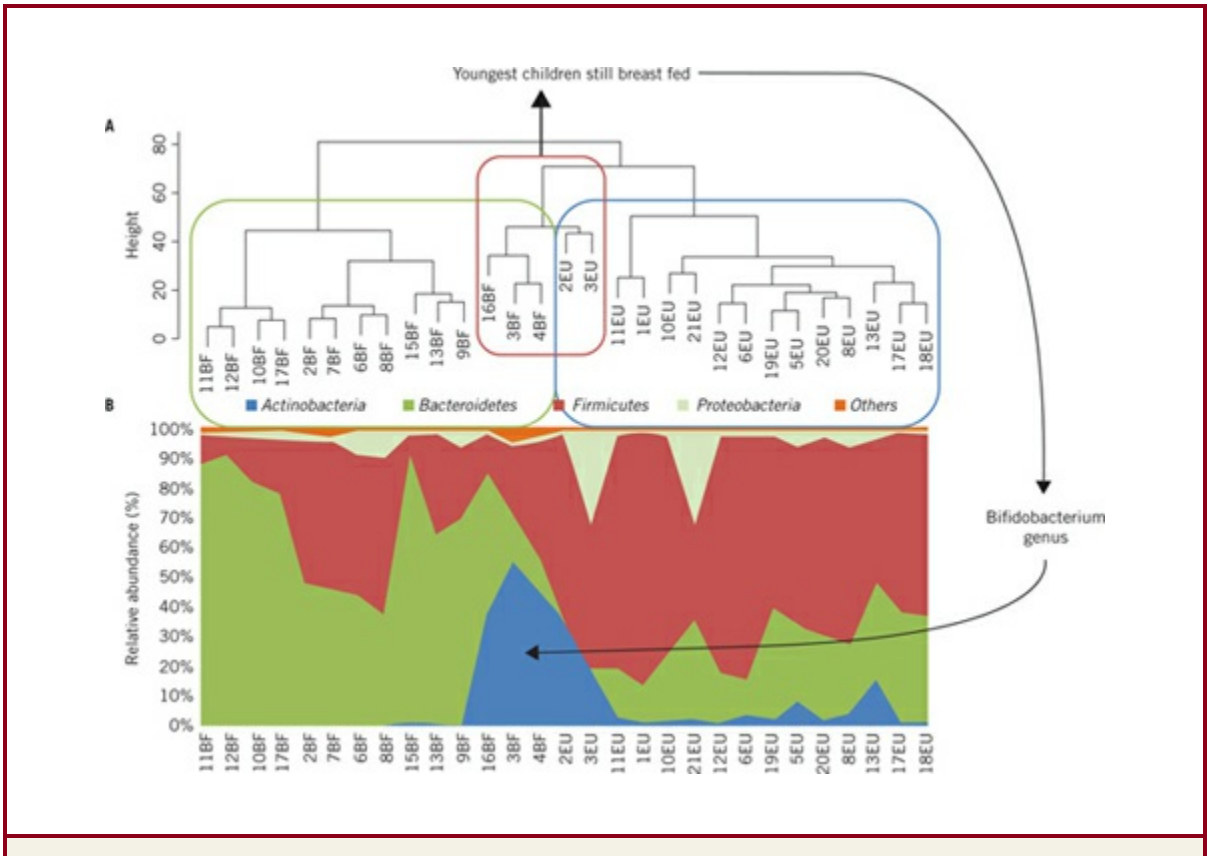


Figure 23-2 (A) Dendrogram obtained with complete linkage hierarchical clustering of the samples from BF and EU populations based on their genera. The subcluster located in the middle of the tree contains samples taken from the five youngest children (1–2 years old) still breast fed (three from BF group and two from EU group). (B) Relative abundances (percentage of sequences) of the four most abundant bacterial phyla in each individual among the BF and EU children. Firmicutes (red) and Bacteroidetes (green) abundances significantly differentiate the BF from the EU children. Blue area in middle shows abundance of *Actinobacteria*, mainly represented by *Bifidobacterium* genus, in the five youngest EU and BF children. BF=Burkina Faso; EU=Italian.

The increased biodiversity and possibly the activation of the immune function could be important also in protection from infections and to outcompete pathogenic bacteria. A diet poor in simple sugars and rich in fibers could in fact explain the marked reduction in potentially pathogenic phyla in the rural Burkinabè children with respect to the Europeans,[38] a paradox if one considers the fact that Burkinabè children drink from polluted water and live in a much less sanitized environment.

Cultural factors, especially diet, may be crucial in shaping the gut microbiota. The ratio of two of the main genera of gut bacteria, *Prevotella* and *Bacteroides*, correlates well with the overall pattern of diversity across healthy adults. Levels of *Prevotella* were enriched in our study on children from a rural African village who had a high-fiber diet, and this phenomenon was confirmed in a study on children and adults from Malawi and Venezuela whose diet was dominated by plant-derived polysaccharide foods such as maize and cassava.[47] By contrast, the microbiota of those in the United States had more *Bacteroides*. For healthy adults in the United States, differences in long-term diet between individuals also were correlated with these bacteria. Bacteroides was associated with a long-term diet rich in animal protein, several amino acids, and saturated fats, and *Prevotella* was associated with carbohydrates and simple sugars. The relative importance of changes in the microbiota as a result of diet, compared with other factors that affect health, remains a subject of active investigation.

DEVELOPMENT OF INTESTINAL MICROBIOTA IN INFANTS AND ITS IMPACT ON HEALTH

Metagenomics research is particularly promising in studies on infants gut microbiota Germ-free while in the womb, babies are exposed to beneficial bacteria in the birth canal and in the anogenital region of the mother's body during and following delivery. Statistically significant reports indicated increased rates of childhood allergies and asthma as related to Caesarean section and the lack of a typical bacterial ecology at birth. Breastfeeding also seems beneficial for bacterial balance. Sartor and Anday in 1987[48] report that formula-fed premature infants are susceptible to necrotizing enterocolitis. Breast milk provides a steady influx of beneficial bacteria (probiotic species such as bifidobacteria and lactobacilli are common in breast milk), with abundant evidence that gut bacteria that colonized early in life induce protective immune responses. The theory is that vaginal birth and breastfeeding help babies to acquire a healthy gut bacterial ecology, which helps the immune system ward off chronic ailments later in life.[49] To date, however, we know little about how the immune system responds to and learns from the successional development of the infant gut microbiota, from sterility before birth to the first early facultative anaerobic colonizers like the Enterobacteria and lactobacilli in the first days of life and later bifidobacteria during breastfeeding and subsequent development of the complex adult-like anaerobic gut microbiota in the young child. These early interactions between the infant's immune system and intestinal microorganisms are critical for development of immune tolerance and for protecting against immune related diseases. The key "educational" processes of exposure to microbial infections as well as appropriate colonization of the gut by a beneficial microorganisms, appears to be critical in the development of immune homeostasis. Importantly, early infections allow the immune system to recognize "friend from foe" by virtue of the fact that pathogenic bacteria distinguish themselves from other microbiota members by invading human tissues, occurring in inappropriate cellular compartments and causing cellular damage that then triggers inflammatory processes. The memory of these early encounters with unfriendly invaders is thought to contribute to the development of a well-functioning immune system, tolerant of the majority of intestinal microbiota members but able to mount rapid and effective inflammatory and antibody responses to invading pathogens. However, we still know very little about how diet, the mothers prenatum and during breast-feeding, and the infants during suckling and subsequently weaning, impacts on these complex interactions between the naive immune system and microbial successional development within the gut. Importantly, most previous studies have focused on only certain often

cultivable bacterial species within the infant gut microbiota or on particular time points in microbiota successional development. Human milk is a source of bacteria to the infant gut, where they may play a variety of antiinfectious, immunomodulatory, and metabolic roles. In fact, recent studies indicate that the human milk contains its own microbiota during lactation.[50] This bacterial community may differ depending on the individual and the health status of the lactating women. It seems that certain bacteria from the maternal gut can use mononuclear immune cells to colonize, first, the mammary gland and, later, the infant gut through breastfeeding.[51] Recently, it has been shown that breast-fed babies have many more Firmicutes bacteria than formula-fed infants.[52] Formula-fed babies have bacterial patterns more similar to adults, with bacterial imbalances, which are being linked to chronic conditions, such as diabetes associated with obesity. The results are indicating that formula probably lacks important components of mother's milk. Other studies have implicated bacteria in type 2 diabetes, which can be a consequence of obesity, reporting that a high-fat diet triggers inflammatory responses in the small intestines and colon long before the animals gain weight.[53] Germ-free animals that don't have bacteria don't get the intestinal inflammation, insulin resistance, or weight gain even when they eat as much fat as regular rats.[54] These researchers also found a strong correlation between bacteria-induced inflammation and insulin resistance, which can lead to diabetes. It is right now widely demonstrated that a bacterial imbalance in our gut plays a role in diabetes, asthma, and allergies. Probably this is just the tip of the iceberg, and the correlations and causations between bacteria and disease are much more than we could predict.

It is estimated that certain periods in life (like the suckling period, introduction of formula and solid foods) offer windows during which nutrition might affect the immunologic development of the young infant. Nutrition may be the source of antigens to which the immune system must become tolerant, provide factors (including nutrients) that themselves might modulate immune maturation and responses, and provide factors that influence intestinal microbiota, which in turn will affect antigen exposure, immune maturation, and immune response.[55] The diverse mixture of bioactive components in human milk influences the immune status of the suckling infant not only by providing protection but also by facilitating the development, tolerance, and appropriate inflammatory response. Maternal milk can be considered as the communication vehicle

between the maternal immune system and the infant.[56–58] The interaction of food compounds with the immune system or indirectly through their effect on the gut microbiota will influence the process of immune maturation and responses.

ROLE OF GUT MICROBIOTA IN OBESITY AND METABOLIC DISEASE

Several studies focused on the ability of different microbiota to differently extract food calories. This trait has indeed been under selection during our evolution; the ability to store energy would be a beneficial attribute for ancient humans who had variable access to food. However, in modern, developed societies, where there is ready access to large portion, high-calorie diets, this benefit becomes a detriment. Many studies described that one manifestation of this symbiotic relationship is microbial processing of components of the diet and deposition the extracted energy in host fat depots. Obesity has reached epidemic proportions in Westernized cultures, and the diseases associated with it, including insulin resistance and type II diabetes mellitus, hepatic steatosis and steatohepatitis, dyslipidemia, and atherosclerotic cardiovascular disease, have become major public health problems.[59] Traditional obesity research has focused on environmental and host genetic factors, including descriptive studies in humans and mechanistic studies using genetically altered rodent models.[60] More recently, research over the last five years using culture-independent methods and NGS has suggested that the pathogenesis of obesity may be influenced by our endogenous gut microbiota.[61–65] A fundamental breakthrough came from 16S rRNA–based analysis of community structure (phylotype abundance distribution) associated to sequencing of fecal microbial communities (metagenomics) of obese and nonobese adult human twins, and their mothers.[2,22,62] Family members had considerable overlap in their gut microbial communities, with the degree of variation being similar between monozygotic and dizygotic twin pairs. Notably, however, obese individuals showed an impressive overall reduction in microbial diversity. One of the most striking results was an altered balance of the ratio between Bacteroidetes and Firmicutes, the two major phyla commensal bacteria residing in the GI tract. An important metabolic difference between these two phyla is the efficiency of energy extraction from undigestible dietary carbohydrates. Therefore, alterations in their relative ratio can result in increased energy accumulation, which is of

special concern in certain pathological conditions. For this reason, several studies have focused in recent years on the composition of human gut microbiota in obese subjects under different dietary regimens.[22,65] The authors liken this reduced diversity to a fertilizer runoff, in which a subset of the microbial community blooms in response to abnormally high-energy input, as opposed to the rainforest- or reef-like community of the lean gut, which displays high species diversity in the face of high-energy flux.

To selectively test the role of microbiota in obesity animal, models have been used as a controlled system to investigate the role of dietary changes on microbiota.[35,63,64]

"Top-down" systems biology comparisons of metabolic profiles of normal human and mice microbiota revealed that absorption, storage, and metabolism of dietary lipids were specifically modulated by the microbiome.[66,67] Moreover, the induction of type 2 diabetes and obesity with a high-fat diet in rats has been shown to correlate with preexisting metabolic patterns associated with differences in gut bacterial activities, indicating causality between microbiome and host predisposition to diseases.[68]

Changes in the gut microbiota of humanized gnotobiotic mice have been studied after the mice are switched from a diet low in fat and rich in plant polysaccharides to a Western diet high in fat and sugar and low in plant polysaccharides.[35] After just one day on the Western diet, mice show changes in their microbial composition, metabolic pathways, and gene expression, and within two weeks they develop more adiposity. Mice on a Western diet show an increase in bacteria of the Firmicutes phylum and a decrease in those of the Bacteroidetes phylum.

These concepts represent an important paradigm shift in understanding health conditions and disease and are likely to have a significant impact on the future of the prevention and therapy of intestinal diseases.[66]

Despite the advantage of selectively and precisely controlling all the variables, mice seems to be a questionable model for studies on human gut microbiota. The conclusions obtained from animal models or from a handful of human individuals can be hardly generalized to human populations.[68]

The challenge we face now is to understand how the different environments and wide range of diets that modern humans around the world experience has affected the microbial ecology of the human gut.

PROBIOTICS AND GUT MICROBIOTA MODULATION

A number of functional foods have been developed with the aim of altering or modulating the human gut microbiota. Probiotics are live microorganisms, which when administered in adequate amount confer a health benefit on the host.

Probiotics as defined by the World Health Organization in 2001[69] are a relatively recent phenomenon. In that, the first commercial preparations appeared in France and Japan in the early 1900s and 1930s, respectively, and aim to improve host health by adding exogenous microorganisms, usually lactic acid bacteria, bifidobacteria, or the yeast *Saccharomyces boulardii* to the hosts' gut microbiota and are gaining scientific backing for protective capabilities against a number of chronic human diseases and GI infections. However, "probiotics" in the human diet probably goes back much further, with long histories of using very similar microorganisms for fermented milk, meats, and cereals going back to the beginnings of agriculture and touching on the "hygiene hypothesis" or "old friends hypothesis,"[70] which endow the health benefits through appropriate education of the immune system in infancy and lifelong regulation of immune homeostasis by regular and diverse interactions with environmental, including food, microorganisms, both beneficial or benign, and those responsible for minor infections. Since modern probiotics do not colonize the human gut microbiota and only on a few occasions have been shown to alter mammalian metabolic processes, the likely mechanism of probiotics effect is through the immune system.[71] Thus, far from being a modern invention of the food industry, probiotics and the probiotic principle has a long history and is part of wider scientific paradigm describing our continued connection with the natural world through the microorganisms we eat, our gut microbiota and our highly evolved immune system.[70]

"A prebiotic is a selectively fermented ingredient that results in specific changes in the composition and/or activity of the GI microbiota, thus conferring benefit(s) upon host health" and represents another functional food approach toward modulating the composition of the human gut microbiota.[72] These fibers, which are selectively fermented by the human gut microbiota, again although recently defined as modern food ingredients, have a scientific basis at the core of gut microbiota functionality, saccharolytic fermentation with its resultant production of SCFAs, and changes within the relative abundance of bacteria known to

play important roles in immune-homeostasis, lipid/fat metabolism, regulation of mucosal integrity, and vitamin production, most notably, the bifidobacteria.[73] Interestingly, certain whole plant foods, especially whole grain cereals are proving to mediate the same alterations within the composition and activity of the human gut microbiota as these modern prebiotic food ingredients, fitting well what we know about the health benefits of both prebiotics and whole grain cereals especially in biomarkers of metabolic health such as blood cholesterol levels.

REFERENCES

1. Gill SR, Pop M, Deboy RT, et al. Metagenomic analysis of the human distal gut microbiome. *Science.* 2006;312:1355–1359.
2. Turnbaugh PJ, Hamady M, Yatsunenko T, et al. A core gut microbiome in obese and lean twins. *Nature.* 2009;457:480–484.
3. Lozupone CA, Stombaugh JI, Gordon JI, et al. Diversity, stability and resilience of the human gut microbiota. *Nature.* 2012;489:220–230.
4. Metzker ML. Sequencing technologies—the next generation. *Nat Rev Genet.* 2010;1(1):31–46.
5. Turnbaugh PJ, Ley RE, Hamady M, et al. The human microbiome project. *Nature.* 2007;449(7164):804–810.
6. Cole JR, Wang Q, Cardenas E, et al. The Ribosomal Database Project: improved alignments and new tools for rRNA analysis. *Nucleic Acids Res.* 2009;37(Database issue):D141–D145.
7. Petrosino JF, Highlander S, Luna RA, et al. Metagenomic pyrosequencing and microbial identification. *Clin Chem.* 2009;55(5):856–866.
8. McKenna P, Hoffmann C, Minkah N, et al. The macaque gut microbiome in health, lentiviral infection, and chronic enterocolitis. *PLoS Pathog.* 2008;4(2):e20.
9. Andersson AF, Lindberg M, Jakobsson H, et al. Comparative analysis of human gut microbiota by barcoded pyrosequencing. *PLoS One*. 2008;3(7):e2836.
10. Qin J, Li R, Raes J, et al. A human gut microbial gene catalogue established by metagenomic sequencing. *Nature.* 2010;464:59–65.
11. Arumugam M, Raes J, Pelletier E, et al. Enterotypes of the human gut microbiome. *Nature.* 2011;473(7346):174–180.
12. De Filippo C, Ramazzotti M, Fontana P, Cavalieri D. Bioinformatic approaches for functional annotation and pathway inference in metagenomics data. *Brief Bioinform.* 2012;13(6):696–710.
13. Elango R, Ball RO, Pencharz PB. Amino acid requirements in humans: with a special emphasis on the metabolic availability of amino acids. *Amino Acids.* 2009;37(1):19–27.
14. Gao X, Pujos-Guillot E, Martin JF, et al. Metabolite analysis of human fecal water by gas chromatography/mass spectrometry with ethyl chloroformate derivatization. *Anal Biochem.* 2009;393(2):163–175.
15. Macfarlane GT, Macfarlane S. Bacteria, colonic fermentation, and gastrointestinal health. *J AOAC Int.* 2012;95(1):50–60.
16. Kaczmarczyk MM, Miller MJ, Freund GG. The health benefits of dietary fiber: beyond the usual suspects of type 2 diabetes mellitus, cardiovascular disease and colon cancer. *Metabolism.* 2012;61(8):1058–1066.

17. Richardson AJ, McKain N, Wallace RJ. Ammonia production by human faecal bacteria, and the enumeration, isolation and characterization of bacteria capable of growth on peptides and amino acids. *BMC Microbiol.* 2013;13:6.
18. Shen Q, Zhao L, Tuohy KM. High-level dietary fibre up-regulates colonic fermentation and relative abundance of saccharolytic bacteria within the human faecal microbiota in vitro. *Eur J Nutr.* 2012;51(6):693–705.
19. Tuohy KM, Gougoulias C, Shen Q, et al. Studying the human gut microbiota in the trans-Omics era—focus on metagenomics and metabonomics. *Curr Pharm Des.* 2009;15(13):1415–1427.
20. Nicholson JK, Holmes E, Wilson ID. Gut microorganisms, mammalian metabolism and personalized health care. *Nat Rev Microbiol.* 2005;3:431–438.
21. Eckburg PB, Bik EM, Bernstein CN, et al. Diversity of the human intestinal microbial flora. *Science.* 2005;308:1635–1638.
22. Ley RE, Turnbaugh PJ, Klein S, et al. Microbial ecology: human gut microbes associated with obesity. *Nature.* 2006;444:1022–1023.
23. Del Rio D, Costa LG, Lean ME, Crozie A. Polyphenols and health: what compounds are involved? *Nutr Metab Cardiovasc Dis.* 2010;20(1):1–6.
24. Cordain L, Eaton SB, Sebastian A, et al. Origins and evolution of the Western diet: health implications for the 21st century. *Am J Clin Nutr*. 2005;81:341–354.
25. Willett WC. Overview and perspective in human nutrition. *Asia Pac J Clin Nutr.* 2008;17(suppl 1):1–4.
26. Willcox DC, Willcox BJ, Todoriki H, Suzuki M. The Okinawan diet: health implications of a low-calorie, nutrient-dense, antioxidant-rich dietary pattern low in glycemic load. *J Am Coll Nutr.* 2009;28(suppl):500S–516S.
27. Issazadeh-Navikas S, Teimer R, Bockermann R. Influence of dietary components on regulatory T cells. *Mol Med.* 2012;18:95–110.
28. Macia L, Thorburn AN, Binge LC, et al. Microbial influences on epithelial integrity and immune function as a basis for inflammatory diseases. *Immunol Rev.* 2012;245(1):164–176.
29. Soerjomataram I, Oomen D, Lemmens V, et al. Increased consumption of fruit and vegetables and future cancer incidence in selected European countries. *Eur J Cancer.* 2010;46(14):2563–2580.
30. Bhupathiraju SN, Tucker KL. Coronary heart disease prevention: nutrients, foods, and dietary patterns. Clin Chim Acta. 2011;412(17–18):1493–1514.
31. Arthur JC, Jobin C. The complex interplay between inflammation, the microbiota and colorectal cancer. *Gut Microbes*. 2013;4(3):253–258.
32. Hehemann JH, Correc G, Barbeyron T, et al. Transfer of carbohydrate-active enzymes from marine bacteria to Japanese gut microbiota. *Nature.* 2010;464:908–912.
33. Tuohy K, Davies M, Rumsby P, et al. Monitoring transfer of recombinant and non-recombinant plasmids between Lactococcus lactis strains and members of the human gastrointestinal microbiota in vivo—impact of donor cell number and diet. *J Appl Microbiol.* 2002;93:954–964.
34. Bäckhed F, Ley RE, Sonnenburg JL, et al. Host-bacterial mutualism in the human intestine. *Science.* 2005;25:1915–1920.
35. Turnbaugh PJ, Ridaura VK, Faith J, et al. The effect of diet on the human gut microbiome: a metagenomic analysis in humanized gnotobiotic mice. *Sci Transl Med.* 2009;1:6ra14.
36. Mueller S, Saunier K, Hanisch C, et al. Differences in fecal microbiota in different European study populations in relation to age, gender, and country: a cross-sectional study. *Appl Environ Microbiol.* 2006;72:1027–1033.

37. Adlerberth I, Jalil F, Carlsson B, et al. High turnover rate of *Escherichia coli* strains in the intestinal flora of infants in Pakistan. *Epidemiol Infect*. 1998;121:587–598.
38. De Filippo C, Cavalieri D, Di Paola M, et al. Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proc Natl Acad Sci USA.* 2010;107(33):14691–14696.
39. Scheppach W. Effects of short chain fatty acids on gut morphology and function. *Gut*. 1994;35(1 suppl):S35–S38.
40. Wolever TM, Brighenti F, Royall D, et al. Effect of rectal infusion of short chain fatty acids in human subjects. *Am J Gastroenterol.* 1989;84:1027–1033.
41. Wolever TM, Spadafora P, Eshuis H. Interaction between colonic acetate and propionate in humans. *Am J Clin Nutr*. 1991;53:681–687.
42. Cummings JH, Pomare EW, Branch WJ, et al. Short chain fatty acids in human large intestine, portal, hepatic and venous blood. *Gut*. 1987;28:1221–1227.
43. Topping DL, Clifton PM. Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. *Physiol Rev.* 2001;81(3):1031–1064.
44. Binder HJ. Role of colonic short-chain fatty acid transport in diarrhea. *Annu Rev Physiol.* 2010;72:297–313.
45. Soret R, Chevalier J, De Coppet P, et al. Short-chain fatty acids regulate the enteric neurons and control gastrointestinal motility in rats. *Gastroenterology*. 2010;138(5):1772–1782.
46. Burkitt DP. Epidemiology of large bowel disease: the role of fibre. *Proc Nutr Soc* 1973;32:145–149.
47. Yatsunenko T, Rey FE, Manary MJ, et al. Human gut microbiome viewed across age and geography. *Nature.* 2012;486:222–227.
48. Sartor O, Anday E. Campylobacter jejuni enteritis in a premature neonate. *South Med J*. 1987;80:1593–1594.
49. Morowitz MJ, Denef VJ, Costello EK, et al. Strain-resolved community genomic analysis of gut microbial colonization in a premature infant. *Proc Natl Acad Sci USA.* 2011;108(3):1128–1133.
50. Ward TL, Hosid S, Ioshikhes I, Altosaar I. Human milk metagenome: a functional capacity analysis. *BMC Microbiol*. 2013;13:116.
51. Donnet-Hughes A, Perez PF, Doré J, et al. Potential role of the intestinal microbiota of the mother in neonatal immune education. *Proc Nutr Soc*. 2010;69(3):407–415.
52. Azcarate-Peril MA, Altermann E, Goh YJ, et al. Analysis of the genome sequence of Lactobacillus gasseri ATCC 33323 reveals the molecular basis of an autochthonous intestinal organism. *Appl Environ Microbiol.* 2008;74:4610–4625.
53. Karlsson FH, Tremaroli V, Nookaew I, et al. Gut metagenome in European women with normal, impaired and diabetic glucose control. *Nature.* 2013;498(7452):99–103.
54. Ding S, Chi MM, Scull BP, et al. High-fat diet: bacteria interactions promote intestinal inflammation which precedes and correlates with obesity and insulin resistance in mouse. *PLoS One.* 2010;5(8):e12191.
55. Calder PC, Krauss-Etschmann S, de Jong EC, et al. Early nutrition and immunity —progress and perspectives. *Br J Nutr.* 2006;96(4):774–790.
56. van Odijk J, Kull I, Borres MP, et al. Breastfeeding and allergic disease: a multidisciplinary review of the literature (1966–2001) on the mode of early feeding in infancy and its impact on later atopic manifestations. *Allergy.* 2003;58(9):833–843.

57. Morrow AL, Ruiz-Palacios GM, Altaye M, et al. Human milk oligosaccharides are associated with protection against diarrhea in breast-fed infants. *J Pediatr*. 2004;145(3):297–303.
58. Martín R, Jiménez E, Olivares M, et al. Lactobacillus salivarius CECT 5713, a potential probiotic strain isolated from infant feces and breast milk of a mother-child pair. *Int J Food Microbiol*. 2006;112(1):35–43.
59. Van Gaal LF, Mertens IL, De Block CE. Mechanisms linking obesity with cardiovascular disease. *Nature.* 2006;444(7121):875–880.
60. Friedman JM. Leptin at 14 y of age: an ongoing story. *Am J Clin Nutr*. 2009;89(3):973S–979S.
61. Bäckhed F, Ding H, Wang T, et al. The gut microbiota as an environmental factor that regulates fat storage. *Proc Natl Acad Sci USA.* 2004;101(44):15718–15723.
62. Ley RE, Bäckhed F, Turnbaugh P, et al. Obesity alters gut microbial ecology. *Proc Natl Acad Sci USA.* 2005;102(31):11070–11075.
63. Bäckhed F, Manchester JK, Semenkovich CF, Gordon JI. Mechanisms underlying the resistance to diet-induced obesity in germ-free mice. *Proc Natl Acad Sci USA.* 2007;104(3):979–984.
64. Turnbaugh PJ, Bäckhed F, Fulton L, Gordon JI. Diet-induced obesity is linked to marked but reversible alterations in the mouse distal gut microbiome. *Cell Host Microbe*. 2008;3(4):213–223.
65. Turnbaugh PJ, Ley RE, Mahowald MA, et al. An obesity-associated gut microbiome with increased capacity for energy harvest. *Nature.* 2006;444(7122):1027–1031.
66. Nicholson JK. Global systems biology, personalized medicine and molecular epidemiology. *Mol Syst Biol*. 2006;2:52.
67. Martin FP, Dumas ME, Wang Y, et al. A top-down systems biology view of microbiome-mammalian metabolic interactions in a mouse model. *Mol Syst Biol.* 2007;3:112.
68. Seok J, Warren HS, Cuenca AG, et al. Inflammation and Host Response to Injury, Large Scale Collaborative Research Program. Genomic responses in mouse models poorly mimic human inflammatory diseases. *Proc Natl Acad Sci USA.* 2013;110(9):3507–3512.
69. Food and Agriculture Organization, World Health Organization. *Guidelines for the Evaluation of Probiotics in Food. Report of a Joint FAO/WHO Working Group on Drafting Guidelines for the Evaluation of Probiotics in Food. London, Ontario, Canada;* April 30 and May 1, 2002. ftp://ftp.fao.org/es/esn/food/wgreport2.pdf.
70. Rook GAW, Brunet LR. Microbes, immunoregulation and the *gut*. Gut. 2005;54:317–320.
71. McNulty NP, Yatsunenko T, Hsiao A, et al. The impact of a consortium of fermented milk strains on the gut microbiome of gnotobiotic mice and monozygotic twins. *Sci Transl Med.* 2011;3(106):106ra106.
72. Gibson GR. Dietary modulation of the human gut microbiota using the prebiotics oligofructose and inulin. *Am Socity Nutr Sci*. 1999;129:1438S–1441S.
73. Hardy H, Harris J, Lyon E, et al. Probiotics, prebiotics and immunomodulation of gut mucosal defences: homeostasis and immunopathology. *Nutrients.* 2013;5(6):1869–1912.

Nutrition and 24 Neuropsychological Development

Maureen M. Black, PhD and Kristen M. Hurley, PhD

Nutritional deprivation is a worldwide public health concern that can result in long-term deficits to children's growth and development. Two series of papers in *The Lancet* on global child development found that more than 200 million children under 5 years do not reach their developmental potential, largely due to poverty, chronic undernutrition (stunting), micronutrient deficiencies, and lack of early stimulation.[1–5] Delayed development early in life increases the likelihood of neuropsychological problems, poor school performance, and subsequently low income-generating potential, high fertility, and poor caregiving capacity, thus contributing to the intergenerational transmission of poverty. However, early home and center-based programs targeted toward young disadvantaged children throughout the world can disrupt the negative cycle and contribute to positive effects on children's neuropsychological development, academic performance, and life chances.[4]

This chapter examines the relationship between nutritional deficiencies and neuropsychological development in children. It reviews the history of research linking nutrition and children's development, methods used to study child development, and the evidence linking nutrition and child development, with an emphasis on micronutrient deficiencies. Finally,

recommendations are made to reduce the burden that children experience from nutritional deficiencies.

HISTORICAL CONSIDERATIONS

The association between nutritional deprivation and child development has been a topic of interest for over 40 years. Initially, investigators focused on protein-energy malnutrition (PEM), caused by a lack of calories. During the acute phase of nutritional deprivation, children have been described as lethargic, apathetic, and inactive, with poor performance on measures of cognitive functioning.[6]

Research on PEM has focused on the long-term consequences associated with early nutritional deprivation. Guided by findings from animal research that nutritional deprivation can lead to structural and functional changes in the developing brain,[7] investigators have used a variety of standardized assessment tools, such as intelligence quotient (IQ) tests or behavioral rating scales, to determine whether nutritionally deprived children recover or experience long-term neurobehavioral consequences. Case-control, retrospective designs have been used to study children with a history of nutritional deprivation and compare their task performance with that of adequately nourished children. Findings generally indicate long-lasting deficits in intelligence and school performance.[6]

Nutritional deprivation often occurs in the context of poverty. Although most investigators handle socioeconomic differences in study populations by recruiting comparison children from similar socioeconomic backgrounds, little attention has been directed to individual differences or how children's response to nutritional rehabilitation varies by environmental characteristics. Advances in the areas of growth, nutrition, animal research, behavioral neuroscience, and child development have influenced recent investigations of nutritional deprivation and child development.

Growth

In the absence of adequate nutrients, the body may conserve energy by initially limiting weight gain and subsequently limiting linear growth. Linear growth or height is influenced by genetic factors, environmental factors, and medical conditions. In 2006, the World Health Organization (WHO) published international growth standards from the Multicountry

Growth Reference Study, which examined how healthy children grow in homes that engage in health-promoting practices (breastfeeding and not smoking). Approximately 8500 children from varying ethnic and cultural groups in six diverse countries (Brazil, Ghana, India, Norway, Oman, and the United States) participated in the study. Children were measured from birth through age 5 years. There was strong similarity in linear growth across the six sites, providing evidence for the use of a single international growth standard and demonstrating that when children's health and environmental needs are met, growth is similar.[8]

Stunting, defined as children who fall below −2SD from the length- or height-for-age WHO Child Growth Standards median, serves as a general indicator of children's chronic nutritional status. Growth stunting is a gradual process that occurs in response to chronic biological insults, including undernutrition and infectious diseases, during periods of linear bone growth. It often begins in utero and extends through the first 2 years, commonly referred to as the first 1000 days.[9] Childhood stunting is closely associated with poverty and is often used as a population-based indicator to compare nutritional adequacy across countries. Early stunting can disrupt permanent growth, resulting in shorter stature among adults with a history of stunting. Recent evidence from United Nations Children's Fund (UNICEF), WHO, and the World Bank, based on 2011 data from 639 national surveys from 142 countries finds that 26% of children under age 5 years (165 million) are stunted (height-for-age Z score of −2 or lower). This figure is a dramatic decline from surveys conducted in 1990, which estimated rates of stunting of 40% (253 million children). Changes in the rates of stunting reveal marked inequalities, particularly related to economic indicators.[10] The highest prevalence of stunting occurs in Africa, but the greatest number of children affected is in Asia.

Mounting evidence from multiple countries suggest that children who experienced nutritional deprivation severe enough to cause stunting early in life have cognitive deficits during school-age years that can interfere with successful academic performance.[4,5] Early stunting undermines children's long-term performance and has been used as an indicator to estimate the number of children worldwide who do not reach their developmental potential.[3]

Animal Research

Animal models have been widely utilized in research linking nutrition to cognitive functioning. Much of the neurological development of rats

occurs postnatally, enabling investigators to study the timing and chronicity of nutritional deprivation.[7] However, many of the techniques used to introduce nutritional deprivation (e.g., increasing the number of infants for a mother to feed, decreasing access to the mother, etc.) altered mother-pup interaction and the social environment, thus confounding the research. Through innovative designs, such as the introduction of artificial feeding, investigators have been able to vary both nutritional intake and environmental stimulation.[11] Animals who experience both nutritional and environmental deprivation tend to have worse performance than animals deprived in only one condition, suggesting that the environmental stimulation may attenuate the adverse consequences of undernutrition.[12] Levitsky and Barnes[13] described the process whereby nutritional deficiencies may alter caregiving behavior as *functional isolation* (Figure 24-1).[13] Thus, in addition to the direct effects of nutritional deprivation, undernourished animals may also be denied the environmental enrichment that promotes development.

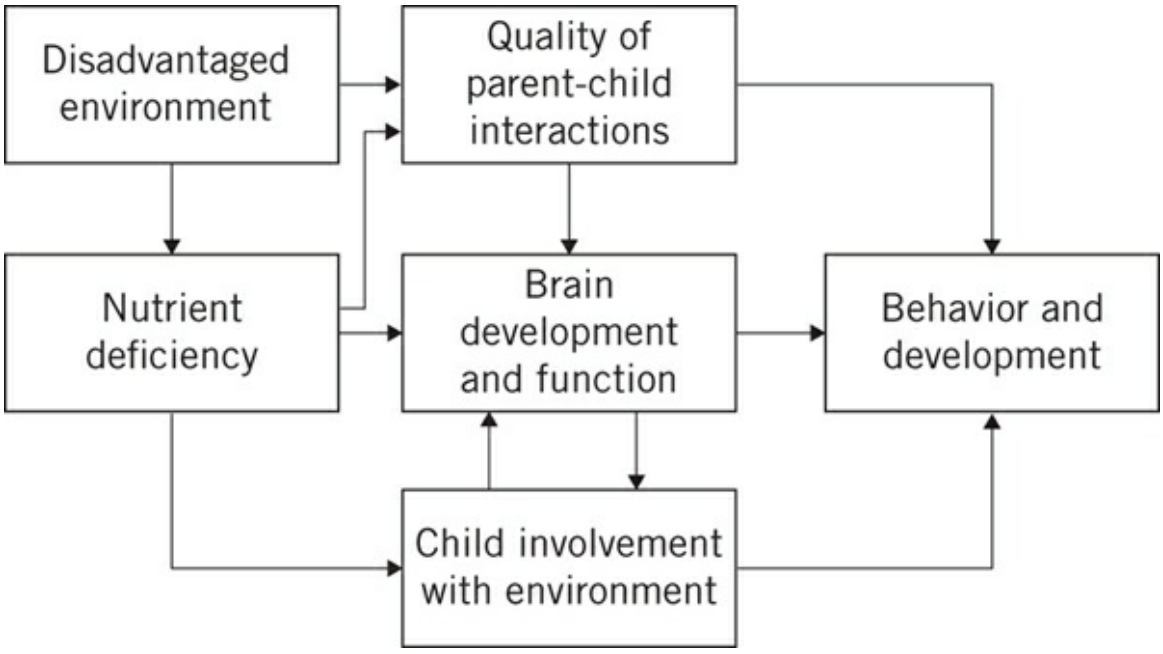


Figure 24-1 Association between nutritional deficiency and children's behavior and development. Adapted from Levitsky and Barnes.[13]

The timing of nutritional deprivation has been a particularly relevant question because early investigations were guided by the sensitive period

hypothesis that developing organs deprived of essential nutrients would experience irreversible structural damage.[7] Although some changes induced by nutritional deprivation remain following rehabilitation, particularly related to myelin (the fatty substance that surrounds nerve cells and aids with transmission), neuronal mitrochondria, and neurotransmitter metabolism,[11] many structural changes can be reversed with nutritional rehabilitation.[14] Evidence suggests that there may be metabolic or neurochemical consequences of nutritional deprivation.[11]

Rehabilitated animals with a history of nutritional deprivation appeared to be at risk for changes in their emotionality, motivation, anxiety, and cognitive flexibility, evidenced by behaviors such as increased spilling, hyperreactivity, and increased aggression. These behaviors may interfere with performance on cognitive tasks, suggesting that early undernutrition may alter neurochemical-receptor systems, resulting in an inability to adapt to novel or stressful situations.[14]

Behavioral Neuroscience

Advances in behavioral neuroscience have shown the important role that experiences on brain development, particularly with respect to pruning, myelination, and synaptic formation.[15] Brain development begins prenatally and continues through adolescence (Figure 24-2).[16,17] It begins with the formation of brain cells, followed by cell migration and differentiation, and the development of synapses to enable cells to communicate with one another. Finally, the supportive tissue (myelin) is formed to protect the nerve cells and facilitate communication.[17,18]

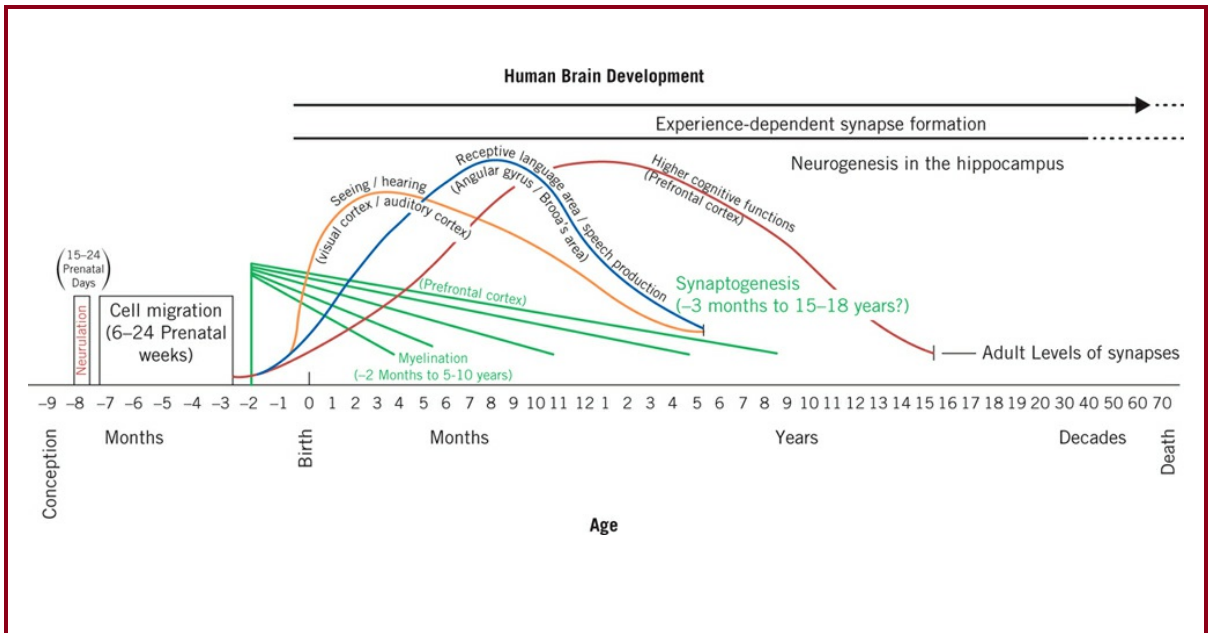


Figure 24-2 Human brain development. Adapted from Thompson and Nelson.[16]

The process of forming and eliminating synapses occurs throughout the developmental period and is influenced by two types of experiences: experience-expectant and experience- dependent.[18] Experience-expectant refers to species-specific development, including maturational influences that guide early sensory and motor development, such as the specific roles played by nutrients in altering cell function and structure through neurochemical changes within the cells. Experience-dependent occurs throughout life as individuals adapt to specific cultural and environmental demands and opportunities. The structural changes in synaptic formation are dependent on neurochemical- receptor systems that, in turn, are influenced by experiences as basic as caregiving.[18]

The timing of early developmental experiences has been a central issue in studying behavioral neuroscience. The critical or sensitive period hypothesis suggests that if an event, such as nutritional deprivation, occurs during a period of rapid development, such as the first 1000 days, it may have long-lasting impact on the organism.[7]

Recent evidence suggests that although the first 1000 days (conception to 24 months of age) is a sensitive period in children's development, it is not the only sensitive period.[19] Children's development is characterized by multiple sensitive periods, depending on specific structures or functions. For example, adolescence is a sensitive period for the ongoing development of the prefrontal cortex, vulnerability to stress, and aspects of executive function, perspective taking, and abstract reasoning.

Child Development

Child development is defined here as the orderly progression of skills that children acquire as they mature and adapt to their specific caregiving settings. Although there are universal aspects of child development, particularly early in life as children gain motor skills and learn to roll over, sit, stand, and walk, there are also individual and cultural differences in the acquisition of skills. For example, children learn to understand and speak the languages they hear in their household.

The recognition that the link between nutritional deprivation and neuropsychological development depends on nonnutritional factors and can be influenced by environmental stimulation has had an important impact on research involving nutritionally deprived children. Caregiving

variables, such as the quality of parent–child interaction, have been incorporated into models of child development, moving away from models that merely control for socioeconomic differences.[20,21]

A transactional system approach provides a useful framework for understanding the link between nutrition and early child development because it emphasizes the interplay that occurs between children and their environment, mediated through relations with their caregivers (Figure 24-3).[22,23] Children's development is influenced by characteristics from the child (e.g., nutritional status), from the caregiver (e.g., maternal education), and from interactions between the child and caregiver (e.g., reciprocity and clarity of communication during feeding).

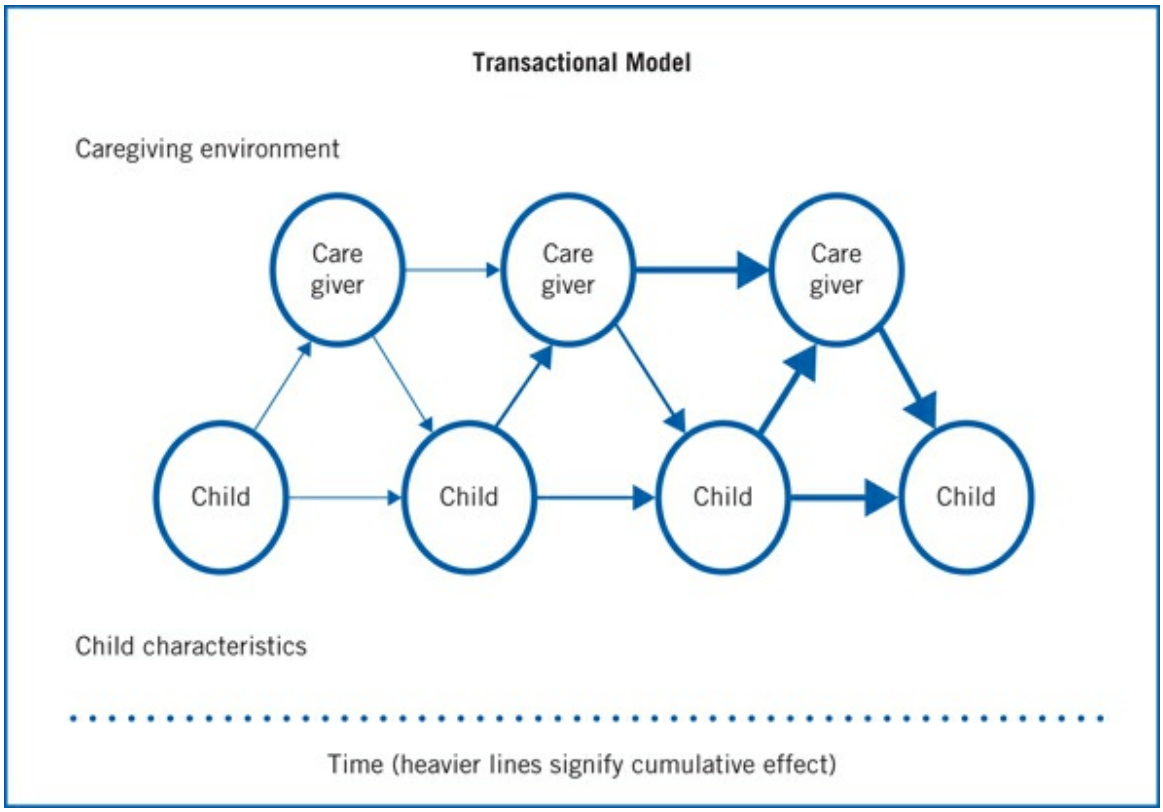


Figure 24-3 The transactional model.

The transactional system forms the theoretical basis for responsive parenting and responsive feeding.[24] Responsive feeding emphasizes the interactive nature of feeding, whereby caregivers set guidelines and gage their reactions to the signals they read from their children, ideally resulting in a respectful give-and-take (serve-and-return) exchange around feeding,

resulting in healthy mealtime behavior and a healthy weight gain.[25] Unresponsive feeding interactions may be characterized by negative, coercive strategies to promote eating or by permissive, overindulgent strategies, such as making food available throughout the day or providing favorite foods with little regard to quality. These strategies are likely to undermine children's appetite, increasing mealtime stress as children hold out for snacks.

The transactional model may also form the basis for intergenerational transmission of disparities. For example, a woman who is undernourished during her pregnancy and does not receive prenatal care is at increased risk to give birth to an infant with low birth weight (LBW) (<2500 g). In comparison with full-term infants, LBW infants are smaller and often have weaker cries and sucking skills.[26] These characteristics make infants with LBW more difficult to feed, particularly for mothers who are undernourished themselves. A cycle emerges whereby the small, weak infant is difficult to feed, does not grow well, may be vulnerable to illnesses, and lags behind in developmental skills. Mothers may feel frustrated and/or frightened by their infants who are small and sickly and may limit their caregiving to holding, rather than talking or encouraging exploration.[27] This process results in a negative cycle whereby infants do not elicit or receive the care and attention they need to promote optimal development, a human example of functional isolation.

Intervention can occur at multiple points within the transactional system.[23] For example, intervention could be directed to the infant with LBW, perhaps through nutritional and developmental intervention that might be available through community nutrition programs, or through early intervention programs. If the child's growth and development improve, the child will be more likely to elicit and receive age-appropriate care and interaction from the caregiver.

Intervention could also be directed toward the caregiver and the interaction between the caregiver and the child. Children's cues can be targeted to ensure that the caregiver understands how the child expresses hunger or satiety, and can read and respond to the child's cues. Using components of the transactional system to promote the development of undernourished children has been a successful strategy.[1,2]

Although, most of the attention has been directed to the impact of inadequate nutritional and caregiving practices on children's nutritional status, children may influence caregiver behavior through their nutritional status, as predicted by the transactional system. Children with PEM have

been described as negative, wary, and lethargic.[28] During home observations, mothers of undernourished infants have been described as less sensitive and responsive than mothers of healthy infants,[29] and tended to hold or carry them, rather than play with them.[27] Caregivers who treat their infants as though they are ill or unable to participate in age-appropriate activities may be inadvertently denying them opportunities for developmental stimulation and enrichment.

In one of the only studies to observe caregiver-child interaction prior to the child's becoming undernourished, investigators in Mexico reported that infants of mothers who lacked sensitivity and affection were more likely to become undernourished, compared to infants of more sensitive mothers.[30] In turn, after infants received nutritional supplements, their mothers were more engaged, both in caregiving and in enrichment activities, compared with mothers of nonsupplemented infants, suggesting that mothers respond positively to their infants' increased energy and activity.

METHODOLOGY

Most research examining associations between early nutritional deprivation and subsequent intellectual skills has been retrospective and observational, with little attention to differences in environmental or caregiving factors. This section examines the choice of outcome and predictor variables, and a consideration of environmental and caregiving variables that may confound findings.

Childhood Measure of Intelligence

The primary outcome measures used in studies of recovery from nutritional deprivation have been generalized tests of intelligence administered during the school-age years, such as the Wechsler Intelligence Scale for Children (WISC). The WISC is a standardized assessment that yields an IQ that is stable over time and predicts both academic performance and occupational status.[31] Although many other factors contribute to academic performance and occupational success (e.g., motivation), the IQ is a useful indicator of successful rehabilitation.

Infant Measures of Development

Infancy is marked by rapidly changing skills, discontinuities in early development, and limited performance options. Early measures of infant

development often focus on sensorimotor processes.

General Tests of Intelligence

Investigators have used either generalized tests of infant intelligence, such as the Bayley Scales of Infant Development Third Edition (Bayley III), or specific information processing tests. The Bayley III is a well-standardized assessment for infants aged 1–42 months that includes three scales administered by trained examiners: cognitive, language, and motor, and two scales completed by caregivers: socioemotional and adaptive behavior.[32] The Bayley III was designed to determine if infants are able to complete age-normed tasks, not to predict future functioning.[32] It has been used globally and has excellent psychometric properties, with internal consistencies measured by coefficient alphas averaging 0.86 and above, and test-retest reliabilities ranging from 0.72 to 0.81 across all ages. Correlations from infant assessments administered prior to 18–24 months of age to later intellectual performance vary and are often improved by models that include socioeconomic variables, such as maternal IQ or education.[33] The low correlations between scores from the Bayley III and specific information processing tests, such as visual habituation and attention to a novel stimulus, suggest that they are tapping specific areas, with the higher order functioning measured in the Bayley III incorporating motivation, attention, and multiple cognitive processes.[34] Beyond age 2, once infants gain symbol formation skills, predictability from infant assessments to subsequent intellectual performances increases.[33]

Information Processing

Several information processing tests administered during infancy (e.g., visual attention, learning, and memory) are predictive of school-age intellectual performance.[35] The paired comparison test has been used to assess short- and long-term visual recognition memory.[36] It includes two phases: familiarization and test. During the familiarization phase, two identical stimuli are presented and the infant has an opportunity to view them. During the test phase, a familiar stimulus is paired with a novel stimulus. The observer notes the proportion of time the infant gazes at each stimulus. Since infants are likely to spend more time gazing at the novel stimulus, a novelty score is calculated by dividing the amount of time the infant looked at the novel stimulus by the time the infant looked at both stimuli. The percentage of infant visual fixation to a novel stimulus from the Fagan test is a widely used indicator of infant visual recognition

memory.[37]

Other infant processing tests include focused attention (concentration devoted to processing information about an object),[38] cross-modal transfer (transfer of learning from one sensory modality to another),[39] and the A-not-B task (a Piagetian task that examines object permanence).[40] Although there has been limited use of infant information processing tests related to infants' nutritional status, they may be helpful in understanding links between nutrients and specific processes. As examples, undernourished infants (5–12 months of age) from India had difficulty with two tasks related to cognitive processing—visual recognition memory and cross-modal transfer,[39] the performance of infants from Chile on the Fagan Test varied by their iron status,[41] and there were no effects of iron-folic acid or zinc supplementation on either the Fagan test or the A-not-B test among infants in Nepal.[42] Most investigators recommend that tasks be selected to assess specific processes and that multiple measures be used, beyond IQ, to gain a comprehensive understanding of how nutritional deprivation and recovery impact processing skills.[33,36]

Measures of Social Emotional and Motor Development

The Ages and Stages Social Emotional questionnaire is frequently used to screen for social and emotional behavior. It is a validated caregiver-completed questionnaire organized into age-appropriate categories.

Undernutrition can have a negative impact on motor development, leading investigators to examine how nutritional interventions affect motor development, measured by standardized tests, such as the Bayley III or the Alberta Infant Motor Scale; maternal reports of the acquisition of motor milestones; or observation. Although motor development promotes independence and potentially advances cognitive development by enabling children to explore their environment, the implications of small differences in the timing of early motor development on intellectual functioning are not clear.

Measures of Brain Function in Children

In recent years, there has been an increased emphasis on the neurophysiological and neuropsychological development of children in response to environmental conditions. An increase in the sophistication of measures used to assess the structure and functioning of the brain allows for an improved understanding of the developmental effects of nutritional deficiencies. Optimal brain development during childhood relies on the

intake of specific nutrients such as fatty acids, proteins, and carbohydrates. Specifically, lipids and proteins are needed for adequate cell membrane formation, neurotransmitter release, and myelination, which enhance neuronal efficiency and processing speed throughout the central nervous system. Carbohydrates also appear to play a key role in the regulation of the hypothalamic- pituitary-adrenal (HPA) axis as it relates to food intake under stressful conditions.[43]

The ability to assess the brain functioning of children has improved substantially in the past decade. Electrophysiological techniques, event-related potentials (ERP), functional magnetic resonance imaging (fMRI), near-infrared spectroscopy (NIRS), diffusion tensor imaging, and magnetic resonance spectroscopy (MRS) may be effective methods for determining associations between nutrition and brain functioning. Each assessment method possesses strengths that make it amenable to particular research questions regarding neurodevelopment.[44,45]

Electrophysiological techniques involve the measurement of electric signals in the body and are generally superior to brain scanning techniques in terms of temporal resolution (when activity in the brain occurs in response to an event), but typically possess poor spatial resolution (where in the brain activity occurs). Since active neurons transmit electric signals through brain tissue, this activity can be recorded by electrodes placed on the scalp surface. Voltage distribution and synchronization are measured by numerous electrodes. Measurement of electrical activity is typically time-locked to a specific sensory or cognitive event and averaged over many trials to determine how the brain usually responds to a certain stimulus. ERP studies may be particularly useful in assessing infants and very young children because they do not depend on motor activity and children do not have to remain perfectly still as with most scanning techniques. As such, ERP recordings are an effective way to assess ongoing brain activity correlates in young children.[45]

Functional MRI is an alternative technique used to identify the location of neural activity. When a specific area of the brain is activated, blood flow to that area increases and oxygen is created. Because oxygenated and deoxygenated hemoglobin behave differently when pulsed with a magnetic field, this contrast in oxygen can be mapped onto consecutive “slices” of the brain. The MRI scanner determines the areas of greatest activation. The fMRI procedure has been used with older children and adults to study the location of neural activity. Using fMRI in combination with behavioral, cognitive, or neuropsychological assessments can provide detailed information about the activation of specific brain regions in

response to certain tasks. Limitations are that hemodynamic response is relatively slow (several seconds) in comparison to ERP techniques (milliseconds); fMRI scanning requires the child to be still, making it appropriate only for children who can be trained to remain immobile; and the cost of fMRI is high. NIRS provides noninvasive assessments of cortical functional connectivity by measuring changes in blood hemoglobin concentrations associated with neural activity. Although NIRS can be portable and used with infants because it does not require the subject to be still, it cannot measure deep structures.[44]

Diffusion tensor imaging (DTI) and MRS represent recent advances in neuroimaging that may be particularly relevant for research involving the neurodevelopmental effects of nutrition. DTI is a technique based on magnetic resonance imaging that produces functional images of the location, orientation, and activity of the brain's white matter tracts. DTI reveals the integrity of white matter tracts that link regions of the brain to each other by assessing water diffusion in the axons and their myelin sheaths.[45] Clinical research applications of DTI involve the localization of white matter lesions and the assessment of the white matter maturation in children.[46] For example, deficiencies in nutrients such as iron or essential fatty acids can interfere with myelination in preterm infants.[47] DTI technology is likely to be increasingly utilized to determine associations between nutritional intake and the efficiency of axonal connections throughout the brain.

MRS goes beyond providing information about tissue water content and provides data regarding the chemical nature of a brain region distinguishing the properties of one area relative to those of surrounding areas. MRS can assess the concentration of common metabolites and biological compounds giving important information about glial processes, membrane phospholipids, and high-energy phosphates.[48] Changes in these chemical compositions could be monitored following dietary supplementation throughout a child's postnatal development or after a period of nutritional deprivation.[44]

STRESS AND NEURODEVELOPMENTAL PROCESSES

Nutritional deprivation frequently occurs in the context of poverty, highlighting the need to consider how contextual variables, including environmental stress and caregiving behavior, relate to brain development.

Although brief periods of moderate stress allow children to develop appropriate stress responses,[49] exposure to high levels of stress (toxic stress) early in life during periods of rapid brain development can lead to numerous physical and developmental problems.[49]

The neurobiological mechanisms proposed to account for the cognitive deficits that occur in children with a history of undernutrition include deficits in the nutrients necessary for adequate physical growth and functioning as well as dysregulated HPA axis functioning.[50] The HPA system coordinates the stress response through multiple mechanisms resulting in the eventual release of glucocorticoids (cortisol) from the adrenal cortex. The brain is rich in receptors for cortisol with concentrations noted in the hippocampus and hypothalamus, prefrontal cortex, amygdala, and anterior cingulate gyrus. As excessive cortisol can impair brain regions housing cortisol receptors, intact HPA regulation is important for early development and ongoing maintenance of cortical systems involved with emotion, learning, memory, and attention and impulse control.[21,51,52]

In addition to an inadequate and/or inconsistent food supply, undernourished children often experience a lack of positive interactions with their primary caregivers. Effective caregivers play a critical role in helping children to modulate their emotional and physiological response to stressful events.[21] In the absence of positive interactions and stimulation, children may experience deficits in the development of their stress-sensitive regulatory systems. If they are also undernourished, they may be in a position of double jeopardy. An overexposure to stress in combination with disruptions in HPA axis functioning may be harmful to children's neurodevelopment. Heavily secreted neuroendocrine agents associated with chronic stress produce neurotoxic effects that may damage the developing brain, especially in areas rich in glucocorticoids such as the hippocampus, corpus callosum, and areas of the prefrontal cortex. Children who are repeatedly exposed to stress in the absence of supportive caregiving are at a higher risk for being chronically hyper-aroused to aspects of their environment, with possible negative effects on their attention and executive cognitive processes.[21,52]

In summary, a nonresponsive caregiving environment may undermine children's neural development.[53] The combination of nutritional and caregiving risk factors early in life may place undernourished children at risk for negative cognitive outcomes, possibly mediated by poorly modulated stress reactivity.

ENVIRONMENTAL AND CAREGIVING FACTORS RELATED TO NUTRITIONAL DEPRIVATION

Nutritional deficiencies often occur in a context of poverty among families who have few economic resources and limited education. Not only do many low-income families have restricted access to nutritionally healthy food, but mothers who are poorly educated are less likely to choose nutritionally appropriate foods for their children, compared with better educated mothers.[54] Thus, poverty and factors associated with poverty, such as low maternal education, may have a direct effect on children's development.

Caregivers contribute to their child's development through their care practices, such as providing food and basic care required by young children.[55] In the past, much attention regarding care practices was directed towards defining the quality and quantity of food that children require. However, evidence has demonstrated that care practices play a role in children's development that extends beyond the provision of food. Care practices include the relationships that caregivers form with their children during feeding and other daily activities.[24] Health care practices include behavior that is responsive to children's needs and is characterized by warmth and affection.

Care practices that influence children's nutritional status can be disrupted by conditions such as maternal mental health problems (e.g., symptoms of depression). For example, a recent meta-analysis found young children of caregivers with depressive symptoms were at significantly increased risk for stunting and underweight.[56] Maternal depression is a global public health concern, with potential risks for the health, nutritional status, and development of young children.[57]

BREASTFEEDING

The American Academy of Pediatrics and the WHO recommend exclusive breast milk for infants during the first 6 months and continued breastfeeding for at least the first year of life.[58,59] While the health and immunological benefits of breast milk for children are well established,[58,60] the effects of breast milk on children's development is less clear. Multiple methodological problems interfere with interpretation of studies that examine associations between breastfeeding and subsequent child development. Maternal factors associated with breastfeeding (e.g., intelligence, motivation, responsivity, and education) are also associated

with child development, the definition of breastfeeding varies across studies, and most experimental designs related to breastfeeding would be unethical. Thus, most breastfeeding studies cannot separate motivation and behavioral factors from the nutritional aspects of breast milk on infant growth and development.

A recent trial in Belarus involving over 13,000 infants overcame the ethical dilemma by randomizing breastfeeding promotion at the hospital level.[61] Infants in the experimental condition were exclusively breastfed longer and at age 6.5 years had higher cognitive scores based on standardized tests and higher academic performance based on teacher reports than children in the control condition. Further studies are needed to confirm and explain the relations between breastfeeding and brain development.

LONG-TERM CONSEQUENCES OF EARLY NUTRITIONAL DEFICIENCIES

Information on the long-term consequences of early nutritional deficiencies is available from several natural experiments, including nutritionally deprived children adopted into middle-class families or nutritional deficiencies occurring among children from middle-class families caused by disasters or chronic illness. In addition, information is available from follow-up studies of children from low-income countries who experienced moderate to severe nutritional deprivation or children from middle-income countries who experienced mild nutritional deprivation. However, data on the long-term consequences of early nutritional deficiencies can be difficult to interpret because children who experience nutritional deficiencies are often raised in low-income families where they also experience environmental deficiencies.

Natural Experiments

Female orphans from Korea who varied in their nutritional status, were adopted by middle-class American families, and tested during their school-age years. The children who had been undernourished early in life achieved scores in the normal range, but lower than those of better nourished Korean adoptees (Figure 24-4).[62] Although these findings demonstrate the impact of an enriched environment on children's intellectual skills, they also suggest that early nutritional deficiencies may limit children's ability to benefit from an enriched environment.

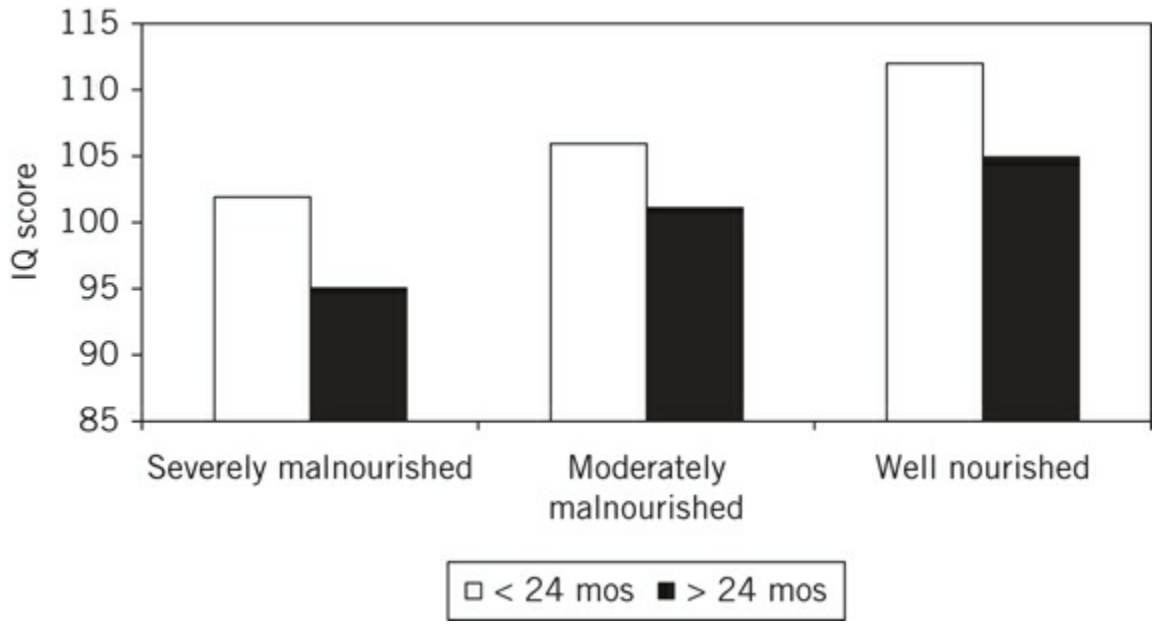


Figure 24-4 IQ scores among female Korean orphans varying by history of undernutrition and age of adoption. Adapted from Winick.[62]

The 1944 famine in Holland affected the entire population, regardless of economic standing. When the youth who experienced the famine were tested at 19 years of age as part of an army physical there was no long-term evidence of intellectual deficiencies, compared with scores from nonfamine years.[63] These findings suggest that environmental opportunities had overcome deficiencies associated with the early nutritional deficiencies.

However, recent evidence has shown that the children of mothers who suffered nutritional deprivation during the Dutch famine experienced reduced cognitive function and multiple health risks, including cardiovascular, vascular, and metabolic problems.[64] These findings suggest an epigenetic effect related to prenatal nutritional deprivation.

Children with chronic diseases, such as cystic fibrosis, are at risk for undernutrition due to malabsorption. Long-term consequences varied by family income. Among children from middle-income families, there were no differences in school-age intellectual scores, based on nutritional history.[65,66] In contrast, among children from low-income families, those with a history of nutritional deficiencies secondary to chronic disease had lower scores than those who had been hospitalized, but were well nourished. These studies illustrate the buffering effects of a nurturing

environment on the intellectual performance of nutritionally deprived children.

LONG-TERM CONSEQUENCES OF MILD NUTRITIONAL DEFICIENCIES

Findings on the long-term consequences of children from middle-income countries diagnosed with early growth faltering (failure to thrive) add to our information on nutritional deprivation and cognition. Although there are no agreed upon criteria for the diagnosis of failure to thrive, the term is generally used to describe children under age 3 years who have experienced a deceleration in growth over time, such that their weight-for-age or weight-for-height is below the fifth or third percentile.[67] Children with early growth faltering usually experience mild forms of deficiency, compared to children identified with nutritional deficiencies in low-income countries. During infancy and toddlerhood, the cognitive scores of children with growth faltering are lower than scores of comparison children with adequate growth, and often remain low through the preschool period.[68,69]

Follow-up studies of infants with growth faltering in infancy report that by age 13 their growth is within the normal range, but the rate of growth varies depending on the timing of the early weight faltering.[70] At school-age, children with a history of early growth faltering differ from children with a history of adequate weight gain by approximately 4 IQ points,[71–73] but in one of the only studies to follow children into adolescence, any educational differences could be explained by confounding.[74]

The relationship between nutritional status and cognitive functioning is often mediated by family, environmental, and cultural variables,[75] making the family an ideal context for prevention of the negative consequences of growth faltering. An intervention trial among children with early growth faltering found that after 1 year infants who received the home intervention had a more child-centered home environment, better cognitive scores, and better expressive language scores, compared to infants who did not receive the home intervention.[68] At age 8, beneficial intervention effects were present on measures of growth, parent-reported internalizing behavior, and teacher-reported work habits.[71] Findings suggest that the early home-visiting intervention attenuated some of the negative effects of early growth faltering, possibly by promoting maternal sensitivity and helping children build strong academic habits that enabled them to benefit

from school.

Intervention Studies

As illustrated by the transactional system,[23] optimal child development depends not only on food quality and availability, but also on the care the family provides to the child, especially during the first few years of life.[76] Evaluations of psychosocial stimulation delivered to undernourished children provide evidence for the importance of caregiving environment in promoting development.[1] In Jamaica, Grantham-McGregor and colleagues[77] demonstrated that stunted children who received either nutritional supplementation or psychosocial stimulation showed improved mental and motor development. The effects of the interventions were additive, children who received *both* supplementation and stimulation achieved mental and motor scores that approximated those of the nonstunted comparison group. Long-term follow-up demonstrated benefits that extended into early adult promoting competence and reductions in violent behavior.[78] These findings demonstrate that the benefits of environmental enrichment are sustainable over time. Recent evidence has shown the benefit of integrating nutrition and early psychosocial stimulation/learning opportunities into intervention trials.[79] In another example of an integrated nutrition and early psychosocial stimulation/learning intervention, zinc supplementation had beneficial effects on general infant development, but only in the context of psychosocial stimulation,[80] suggesting that the beneficial effects of the supplement may only be apparent in the context of a stimulating environment. Thus, more work is necessary to clarify the interactive role between nutrients and the caregiving environment.

MICRONUTRIENTS

Micronutrient deficiencies have been linked to delays in children's development, especially iodine, iron, and zinc deficiencies.[81] Micronutrients represent small quantities of essential vitamins and minerals that promote healthy growth and development. Since they are not produced by the body, they must be acquired either through the diet, fortification, or supplementation. The relationship between micronutrient deficiency and early cognitive development has captured global attention because micronutrients are related to specific physiological and neurological processes.[82]

Fatty Acids

The *n*-3 and *n*-6 polyunsaturated fatty acids (PUFAs) in the human brain are predominantly docosahexaenoic acid (DHA) and arachidonic acid (AA). Both DHA (*n*-3 PUFA) and AA (*n*-6 PUFA) accumulate in neural tissues during gestation and the first year of life. There is evidence from animal studies that DHA in essential to brain structure and function.[83,84] Studies in rats have shown that parental restriction results in reduced brain DHA concentration in the offspring, which is accompanied by poor cognitive and behavioral test performance.[84] The evidence from human studies on the role of DHA has focused largely on maternal depression and child development and behavioral disorders.[84–86] For example, an ecological analysis found an association between low seafood consumption, a key source of *n*-3 PUFAs, and the prevalence of postpartum depression.[85] In addition, maternal seafood intake during pregnancy of less than 340 g per week was associated with increased risk of their children being in the lowest quartile for verbal IQ, compared with mothers who consumed more than 340 g per week. Low maternal seafood intake was also associated with increased risk of poor outcomes for prosocial behavior, fine motor, communication, and social development scores.[86] Although the exact mechanism by which PUFAs, especially DHA, affect development and behavior remains unclear, it is thought that deficits in frontal cortex dopamine neurotransmission seen in patients with attention-deficit hyperactivity disorder (ADHD) may be associated with lower brain DHA concentrations.[84] Children with ADHD have also been shown to have lower plasma or red blood cell DHA concentrations.[87]

Fatty Acid Interventions

Because PUFAs are most actively accreted into the fetal brain during the last trimester of pregnancy, preterm infant are especially vulnerable for PUFA deficiency.[88] Many of the intervention studies have been conducted to investigate the effect of *n*-3 long-chain polyunsaturated fatty acid (LCPUFA) supplementation during the perinatal period on neurobehavioural development of young children.[89] Most of these trials have involved infants from high-income countries, and a significant proportion has investigated supplementation of infant formulas. Generally, supplementation of infant formula for preterm rather than term infants has demonstrated more consistent, positive effects on aspects of neurobehavioral development. The effect of maternal supplementation on global neurobehavioral outcomes for children born at term remains

unclear, although *n*-3 LCPUFA supplementation of women expressing milk for their preterm infants does improve their performance on tests of global neurodevelopment. Further work is required to determine whether dietary *n*-3 LCPUFA is neuroprotective for term infants and for children from disadvantaged or low-income backgrounds.

Iodine Deficiency

Iodine deficiency is a major problem that affects children in areas where iodine is depleted from the soil, primarily mountainous regions, such as the Himalayas and the Andes, and flood plains. Iodine is an essential component of at least two thyroid hormones that are necessary for skeletal growth and neurological development.[90] When iodine is deficient, hypothyroidism occurs, resulting in increased production of thyrotropin, which stimulates production of thyroid hormones. The most common manifestation of iodine deficiency is goiter, which is an enlarged thyroid gland.

When iodine deficiency occurs in utero, it leads to fetal hypothyroidism and irreversible neurological and cognitive deficits, manifested as cretinism. Neurological cretinism includes mental retardation, primitive reflexes, visual problems, facial deformities, stunted growth, and diplegia.[91] When iodine deficiency occurs postnatally, the child may experience thyroid failure that can lead to hypothyroidism. Observational studies that have compared children with and without a goiter have had mixed results; some have reported cognitive deficits among children with goiters and others have not. One explanation for the lack of clarity may be that differing levels of hypothyroidism can lead to a goiter. In addition, families who live in iodine- deficient areas are often more impoverished than families in areas where iodine is adequate.

Iodine Interventions

Several randomized trials have been conducted to examine the impact of iodine supplementation on the cognitive performance of children in iodine-deficient areas. However, results have not been consistent. In a longitudinal follow-up of school-age children, all of whom received iodine, those who received iodine in utero prior to the third trimester had better scores on a measure of psychomotor performance than children who received iodine later in pregnancy or at age 2 years.[92] There was a similar trend when measures of cognitive performance were considered; however, the differences did not reach significance. Thus, the effects of postnatal

iodine deficiency on children's cognitive performance are less clear than the effects of prenatal iodine deficiency.

There has been significant progress in global programs to reduce iodine deficiency, including a decrease in the number of iodine deficient countries (54–30) and an increase in the number of iodine sufficient countries (67–112). However, approximately 30% of households worldwide do not have adequate access to iodized salt, making iodine deficiency and associations with cognitive deficits an ongoing concern.[93]

Iron Deficiency

Iron deficiency (ID) is the most common nutritional deficiency in the world. UNICEF and WHO committees estimate that worldwide there are 2 billion individuals with anemia and up to 5 billion who are iron deficient. The highest risk of ID occurs during times of rapid growth and nutritional demand, during infancy (6–24 months of age), early childhood, adolescence, and pregnancy.[94]

Iron is necessary for hemoglobin synthesis. ID leads to reduced oxygen carrying capacity and can impact immunity, growth, and development. Although iron status is often assessed by anemia (low blood hemoglobin concentration), only 50% of anemia is caused by ID.[92] Iron has multiple roles in neurotransmitter systems and may affect behavior through its effects on dopamine metabolism.[95] This is highly relevant to children's cognitive development as dopamine clearance has strong effects on attention, perception, memory, motivation, and motor control.[96]

Approximately 47% of the world's children under age 3 have anemia,[94] with higher rates in low-income countries. ID is the most common cause of anemia and has been associated with multiple adverse health conditions, including low scores on tests of mental performance and were fearful, inattentive, and solemn with low levels of initiation and exploration, and many continued to demonstrate lower academic performance during their school-age years, even when the anemia had been treated.[97] For example, a follow-up of Costa Rican adolescents who had been born at term and were free of health problems in infancy other than ID, showed persisting motor differences, more grade repetition, increased anxiety/depression, social problems and inattention rated by parents and teachers, and a widening gap in cognitive scores to 19 years, despite iron therapy that corrected Iron-deficiency anemia (IDA) in infancy.[98]

Iron Interventions

Prevention of iron deficiency is a primary public health goal in many counties. However, evidence from prevention trials has been mixed. Several recent reviews of randomized controlled trials of iron supplementation provide evidence for improved motor and social-emotional development in infants who received iron.[99–101] However, other reviews have suggested that although supplementation trials reduce anemia, the effects on development are inconsistent and uncertain.[97] The variability in the quality of the trials, including the timing, dose, and duration of the supplement, make comparisons across studies.

Zinc Deficiency

Zinc is a trace mineral that is involved with DNA and RNA synthesis and the metabolism of proteins, carbohydrates, and fat. There have been at least 15 trials of the association between zinc supplementation and cognitive performance among infants and young children. Although zinc is essential for neurological functioning and observational studies have shown associations between low zinc concentration and negative performance and mental health status, randomized trials of zinc alone or in combination with other micronutrients have yielded inconsistent results linking supplementation with children's early development.[102] Many factors could interfere with clear interpretation of the zinc supplementation trials. Zinc deficiency is difficult to document, interfering with the ability to ensure that participants are zinc deficient. Micronutrient deficiencies often co-occur, thus providing zinc alone or with iron may not alleviate other nutritional problems. There may be interactive effects between zinc and iron, resulting in adverse effects. Nutritionally deficient infants, who are smaller, less active, and less competent than better nourished infants, may be less able to elicit age-appropriate expectations from their caregivers. It is also possible that the intervention trials were too brief or the dose of zinc supplement was insufficient. Finally, the low-income context surrounding zinc deficiency may interfere with children's developmental progress, even in the context of zinc supplementation.

Multiple Micronutrients

Infants in low-income populations are at risk for multiple micronutrient deficiencies due to inadequate dietary intakes, poor bioavailability in foods, and delayed introduction of complementary foods. Thus supplementation of multiple micronutrients from 6 months through early

childhood has been proposed as a strategy to prevent multiple deficiencies and promote brain development. While several studies have assessed the effects of iron and zinc supplementation on infant development, findings have been inconsistent. In Bangladesh beneficial effects of the two micronutrients were reported on infants' motor development and behavior.[103] However, in Indonesia, infants showed benefits on psychomotor development from iron but not zinc or iron plus zinc supplementation.[104] In Zanzibar, infants aged 5–11 months given either iron plus folic acid or iron plus folic acid plus zinc walked unassisted sooner than did those who received zinc alone or a placebo.[105] Conversely, iron plus folic acid and zinc supplementation, alone or in combination, did not affect the age at first walking unassisted in Nepal.[106] Overall, these studies suggest a possible benefit of iron supplementation, but are inconsistent as to the effects of multiple micronutrients on infant development.

The long-term effects of multiple micronutrient supplementation on child development remains unclear. While one study reported that intellectual function of school-aged Nepalese children was positively associated with prenatal iron plus folic acid supplementation,[107] at least three other longitudinal randomized controlled intervention trials have reported no long-term developmental benefits of multiple micronutrient supplementation during infancy, toddlerhood, or the preschool period. In rural Nepal, zinc and iron plus folic acid supplementation during infancy (4–17 months of age) did not improve motor and language milestone scores of toddlers,[108] and iron plus folic acid during the preschool years (12–35 months of age) had no effect on aspects of intellectual, executive, and motor function at 7 to 9 years of age.[109] In Thailand, supplementation with iron or zinc or both during infancy did not lead to long-term cognitive improvement in 9-year-old children.[110] There is little evidence to suggest routine prophylactic multiple micronutrient supplementation of children in low-income populations at high risk for deficiencies improves developmental outcomes. Further studies are needed to evaluate the effect of timing, type, and duration of single and multiple micronutrient supplementation on developmental outcomes.

NEUROTOXINS

Lead Exposure

Numerous studies have documented that lead exposure has a neurotoxic

effect on infants and young children and is associated with impairments in intellectual and behavioral functioning.[111,112] Although significant progress has been made in reducing the average blood lead level in many parts of the world, lead remains a major cause of encephalopathy in children. Even in the absence of encephalopathy, lead poisoning contributes to permanent neuropsychological impairments including deficits in general intellectual functioning, ability to sustain attention on tasks, aspects of speech production and language comprehension, motor coordination, flexible problem solving, and behavioral self-regulation. Several recent reviews that focused on the impact of low levels of lead exposure (blood lead levels < 10 μg/dL) among children indicated that even low-level lead exposure can lead to a decline in IQ scores.[113,114] An international pooled analysis found a dose–response effect associated with the amount of lead found in children's blood.[114] An increase in blood lead levels from 2.4 μg/dL to 10 μg/dL resulted in a 1.1 point decline in IQ, 10 μg/dL to 20 μg/dL resulted in an additional 1.9 point decline, and for blood lead concentrations from 20 μg/dL to 30 μg/dL there was an additional 3.9 point decline in IQ scores (a total decrement of 6.9 IQ points for the 20–30 μg/dL group).

In general, the long-term effects of prenatal or postnatal lead exposure appear to be nonspecific, impacting multiple areas of the brain and making it difficult to pinpoint the specific neurodevelopmental mechanisms responsible for cognitive and behavioral changes in lead-exposed children. However, there is evidence that lead alters neuropsychological development by disrupting processes of neuronal migration, synaptogenesis, and myelination.[115] Many of the neurotoxic effects of lead can be traced to lead's ability to mimic or inhibit the action of calcium as a regulator of cell function. As such, exposure to lead causes changes in how neurotransmitters are stored and released, which may cause an inhibition of synaptic growth and detract from the efficiency of the developing brain. Lead also disrupts the main structural components of the blood–brain barrier by damaging astrocytes and the endothelial microvasculature. This damage allows for the passage of certain molecules (e.g., albumin) that are toxic and typically restricted by the blood–brain barrier.[116] Other researchers note that lead exposure alters second and third messenger activity,[117] which also interferes with cognitive efficiency.

Children are typically more vulnerable to lead exposure than adults because they are more likely to ingest lead, resulting in severe consequences. Hand to mouth activity and behaviors such as pica (which

peaks at around 2 years of age) increase the likelihood of ingesting lead-contaminated substances.[113] The most common ways that children are exposed to lead are through the consumption of soil, lead-based paint, water from pipes with lead-soldered joints, and contaminants brought home from parents engaged in mining/industrial activities. Once ingested, lead may have a more deleterious effect on the developing central nervous system of children than on adults. Children's increased vulnerability to lead is attributable to several factors. First, a greater proportion of ingested lead is absorbed from the gastrointestinal tract of children than of adults. Second, a greater proportion of circulating lead crosses the blood–brain barrier in children, especially children under the age of five, than in adults.[115] Finally, the developing brain seems to be more vulnerable to the neurotoxic effects of lead than the mature brain. The neurotoxic effects of lead are particularly evident in the prefrontal cortex, the cerebellum, and the hippocampus.[117] Investigators speculate that because regions such as the prefrontal cortex are in a phase of rapid development during the first few years of life, they may be particularly susceptible to the harmful effects of neurotoxins. The vulnerability of the prefrontal cortex could also explain the consistent associations between elevated blood lead levels and poor performance on tasks involving sustained attention and other executive functioning.

SUMMARY

Most of the research examining the impact of nutritional deficiencies on children's intellectual development has hypothesized a direct effect, possibly through changes in neuroanatomy or neurotransmission. However, evidence from both animal and human studies has shown that children's development is influenced by environmental factors, especially the quality of interactions with primary caregivers. Thus, behavioral changes associated with nutritional deficiencies may alter the caregiving that the child receives, thereby compromising the child's development even further. The result could be a child who experiences both the neurochemical changes that have been associated with nutritional deficiency, together with limited environmental enrichment. Initial findings from integrated trials that include both nutritional supplementation and early learning/stimulation opportunities have been promising. Future research should consider whether the caregiving system mediates the effects of nutritional deficiencies on child development, and how integrated interventions that include nutrition and early

learning/stimulation relate to child development.

Observational studies have consistently shown associations between single nutrient deficiencies and children's intellectual development. Yet, evidence from intervention trials of single nutrients has been inconsistent. In many low and middle-income countries, children experience deficiencies in multiple nutrients, often in the context of poverty. Trials are needed that correct both macro and micronutrient deficiencies, and provide opportunities for responsive learning/stimulation often lacking in low-income contexts.

The findings linking nutritional deficiencies and child development point to the importance of effective prevention programs that begin prenatally or early in life and extend through the periods of vulnerability, which may include adolescence. There are many unanswered questions regarding nutritional deficiencies and child development that require further research to guide prevention, including the timing of interventions to prevent deficiencies, the specific processes involved in effective prevention, and the long-term benefits on academic achievement and human capital.

REFERENCES

1. Engle PL, Black MM, Behrman JR, et al. Strategies to avoid the loss of developmental potential in more than 200 million children in the developing world. *Lancet.* 2007;369(9557):229–242.
2. Engle PL, Fernald LC, Alderman H, et al. Strategies for reducing inequalities and improving developmental outcomes for young children in low- income and middle-income countries. *Lancet*. 2011;378(9799):1339–1353.
3. Grantham-McGregor S, Cheung Y, Cueto S, et al. Over two hundred million children fail to reach their developmental potential in the first five years in developing countries. *Lancet*. 2007;369:60–70.
4. Walker SP, Wachs TD, Gardner JM, et al. Child development: risk factors for adverse outcomes in developing countries. *Lancet*. 2007;369(9556):145–157.
5. Walker SP, Wachs TD, Grantham-McGregor S, et al. Inequality in early childhood: risk and protective factors for early child development. *Lancet*. 2011;378(9799):1325–1338.
6. Grantham-McGregor S, Walker S, Powell C. Nutritional supplementation and mental development. *Lancet*. 1991;338(8769):758.
7. Dobbing J, ed Vulnerable periods of brain development. Lipids, Malnutrition & the Developing Brain. Amsterdam, North Holland1972. Knight E, ed.
8. WHO Multicentre Growth Reference Study Group. WHO Child Growth Standards based on length/height, weight and age. *Acta Paediatr*. 2006;95(suppl 450):76–85.
9. Victora CG, de Onis M, Hallal PC, et al. Worldwide timing of growth faltering: revisiting implications for interventions. *Pediatrics*. 2010;125(3):e473–480.
10. Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and

overweight in low-income and middle-income countries. *Lancet*. 2013;382(9890):427–451.
11. Smart J. *Malnutrition, Learning, and Behavior: Recent Advances in Laboratory Animal Research*. Washington, DC: Pan American Health Organization;1998.
12. Tonkiss J, Smart JL, Massey RF. Effects of early life undernutrition in artificially-reared rats. 2. Subsequent behaviour. *Physiol Behav*. 1987;41(6):555–562.
13. Levitsky DA, Barnes RH. Nutritional and environmental interactions in the behavioral development of the rat: long-term effects. *Science*. 1972;176(30):68–71.
14. Levitsky DA, Strupp BJ. Malnutrition and the brain: changing concepts, changing concerns. *J Nutr*. 1995;125(suppl 8):2212S–2220S.
15. Fox SE, Levitt P, Nelson CA, 3rd. How the timing and quality of early experiences influence the development of brain architecture. *Child Dev*. 2010;81(1):28–40.
16. Thompson RA, Nelson CA. Developmental science and the media. Early brain development. *Am Psychol*. 2001;1:5–15.
17. Nyaradi A, Li J, Hickling S, et al. The role of nutrition in children's neurocognitive development, from pregnancy through childhood. *Front Hum Neurosci*. 2013;7:97.
18. Shonkoff JP, Phillips DA. *From Neurons to Neighborhoods: The Science of Early Childhood Development*. Washington, DC: National Academy Press; 2000.
19. Wachs T, Georgieff M, Cusick S, McEwen B. Issues in the timing of integrated early interventions: contributions from nutrition, neuroscience and psychological research. *Ann N Y Acad Sci*. 2014;1308(1):89–106.
20. Wachs TD. *Necessary But Not Sufficient. The Respective Roles of Single and Multiple Influences on Individual Development*. Washington, DC: American Psychological Association; 2000.
21. Johnson SB, Riley AW, Granger DA, Riis J. The science of early life toxic stress for pediatric practice and advocacy. *Pediatrics*. 2013;131(2):319–327.
22. Bronfenbrenner U, Ceci SJ. Nature-nurture reconceptualized in developmental perspective: a bioecological model. *Psychol Rev*. 1994;101(4):568–586.
23. Sameroff A. *Transactional Model of Development*. Washington, DC: American Psychological Association; 2009.
24. Black M, Aboud F. Responsive feeding is embedded in a theoretical framework of responsive parenting. *J Nutr*. 2011;141(3):490–494.
25. Black M, Hurley K. Helping children develop healthy eating habits. In: Tremblay R, Barr RG, Peters, R, Bolvin, M, eds. *Encyclopedia on Early Childhood Development*. 2nd ed. Montreal, Quebec: Centre of Excellence for Child Development; 2013.
26. Hack M, Taylor HG. Perinatal brain injury in preterm infants and later neurobehavioral function. *JAMA*. 2000;284(15):1973–1974.
27. Graves PL. Nutrition, infant behavior, and maternal characteristics: a pilot study in West Bengal, India. *Am J Clin Nutr*. 1976;29(3):305–319.
28. Meeks Gardner J, Grantham-McGregor SM, Chang SM, Himes JH, Powell CA. Activity and behavioral development in stunted and nonstunted children and response to nutritional supplementation. *Child Dev*. 1995;66(6):1785–1797.
29. Valenzuela M. Maternal sensitivity in a developing society: the context of urban poverty and infant chronic undernutrition. *Dev Psychol*. 1997;33(5):845–855.
30. Cravioto J, DeLicardie E. Mother–infant relationship prior to the development of clinically severe malnutrition in the child. In: White P, Selve N, eds. *Proceedings of the Western Hemisphere Nutrition Congress IV*. Boston: Massachusetts

Publishing Sciences Group; 1975. pp. 126–137.
31. McCall R. Nature-nurture and the two realms of development: a proposed integration with respect to mental development. *Child Dev* 1981;52:1–12.
32. Bayley N. *Bayley Scales of Infant and Toddler Development*. 3rd ed. San Antonio, TX: Harcourt Assessment, Inc.; 2006.
33. McCall R, Mash C. Long chain polyunsaturated fatty acids and the measurement and prediction of intelligence. In: Dobbing J, ed. *Developing Brain and Behaviour: The Role of Lipids in Infant Formula*. New York: Academic Press; 1997: pp. 295–338.
34. Lewis M, Brooks-Gunn J. Visual attention at three months as a predictor of cognitive functioning at two years of age. *Intelligence*. 1981;5:131–140.
35. Bornstein MH, Sigman MD. Continuity in mental development from infancy. *Child Dev*. 1986;57(2):251–274.
36. Columbo J. Individual differences in infant cognition: Methods, measures, and models. In: Dobbing J, ed. *Developing Brain and Behaviour: The Role of Lipids in Infant Formula*. New York: Academic Press; 1997:339–385.
37. Singer L. Methodological considerations in longitudinal studies of infant risk. In: Dobbing J, editor. *Developing Brain and Behaviour: The Role of Lipids in Infant Formula*. New York: Academic Press; 1997:209–252.
38. Lawson K, Ruff H. Focused attention: Assessing a fundamental cognitive process in infancy. In: Singer L, Zeskind P, eds. *Biobehavioral Assessment of the Infant*. New York: Guilford Press; 2001:293–311.
39. Rose S, Orlin E. Visual information processing. In: Singer L, Zeskind P, editors. *Biobehavioral Assessment of the Infant*. New York: Guilford Press; 2001:274–292.
40. Noland J. The A-not-B task. In: Singer L, Zeskind P, eds. *Biobehavioral Assessment of the Infant*. New York: Guilford Press; 2001:312–322.
41. Lozoff B, De Andraca I, Castillo M, et al. Behavioral and developmental effects of preventing iron-deficiency anemia in healthy full-term infants. *Pediatrics*. 2003;112(4):846–854.
42. Siegel EH, Kordas K, Stoltzfus RJ, et al. Inconsistent effects of iron-folic acid and/or zinc supplementation on the cognitive development of infants. *J Health Popul Nutr*. 2011;29(6):593–604.
43. Walker CD. Nutritional aspects modulating brain development and the responses to stress in early neonatal life. *Prog Neuropsychopharmacol Biol Psychiatry*. 2005;29(8):1249–1263.
44. Lloyd-Fox S, Papademetriou M, Darboe MK, et al. Functional near infrared spectroscopy (fNIRS) to assess cognitive function in infants in rural Africa. *Sci Rep*. 2014;4:4740.
45. Sizonenko SV, Babiloni C, de Bruin EA, et al. Brain imaging and human nutrition: which measures to use in intervention studies? *British J Nutr*, 2013;110:S1–S30.
46. Schmithorst VJ, Wilke M, Dardzinski BJ, Holland SK. Correlation of white matter diffusivity and anisotropy with age during childhood and adolescence: a cross-sectional diffusion-tensor MR imaging study. *Radiology*. 2002;222(1):212–218.
47. Georgieff MK, Innis SM. Controversial nutrients that potentially affect preterm neurodevelopment: essential fatty acids and iron. *Pediatr Res*. 2005;57:99–103.
48. Urenjak J, Williams SR, Gadian DG, Noble M. Proton nuclear magnetic resonance spectroscopy unambiguously identifies different neural cell types. *J Neurosci*. 1993;13(3):981–989.
49. Shonkoff JP. Building a new biodevelopmental framework to guide the future of

early childhood policy. *Child Dev*. 2010;81(1):357–367.
50. Fernald LC, Grantham-McGregor SM, Manandhar DS, Costello A. Salivary cortisol and heart rate in stunted and nonstunted Nepalese school children. *Eur J Clin Nutr*. 2003;57(11):1458–1465.
51. Bremner J, Vermetten E. Stress and development: behavioral and biological consequences. *Dev Psychopathol*. 2001;13(3):473–489.
52. Hostinar CE, Sullivan RM, Gunnar MR. Psychobiological mechanisms underlying the social buffering of the hypothalamic-pituitary-adrenocortical axis: a review of animal models and human studies across development. *Psychol Bull*. 2014;140(1):256–82.
53. Pollak S, Cicchetti D, Klorman R. Stress, memory, and emotion: developmental considerations from the study of child maltreatment. *Dev Psychopathol*. 1998 Fall;10(4):811–828.
54. Wachs T, McCabe G. Relation of maternal intelligence and schooling to offspring nutritional intake. *Intl J Behav Dev*. 2001;25:444–449.
55. Engle PL, Bentley M, Pelto G. The role of care in nutrition programmes: current research and a research agenda. *Proc Nutr Soc*. 2000;59(1):25–35.
56. Surkan PJ, Kennedy CE, Hurley KM, Black MM. Maternal depression and early childhood growth in developing countries: systematic review and meta-analysis. *Bull World Health Organ*. 2011;89(8):608–615.
57. Wachs TD. Models linking nutritional deficiencies to maternal and child mental health. *Am J Clin Nutr*. 2009;89(3):935S–939S.
58. American Academy of Pediatrics Work Group on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2005;115:496–506.
59. WHO. The *Optimal Duration of Exclusive Breastfeeding*: *Report of an Expert Consultation*. Geneva, Switzerland: World Health Organization; 2001.
60. James DC, Dobson B. Position of the American Dietetic Association: Promoting and supporting breastfeeding. *J Am Diet Assoc*. 2005;105(5): 810–818.
61. Kramer MS, Aboud F, Mironova E, et al. Breastfeeding and child cognitive development: new evidence from a large randomized trial. *Arch Gen Psychiatry*. 2008;65(5):578–584.
62. Winick M, Meyer KK, Harris RC. Malnutrition and environmental enrichment by early adoption. *Science*. 1975;190(4220):1173–1175.
63. Stein Z, Susser M, Saenger G, Marolla F. Nutrition and mental performance. *Science*. 1972;178(4062):708–713.
64. de Rooij S, Wouters H, Yonker J, et al. Prenatal undernutrition and cognitive function in late adulthood. *Proc Natl Acad Sci USA*. 2010;107(39):16881–16886.
65. Beardslee WR, Wolff PH, Hurwitz I, et al. The effects of infantile malnutrition on behavioral development: a follow-up study. The *Am J Clin Nutr*.1982;35(6):1437–1441.
66. Lloyd-Still JD, Hurwitz I, Wolff PH, Shwachman H. Intellectual development after severe malnutrition in infancy. *Pediatrics*. 1974;54(3):306–311.
67. Wright CM. Identification and management of failure to thrive: a community perspective. *Arch Dis Child*. 2000;82(1):5–9.
68. Black MM, Dubowitz H, Hutcheson J, et al. A randomized clinical trial of home intervention for children with failure to thrive. *Pediatrics*. 1995;95(6):807–814.
69. Singer LT, Fagan JF, 3rd. Cognitive development in the failure-to-thrive infant: a three-year longitudinal study. *J Pediatr Psychol*. 1984;9(3):363–383.
70. ud Din Z, Emmett P, Steer C, Emond A. Growth outcomes of weight faltering in infancy in ALSPAC. *Pediatrics*. 2013;131(3):e843–849.
71. Black MM, Dubowitz H, Krishnakumar A, Starr RH, Jr. Early intervention and

recovery among children with Failure to Thrive: follow-up at age 8. *Pediatrics*. 2007;120(1):59–69.
72. Boddy J, Skuse D, Andrews B. The developmental sequelae of nonorganic failure to thrive. *J Child Psychol Psychiatry*. 2000;41(8):1003–1014.
73. Drewett RF, Corbett SS, Wright CM. Cognitive and educational attainments at school age of children who failed to thrive in infancy: a population-based study. *J Child Psychol Psychiatry*. 1999;40(4):551–561.
74. Holme AR, Blair PS, Emond AM. Psychosocial and educational outcomes of weight faltering in infancy in ALSPAC. *BMJ Open*. 2013;3(7).
75. Frank D, Drotar D, Cook J, Bleiker J, Kasper D. Failure to thrive. In: Ludwig RMRS, ed. *Child Abuse: Medical Diagnosis and Management*. 2nd ed. Philadelphia, PA: Lea & Feiber; 2001:307–338.
76. Shonkoff JP, Boyce WT, McEwen BS. Neuroscience, molecular biology, and the childhood roots of health disparities: building a new framework for health promotion and disease prevention. *JAMA*. 2009;301(21):2252–2259.
77. Grantham-McGregor S, Stewart M, Powell C. Behaviour of severely malnourished children in a Jamaican hospital. *Dev Med Child Neurol*. 1991;33(8):706–714.
78. Walker SP, Chang SM, Vera-Hernandez M, Grantham-McGregor S. Early childhood stimulation benefits adult competence and reduces violent behavior. *Pediatrics*. 2011;127(5):849–857.
79. Grantham-McGregor S, Fernald L, Kagawa R, Walker S. Effects of integrated child development, nutrition and health interventions on child development and nutritional status. *Ann N Y Acad Sci*. 2013:1309(1);11–32.
80. Gardner JM, Powell CA, Baker-Henningham H, Walker SP, Cole TJ, Grantham-McGregor SM. Zinc supplementation and psychosocial stimulation: effects on the development of undernourished Jamaican children. *Am J Clin Nutr* 2005;82(2):399–405.
81. Ramakrishnan U, Goldenberg T, Allen LH. Do multiple micronutrient interventions improve child health, growth, and development? *J Nutr*. 2011;141(11):2066–2075.
82. Grantham-McGregor SM, Ani CC. The role of micronutrients in psychomotor and cognitive development. *Br Med Bull*. 1999;55(3):511–527.
83. McCann JC, Ames BN. Is docosahexaenoic acid, an n-3 long-chain polyunsaturated fatty acid, required for development of normal brain function? An overview of evidence from cognitive and behavioral tests in humans and animals. *Am J Clin Nutr*. 2005;82(2):281–295.
84. McNamara RK, Carlson SE. Role of omega-3 fatty acids in brain development and function: potential implications for the pathogenesis and prevention of psychopathology. *Prostaglandins Leukot Essent Fatty Acids*. 2006;75(4–5):329–349.
85. Hibbeln JR. Seafood consumption, the DHA content of mothers' milk and prevalence rates of postpartum depression: a cross-national, ecological analysis. *J Affect Disord*. 2002;69(1–3):15–29.
86. Hibbeln JR, Davis JM, Steer C, et al. Maternal seafood consumption in pregnancy and neurodevelopmental outcomes in childhood (ALSPAC study): an observational cohort study. *Lancet*. 2007;369(9561):578–585.
87. Antalis CJ, Stevens LJ, Campbell M, et al. Omega-3 fatty acid status in attention-deficit/hyperactivity disorder. *Prostaglandins Leukot Essent Fatty Acids*. 2006;75(4–5):299–308.
88. Clandinin MT, Chappell JE, Leong S, et al. Intrauterine fatty acid accretion rates

in human brain: implications for fatty acid requirements. *Early Hum Dev*. 1980;4(2):121–129.
89. Makrides M, Collins CT, Gibson RA. Impact of fatty acid status on growth and neurobehavioural development in humans. *Matern Child Nutr*. 2011;7 (suppl 2):80–88.
90. Delange F, de Benoist B, Pretell E, Dunn J. Iodine deficiency in the world: where do we stand at the turn of the century? *Thyroid*. 2001;11(5):437–447.
91. Halpern J. The neuromotor deficit in endemic cretinism and its implications for the pathogenesis of the disorder. In: Stanbury J, ed. *The Damaged Brain of Iodine Deficiency*. New York: Cognizant Communication; 1994:15–24.
92. Yip R. Iron deficiency: contemporary scientific issues and international programmatic approaches. *J Nutr*. 1994;124(Suppl 8):1479S–1490S.
93. Zimmermann M. Iodine Deficiency and Excess in children: worldwide status in 2013. *Endocr Pract*. 2013;19(5):839–846.
94. McLean E, Cogswell M, Egli I, et al. Worldwide prevalence of anaemia, WHO Vitamin and Mineral Nutrition Information System, 1993–2005. *Public Health Nutr*. 2009;12(4):444–454.
95. Beard JL. Iron biology in immune function, muscle metabolism and neuronal functioning. *J Nutr*. 2001;131(2S-2):568S–579S; discussion 580S.
96. Lozoff B. Early iron deficiency has brain and behavior effects consistent with dopaminergic dysfunction. *J Nutr*. 2011;141(4):740S–746S.
97. Pasricha S, Hayes E, Kalumba K, Biggs B. Effect of daily iron supplementation on health in children aged 4–23 months: a systematic review and meta-analysis of randomised controlled trials. *Lancet*. 2013;1:77–86.
98. Lozoff B, Jimenez E, Smith JB. Double burden of iron deficiency in infancy and low socioeconomic status: a longitudinal analysis of cognitive test scores to age 19 years. *Arch Pediatr Adolesc Med*. 2006;160(11):1108–1113.
99. Falkingham M, Abdelhamid A, Curtis P, et al. The effects of oral iron supplementation on cognition in older children and adults: a systematic review and meta-analysis. *Nutr J*. 2010;9:4.
100. Hermoso M, Vucic V, Vollhardt C, et al. The effect of iron on cognitive development and function in infants, children and adolescents: a systematic review. *Ann Nutr Metab*. 2011;59(2–4):154–165.
101. Iannotti LL, Tielsch JM, Black MM, Black RE. Iron supplementation in early childhood: health benefits and risks. *Am J Clin Nutr*. 2006;84(6):1261–1276.
102. Black M. Zinc deficiency and cognitive development. In: Benton D, ed. *Lifetime Nutritional Influences on Cognition, Behaviour and Psychiatric Illness*. Cambridge, UK: Woodhead Publishing Limited; 2011:79–93.
103. Black MM, Baqui AH, Zaman K, et al. Iron and zinc supplementation promote motor development and exploratory behavior among Bangladeshi infants. *Am J Clin Nutr*. 2004;80(4):903–910.
104. Lind T, Lonnerdal B, Stenlund H, et al. A community-based randomized controlled trial of iron and zinc supplementation in Indonesian infants: effects on growth and development. *American J Clinl Nutr*. 2004;80(3):729–736.
105. Olney DK, Pollitt E, Kariger PK, et al. Combined iron and folic acid supplementation with or without zinc reduces time to walking unassisted among Zanzibari infants 5- to 11-mo old. *J Nutr*. 2006; 136(9):2427–2434.
106. Katz J, Khatry SK, Leclerq SC, et al. Daily supplementation with iron plus folic acid, zinc, and their combination is not associated with younger age at first walking unassisted in malnourished preschool children from a deficient population in rural Nepal. *J Nutr*. 2010;140(7):1317–1321.

107. Christian P, Murray-Kolb LE, Khatry SK, et al. Prenatal micronutrient supplementation and intellectual and motor function in early school-aged children in Nepal. *JAMA*. 2010;304(24):2716–2723.
108. Surkan PJ, Siegel EH, Patel SA, et al. Effects of zinc and iron supplementation fail to improve motor and language milestone scores of infants and toddlers. *Nutrition*. 2013;29(3):542–548.
109. Murray-Kolb LE, Khatry SK, Katz J, et al. Preschool micronutrient supplementation effects on intellectual and motor function in school-aged Nepalese children. *Arch Pediatr Adolesc Med*. 2012;166(5):404–410.
110. Pongcharoen T, DiGirolamo A, Ramakrishnan U, et al. Long-term effects of iron and zinc supplementation during infancy on cognitive function at 9 years of age in northeast Thai children: a follow-up study. *Am J Clin Nutr*. 2011;93(3):636–643.
111. Canfield RL, Gendle MH, Cory-Slechta DA. Impaired neuropsychological functioning in lead-exposed children. *Dev Neuropsychol*. 2004;26(1):513–540.
112. Needleman HL, Riess JA, Tobin MJ, et al. Bone lead levels and delinquent behavior. *JAMA*. 1996;275(5):363–369.
113. Koller K, Brown T, Spurgeon A, Levy L. Recent developments in low-level lead exposure and intellectual impairment in children. *Environ Health Perspect*. 2004;112(9):987–994.
114. Lanphear BP, Hornung R, Khoury J, et al. Low-level environmental lead exposure and children's intellectual function: an international pooled analysis. *Environ Health Perspect*. 2005;113(7):894–899.
115. Lidsky TI, Schneider JS. Lead neurotoxicity in children: basic mechanisms and clinical correlates. *Brain*. 2003;126(Pt 1):5–19.
116. Bressler JP, Goldstein GW. Mechanisms of lead neurotoxicity. *Biochem Pharmacol*. 1991;41(4):479–484.
117. Finkelstein Y, Markowitz ME, Rosen JF. Low-level lead-induced neurotoxicity in children: an update on central nervous system effects. *Brain Res Brain Res Rev*. 1998;27(2):168–176.

Maternal Nutrition and Pregnancy Outcome

25

Theresa O. Scholl, PhD, MPH and Xinhua Chen, MD

Poor maternal nutrition is associated with an increased risk of serious complications and adverse outcomes during pregnancy. However, in humans, poor nutrition is often coupled with poverty and disadvantage, inadequate prenatal care, smoking, and drinking, making its effects difficult to disentangle.

In the classic study of a rare event, a World War II famine where there was a serious food shortage in a previously well-nourished population, it was noted that pregnant women lost weight, the birth rate declined, infant mortality increased, and the rate of infant low birth weight increased. When the peak of famine occurred during the first trimester, there was an excess of preterm births and an increase in infants weighing <2000g.[1] In addition to reducing the total amount of food that was eaten, famine would also have reduced many micronutrients and nutrients important to maternal–fetal being iron, folate, and calcium among others. Current recommendations for nutrient intakes that are important for pregnant women, along with information about the upper limit considered safe, are given in Appendix B.

Long-term consequences of poor maternal nutrition and other factors are discussed in Chapter 27. But, one of the immediate consequences of the famine would have been an increase in maternal anemia. Anemia, measured by low hemoglobin or hematocrit, is the most prevalent

nutritional deficiency in the world affecting 40% of pregnant women.[2] By Centers for Disease Control and Prevention estimates, anemia increases from 7.6 % in the first trimester to 33.9% during the third trimester.[3] The etiology of anemia is multifactorial, including hemoglobiopathies, acute infections, chronic inflammation, and diets poor in nutrients such as folate, vitamin B12, and vitamin A, along with iron, the most commonly recognized nutritional cause.

IRON, IRON DEFICIENCY, AND ANEMIA

During pregnancy, the maternal body requirement for iron increases to approximately 1000 mg.[4] Of this 350 mg is associated with fetal and placental growth, 500 mg with expansion in the red blood cell mass, and 250 mg with blood loss at delivery. The increased iron requirement needs to be supported by higher maternal iron intake - increasing from 6 mg/d in the first trimester, to 19 mg/d in the second trimester, to 22 mg/d in the third trimester.[4] To meet these increased requirements, gravidae must draw upon stores, thus increasing the risk of iron deficiency and iron-deficiency anemia (IDA).

Iron deficiency and anemia are prevalent among women who are pregnant or in their reproductive years usually because of a low intake of absorbable iron. Data from National Health and Nutrition Examination Survey (NHANES) III show a median iron intake of 14.7 mg/d of for pregnant women, with approximately 90% below the estimated average requirement for pregnancy (22 mg/d).[5] In addition to a poor-quality diet, poor iron absorption and blood loss—from menstruation, during labor and delivery, or from rapid repeat pregnancy—are other important causes of iron deficiency in women.[4] Among reproductive age US women who are not pregnant, IDA estimated from low hemoglobin and two of three iron indices was present in 2%–5%, while iron deficiency occurred in 11%–16%.[5]

Iron deficiency is defined by three stages of increasing severity: depletion of iron stores (stage 1), iron deficiency without anemia (stage 2), and IDA (stage 3). Low hemoglobin and hematocrit assess anemia in general but not specifically iron deficiency or IDA. Due to hemodilution during pregnancy, the optimal time to study effects may be before pregnancy or early in gestation before the plasma volume is fully expanded. This was originally examined in Camden by separating anemia by time (early vs. later gestation) and etiology (IDA and anemia from

other causes). IDA detected early in pregnancy was related to inadequate gestational weight gain from a diet deficient in energy and iron and resulted in a greater than twofold increase in the risks of low birth weight and preterm delivery, whereas anemia from other causes was unrelated to increased risk. In the third trimester, preterm delivery was less frequent for women with IDA.[6,7] Similar results were reported from Chinese workers tested for anemia before they became pregnant; 83% were anemic; but only 8% had IDA.[8] Pregravid anemia was associated with lower birth weight (−100 to −125 g), whereas IDA was associated with a birth weight deficit of −240 g.[8] Likewise, anemia detected early in pregnancy—during the first or second trimester—is usually associated with a higher risk of preterm delivery and low birth weight,[9,10] whereas anemia during the third trimester generally is related to a decreased risk of these adverse outcomes.[9]

Plausible mechanisms underlying the effect of anemia in pregnancy include (1) chronic hypoxia from anemia that initiates a stress response; (2) increased oxidative stress that damages the maternal–fetal unit; and (3) reduced maternal immune function with increased infection leading to preterm delivery and reduced fetal growth.[11]

While anemia is a risk factor, clinical trials that involve supplementation with iron or iron/folic acid have been equivocal. A recent Cochrane review found that women receiving iron or iron/folic acid supplements had infants significantly heavier (+58 g) than women on placebo or other regimens with no difference in preterm delivery, small-for-gestational-age (SGA) infant, or low birth weight.[12] A more recent meta-analysis also found difference in low birth weight (25% reduction)[13] after including a recent trial from rural China where iron/folic acid supplements increased gestation duration (+0.23 weeks) and reduced risk of very preterm delivery <34 weeks (0.98% vs. 1.8%).[14] Thus, supplementation may more effectively reduce the risk of adverse outcomes when the proportion of women who are iron deficient or anemic is high.[15] It is also plausible that duration is an issue. Supplements may need to be started early in pregnancy if not beforehand and continued throughout the reproductive years to reduce risk.[12,15]

Although clinical trials have shown almost uniformly that iron supplementation results in a reduction in anemia, they have raised the possibility that giving too much iron to nonanemic women, that is, daily administration of ≥50–60 mg/d, can increase blood viscosity and associated poor outcomes.[12,15] Thus, selective supplementation of women

with poor iron stores may be preferable to routine supplementation. Iron is potentially toxic and could expose women who are not deficient to high levels of oxidative stress and has no benefit for mother or fetus.[11,15] High maternal hemoglobin has consistently been associated with failure of the plasma volume to expand and adverse outcomes related to maternal pathology—gestational hypertension, preeclampsia, and diabetes.[12] Increased levels of the iron storage, ferritin, also are associated with preterm delivery but likely via chronic inflammatory changes.[16] Thus, the relationship between maternal hemoglobin and pregnancy outcome is said to be U shaped with low hemoglobin reflecting a mix of true and physiologic anemia and high hemoglobin reflecting failure of the plasma volume to expand or hemoconcentration related to an iron excess.

FOLIC ACID

Folic acid is a water-soluble B vitamin (B9) present in leafy green vegetables, dried beans, and fruits that is used to fortify flour and cereals in the United States. Humans cannot synthesize folate and are entirely dependent on dietary sources or folic acid supplements for their supply. Folate is important for the production of S-adenosyl methionine, a universal donor of methyl groups that transfers one-carbon units to be used for DNA methylation. Folate is required for cell division because of its role in DNA synthesis and is a substrate for reactions that affect metabolism of amino acids on the transmethylation and transsulfuration pathways.

A central feature of fetal development is widespread and sustained cell division. Because of its role in nucleic acid synthesis, the need for folate increases during times of rapid tissue growth. During pregnancy, folate-dependent processes include increase in the red blood cell mass, enlargement of the uterus, and growth of the placenta and fetus along with epigenetic regulation of differentially methylated regions (DMRs) of the fetal genome. During the aforementioned Dutch Famine when periconceptional exposure to the famine occurred, there was an increase in the frequency of malformations of the central nervous system, including spina bifida,1 which is consistent with a deficiency in folate and other methyl donors. A long-term effect was noted six decades later in the lower methylation status of the *IGF2* gene with fetal exposure to famine during early gestation.[17] The *IGF2* gene is maternally imprinted and produces insulin-like growth factor 2 that is important for growth and development. In the case of *IGF2*, the imprinted allele from the mother normally is

silenced and only the father's nonimprinted allele is expressed. Lower methylation of the IGF2 DMR would be expected to lead to increased expression of both maternal and paternal IGF2 alleles.[17]

It is recommended that reproductive age women consume 400 μg folic acid from supplements or fortified foods, in addition to folate-containing foods from their diet, to reduce the risk of neural tube defects, increasing to 600 μg/d during pregnancy (see Appendix B). Approximately one-third of women in their reproductive years consume the recommended amount.[18] Fortification of flour and cereal products in the United States with folic acid (since 1998) has been associated with a 19% decline in the risk of live born infants with neural tube defects, along with a 27% decline in anemia in women along with increases in hemoglobin, hematocrit, serum folate, and a decline in homocysteine levels.[19,20]

Although these are positive changes, maternal periconceptional supplementation with folic acid may also have led to epigenetic modifications and changed expression of some fetal genes. In one study, maternal periconceptional use of ≥400 μg folic acid/d was linked to increased methylation of IGF2 DMRs among infants[21]; the findings that are opposite to the Dutch Famine. In another study, maternal use of folic acid supplements before and during pregnancy was associated with increased methylation of H19 DMR but not IGF2 DMR in cord blood.[22] The long-term consequences of these epigenetic changes, if any, are not known. Current knowledge about early intrauterine exposures, epigenetics, gene expression, and their sequelae are reviewed in Chapters 19 and 20.

Serum folate is a sensitive indicator of the folate available to replicating cells with high turnover rates, whereas red blood cell folate reflects folate status over preceding months. A metabolic effect of a deficiency in folate (B_9) and other B vitamins (B_6 and B_{12}) is an elevation of homocysteine.[23] Hyperhomocysteinemia, a risk factor for cardiovascular disease, is a known cause of vascular damage and endothelial dysfunction. If homocysteine was related to poor placental development or attachment, then high levels might do injury in the course of pregnancy, increasing serious complications like placental abruption and preeclampsia where endothelial damage is a significant factor. The consequences for pregnancy would be expected to reflect placental vascular compromise, that is, reduced infant birth weight and increased rates of fetal growth restriction and preterm delivery.[23–25]

Homocysteine, when measured in more than 5800 Norwegian women aged 40–42 years, was positively related to a past reproductive history that

included preeclampsia, preterm delivery, low birth weight, and fetal growth restriction.[26] High homocysteine, examined in Chinese workers before conception, was associated with a fourfold increased risk of preterm delivery, not SGA.[27] A longitudinal study, with repeated measurement of maternal homocysteine from before conception, reported lower weight infants (~200 g) born to mothers with higher homocysteine; whether this was attributable to shorter gestation duration is unclear.[28] More recently, investigators from the Generation R study, which assayed both homocysteine and folate early in pregnancy (13 weeks gestation on average) reported that low folate and high homocysteine were each related to lower placental weight (~−26 to −30 g) and infant birth weight (−110g to −125 g). Twofold increases in risks of preterm delivery and preeclampsia were evidenced with low folate (lowest quintile) but not high homocysteine (highest quintile); SGA risk was increased twofold with each. When both folate (low) and homocysteine (high) were examined jointly, there was a fourfold increased risk for SGA and preeclampsia.[25] That low folate was more regularly associated with increased placental vascular resistance during mid to late gestation than homocysteine, suggesting a direct effect of folate on the placental vasculature with homocysteine being a response to or marker for poor folate status.[25]

Inadequate intake of folate leads to impaired cell division and alterations in protein synthesis. An absolute deficiency of folate—from a diet inadequate to meet the needs of pregnancy—will interfere with growth of the fetus and duration of gestation.[23] We have shown in the Camden study that low dietary and circulating folate, measured early in gestation, tripled the risk of low-birth-weight infant and preterm delivery and interacted with a deletion allele for a folate-metabolizing gene enzyme (dihydrofolate reductase [DHFR]) that converts folic acid that is used in supplements and to fortify the US food supply to the reduced folate forms used by cells. Presence of the DHFR deletion allele increased the risk of preterm delivery threefold, and low intake of folate increased the risk of low birth weight eightfold and risk of preterm birth fivefold.[29,30] Lower folate intake (<500 μg/d) at mid-gestation was associated with approximately twofold increased risk of preterm delivery in women from North Carolina as were low levels of serum and red blood cell folate[31] and the folate metabolite 5 methyltetrafolate.[32] Others have reported increased infant birth weight for mothers whose folate intake was high and positive correlations between maternal serum and red cell folate with birth weight or gestation duration.[33]

In particular, effects of folic acid supplementation before and during the course of pregnancy are unclear. A large cohort of women who supplemented for at least 1 year with folic acid had a 70% reduction in risk of delivering before week 28 and a 50% reduction for delivery between 28 and 32 weeks.[34] After folic acid fortification of the US food supply, rates of preterm delivery, very low birth weight, and low birth weight declined between 4% and 10% among Californian liveborn infants[35] and elsewhere.[36] A recent meta-analysis found an overall effect of folic acid on birth weight corresponding to a 2% increase for each doubling of the folic acid supplement.[37] While smaller scale trials have and have not reported reductions in risk,[33] a large randomized study found no effect of folic acid supplements on mean birth weight, gestation at delivery, or placental weight.[38] After pooling with data from another trial, a high dose of folic acid (5 mg/d) was associated with a 37% reduction in risk of low-birth-weight infant.[38] Likewise, although supplementation with folic acid (with/without iron) showed no effects on the white South African women, the risk of bearing an infant of less than 1870 g was reduced fourfold among the Bantu participants, who subsisted primarily on maize porridge.[39]

In summary, although observational studies of folate and pregnancy suggest a potential benefit of folate supplementation during pregnancy—a decrease in serious complications and an improvement in birth weight and gestation—randomized trials imply that women who are at risk, either because of common genetic polymorphisms that alter folate metabolism or because of environmental factors associated with a diet low in folate, would seem to benefit the most. Many of the investigators who did not find lower folate (circulating levels, diet, or supplements) to be related to preterm delivery or gestation duration did not confirm the mother's last menstrual period (LMP) by ultrasound with resulting misclassification and loss of precision to detect an effect.[32]

While supplementation with folic acid before pregnancy carries an enormous benefit—a reduction in risk of neural tube and other congenital defects—it may also alter the methylation patterns in DMR for some fetal genes. It is now an open question whether supplementation with folic acid before and during gestation will have consequences for the exposed fetus as it grows and develops.

CALCIUM

The skeleton acts as the calcium reservoir of the body (99% is stored there), and when dietary calcium intake is low or poorly absorbed, calcium is withdrawn from the bone to support serum calcium homeostasis.[40] During pregnancy, retention of an estimated 30 g of calcium, most of which is transferred during the third trimester, is needed to mineralize the fetal skeleton.[40] Although it is often assumed that pregnancy, like lactation, might be a setting for bone mass loss because of this requirement, this might be buffered by a 60%–70% enhancement in intestinal calcium absorption during pregnancy which is mediated by 1,25 dihydroxyvitamin D, the active form of vitamin D and other calcitropic hormones. This view is supported by cross-sectional studies of bone mass and parity, usually confined to mature white women, which show increased bone mass and decreased fracture risk or no effect of parity on bone mass.[41]

The few available longitudinal studies have come to a different conclusion, they suggest that trabecular bone, which has a larger surface area, is softer than cortical bone, and more readily mobilized may be another source of the calcium needed to ossify the fetal skeleton.[41] Longitudinal research on change in bone mass with pregnancy usually report small gains in cortical bone but significant decreases in sites (spine and pelvis) where trabecular bone predominates.[41,42] One reason for the limited research is risk of radiation exposure. Measurement of the mother's appendicular skeleton by other means—ultrasound or peripheral quantitative computed tomography—also shows decreases in bone at some sites. One study reported a quantitative ultrasonographic index 3.6% lower at six weeks postpartum compared to early pregnancy.[43] In another, measurements of the distal radius on the nondominant arm showed no loss in cortical bone but a 1.6% decrease in trabecular bone density between first and third trimesters compared to nonpregnant controls.[44] Although the amount of bone lost is typically small, it is consistent with the fetal demand for calcium.

Low calcium intake, which is an important determinant of bone mineralization, could also influence bone changes. Higher calcium intakes during childhood and adolescence are associated with higher bone mass at maturity.[45] If dietary calcium intake is very low (<500 mg/d), then decreases in serum calcium and calcium absorption would be expected.[41] Lower serum calcium increases parathyroid hormone (PTH) that stimulates production of 1,25(OH)2D, the active form of vitamin D. Increased production of $1,25(OH)_2D$ enhances calcium absorption from

the gut, mobilizes skeletal calcium, and reduces urinary calcium excretion to maintain calcium balance. When calcium intake is chronically low and calcium is added to the maternal diet, positive changes in bone mineral density have been noted as well as reduction in markers of bone turnover.[41]

During adolescence, the bone mineral content of the femur and the lumbar spine increase by 50%–150% and volumetric bone mineral density increases by 10%–30%.[46] Factors that interfere with normal accrual may be related to later osteoporosis,[47] and regions that are actively growing may be more severely affected.[46,47] During adolescent pregnancy, calcium nutriture could be limited by maternal diet but simultaneously driven by the need to retain enough calcium to mineralize two skeletons. Approximately 50% of teenagers continue to grow while pregnant.[48] Ultrasonographic bone measures of the os calcis showed that bone loss was greater for growing teenage gravidae (−5.5%) than for mature women (−1.9%).[43] Iowa women with an adolescent pregnancy (<19 years) had, at maturity, a bone mass at the mid distal forearm that was below controls.[49] Calcium consumption was also related to bone and the joint effect of an early pregnancy, and lower calcium intake was substantial.[49] This is consistent with a recent study which suggested that an adolescent pregnancy resulted in a twofold increase in the risk of osteoporosis and a tendency to more osteoporotic fractures in postmenopausal Korean woman.[47]

Diets low in calcium, especially during pregnancy, are associated with increased blood pressure levels through heightened smooth muscle reactivity (see Appendix B for recommended dietary intake of calcium and other nutrients during pregnancy). Belizan and Villar[50] reported an inverse relationship between eclampsia and dietary calcium from ecologic-level data that showed countries where calcium intake was low had a higher frequency of eclampsia than countries where calcium intake was higher.

A meta-analysis of 14 randomized, controlled trials of calcium supplementation involving more than 2400 pregnant women showed statistically significant reductions in systolic and diastolic blood pressure with the administration of calcium salts (375–2000 mg/d elemental calcium).[51] Risk of preeclampsia was reduced more than twofold among women supplemented with calcium during these trials. Duration of treatment ranged from 10 to 22 weeks, with most studies starting the supplement during the second trimester. Although differences in the risk of preeclampsia were significant in the aggregate, only two of the nine

studies showed statistically significant differences.

A large, randomized, double-blind, placebo-controlled trial of 5489 low-risk nulliparous women showed no effect of calcium supplementation.[52] Pregnant women were enrolled at centers throughout the United States, supplemented with 2 g of elemental calcium (or placebo) beginning at 13–21 weeks gestation and continuing until they delivered. Calcium supplementation did not significantly reduce the risk of preeclampsia, gestational hypertension without preeclampsia (either mild or severe), or pregnancy-associated proteinuria without hypertension. Blood pressure was also unaffected; however, participants' dietary intake of calcium was high, averaging about 1100 mg/d.[52]

It seemed likely that an effect of calcium on preeclampsia might be confined to high-risk populations in which there was either a severe dietary restriction of calcium or a high demand for calcium as in adolescent pregnancy. The World Health Organization (WHO) trial that involved more than 8300 pregnant women from populations where dietary calcium intakes were low (<600 mg/d) supported the original ecologic observations made on eclampsia.[53] Calcium supplements (1.5 g/d) did indeed reduce the relative risk of eclampsia (by 47%), severe gestational hypertension (by 40%), severe preeclamptic complications (by 32%), the severe maternal morbidity and mortality index (by 25%), and neonatal mortality (by 43%). The overall risk of preeclampsia was not lower on the calcium regimen than placebo (4.1% vs. 4.5%). However, a meta-analysis pooling data from clinical trials that randomized more than 15,000 women concluded that calcium supplementation did in fact reduce the overall risk of preeclampsia (Relative Risk (RR) = 0.45, 95% confidence interval [CI] 0.31–0.65) and by 65%–75% for women at high risk for preeclampsia (RR = 0.22, 95% CI 0.12–0.42) or with low calcium intake (RR = 0.36, 95% CI 0.20–0.65).[54]

During pregnancy, an increased risk of gestational hypertension or preeclampsia associated with lower calcium intake could result in an increased risk of an indicated preterm delivery. Consequently, supplementation with calcium during pregnancy could have a side effect of reducing preterm birth. The WHO trial showed that for women aged 20 years or younger, very preterm delivery (<32 weeks) was significantly reduced (by 56%) and preterm delivery showed a nonsignificant trend in the same direction.[53] The aforementioned clinical trial performed by Levine et al. found no effect of calcium supplementation on obstetric outcomes, including preterm delivery, birth weight, low birth weight, or

fetal growth restriction.[52] A meta-analysis of trials that randomized more than 15,000 women found that calcium reduced the risk of preterm delivery by ~30% (RR = 0.76, 95% CI 0.60–0.97) and by 50% in 4 trials (*N* = 568) where women were at high risk for preeclampsia (RR = 0.45, 95% CI 0.24–0.83).[54]

VITAMIN D

Vitamin D, a collection of fat-soluble secosterols, is present in few foods naturally (fatty fish, fish liver oils), used for food fortification (fortified milk, breakfast cereals, orange juice), and included in nutritional supplements (prenatal multivitamins). It is also synthesized photochemically by the skin after exposure to ultraviolet B (UVB) radiation from the sun. The circulating level of 25(OH)D is the best indicator of vitamin D status representing the amount of vitamin D produced cutaneously and ingested from diet and supplements.[40] Prevalence of vitamin D deficiency and insufficiency is modified by skin pigmentation. More pigment-intense skin will block UVB absorption and increase the risk of poor vitamin D status so that the risk of vitamin D deficiency and insufficiency is higher among African Americans and intermediate among Hispanics compared to whites.[40]

$1,25(OH)_2$ vitamin D is the active form of the vitamin related to calcium and homeostasis, bone health, and other physiologic functions.[40] Several recent studies found an increased risk of preeclampsia with maternal 25(OH)D consistent with insufficiency deficiency as well as with low intake of total vitamin D although others have not.[55] Two prospective studies, one of them longitudinal, reported lower serum 25(OH)D in gravidae who subsequently developed preeclampsia[56,57] as well as in their infants.[56] However, they disagreed on when risk was detectable, either early in gestation (10 weeks on average) or later (weeks 24–26) without an earlier window.

Neither the National Institutes of Health (NIH)[52] nor the WHO trial[53] of calcium and preeclampsia measured 25(OH)D. Many of the participants from the WHO trial were from the tropics, likely exposed to more sunshine with a lower expected prevalence of vitamin D insufficiency than the North American women studied by the NIH. There is an interaction between vitamin D and calcium such that when vitamin D is deficient or insufficient, even high calcium intakes may be inadequate to maintain calcium metabolism.[58] Thus, it is plausible that an effect might have been

detected had the supplement combined calcium with vitamin D.

Overweight and obesity increase the risk of type 2 diabetes and preeclampsia as well as poor vitamin D status. Vitamin D is a fat-soluble vitamin.[40] Adiposity measures, such as skinfolds, body mass index (BMI), total body fat, all correlate inversely with 25(OH)D, and it is likely that there is an increased storage of vitamin D in adipose tissue with consequent reductions in bioavailability.[40] Poor vitamin D status likely alters the insulin response to a glucose load, is associated with increased insulin resistance, and amplifies the systemic inflammatory response characteristic of type 2 diabetes.[40] After adjustment for BMI and other factors, lower circulating levels of 25(OH)D has been implicated in type 2 diabetes.[40] Observational data suggest that circulating levels of 25(OH)D were significantly lower in gestational diabetics compared to normoglycemic women; overall risk of gestational diabetes was increased by 50% with 25(OH)D in the deficient or insufficient range.[59]

In observational studies, Canadian gravidae who restricted drinking milk had infants with lower weights at birth (~80 g); both milk consumption and vitamin D intake were significant birth weight predictors.[60] Vitamin D deficiency is characterized by elevations in PTH. A small study (N = 30) of vitamin D-deficient Pakistani gravidae showed an inverse correlation (r = −0.39, $P < 0.05$) between lower birth weight and higher maternal PTH.[61] Data from Camden also suggested that total intake of vitamin D (diet plus supplements) was a significant predictor of infant birth weight for gestation; the difference in birth weight between the lowest quintile and the highest quintiles of vitamin D amounted to 65 g.[62]

More than two decades ago, it was reported that vitamin D-supplemented Asian women residing in India or living in London gained significantly more weight and had infants with higher weights at birth. The increases ranged from 80 to 410 g birth weight; one study showed a twofold reduction in low birth weight.[63] However, a trial conducted among French women did not show positive effects of vitamin D supplements on birth weight.[63] The overall effect of supplementation was consistent with no change in infant birth weight but a trend toward lower risk of low-birth-weight infant (RR = 0.48, 95% CI 0.23–1.01).[63] Most studies have found no association between vitamin D supplementation and gestation duration or preterm birth[55] with one exception. Australian gravidae with very low levels of 25(OH)D (<28 nmol/L) had gestations that were 0.7 week (4.9 days) shorter than women with higher circulating levels; birth weight was also reduced but the difference (–157 g) was not statistically significant.[64]

Mode of delivery may differ for vitamin D-deficient women. Vitamin D is related to muscle mass and strength.[40] Two studies now suggest an increased risk for cesarean delivery in vitamin D-deficient women.[65,66] When specific indications were examined, vitamin D deficiency was linked to a 2-fold increased risk of Cesarean for prolonged labor in Camden gravidae.[66] Thus, it is plausible that poor maternal vitamin D status increases the risk of Cesarean delivery by reducing pelvic muscle strength and control leading a reduced ability to push and to a longer and more difficult labor.

MULTIVITAMIN AND MINERAL SUPPLEMENTS

In the United States, the use of vitamin and mineral supplements has been increasing, and currently, more than 40% of US women use multivitamins/mineral supplements; 34% of women in their reproductive years regularly take one or more supplements that contain folic acid.[18,67]Appendix B contains information on recommended dietary intakes for pregnant women in the United States including the upper limit that is presumed to be safe.

Multivitamin/mineral supplements contain at least three vitamins and may or may not contain minerals.[67] Although the value individual micronutrients like folic acid and iron have been long recognized, the import of others (vitamin D) has only recently become apparent. Deficiencies of more than one micronutrient are found within as well as between populations depending upon dietary intake, supplement use, food preferences, income, and residence. In poor urban areas in the United States, there are neighborhoods where fresh fruits and vegetables are hard to find and thus rarely eaten. For other nutrients, like vitamin D, there can be wide seasonal variation because of their dependence on seasonal UVB radiation from the sun.

In the developing world, pregnant women are often deficient in a variety of vitamins and minerals, and iron or iron/folic acid supplements alone are less likely to be effective. In one of the first studies, Fawzi et al. examined the effect of supplementing more than 1000 HIV-positive women from Tanzania with multivitamins, vitamin A, and placebo.[68] Multivitamin supplementation decreased the risk of very preterm delivery (approximately twofold reduction) along with risk of low-birth-weight infant and fetal growth restriction. The multivitamin supplement had the bonus of increasing CD4, CD8, and CD3 counts in the women.

Meta-analyses compared outcomes of pregnant women from the developing world presumed to be HIV negative who participated in trials where similar protocols were used. Women were supplemented with multiple micronutrients (usually 1 Recommended Dietary Allowance (RDA)) and controls with iron, iron/folic acid, iron/folic acid/vitamin A, or placebo. Multiple micronutrients reduced low birth weight and SGA by approximately 10%.[69,70] In one of the analyses, multiple micronutrients significantly increased birth weight (+22 g), but depending on the mothers BMI.[69] In gravidae where BMI was <20, there was no effect of the supplement on birth weight; when BMI was 20 or more, birth weight increased by +39 g.[69] There was no effect on gestation duration or on preterm birth; however, gestation was usually based on the mother's LMP, often resulting in implausible combinations of birth weight and gestation,[69] which implies substantial misclassification. Use of prenatal vitamins by women from North America and Europe before and during pregnancy is associated with reductions in the risk of preterm delivery, low-birth-weight infant, and SGA.[71,72]

HERBAL PRODUCTS

Use of herbs or herbal products during pregnancy is increasing worldwide and in English-speaking counties ranges from 7% to 56%.[73–75] Data from the US National Birth Defects Prevention Study showed that 10.9% of women (N = 4239 with nonmalformed infants) used herbals before and during pregnancy amounting to 395,000 US births per year.[74] Actual use is probably higher than the published literature; this may be because of failure to admit use to health care providers or to participate in surveys.

Common herbs along with the indication for their use are listed in Table 25-1.[73–75] Herbal products are used for a wide range of maternal complaints particularly morning sickness, anemia, and infection. Most herbal products are perceived as natural, with benign effects and as safe for use during pregnancy; this may or may not be true.[76] Ginger, which is commonly used for morning sickness and other conditions, has multiple therapeutic properties including antioxidant, antimicrobial, and anti-inflammatory actions.[77,78] Apart from pregnancy, ginger may prevent the development of cardiovascular diseases or cancer.[78] A retrospective study reported that taking ginger and the iron-rich herb echinacea was associated with high-birth-weight infant (3663 g vs. 3508 g, $P < 0.001$).[73] A large study with 68,522 Norwegian mothers suggested that ginger was not

associated with an increased risk of adverse perinatal outcomes including congenital malformation, preterm birth, low birth weight, stillbirth, or low Apgar score.[77]

CHINESE HERBAL MEDICINE

Herbal medicine is the most important practice of traditional Chinese medicine. It is well accepted as mainstream medical care throughout East Asia but is considered to be complementary or alternative medicine in the West.[79] Typical treatment with Chinese herbal medicine is based on several combined remedies or formulas that have been used in the prevention and treatment of severe nausea and vomiting, infection, threatened abortion, and ectopic pregnancy.[80–82] Hundreds of publications, especially in Chinese medical journals, report the use of Chinese herbal medicine in complicated pregnancies, examples of which are listed in Table 25-1.

TABLE 25-1 Herbal Supplement Use in Pregnancy

	Common Reason for Use
Herbs	
Ginger	Morning sickness and various conditions
Chamomile	Morning sickness, relaxation, and sleep
Cranberry juice	Preventing or treating urinary tract infection
Echinacea or Elderberry	Treating cold or influenza symptoms
Slippery elm	Digestive disorders
Rose hips	Fighting infection
Raspberry leaf	Toning the uterus to prepare for labor
Iron-rich herbs, i.e., oat straw or Echinacea	Preventing anemia
Chinese herbal medicines	
Xiaochaihu plus Fulonggan formula[a]	Treating severe nausea and vomiting
Shapei Huayu formula[b]	Nonsurgical treatment for ectopic pregnancy
Bushenantai drink[c]	Treating threatened abortion
Other supplements	
Fish oil	Increase gestation duration

[a] Contain eight herbs such as Chai hu, Huang qin, Ban xia, Dang shen, and Gan cao.

[b] Contain 11 herbs such as Dan Shen, Tao ren, San leng, and Chi shao.

[c] Contains 12 herbs such as To si zi, Gou qi zi, Du zhong, and Huang qin.

Supplements made from Chinese herbs are marketed and used globally. For example, the Bushenantai drink made from 12 herbs is used to treat threatened abortion[82] and a modified Xiaochaihu formula with 8 herbs is commonly used to treat severe nausea and vomiting during early pregnancy.[80] Chinese herbal medicine also has been used as an adjunct to reduce side effects and toxicity from Western pharmaceuticals. Patients with an ectopic pregnancy were randomly assigned to methotrexate plus mifepristone (N = 125) or to a combination of mifepristone with Shapei Huayu (N = 125), a mixture of 11 Chinese herbs.[81] The success of treatment was evaluated by normal blood β-HCG concentrations, no vaginal bleeding or abdominal pain, and reduced or no abdominal mass by ultrasound. There was no significant difference in the success rate; however, side effects including digestive symptoms, decreased peripheral white cell counts, and liver damage were significantly fewer in the group on the combination treatment. During follow-up, the subsequent successful intrauterine pregnancy rate was higher in the combination treatment (69%) compared to Western medicine alone (49.5%, $P < 0.01$).

Some data suggest that herbal treatments alone or the combination of Chinese herbal medicine with Western medicine carries a significantly lower risk of side effects as does the treatment of ectopic pregnancy described earlier.[81] Other studies have suggested that use of certain common herbal remedies during pregnancy may have untoward consequences.[83] For example, a survey in 3191 Canadian women found that use of flax increased the risk of preterm birth (Adjusted Odds Ratio (AOR) 3.91, 95% CI 1.16–13.14), whereas use of several other herbs (chamomile, peppermint, or green tea) was not associated with increased risk.[83] The problem is that most observational or randomized studies have small sample sizes with inadequate statistical power to detect adverse effects, most of which are relatively uncommon so that case reports are often the only indicator of an adverse experience.[76,84] The recommendation to use herbal products during pregnancy remains controversial.

FISH OIL SUPPLEMENTS

Maternal fatty acids (FAs) are important for fetal growth and development. Both omega (n)-3 and omega (n)-6 polyunsaturated fatty acids (PUFAs)

are essential FAs meaning that they must be consumed in the diet. The essential FA α-linolenic acid gives rise to n 3-PUFA and linoleic acid to n 6-PUFA. The most bioactive forms of n-3 PUFA are eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), and the key n-6 PUFA is arachidonic acid. All of the n-3 and n-6 PUFA structures acquired by the fetus must come from the mother across the placenta, either in the form of the essential FAs or their derivatives EPA and DHA.[85] Thus, an enhanced maternal dietary intake of DHA increases the fetal supply and leads to a higher DHA concentration in cord blood.[86] An adequate supply of bioactive FAs needs to be transferred from the mother to the fetus; they are important during the perinatal period for the rapid growth and development of new tissues and organ systems.

Consumption of supplements that contain fish oil as well as eating a diet rich in marine foods that contain n-3-PUFA are associated with beneficial effects including longer gestation duration, improved fetal growth and developmental of visual acuity, cognitive function, and immune phenotype in cohort studies and randomized clinical trials.[85,87] Canadian Inuit with high n-3 PUFA intake have longer gestations (+5.4 days) and increased infant birth weights (+77 g).[88] A trial of 533 Danish women who received fish oil, olive oil, or no supplement by 30 weeks gestation showed taking the fish oil supplements resulted in longer average gestations (+4 days) and infants with higher average weights (+107 g), mostly attributable to increased gestation.[89] The effect of fish oil was strongest with low fish consumption and amounted to +7.4 days longer gestation in this group. A clinical trial of a high-risk, low-income population that was primarily African American showed that the consumption of eggs fortified with DHA late in second trimester increased gestation duration by six days on average.[90] However, a recent meta-analysis of several randomized trials suggested that although the effect of n-3 PUFAs on gestation duration was present, it was weak (+1.5 days).[91] There was little effect of the fish oils when fish consumption was high (−1.6 days).[89] Thus, the hypothesized effect of n-3 PUFA on gestation appears to have a threshold and to be specific to the initiation of idiopathic preterm labor.

Fish oil or use of n-3 PUFA supplements during pregnancy seem clearly to increase the length of gestation, whether such supplements could prevent preterm delivery or other adverse pregnancy outcomes have not been confirmed. A multicenter trial enrolled high-risk women with a history of preterm delivery, fetal growth restriction, or preeclampsia.

Women were randomized to either fish oil or olive oil. Fish oil reduced the recurrence risk of preterm delivery approximately twofold but did not alter the recurrence of the other poor outcomes.[92] Higher doses of n-3 PUFAs (6.1 g/d) in women with threatened preeclampsia or suspected fetal growth restriction showed no effect on these outcomes.[92]

Ecologic studies have indicated that the consumption of large quantities of fish during pregnancy is related to lower blood pressure and thus, in theory, with a lower incidence of pregnancy-induced hypertension. EPA and DHA have been shown to modulate inflammatory and vascular effects and subsequently reduce inflammation-related diseases like cardiovascular disease[93]; it is plausible that n-3 PUFAs could effectively improve vasoconstriction and the endothelial damage related to gestational hypertension and preeclampsia.[85,91] One of the first studies on the topic, The People's League of Health, showed a reduction in risk of preeclampsia, which, because multiple supplementation regimens were used, could not be attributed to the use of the fish oil.[94] Another trial, a multicenter study of fish oil supplements, showed no effect on high blood pressure or the risk of preeclampsia.[91]

An increased n-3 PUFA is needed for fetal growth and development. However, the specific amount of n-3 PUFA to maintain the optimal benefit for the mother and fetus has been difficult to determine, due to the complex metabolism of the PUFA and the preferential use of n-3 PUFA mobilized from the n-3 stored in maternal adipose tissue.[85,95]

Fish provide a source of easily digestible protein and other nutrients, and many fish provide a uniquely rich food source of long-chain PUFA, especially DHA and EPA. The benefit of maternal seafood intake and the competing risk of maternal mercury exposure have been evaluated.[96] Methyl mercury crosses the placenta. The US Food and Drug Administration advised all pregnant women to limit seafood consumption to 340 g/wk to reduce fetal exposure to trace amounts of neurotoxins from methylmercury.[96] This amount of fish should provide approximately 200 mg of DHA per day. Alternative source of DHA including fish oil capsules, which provide variable amounts of DHA ranging from 150 to 1200 mg/d, is low in contaminants like mercury. Many prenatal vitamins contain up to 200–300 mg of DHA/d.[85] Similarly, the consensus of the European Commission on the topic was that dietary intake should be at least 200 mg/d DHA during pregnancy. The commission also recommended screening for maternal dietary inadequacies during first trimester and offering individual counseling during pregnancy and

lactation to women with less desirable diets.[95]

REFERENCES

1. Stein Z, Susser M, Saenger G, Marolla F. *Famine and Human Development: The Dutch Hunger Winter of 1944–1945*. New York, NY: Oxford University Press; 1975.
2. McLean E, Cogswell M, Egli I, et al. Worldwide prevalence of anemia, WHO Vitamin and Mineral Nutrition Information System, 1993–2005. *Publ Hlth Nutr.* 2008;12:444–454.
3. Dalenius K, Brindley P, Smith B, et al. *Pregnancy Nutrition Surveillance 2010 Report*. Atlanta, GA: U.S. Department of Health and Human Services, Centers for Disease Control and Prevention; 2012.
4. IOM (Institute of Medicine). *Nutrition During Pregnancy*. Washington, DC: National Academy Press; 1990.
5. Cogswell ME, Looker AC, Pfeiffer CM, et al. Assessment of iron deficiency in US preschool children and nonpregnant females of childbearing age: National Health and Nutrition Examination Survey 2003–2006. *Am J Clin Nutr.* 2009;89:1334–1342.
6. Scholl TO, Heldiger ML, Fischer RL, et al. Anemia vs iron deficiency: increased risk of preterm delivery in a prospective study. *Am J Clin Nutr.* 1992;55:985–988.
7. Scholl TO, Heldiger ML. Anemia and iron deficiency anemia, compilation of data on pregnancy outcome. *Am J Clin Nutr.* 1994;59:492S–501S.
8. Ronnenberg AG, Wood RJ, Wang X, et al. Preconception hemoglobin and ferritin concentrations are associated with pregnancy outcome in a prospective cohort of Chinese women. *J Nutr*. 2004;134:2586–2591.
9. Scanlon KS, Yip R, Schieve LA, et al. High and low hemoglobin level during pregnancy: differential risks for preterm birth and small for gestational age. *Obstet Gynecol.* 2000;96:741–748.
10. Ren A, Wang J, Ye RW, et al. Low first trimester hemoglobin and low birth weight, preterm birth and small for gestational age newborns. *Int J Gynaecol Obstet.* 2007;98:124–128.
11. Allen LH. Anemia and iron deficiency: effects on pregnancy outcome. *Am J Clin Nutr*. 2000;71:1280S–1284S.
12. Pena-Rosas JP, Viteri FE. Effects and safety of preventive oral iron or iron+folic acid supplementation for women during pregnancy. *Cochrane Database Syst Rev.* 2009;4:CD004736.
13. Imdad A, Bhutta ZA. Routine iron/folate supplementation during pregnancy: effect on maternal anemia and birth outcomes. *Paediatric Perinatal Epidemiol.* 2012;26:168–177.
14. Zeng L, Cheng Y, Dang S, et al. Impact of micronutrient supplementation during pregnancy on birth weight, duration of gestation, and perinatal mortality in rural western China: double blind cluster randomized controlled trial. *BMJ.* 2008;337:a2001.
15. Aranda N, Ribot B, Garcia E, et al. Pre-pregnancy iron reserves, iron supplementation during pregnancy and birth weight. *Early Hum Dev.* 2011;87:791–797.
16. Scholl TO. High third-trimester ferritin concentration: associations with very preterm delivery, infection, and maternal nutritional status. *Obstet Gynecol.* 1998;92:161–165.

17. Heijmans BT, Tobi EW, Stein AD, et al. Persistent epigenetic differences associated with prenatal exposure to famine in humans. *Proc Natl Acad Sci USA.* 2008;105:17046–17049.
18. Yang QH, Carter HK, Mulinare J, et al. Race-ethnicity differences in folic acid intake in women of childbearing age in the United States after folic acid fortification: findings from the National Health and Nutrition Examination Survey, 2001–2002. *Am J Clin Nutr.* 2007;85:1409–1416.
19. Ganji V, Kafai MR. Hemoglobin and hematocrit values are higher and prevalence of anemia is lower in the post-folic acid fortification period than in the pre-folic acid fortification period in US adults. *Am J Clin Nutr.* 2009;89:363–371.
20. Jacques PF, Selhub J, Bostom AG, et al. The effect of folic acid fortification on plasma folate and total homocysteine concentrations. *N Engl J Med.* 1999;340:1449–1454.
21. Steegers-Theunissen RP, Obermann-Borst SA, Kremer D, et al. Periconceptional maternal folic acid use of 400 microg per day is related to increased methylation of the IGF2 gene in the very young child. *PloS One.* 2009;4:e7845.
22. Hoyo C, Murtha AP, Schildkraut JM, et al. Methylation variation at IGF2 differentially methylated regions and maternal folic acid use before and during pregnancy. *Epigenetics.* 2011;6:928–936.
23. Tamura T, Picciate MF. Folate and human reproduction. *Am J Clin Nutr.* 2006;83:993–1016.
24. Hogeveen M, Blom HJ, den Heijer M. Maternal homocysteine and small-for-gestational-age offspring: systematic review and meta-analysis. *Am J Clin Nutr.* 2012;95(1):130–136.
25. Bergen NE, Jaddoe VW, Timmermans S, et al. Homocysteine and folate concentrations in early pregnancy and the risk of adverse pregnancy outcomes: the Generation R Study. *BJOG.* 2012;119:739–751.
26. Vollset SE, Refsum H, Emblem BM, et al. Plasma total homocysteine, pregnancy complications and adverse pregnancy outcomes: the Hordaland Homocysteine Study. *Am J Clin Nutr.* 2001;71:962–968.
27. Ronnenberg AG, Goldman MB, Chen D, et al. Presonception homocysteine and B vitamin status and birth outcomes in Chinese women. *Am J Clin Nutr.* 2002;76:1385–1391.
28. Murphy MM, Scott JM, Arija V, et al. Maternal homocysteine before conception and throughout pregnancy predicts fetal homocysteine and birth weight. *Clin Chem.* 2004;50:1406–1412.
29. Scholl TO, Hediger ML, Schall JI, et al. Dietary and serum folate: their influence on the outcome of pregnancy. *Am J Clin Nutr.* 1996;63:520–525.
30. Johnson WG, Scholl TO, Spychala JR, et al. Common dihydrofolate reductase 19–base deletion allele: a novel risk factor for preterm delivery. *Am J Clin Nutr.* 2005;81:664–668.
31. Siega-Riz AM, Savitz SA, Zeisel SH, et al. Second trimester folate status and preterm birth. *Am J Obstet Gynecol.* 2004;191:1851–1857.
32. Bodnar LM, Himes KP, Venkataramanan R, et al. Maternal serum folate species in early pregnancy and risk of preterm birth. *Am J Clin Nutr.* 2010;92(4):864–871.
33. Scholl TO, Johnson WG. Folic acid: influence on the outcome of pregnancy. *Am J Clin Nutr.* 2000;71:1295S–1303S.
34. Bukowski R, Malone FD, Porter FT, et al. Preconceptional folate supplementation and the risk of spontaneous preterm birth: a cohort study. *PLoS Med.* 2009;6:e1000061.
35. Shaw GM, Carmichael SL, Nelson V, et al. Occurrence of low birthweight and

preterm delivery among California infants before and after compulsory food fortification with folic acid. *Public Health Rep.* 2004;119:170–173.
36. Czeizel AE, Puho EH, Langmar Z, et al. Possible association of folic acid supplementation during pregnancy with reduction of preterm birth: a population-based study. *Eur J Obstet Gynecol Reprod Biol.* 2010;148:135–140.
37. Fekete K, Berti C, Trovata M, et al. Effect of folate intake on health outcomes in pregnancy: a systematic review and meta-analysis on birth weight, placental weight and length of gestation. *Nutr J.* 2012;11:75
38. Charles DHM, Ness AR, Campbell D, et al. Folic acid supplements in pregnancy and birth outcome: re-analysis of a large randomized controlled trial and update of Cochrane review. *Paediatr Perinat Epidemiol.* 2005;19:112–124.
39. Baumslag N, Edelstein T, Metz J. Reduction of incidence of pre-maturity by folic acid supplementation in pregnancy. *BMJ.* 1970;1:16–17.
40. IOM (Institute of Medicine). *Dietary Reference Intakes for Calcium and Vitamin D*. Washington, DC: The National Academies Press; 2011.
41. Hacker AN, Fung EB, King JC. Role of calcium during pregnancy: maternal and fetal needs. *Nutr Rev.* 2012;70:397–409.
42. Oliveri B, Parisi MS, Zeni S, et al. Mineral and bone mass changes during pregnancy and lactation. *Nutrition.* 2004;20:235–240.
43. Sowers MF, Scholl TO, Harris L, et al. Bone loss in adolescent and adult pregnant women. *Obstet Gynecol.* 2000;96:189–193.
44. Wisser J, Florio I, Neff M, et al. Changes in bone density and metabolism in pregnancy. *Acta Obstet Gynecol Scand.* 2005;84:349–354.
45. Sandler RB, Slemenda CW, LaPorte RE, et al. Postmenopausal bone density and milk consumption in childhood and adolescence. *Am J Clin Nutr.* 1985;42:270–274.
46. Bass S, Delmas PD, Pearce G, et al. The differing tempo of growth in bone size, mass, and density in girls is region specific. *J Clin Invest.* 1999;104:795–804.
47. Cho GJ, Shin JH, Yi KW, et al. Adolescent pregnancy is associated with osteoporosis in postmenopausal women. *Menopause.* 2012;19:456–460.
48. Scholl TO, Hediger ML, Schall JI. Maternal growth and fetal growth: pregnancy course and outcome in the Camden Study. *Ann NY Acad Sci.* 1997;817:281–291.
49. Sowers MF, Wallace RB, Lemke JH. Correlates of forearm bone mass among women during maximal bone mineralization. *Prev Med.* 1985;14:585–596.
50. Belizan JM, Villar J. The relationship between calcium intake and edema-, proteinuria-, and hypertension-gestosis: an hypothesis. *Am J Clin Nutr.* 1980;33:2202–2210.
51. Buchler HC, Guyatt GH, Cook RJ, et al. Effect of calcium supplementation on pregnancy-induced hypertension and preeclampsia. *JAMA.* 1996;275:1113–1117.
52. Levine RJ, Hauth JC, Curet LB, et al. Trial of calcium to prevent preeclampsia. *N Engl J Med.* 1997;337:69–76.
53. Villar J, Abdel-Aleem H, Merialdi M, et al. World Health Organization randomized trial of calcium supplementation among low calcium intake pregnant women. *Am J Obstet Gynecol.* 2006;194:639–649.
54. Hofmeyr GJ, Lawrie TA, Atallah AN, et al. Calcium supplementation during pregnancy for preventing hypertensive disorders and related problems. *Cochrane Database Syst Rev*. 2010;8:CD001059.
55. Christesen HT, Falkenberg T, Lamont RF, et al. The impact of vitamin D on pregnancy: a systematic review. *Acta Obstet Gynecol Scand.* 2012;91:1357–1367.
56. Bodnar LM, Catov JM, Simhan HN, et al. Maternal vitamin D deficiency increases the risk of preeclampsia. *J Clin Endocrinol Metab.* 2007;92:3517–3522.

57. Wei SQ, Audibert F, Hidiroglou N, et al. Longitudinal vitamin D status in pregnancy and the risk of pre-eclampsia. *BJOG.* 2012;119:832–839.
58. Steingrimsdottir L, Gunnarsson O, Indridason OS, et al. Relationship between serum parathyroid hormone levels, vitamin D sufficiency, and calcium intake. *JAMA.* 2005;294:2336–2341.
59. Poel YHM, Hummel P, Lips P, et al. Vitamin D and gestation diabetes: a systematic review and meta-analysis. *Eur J Intern Med.* 2012;23:465–469.
60. Mannion CA, Gray-Donald K, Koski KG. Association of low intake of milk and vitamin D during pregnancy with decreased birth weight. *CMAJ.* 2006;174:1273–1277.
61. Brunvand L, Quigstad E, Urdal P, et al. Vitamin D deficiency and fetal growth. *Early Hum Dev.* 1996;45:27–33.
62. Scholl TO, Chen X. Vitamin D intake during pregnancy: association with maternal characteristics and infant birth weight. *Early Hum Dev.* 2009;85:231–234.
63. De-Regil LM, Palacios C, Ansary A, et al. Vitamin D supplementation for women during pregnancy (*Review)*. *Cochrane Database Syst Rev.* 2012;8:CD008873.
64. Morley R, Carlin JB, Pasco JA, et al. Maternal 25–hydroxyvitamin D and parathyroid hormone concentrations and offspring birth size. *J Clin Endo Metab.* 2006;91:906–912.
65. Merewood A, Mehta SD, Chen TC, et al. Association between vitamin D deficiency and primary cesarean section. *J Clin Endocrinol Metab.* 2009;94:940–945.
66. Scholl TO, Chen X, Stein TP. Maternal vitamin D status and delivery by cesarean (Camden Study). *Nutrients.* 2012;4:319–330.
67. Gahche J, Bailey R, Burt V, et al. *Dietary Supplement Use Among U.S. Adults Has Increased Since NHANES III (1988–1994).* NCHS data brief, No 61. Hyattsville, MD: National Center for Health Statistics; 2011.
68. Fawzi WW, Msamanga GI, Spiegelman D, et al. Randomised trial of effects of vitamin supplements on pregnancy outcomes and T cell counts in HIV-1-infected women in Tanzania. *Lancet.* 1998;351:1477–1482.
69. Fall CH, Fisher DJ, Osmond C, et al. Multiple micronutrient supplementation during pregnancy in low-income countries: a meta-analysis of effects on birth size and length of gestation. *Food Nutr Bull.* 2009;30:S533–S546.
70. Haider BA, Bhutta ZA. Multiple-micronutrient supplementation for women during pregnancy. *Cochrane Database Syst Rev.* 2006;18:CD004905.
71. Catov JM, Bodnar LM, Olsen J, et al. Periconceptional multivitamin use and risk of preterm or small-for-gestational-age births in the Danish National Birth Cohort. *Am J Clin Nutr.* 2011;94:906–912.
72. Shah PS, Ohlsson A. Effects of prenatal multimicronutrient supplementation on pregnancy outcomes: a meta-analysis. *CMAJ.* 2009;180:E99–E108.
73. Nordeng H, Bayne K, Havnen GC, et al. Use of herbal drugs during pregnancy among 600 Norwegian women in relation to concurrent use of conventional drugs and pregnancy outcome. *Complement Ther Clin Pract.* 2011;17:147–151.
74. Broussard CS, Louik C, Honein MA, et al. Herbal use before and during pregnancy. *Am J Obstet Gynecol.* 2010;202:443.e1–443.e6.
75. Forster DA, Denning A, Wills G, et al. Herbal medicine use during pregnancy in a group of Australian women. *BMC Pregnancy Childbirth.* 2006;6:21.
76. Johns T, Sibeko L. Pregnancy outcomes in women using herbal therapies. *Birth Defects Res B Dev Reprod Toxicol.* 2003;68:501–504.
77. Heitmann K, Nordeng H, Holst L. Safety of ginger use in pregnancy: results from

a large population-based cohort study. *Eur J Clin Pharmacol.* 2013;69(2):269–277.
78. Butt MS, Sultan MT. Ginger and its health claims: molecular aspects. *Crit Rev Food Sci Nutr.* 2011;51:383–393.
79. Wen-jing R, Mao-de L, Jian-guang Z. Anticancer effects of Chinese herbal medicine, science or myth? *J Zhejiang Univ Sci B.* 2006;7:1006–1014.
80. Bie YY. Combination of Chinese and western medicine in the treatment of hyperemesis gravidarum in 49 patients. *J Chinese Prac Med*. 2004;4:20. http://journal.9med.net/qikan/guokanll_zzxq.php?id=6&eid=1095
81. Shi GY. Application research of treating ecotopic pregnancy by the Shapei Huayurecipe plus mifepristone. *Chinese J Clin Med*. 2012;4:23–25.
82. Luo SY. The drink of Bushenantai treatment of 56 cases of threatened abortion. *China Modern J Obstet Gynecol*. 2004;1:160–161.
83. Moussally K, Bérard A. Exposure to herbal products during pregnancy and the risk of preterm birth. *Eur J Obstet Gynecol Reprod Biol*. 2010;150:107–108.
84. Marcus DM, Snodgrass WR. Do no harm: avoidance of herbal medicines during pregnancy. *Obstet Gynecol*. 2005;105:1119–1122.
85. Coletta JM, Bell SJ, Roman AS. Omega-3 fatty acids and pregnancy. *Rev Obstet Gynecol.* 2010;3:163–171.
86. Velzing-Aarts FV, van der Klis FR, van der Dijs FP, et al. Effect of three low-dose fish oil supplements, administered during pregnancy, on neonatal long-chain polyunsaturated fatty acid status at birth. *Prostaglandins Leukot Essent Fatty Acids.* 2001;65:51–57.
87. Innis SM, Friesen RW. Essential n-3 fatty acids in pregnant women and early visual acuity maturation in term infants. *Am J Clin Nutr.* 2008;87:548–557.
88. Lucas M, Dewailly E, Muckle G, et al. Gestational age and birth weight in relation to n-3 fatty acids among Inuit (Canada). *Lipids.* 2004;39(7):617–626.
89. Olsen SF, Sørensen JD, Secher NJ, et al. Randomised controlled trial of effect of fish-oil supplementation on pregnancy duration. *Lancet.* 1992;339:1003–1007.
90. Smuts CM, Huang M, Mundy D, et al. A randomized trial of docosahexaenoic acid supplementation during the third trimester of pregnancy. *Obstet Gynecol.* 2003;101:469–479.
91. Szajewska H, Horvath A, Koletzko B. Effect of n-3 long-chain polyunsaturated fatty acid supplementation of women with low-risk pregnancies on pregnancy outcomes and growth measures at birth: a meta-analysis of randomized controlled trials. *Am J Clin Nutr.* 2006;83:1337–1344.
92. Olsen SF, Secher NJ, Tabor A, et al. Randomised clinical trials of fish oil supplementation in high risk pregnancies. Fish Oil Trials In Pregnancy (FOTIP) Team. *BJOG.* 2000;107:382–395.
93. Din JN, Newby DE, Flapan AD. Omega 3 fatty acids and cardiovascular disease—fishing for a natural treatment. *BMJ.* 2004;328:30–35.
94. People's League of Health. Nutrition of expectant and nursing mothers. *Lancet.* 1942;240;10–12.
95. Koletzko B, Cetin I, Brenna JT, et al. Dietary fat intakes for pregnant and lactating women. *Br J Nutr.* 2007;98:873–877.
96. *What You Need to Know About Mercury in Fish and Shellfish. U.S. Food and Drug Administration.* http://www.fda.gov/food/foodsafety/product-specificinformation/seafood/foodbornepathogenscontaminants/methylmercury/ucm Updated November 21, 2011. Accessed December 17, 2012.

Human Growth and Disease in Later Life

26

David J.P. Barker, MD, PhD, FRS

PROGRAMING

There is now clear evidence that the pace and pathway of early growth and development are major risk factor for the development of chronic diseases. The list of chronic disease that are now suspected of being initiated during early development includes coronary heart disease and type 2 diabetes, which are the focus of this chapter. The list, however, now extends to a wide range of chronic disorders, including certain cancers and chronic infections. This has led to a new "developmental" model for the origins of disease that proposes that nutrition during fetal life, infancy, and early childhood change gene expression and thereby establish functional capacity, metabolic competence, and responses to the later environment, a phenomenon known as "programing."[1–4]

To explore the developmental model for chronic disease required studies of a kind that had not hitherto been carried out. It was necessary to identify groups of men and women now in middle or late life whose size at birth had been recorded at the time. Their birth weight could thereby be related to the later occurrence of disease. In the county of Hertfordshire, UK, from 1911 onwards, when women had their babies they were attended by a midwife, who recorded the birth weight. A health visitor went to the baby's home at intervals throughout infancy, and the weight at 1 year was recorded. Figure 26-1, based on 15, 726 men and women born in the county, shows that mortality from coronary heart disease fell progressively across the normal range of birth weight.[5] In Figure 26-1, mortality rates in each birth weight group are expressed as a percentage of national rates in England and Wales. Similarly to the United States, there is a large

variation in rates between different counties; and Hertfordshire is one of the groups of counties in the south and east that have below average rates. Table 26-1 shows findings for a sample of men in Hertfordshire who had glucose tolerance tests.[6] The percentage with impaired glucose tolerance or type 2 diabetes fell steeply with increasing birth weight. The association between low birth weight and coronary heart disease has now been replicated among men and women in Europe, North America, and India.[7–12] Low birth weight has been shown to predict altered glucose tolerance in studies of men and women around the world.[13–17]. The associations between low birth weight and later disease depend on slow fetal growth rather than premature birth. Among the different cohorts that have been assembled around the world in order to study programing the Helsinki Birth Cohort has some of the most detailed information. The cohort comprises 20,000 men and women who were born in Helsinki, Finland, during 1924–1944. Each of them has detailed birth records and school health records document their growth and health in childhood. Those born after 1933 have records of their growth from birth to seven years, the age of school entry. Health care is free in Finland and any serious illness within the cohort has been documented through the National Hospital Discharge Register. Deaths and their causes are shown in the National Mortality Register. Information about occupation and income is available through the five-year national censuses. Use of medication for chronic illness is also recorded.

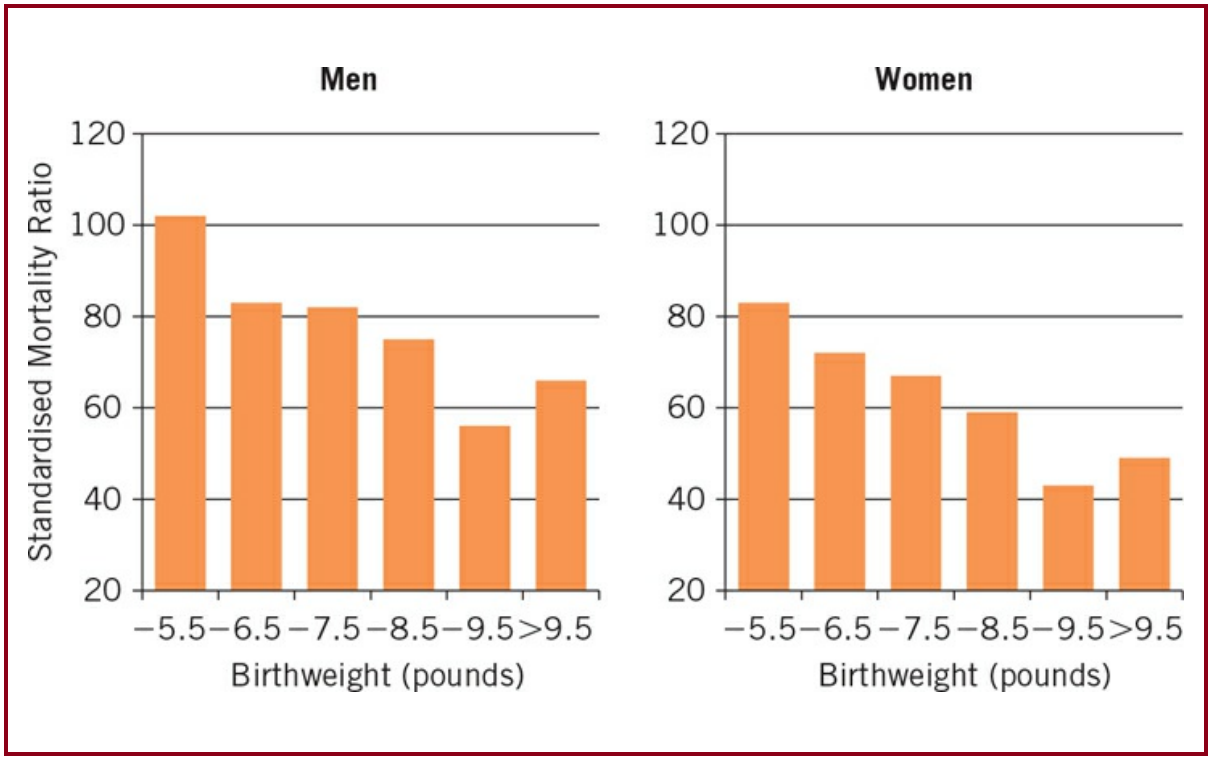


Figure 26-1 Mortality from coronary heart disease in 15,726 men and women in Hertfordshire.

TABLE 26-1 Percentage of Men Aged 64 Years With Impaired Glucose Tolerance or Diabetes According to Weight at Birth in 370 Men in Hertfordshire

Weight (lb)	% of Men With 2-h Glucose of ≥ 7.8 mmol/L	Odds Ratio (95% CI)[a]
≤5.5	40	6.6 (1.5–28)
–6.5	34	4.8 (1.3–17)
–7.5	31	4.6 (1.4–16)
–8.5	22	2.6 (0.8–8.9)
–9.5	13	1.4 (0.3–5.6)
9.5	14	1.0
P for trend	<0.001	

[a] Adjusted for current body mass index.

BIOLOGICAL BASIS

Like other living creatures in their early life human beings are "plastic" and able to adapt to their environment.[18] The development of the sweat glands provides a simple example of this. All humans have similar numbers of sweat glands at birth but none of them function. In the first three years after birth, a proportion of the glands become functional, depending on the temperature to which the child is exposed. The hotter the conditions, the greater the number of sweat glands that are programed to function. After three years, the process is complete and the number of sweat glands is fixed. Thereafter, the child who has experienced hot conditions will be better equipped to adapt to similar conditions in later life, because people with more functioning sweat glands cool down faster.

This brief description encapsulates the essence of developmental plasticity: a critical period when a system is plastic and sensitive to the environment, followed by loss of plasticity and a fixed functional capacity. For most organs and systems, the critical period occurs in utero. Among the major organs only the brain, liver, and immune system remain plastic. There are good reasons why it may be advantageous in evolutionary terms for the body to remain plastic during development. It enables the

production of phenotypes that are better matched to their environment than would be possible if the same phenotype was produced in all environments. Developmental plasticity is defined as the phenomenon by which one genotype can give rise to a range of different physiological or morphological states in response to different environmental conditions during development.[19] A baby's responses to malnutrition include slowing of growth and altered metabolism, which enable it to survive. As Mellanby noted long ago, the ability of a human mother to nourish her baby is partly determined when she herself is *in utero*, and by her childhood growth.[20] The human fetus therefore responds not only to conditions at the time of the pregnancy but to conditions a number of decades before.

Until recently, we have overlooked a growing body of evidence that systems of the body that are closely related to adult disease, such as the regulation of blood pressure, are also plastic during early development. In animals, it is surprisingly easy to produce lifelong changes in the physiology and metabolism of a fetus by minor modifications to the diet of the mother before and during pregnancy.[21–23]

The different size of newborn human babies exemplifies plasticity. The growth of babies has to be constrained by the size of the mother, otherwise, normal birth could not occur. Small women have small babies: in pregnancies after ovum donation they have small babies even if the woman donating the egg is large.[24] Babies may be small because their growth is constrained in this way or because they lack the nutrients for growth. As McCance wrote, "The size attained *in utero* depends on the services which the mother is able to supply. These are mainly food and accommodation."[25] Research into the developmental origins of disease has focused on the nutrient supply to the baby, while recognizing that other influences, such as hypoxia, stress, and maternal size also influence fetal growth.[26] The availability of nutrients to the fetus is influenced by the mother's nutrient stores and metabolism, as well as by her diet during pregnancy. In developing countries many babies are undernourished because their mothers are chronically malnourished. Despite current levels of nutrition in Western countries, the nutrition of many fetuses and infants remains suboptimal because the nutrients available are unbalanced or because their delivery is constrained by maternal metabolism. Globally, size at birth in relation to gestational age is a marker of fetal nutrition.[26]

DEVELOPMENTAL ORIGINS HYPOTHESIS

The developmental origins hypothesis proposes that a range of chronic diseases, including coronary heart disease, stroke, hypertension, and type 2 diabetes, together with certain cancers and chronic infections, originate in developmental plasticity, in response to under nutrition during fetal life and infancy.[2,3] Why should fetal responses to under nutrition lead to disease in later life? The general answer is clear: "life history theory", which embraces all living things, states that during development there is never enough resource to perfect every trait. If a plant in your garden seeks more moisture by growing deeper roots, it will do so at the expense of its stem and leaves. In humans, increased allocation of energy to one trait, such as brain growth necessarily reduces allocation to one or more other traits, such as tissue repair processes. Smaller babies, who have had a lesser allocation of energy, must incur higher costs and these, it seems, include disease in later life. A more specific answer to the question is that people who were small at birth are vulnerable to later disease through three kinds of process. First, they have less functional capacity in key organs, such as the kidney: one theory holds that hypertension is initiated by the reduced number of nephrons, the functional units of the kidney that are found in people who were small at birth.[27] A second process is the setting of hormones and metabolism. An undernourished baby may establish a "thrifty" way of handling food. Insulin resistance, which is associated with low birth weight, may be viewed as persistence of a fetal response by which blood glucose concentrations were maintained for the benefit of the brain but at the expense of glucose transport into the muscles and muscle growth.[28]

A third link between low birth weight and later disease is that people who were small at birth are more vulnerable to adverse environmental influences in later life. Observations on animals show that the environment during development permanently changes not only the body's structure and function but also its responses to environmental influences encountered in later life.[19] Table 26-2 shows the effect of low income in adult life on coronary heart disease among men in Helsinki.[29] As expected, men who had a low taxable income had higher rates of the disease. There is no agreed explanation for this but the association between poverty and coronary heart disease is a major component of the social inequalities in health in many western countries. Among the men in Helsinki, however, the association was confined to one quarter of the men who had slow fetal growth and were thin at birth, defined by a index (birth weight/length^3) of less than 26 kg/m^3 (Table 26-2). Among men who were not thin at birth,

coronary heart disease was not associated with income, which implies that these men, the majority, were resilient to the effects of low income.

TABLE 26-2 Hazard Ratios (95% CI) for Coronary Heart Disease in 3629 Men in Helsinki According to Ponderal Index at Birth (Birth Weight/Length^3) and Household Income in Adult Life

Household Income Pounds Sterling per Year	Ponderal Index ≤ 26.0 kg/m^3 (*n* = 1475)	Ponderal Index < 26.0 kg/m^3 (*n* = 2154)
<15,700	1.00	1.19 (0.65–2.19)
15,700	1.54 (0.83–2.87)	1.42 (0.78–2.57)
12,400	1.07 (0.51–2.22)	1.66 (0.90–3.07)
10,700	2.07 (1.13–3.79)	1.44 (0.79–2.62)
≤8,400	2.58 (1.45–4.60)	1.37 (0.75–2.51)
P for trend	<0.001	0.75

One explanation for these findings emphasizes the psychosocial consequences of a low position in the social hierarchy, as indicated by low income and social class, and suggests that perceptions of low social status and lack of success lead to changes in neuro endocrine pathways and hence to disease.[30] The findings in Helsinki seem consistent with this. People who were small at birth are known to have persisting alterations in responses to stress, including raised serum cortisol concentrations.[31] It is suggested that persisting small elevations of cortisol concentrations for many years may have effects similar to those seen when tumors lead to more sudden, large increases in glucocorticoid concentrations. People with Cushing's syndrome, the result of over activity of the adrenal cortex, are insulin-resistant and have raised blood pressure, both of which predispose to coronary heart disease.

INFANT AND CHILDHOOD GROWTH

Table 26-2 shows the early growth of men and women who were either admitted to hospital with coronary heart disease or died from it.[32] They belong to a cohort of 8760 people who were born in Helsinki, Finland during 1934–1944. Their mean height, weight, and body mass index (BMI,

weight/height^2) at each month from birth to 2 years of age, and at each year from 2 to 11 years of age, are expressed as standard deviations (*z*-scores). The mean *z*-score for the cohort is set at zero and a child maintaining a steady position as tall or short, or fat or thin, in relation to other children would follow a horizontal path on the figure. At birth the mean body size of the boys who developed coronary heart disease in their later lives was approximately 0.2 standard deviations below the average and they were thin. Between birth and two years of age, mean *z*-scores for each measurement fell, so that at two years the boys were thin and short. After 2 years of age their *z*-scores for body mass index began to increase and continued to do so. In a simultaneous regression, both low body mass index at birth and at 2 years of age, and high body mass index at 11 years of age were associated with later coronary events.[32] This suggests that growth during fetal life, infancy, and childhood each contribute to the genesis of coronary heart disease.

As with the boys, the mean body size of the girls who later had coronary events was below the average (Figure 26-2). They tended to be short at birth rather than thin, but their mean *z*-scores for body mass index fell progressively after birth so that, like the boys, they were thin at 2 years of age, though not short. After 4 years of age their *z*-scores for weight and body mass index began to increase and continued to do so, reaching the average at approximately 8 years of age, and exceeding it thereafter. Similarly to the boys, in a simultaneous regression body size at each of the three ages, birth two and eleven years were independently associated with later coronary events.[32]

In Table 26-3 findings for boys and girls have been combined to show the simultaneous effect of birth weight and body mass index at 2 years of age, divided into thirds, on hazard ratios for coronary events. The highest hazard ratios are among subjects with birth weights below 3.0 kg and body mass indices at 2 years of age of 17 kg/m^2 or less. Table 26-4 shows the simultaneous effects of body mass index at 2 and 11 years of age. The highest hazard ratios were among people with body mass indices below 16 kg/m^2 at 2 years of age and above 17.5 kg/m^2 at 11 years of age. The hazard ratios in Table 26-3 and Table 26-4 were little changed if they were adjusted for socio-economic status or income in adult life; and they are a further demonstration of that coronary heart disease is associated with both prenatal and postnatal growth.[33]

The rapid weight gain after the age of 2 years that characterizes the growth of children who later develop coronary heart disease (Figure 26-2)

is thought to reflect “compensatory” growth.

If the growth of a fetus, infant or child falters because of malnutrition or other adversity it has the ability, once the adversity has ceased, to return to its growth trajectory by accelerated growth. The ability to mount rapid “compensatory” growth following growth faltering is common in animals and familiar to farmers. It necessarily has costs. If energy is allocated to rapid growth, the allocation to some other developmental activity must be reduced. In animals, compensatory growth has a wide range of physiological and metabolic costs that include reduced quality of tissues and organs, such as bone, heart, kidneys, and premature death. Little is known about these costs in humans.[3]

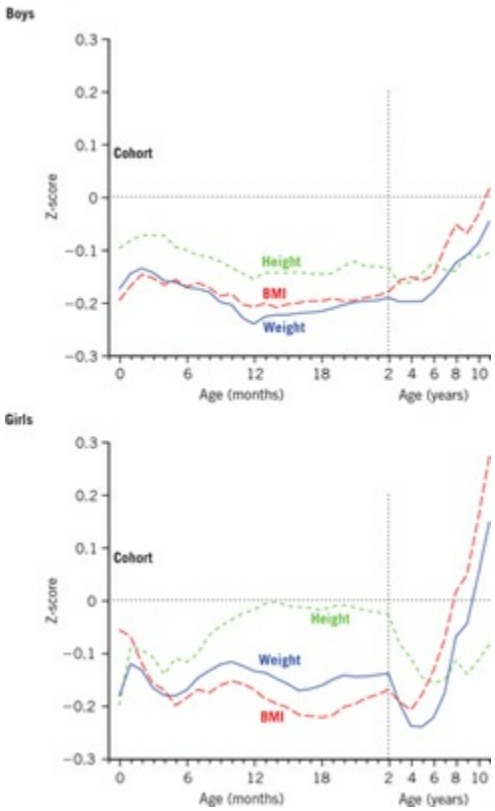


Figure 26-2 Mean *z*-scores for height, weight, and body mass index in the first 11 years after birth among boys and girls who had coronary heart disease as adults. The mean values for all boys and all girls are set at zero, with deviations from the mean expressed as standard deviations (*z*-scores).

TABLE 26-3 Hazard Ratios (95% confidence

intervals) for Coronary Heart Disease According to Birth Weight and Body Mass Index (BMI) at 2 Years of Age for Boys and Girls Combined

	BMI at Age 2 (kg/m^2)		
Birth Weight (kg)	**<16**	**16–17**	**>17**
<3.0	1.9 (1.3–2.8)	1.9 (1.2–3.0)	1.3 (0.7–2.2)
3.0–3.5	1.5 (1.0–2.1)	1.6 (1.1–2.2)	1.2 (0.8–1.8)
>3.5	1.7 (1.2–2.5)	1.5 (1.1–2.2)	1.0

TABLE 26-4 Hazard Ratios (95% confidence intervals) for Coronary Heart Disease According to Body Mass Index (BMI) at 2 and 11 Years of Age for Boys and Girls Combined

	BMI at Age 11 (kg/m^2)		
BMI at Age 2 (kg/m^2)	**<16**	**16–17.5**	**>17.5**
<16	1.6 (0.8–3.3)	2.4 (1.2–4.9)	3.0 (1.4–6.3)
16–17	1.4 (0.7–3.1)	1.6 (0.8–3.3)	1.9 (0.9–3.9)
>17	1.0	1.3 (0.6–2.7)	1.1 (0.5–2.3)

One explanation of the associations between coronary heart disease and small body size at birth and thinness at 2 years of age is that babies who are thin or short at birth and during infancy lack muscle, a deficiency that will persist into childhood as there is little cell replication in muscle after around one year of age.[34] Rapid weight gain in childhood may lead to a disproportionately high fat mass in relation to muscle mass. This could underlie the strong associations between low birth weight, low body mass index at 2 and high body mass index at 11 and later insulin resistance, which was found when a sub-sample of 2,003 subjects in the Helsinki cohort were examined at the age of 62 years.[32]

TYPE 2 DIABETES AND HYPERTENSION

Through their lives people who were small at birth remain biologically different to people who were large: these differences include an increased

susceptibility to type 2 diabetes and hypertension. Table 26-5 shows odds ratios for these two disorders according to birth weight and fourths of body mass index at age 11 years.[33] The two disorders are associated with the same general pattern of growth as coronary heart disease.[32] Risk of disease falls with increasing birth weight and rises with increasing childhood body mass index. The Helsinki Birth Cohort allows estimation of the strength of these effects.[33] If each individual in the Helsinki Birth Cohort had been in the highest third of birth weight and had decreased their standard deviation score for body mass index between ages 3 and 11 years, the incidence of type 2 diabetes would have been halved. This demonstrates the potential power of interventions during development.

The associations between low birth weight and type 2 diabetes shown in Table 26-5 have been found in other studies.[6,13–17] The association with hypertension has also been found in other studies.[35] There is a substantial literature showing that birth weight is associated with differences in insulin sensitivity and blood pressure within the normal range.[6,13,17,36] These differences are found in children and adults but they tend to be small. A 1kg difference in birth weight is associated with around 3mmHg difference in systolic pressure. The contrast between this small effect and the large effect on hypertension (Table 26-5) suggests that structural or functional changes that are associated with poor fetal growth, and that tend to elevate blood pressure after birth, have a small influence on blood pressure within the normal range because counter-regulatory mechanisms maintain normal levels. Over time, however, as the initial changes progress with age, these mechanisms are no longer able to maintain homeostasis. This may initiate a cycle of rising blood pressure, further progression of the initial changes and further rise in blood pressure.[27,37] Rapid increase in body size after birth may exacerbate glomerular injury because greater body size leads to increased excretory load and glomerular hyperfiltration.[38] Direct evidence in support of this has come from a study of the kidneys of people killed in road accidents. Those being treated for hypertension had fewer, but larger, glomeruli.[39]

TABLE 26-5 Odds Ratios (95% confidence intervals) for Type 2 Diabetes and Hypertension According to Birth Weight and Body Mass Index (BMI) at Age 11 Years

	BMI at Age 11 (kg/m^2)

Birth Weight (kg)	<15.7	–16.6	–17.6	>17.6
Type 2 diabetes (698 cases)				
<3.0	1.3 (0.6–2.8)	1.3 (0.6–2.8)	1.5 (0.7–3.4)	2.5 (1.2–5.5)
−3.5	1.0 (0.5–2.1)	1.0 (0.5–2.1)	1.5 (0.7–3.2)	1.7 (0.8–3.5)
−4.0	1.0 (0.5–2.2)	0.9 (0.4–1.9)	0.9 (0.4–2.0)	1.7 (0.8–3.6)
>4.0	1.0	1.1 (0.4–2.7)	0.7 (0.3–1.7)	1.2 (0.5–2.7)
Hypertension (2997 cases)				
<3.0	2.0 (1.3–3.2)	1.9 (1.2–3.1)	1.9 (1.2-3.0)	2.3 (1.5–3.8)
−3.5	1.7 (1.1–2.6)	1.9 (1.2–2.9)	1.9 (1.2-3.0)	2.2 (1.4–3.4)
–4.0	1.7 (1.0–2.6)	1.7 (1.1–2.6)	1.5 (1.0-2.4)	1.9 (1.2–2.9)
>4.0	1.0	1.9 (1.1–-3.1)	1.0 (0.6-1.7)	1.7 (1.1–2.8)

FETAL NUTRITION

Size at birth is the product of the fetus's trajectory of growth, which is set at an early stage in development, and the mater no-placental capacity to supply sufficient nutrients to maintain this trajectory. A rapid trajectory of growth increases the fetus's demand for nutrients.[26] This demand is greatest late in pregnancy but the trajectory is thought to be primarily determined by genetic and environmental effects in early gestation. Experiments in animals have shown that alterations in maternal diet around the time of conception can change the fetal growth trajectory.[22] The sensitivity of the human embryo to its environment is being increasingly recognized with the development of assisted reproductive technology.[40] The trajectory of fetal growth is thought to increase with improvements in periconceptional nutrition, and is faster in male fetuses. The consequent greater vulnerability of male fetuses to under-nutrition may contribute to the shorter lives of men.[41]

MATERNAL NUTRITION

The graded relation between birth weight and later disease (Figure 26-1)

implies that variations in the supply of food from normal healthy mothers to normal healthy babies have major implications for the long-term health of the babies.[4] A baby does not depend on the mother's diet during pregnancy: that would be too dangerous a strategy. Rather it lives off her stored nutrients and the turnover of protein and fat in her tissues.[42] These are related to her body composition and therefore reflect her lifetime nutrition.

Studies in Europe and India have shown that high maternal weight and adiposity are associated with the development of insulin deficiency, type 2 diabetes, and coronary heart disease in the offspring.[43–45] There is also an increasing body of evidence showing that low maternal weight, body mass index and skin fold thickness are associated with insulin resistance and raised blood pressure in the offspring.[46–52] One of the metabolic links between maternal body composition and birth size is protein turnover. Women with a low lean body mass have low rates of protein turnover in pregnancy.[53]

Although a mother's diet during pregnancy is not closely linked to the birth weight of her baby it can program the baby. Follow-up studies of people who were in utero during the war-time famine in Holland have shown that, although the babies' birth weights were little affected, the severe maternal caloric restriction at different stages of pregnancy was variously associated with obesity, dyslipidemia, insulin resistance and coronary heart disease in the offspring.[46] In these studies maternal rations with a low protein density were associated with raised blood pressure in the adult offspring.[54] This adds to the findings of studies in Aberdeen and Mother well, UK, which showed that maternal diets with either a low or a high ratio of animal protein to carbohydrate were associated with raised blood pressure in the offspring during adult life.[47,55] While it may seem counterintuitive that a high-protein diet should have adverse effects, these findings are consistent with the results of controlled trials of protein supplementation in pregnancy, which show that high protein intakes are associated with reduced birth weight.[56] One possibility is that these adverse effects are a consequence of the metabolic stress imposed on the mother by an unbalanced diet in which high intakes of essential amino acids are not accompanied by the micronutrients required to utilize them.

THE ROLE OF THE PLACENTA

A baby's birth weight depends not only on the mother's nutrition but also

on the placenta's ability to transport nutrients to it from its mother. The placenta seems to act as a nutrient sensor regulating the transfer of nutrients to the fetus according to the mother's ability to deliver them, and the demands of the fetus for them.[57] The weight of the placenta, and the size and shape of its surface, reflect its ability to transfer nutrients. The shape and size of the placental surface at birth has become a new marker for chronic disease in later life.[58] The predictions of later disease depend on combinations of the size and shape of the surface and the mother's body size. Particular combinations have been shown to predict coronary heart disease,[59] hypertension,[60] chronic heart failure[61] and certain forms of cancer.[62] The placenta is the most varied of all human organs. Variations in placental size and shape reflect variations in the normal processes of its development, including implantation, unplugging of the spiral arteries, and the growth and compensatory expansion of the chorionic plate(Figure 26-3). These variations are accompanied by variations in nutrient delivery to the fetus. Figure 26-3 also shows potential effects of exposure to maternal hormones during gestation, but this is outside the scope of this review.

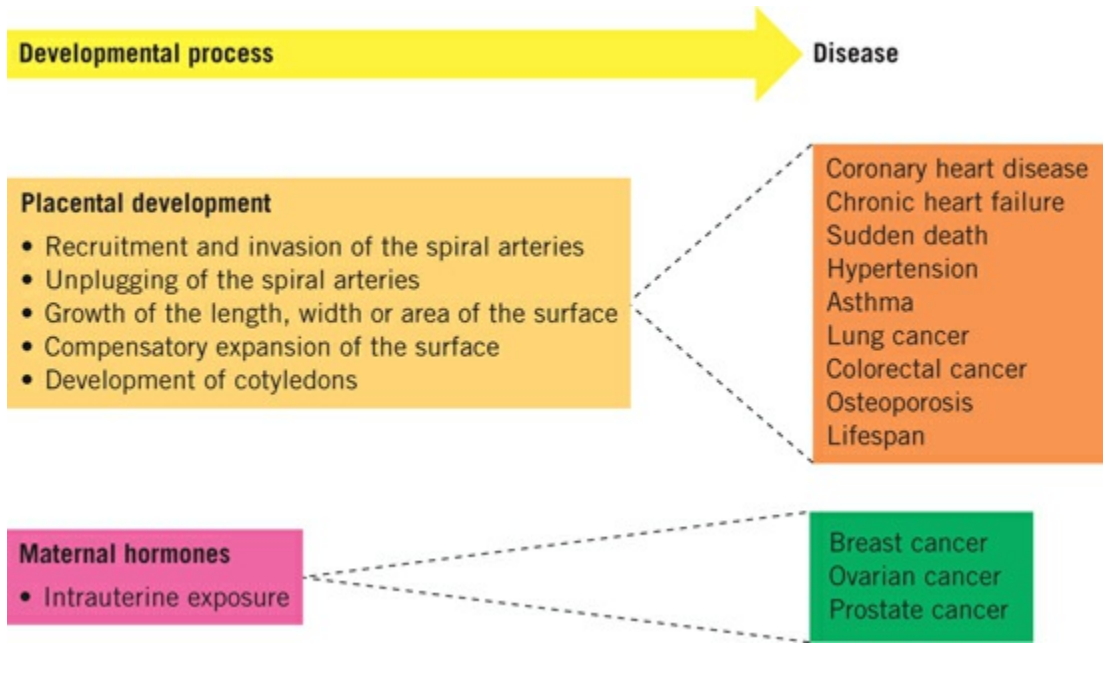


Figure 6-3 Variations in the normal processes of development and later chronic disease.

TABLE 26-6 Mean Systolic Blood Pressure (mmHg)

of Men and Women Aged 50, Born After 38 Weeks Completed Weeks of Gestation, According to Placental Weight and Birth Weight					
	Placental Weight, lb (g)				All
Birth Weight, lb (kg)	≤1.0 (454)	–1.25 (568)	–1.5 (681)	>1.5 (681)	
−6.5 (2.9)	149	152	151	167	152
−7.5 (3.4)	139	148	146	159	148
>7.5 (3.4)	131	143	148	153	149
All	144	148	148	156	149

Table 26-6 shows the systolic pressures of a group of men and women who were born at term in Preston, UK.[63] They are grouped according to their birth weight and placental weight. As expected systolic pressures fall between those with low and high birth weight. In addition the pressures increase with increasing placental weight. People with a mean systolic pressure of 150 mmHg or more, a level used to define hypertension, comprise a group who as babies were relatively small in relation to the size of their placentas. In Table 26-6, the fall in pressures of 10 mmHg across the range of birth weight is statistically opposed by the rise of 12 mmHg associated with increasing placental weight. These large trends are concealed when all pressures at a given birth weight are combined as in the right-hand column.

In studies of children and adults, the association between placental enlargement and raised blood pressure has been inconsistent.[64] Animal studies offer a possible explanation of this. In sheep the placenta enlarges in response to moderate under nutrition in mid-pregnancy.[65] This is thought to be an adaptive response to extract more nutrients from the mother. It is not, however, a consistent response but occurs only in ewes that were well nourished when they conceived. In the Helsinki, birth cohort people who had both hypertension and type 2 diabetes had low placental weight at birth, while those with hypertension alone had large placental weight in relation to birth weight.[65] These statistically opposing trends may also explain why some studies have failed to find associations between placental weight and later blood pressure.

PATHWAYS TO DISEASE

New studies, especially the Helsinki studies with their detailed information on child growth and socioeconomic circumstances, increasingly suggest that the pathogenesis of chronic disease may depend on a series of interactions occurring at different stages of development.[66] To begin with, the effects of the genes acquired at conception may be conditioned by the early environment. Table 26-7 is based on a study of 476 elderly people in Helsinki.[67] It shows mean fasting plasma insulin concentrations according to which of two polymorphisms of the peroxisome proliferators-activated receptor (PPAR)-γ gene is present. The Pro12Pro polymorphism is known to be associated with insulin resistance, indicated by elevated fasting plasma insulin concentrations. Table 26-7 shows, however, that this effect occurs only among men and women who had low birth weight. Conversely, low birth weight has been consistently linked to later insulin resistance,[28] but Table 26-7 shows that this effect occurs only among people with the Pro12Pro polymorphism. As birth weight serves as a marker of fetal nutrition,[26] this gene birth weight interaction may reflect a gene-nutrient interaction during development. The rise of the science of epigenetics is confirming that the expression of genes is contingent on the environment.

The effects of the intrauterine environment on later disease are conditioned not only by events at conception but also by events after birth. Table 26-5 shows how the effects are conditioned by childhood body mass index. Table 26-2 shows that the effects of low ponderal index at birth are conditioned by living conditions in adult life. Table 26-8, based on findings in the Helsinki Birth Cohort, shows how the effects of low birth weight on later hypertension are conditioned by living conditions in childhood, indicated by the occupational status of the father.[68] Among all the men and women, low birth weight was associated with an increased incidence of hypertension, as has been shown before.[36] This association, however, was present only among those who were born into families where the father was a laborer or of lower middle class.

TABLE 26-7 Mean Fasting Insulin Concentrations (pmol/L) in Elderly People in Helsinki According to PPAR-γ Gene Polymorphism and Birth Weight

	Birth Weight (kg)

	<3.0	3.0–3.5	<3.5	*P* for Trend
Pro12Pro	84 (56)[a]	71 (161)	65 (107)	0.003
Pro12Ala/Ala12Ala	60 (37)	60 (67)	65 (48)	0.31
P for difference	0.008	0.02	0.99	

[a] Figures in parentheses are number of subjects.

TABLE 26-8 Cumulative Incidence (%) of Hypertension According to Birth Weight and Father's Social Class in 8760 Men and Women in Helsinki

	Father's Social Class			
Birth Weight (g)	**Laborer**	**Lower Middle Class**	**Upper Middle Class**	***P* for Trend**
<3000	22.2	20.2	10.5	0.002
−3500	18.8	15.2	10.6	<0.001
−4000	14.5	12.5	10.3	0.04
>4000	11.1	15.6	15.7	0.11
P for trend	<0.001	0.05	0.79	

It seems that the pathogenesis of chronic disease cannot be understood within a model in which risks associated with adverse influences at different stages of life add to each other. Rather, disease is the product of branching paths of development. The environment triggers the branching. The pathways determine the vulnerability of each individual to what lies ahead.[38,69]

A clinical study within the Helsinki Birth Cohort showed that two different paths of fetal, infant, and childhood growth preceded the development of hypertension in adult life. In one, which was associated with more severe hypertension in people who tended to be overweight, small body size at birth and during infancy were followed by rapid weight gain, so that at age 11 years the children's body size was around the average. This is the same path of growth that leads to insulin resistance and coronary heart disease (Figure 26-2). In the other path of growth, which was associated with less severe hypertension, slow linear growth in utero and during infancy were followed by persisting small body size so that at age eleven years the children were short and thin. A similar path of growth leads to stroke.[70] One possible process underlying this is that slow growth is associated with impaired development of the cerebral

vasculature during a period of rapid brain growth or with altered lipid metabolism and the development of an atherogenic liver profile.[71] The two different paths of growth may lead to hypertension through different biological mechanisms and may produce two groups of patients who respond differently to medication.

We are beginning to understand the processes through which different paths of development initiate hypertension.[38] The changes occur at different levels and include allocation of stem cells and alteration of gene expression in the embryo, changes in renal growth, and alteration in haemostatic set-points that control blood pressure. These changes can make the affected systems more vulnerable to disruptive influences in postnatal life, which include rapid weight gain, oxidative stress, environmental stress, and a high salt intake.

CONCLUSION

Under the new developmental model for the origins of chronic disease, the causes to be identified are linked to normal variations in the processes of development that lead to variations in the supply of nutrients to the baby. These variations program the function of a few key systems that are linked to chronic disease-the immune system, anti-oxidant defences, inflammatory responses, the number and quality of stem cells, neuro endocrine settings, and the balance of the autonomic nervous system. There is not a separate cause for each different disease. Rather one cause can have many different disease manifestations. Which chronic disease originates during development may depend more on timing than on qualitative differences in exposures.

Exploration of the developmental model will illuminate people's differing responses to the environment through their lives. As René Dubos wrote long ago "The effects of the physical and social environments cannot be understood without knowledge of individual history."[72] The model will also illuminate geographical and secular trends in disease. Because human growth has changed over the past two hundred years so different chronic diseases have risen and then fallen, to be replaced by other diseases.[73,74] Much remains to be done. We are only at the beginnings of an understanding of the costs of compensatory growth in humans. Our knowledge of the structure and function of the normal human placenta is fragmentary.

Coronary heart disease, type 2 diabetes, breast cancer, and other chronic diseases are unnecessary. Their occurrence is not mandated by

genes passed down to us through thousands of years of evolution. Chronic diseases are not the inevitable lot of humankind. They are the result of the ever-changing pattern of human development. We could readily prevent them, had we the will to do so. Prevention of chronic disease and an increase in healthy aging require improvement in the nutrition of girls and young women. Many babies in the womb in the western world today are receiving unbalanced and inadequate diets. Many babies in the developing world are malnourished because their mothers are chronically malnourished. Protecting the nutrition and health of girls and young women should be the corner stone of public health. Not only will it prevent chronic disease but also it will produce new generations who have better health and wellbeing through their lives.

REFERENCES

1. Barker DJP, Osmond C, Winter PD, et al. Weight in infancy and death from ischaemic heart disease. *Lancet*. 1989;2:577–580.
2. Barker DJP. Fetal origins of coronary heart disease. *BMJ*. 1995;311:171–174.
3. Barker DJ. Human growth and chronic disease: a memorial to Jim Tanner. *Ann Hum Biol*. 2012:39:335–341.
4. Jackson AA. All that glitters. *Br Nutr Foundation Nutr Bull*. 2000;25:11–24.
5. Osmond C, Barker DJP, Winter PD, et al. Early growth and death from cardiovascular disease in women. *BMJ*. 1993;307:1519–1524.
6. Hales CN, Barker DJP, Clark PMS, et al. Fetal and infant growth and impaired glucose tolerance at age 64. *BMJ*. 1991;303:1019–1022.
7. Frankel S, Elwood P, Sweetnam P, et al. Birthweight, body mass index in middle age, and incident coronary heart disease. *Lancet*. 1996;348:1478–1480.
8. Stein CE, Fall CHD, Kumaran K, et al. Fetal growth and coronary heart disease in South India. *Lancet*. 1996;348:1269–1273.
9. Rich-Edwards JW, Stampfer MJ, Manson JE, et al. Birth weight and risk of cardiovascular disease in a cohort of women followed up since 1976. *BMJ*. 1997;315:396–400.
10. Forsén T, Eriksson JG, Tuomilehto J, et al. Mother's weight in pregnancy and coronary heart disease in a cohort of Finnish men: follow up study. *BMJ*. 1997;315:837–840.
11. Leon DA, Lithell HO, Vagero D, et al. Reduced fetal growth rate and increased risk of death from ischaemic heart disease: cohort study of 15,000 Swedish men and women born 1915–29. *BMJ*. 1998;317:241–245.
12. Forsén T, Eriksson JG, Tuomilehto J, et al. Growth in utero and during childhood among women who develop coronary heart disease: longitudinal study. *BMJ*. 1999;319:1403–1407.
13. Lithell HO, McKeigue PM, Berglund L, et al. Relation of size at birth to non-insulin dependent diabetes and insulin concentrations in men aged 50–60 years. *BMJ*. 1996;312:406–410.
14. McCance DR, Pettitt DJ, Hanson RL, et al. Birth weight and non-insulin dependent diabetes: thrifty genotype, thrifty phenotype, or surviving small baby genotype? *BMJ*. 1994;308:942–945.

15. Forsén T, Eriksson J, Tuomilehto J, et al. The fetal and childhood growth of persons who develop type 2 diabetes. *Ann Intern Med*. 2000;133:176–182.
16. Rich-Edwards JW, Colditz GA, Stampfer MJ, et al. Birthweight and the risk for type 2 diabetes mellitus in adult women. *Ann Intern Med*. 1999;130:278–284.
17. Newsome CA, Shiell AW, Fall CHD, et al. Is birthweight related to later glucose and insulin metabolism? A systematic review. *Diabet Med*. 2003;20:339–348.
18. Barker DJP. *Mothers, Babies and Health in Later Life*. Edinburgh, UK: Churchill Livingstone; 1998.
19. West-Eberhard MJ. *Developmental Plasticity in Evolution*. Oxford, UK: Oxford University Press; 2003.
20. Mellanby E. Nutrition and child-bearing. *Lancet*. 1933;2:1131–1137.
21. Widdowson EM, McCance RA. The effect of finite periods of under nutrition at different ages on the composition and subsequent development of the rat. *Proc R Soc Lond B Biol Sci*. 1963;158:329–342.
22. Kwong WY, Wild A, Roberts P, et al. Maternal under nutrition during the pre-implantation period of rat development causes blastocyst abnormalities and programming of postnatal hypertension. *Development*. 2000;127:4195–4202.
23. Gluckman P, Hanson M, eds. *Developmental Origins of Health and Disease*. Cambridge, UK: Cambridge University Press; 2006;207–220.
24. Brooks AA, Johnson MR, Steer PJ, et al. Birth weight: nature or nurture? *Early Hum Dev*. 1995;42:29–35.
25. McCance RA. Food, growth and time. *Lancet*. 1962;2:621–626.
26. Harding JE. The nutritional basis of the fetal origins of adult disease. *Int J Epidemiol*. 2001;30:15–23.
27. Brenner BM, Chertow GM. Congenital oligonephropathy: an inborn cause of adult hypertension and progressive renal injury? *Curr Opin Nephrol Hypertens*. 1993;2:691–695.
28. Phillips DIW. Insulin resistance as a programmed response to fetal under nutrition. *Diabetologia*. 1996;39:1119–1122.
29. Barker DJP, Forsén T, Uutela A, et al. Size at birth and resilience to the effects of poor living conditions in adult life: longitudinal study. *BMJ*. 2001;323:1273–1276.
30. Marmot M, Wilkinson RG. Psychosocial and material pathways in the relation between income and health: a response to Lynch et al. *BMJ*. 2001;322:1233–1236.
31. Phillips DIW, Walker BR, Reynolds RM, et al. Low birth weight predicts elevated plasma cortisol concentrations in adults from 3 populations. *Hypertension*. 2000;35:1301–1306.
32. Barker DJP, Osmond C, Forsén TJ, et al. Trajectories of growth among children who have coronary events as adults. *N Engl J Med*. 2005;353:1802–1809.
33. BarkerDJP, Eriksson JG, Forsen T, et al. Fetal origins of adult disease: strength of effects and biological basis. *Int J Epidemiol*. 2002;31:1235–1239.
34. Widdowson EM, Crabb DE, Milner RDG. Cellular development of some human organs before birth. *Arch Dis Child*. 1972;47:652–655.
35. Curhan GC, Chertow GM, Willett WC, et al. Birth weight and adult hypertension and obesity in women. *Circulation*. 1996;94:1310–1315.
36. Huxley RR, Shiell AW, Law CM. The role of size at birth and postnatal catch-up growth in determining systolic blood pressure: a systematic review of the literature. *J Hypertension*. 2000;18:815–831.
37. Ingelfinger JR. Is microanatomy destiny? *N Engl J Med*. 2003;348:99–100.
38. Barker DJP, Bagby S, Hanson M. Mechanisms of disease: in utero programming in the pathogenesis of hypertension. *Nat Clin Pract Nephrol*. 2006;2:700–707.

39. Keller G, Zimmer G, Mall G, et al. Nephron number in patients with primary hypertension. *N Engl J Med*. 2003;348:101–108.
40. Walker SK, Hartwick KM, Robinson JS. Long-term effects on offspring of exposure of oocytes and embryos to chemical and physical agents. *Hum Reprod Update* 2000;6:564–567.
41. Eriksson JG, Kajantie E, Osmond C, et al. Boys live dangerously in the womb. *Am J Hum Biol*. 2010;22:330–335.
42. James WPT. Long-term fetal programming of body composition and longevity. *Nutr Rev*. 1997;55:S41–S43.
43. Forsén T, Eriksson JG, Tuomilehto J, et al. Mother's weight in pregnancy and coronary heart disease in a cohort of Finnish men: follow up study. *BMJ*. 1997;315:837–840.
44. Forsén T, Eriksson J, Tuomilehto J, et al. The fetal and childhood growth of persons who develop type 2 diabetes. *Ann Intern Med*. 2000;133:176–182.
45. Fall CHD, Stein CE, Kumaran K, et al. Size at birth, maternal weight, and type 2 diabetes in South India. *Diabet Med*. 1998;15:220–227.
46. Ravelli ACJ, van der Meulen JHP, Michels RPJ, et al. Glucose tolerance in adults after prenatal exposure to famine. *Lancet*. 1998;351:173–177.
47. Shiell AW, Campbell-Brown M, Haselden S, et al. A high meat, low carbohydrate diet in pregnancy: relation to adult blood pressure in the offspring. *Hypertension*. 2001;38:1282–1288.
48. Mi Jie, Law C, Zhang K-L, et al. Effects of infant birthweight and maternal body mass index in pregnancy on components of the insulin resistance syndrome in China. *Ann Int Med*. 2000;132:253–260.
49. Margetts BM, Rowland MGM, Foord FA, et al. The relation of maternal weight to the blood pressures of Gambian children. *Int J Epidemiol*. 1991;20:938–943.
50. Godfrey KM, Forrester T, Barker DJP, et al. Maternal nutritional status in pregnancy and blood pressure in childhood. *Br J Obstet Gynaecol*. 1994;101:398–403.
51. Clark PM, Atton C, Law CM, et al. Weight gain in pregnancy, triceps skinfold thickness and blood pressure in the offspring. *Obstet Gynaecol*. 1998;91:103–107.
52. Adair LS, Kuzawa CW, Borja J. Maternal energy stores and diet composition during pregnancy program adolescent blood pressure. *Circulation*. 2001;104:1034–1039.
53. Duggleby SL, Jackson AA. Relationship of maternal protein turnover and lean body mass during pregnancy and birth length. *Clin Sci*. 2001;101:65–72.
54. Roseboom TJ, van der Meulen JH, van Montfrans GA, et al. Maternal nutrition during gestation and blood pressure in later life. *J Hypertens*. 2001;219:29–34.
55. Campbell DM, Hall MH, Barker DJP, et al. Diet in pregnancy and the offspring's blood pressure 40 years later. *Br J Obstet Gynaecol*. 1996;103:273–280.
56. Rush D. Effects of changes in maternal energy and protein intake during pregnancy, with special reference to fetal growth. In: Sharp F, Fraser RB, Milner RDG, eds. *Fetal Growth. Royal College of Obstetricians and Gynaecologists*, London, UK: Wiley & Co.; 1989:203–233.
57. Jansson T, Powell TL. Role of the placenta in fetal programming: underlyingmechanisms and potential interventional approaches. *Clin Sci (Lond)*. 2007;113:1–13.
58. Burton GJ, Barker DJP, Moffett A, Thornburg K, eds. *The Placenta and Human Developmental Programming*. Cambridge: Cambridge University Press; 2010.
59. Eriksson JG, Kajantie E, Thornburg KL, et al. Mother's body size and placental size predict coronary heart disease in men. *Eur Heart J*. 2011;32; 2297–303.

60. Barker DJP, Thornburg KL, Osmond C, et al. The surface area of the placenta and hypertension in the offspring in later life. *Int J Dev Biol*. 2010;54:525–553
61. Barker DJ, Gelow J, Thornburg K, et al. The early origins of chronic heart failure: impaired placental growth and initiation of insulin resistance in childhood. *Eur J Heart Fail*. 2010;12:819–825.
62. Barker DJ, Thornburg KL, Osmond C, et al. The prenatal origins of lung cancer. II. The placenta. *Am J Hum Biol*. 2010; 22:512–516.
63. Barker DJP, Bull AR, Osmond C, Simmonds SJ. Fetal and placental size and risk of hypertension in adult life. *BMJ*. 1990;301:259–262.
64. Whincup PH, Cook D, Papacosta O, et al. Birthweight and blood pressure: cross-sectional and longitudinal relations in childhood. *BMJ*. 1995;311:773–776.
65. McCrabb GJ, Egan AR, Hosking BJ. Maternal under nutrition during mid-pregnancy in sheep; variable effects on placental growth. *J Agric Sci*. 1992;118:127–132.
66. Eriksson J, Forsen T, Tuomilheto J, et al. Fetal and childhood growth and hypertension in adult life. *Hypertension*. 2000;36:790–794.
67. Eriksson JG, Lindi V, Uusitupa M, et al. The effects of the Pro12Ala polymorphism of the peroxisome proliferators-activated receptor-γ2 gene on insulin sensitivity and insulin metabolism interact with size at birth. *Diabetes*. 2002;51:2321–2324.
68. Barker DJP, Forsén T, Eriksson JG, et al. Growth and living conditions in childhood and hypertension in adult life: longitudinal study. *J Hypertens*. 2002;20:1951–1956.
69. Barker DJP. Birthweight and hypertension. *Hypertension*. 2006; 48:357–358.
70. Osmond C, Kajantie E, Forsén T, et al. Infant growth and stroke in adult life: the Helsinki birth cohort. *Stroke*. 2007;38:264–270.
71. Kajantie E, Barker DJP, Osmond C et al. Growth before 2 year of age and serum lipids 60 years later: the Helsinki Birth Cohort Study. *Int J Epidemiol*. 2008;37:280–289.
72. Dubos R. *Mirage of Health*. London, UK: Allen & Unwin; 1960.
73. Barker DJP. The rise and fall of Western diseases. *Nature*. 1989; 338:371–372.
74. Floud R, Fogel RW, Harris B, Hong SC. *The changing body*. Cambridge, UK: Cambridge University Press; 2011.

Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism

27

William W. Hay, Jr, MD, Laura D. Brown, MD, Stephanie R. Wesolowski, PhD, Paul J. Rozance, MD

NUTRITION AND DEVELOPMENT OF THE FETUS: CARBOHYDRATE AND LIPID METABOLISM

The rapid rates of metabolism and growth of the placenta and fetus require ready sources of carbon for energy production and energy storage. Maternal plasma glucose provides most of the carbon for placental and fetal energy requirements. As much as 20%–30% of the amino acids taken up by the fetus from the placenta may be oxidized, providing the additional carbon necessary to meet the fetal requirements for oxidation. Lactate that is produced in the placenta (mostly from glucose) also serves as a carbon source for the fetus. Twenty to forty percent of glucose carbon is deposited in growing body tissues; most of this goes to glycogen, some to protein, and some to adipose tissue. Lipid, on the other hand, appears to

be directed primarily to growth and development of adipose tissue, as there is little capacity to oxidize long-chain fatty acids in fetal tissues. Lipids also account for fetal growth through the provision of essential fatty acids supplied to developing structural membranes.

Based on experimental data from late-gestation fetal sheep and estimated data from term human fetuses, it is possible to construct reasonable estimates for the rates of carbon supply to the fetus from glucose (including products of glucose metabolism in the placenta such as lactate and fructose) and amino acids, as well as for fatty acids, and to compare these rates with requirements for energy production and storage. Such estimates are shown in Table 27-1[1–3]

TABLE 27-1 Nutrient Substrate Balance in Late Gestation Fetal Sheep and Humans

Carbon-Calorie Balance	Carbon (g/kg/d)	Calories (kcal/kg/d)
Requirement		
Accretion in carcass: nonfat (sheep)	3.2	32
Accretion in carcass: nonfat (human)	3.2	32
Accretion in carcass: fat (human)	3.5	33
Excretion as CO_2	4.4	0
Excretion as urea	0.2	2
Excretion as glutamate	0.3	2
Heat (measured as O_2 consumption)	0.0	50
Total		
Without fat (sheep)	*8.1*	*86*
With fat (human)	*11.6*	*119*
Uptake		
Amino acids (sheep and human)	3.9	45
Glucose (sheep)	2.4	17
Glucose (human)	3.7	26
Lactate (sheep)	1.4	14
Lactate (human)	1.7	21
Fructose (sheep)	1.0	7
Acetate (sheep)	0.2	3
Fatty acids (human)	1.1–2.2	17–34
Total		

Sheep	*8.9*	*86*
Human	*10.4–11.5*	*109–126*

Source: Adapted from Refs. 5–7.

CARBOHYDRATES

Sources of Glucose for the Placenta and Fetus

Maternal Glucose Supply

Maternal plasma glucose provides all of the glucose for the placenta and fetus, except under pathological conditions of prolonged maternal hypoglycemia, placental insufficiency, or fetal hypoxemia when the fetus can produce its own glucose by glycogenolysis or gluconeogenesis. The placenta also can use its own glycogen for glycolysis, but there is insufficient glucose-6-phosphatase in the placenta to produce significant amounts of glucose from placental glycogen into the fetal circulation. Qualitative evidence demonstrating that maternal glucose is the sole source of glucose for the placenta and fetus under normal conditions comes from studies in which tracers of glucose that are infused into the mother have the same specific activity or enrichment (ratio of tracer glucose to unlabeled glucose) in blood sampled from the fetus, indicating no dilution of fetal glucose with glucose other than from the maternal circulation.[4] Quantitative evidence to support these observations has come from tracer studies in the fetal sheep during fed, normoglycemic conditions.[5] In these studies, net uptake of glucose by the fetus from the placenta equals fetal glucose utilization; that is, there is no source of glucose for fetal glucose utilization other than what comes from the mother by way of the placenta.

Fetal Glucose Production

Only after prolonged periods (several days to weeks) of decreased fetal glucose supply, producing sustained fetal hypoglycemia and hypoinsulinemia, does fetal glucose utilization exceed supply, demonstrating a new source of fetal glucose production.[6,7] However, most species appear to have sufficient functional activities of hepatic gluconeogenic enzymes in late fetal life such that they are prepared to initiate gluconeogenesis, except the rat, in which cytosolic phosphoenolpyruvate carboxykinase (PEPCK) develops only after birth.[8] Thyroid hormone is essential for gluconeogenesis, acting on both the hepatic gluconeogenic pathways and the mechanisms activating

gluconeogenesis. Thyroid hormone effects include up-regulation of catecholamine secretion and increased or maintained concentrations of enzymes necessary for glucose production such as glucose-6-phosphatase, fructose diphosphatase, and alanine aminotransferase.[9] Cortisol concentrations increase approximately 48 hours before birth and correlate with increased endogenous glucose production rates and catecholamine concentrations in fetal sheep.[10] An increase in fetal cortisol concentration, in addition to increased catecholamines, is required for activating fetal gluconeogenesis in response to maternal fasting. A variety of models have shown that normal fetal concentrations of insulin, in addition to glucose, suppress fetal glucose production, but this also requires intact hormonal regulation, including glucagon (necessary for gluconeogenesis) as well as cortisol and catecholamines (necessary for glycogenolysis).[10]

As at all stages of life, fetal glucose production also can come from glycogenolysis. The principal source of fetal hepatic glycogen is fetal plasma glucose. Nonglucose precursors such as circulating lactate and certain amino acids such as alanine, serine, and glutamine contribute to about 10% of glycogen formation (at least in the fetal sheep) by the indirect pathway of reverse glycolysis. Glycogen accumulates progressively toward term in most organs.[11] Lung glycogen content decreases late in gestation, however, presumably as a part of surfactant production, although this issue is controversial. Glycogenolysis in the fetus is rapidly activated by catecholamine infusion, glucagon injection, and hypoxemia/ischemia; all of the enzymes necessary for glycogenolysis are present early in gestation.[12]

Mechanisms of Placental Glucose Uptake and Transport

A principal action of fetal glucose production, regardless of cause or mechanism, is to divert uterine glucose supply to the placenta. As shown in (Figure 27-1),[1] an increase in fetal glucose concentration that is independent of the maternal glucose concentration can separately regulate the partition of uterine glucose uptake into glucose transfer to the fetus and uteroplacental glucose consumption. This observation demonstrates complex regulation of glucose uptake, metabolism, and transfer to the fetus by the placenta.

Maternal Glucose Concentration

Glucose enters the placenta and fetus from the maternal circulation according to concentration-dependent mechanisms, making the maternal

plasma glucose concentration the principal regulator of placental and fetal glucose supply.[1] Increasing consumption of glucose by the growing placenta and fetus tends to decrease maternal glucose concentrations as their growth increases over gestation, but this is balanced by the development of physiological maternal insulin resistance and glucose intolerance caused by certain hormones (e.g., placental lactogen, progesterone, prolactin, estrogen, and glucocorticoids) and certain competitive substrates (e.g., fatty acids) that tend to increase maternal glucose concentrations.[13] This balance is variable, but generally involves a gradual decline in maternal glucose concentration in late second trimester to early third trimester, followed by a gradual increase in late third trimester. In women who develop overt diabetes mellitus with insulin resistance and glucose intolerance during late gestation, insulin resistance appears to be the primary defect. The mechanism for this insulin resistance involves reduction in the insulin signal transduction cascade independent of hormonal influence, as well as potentially significant inhibitory effects of placental growth hormone.[14,15]

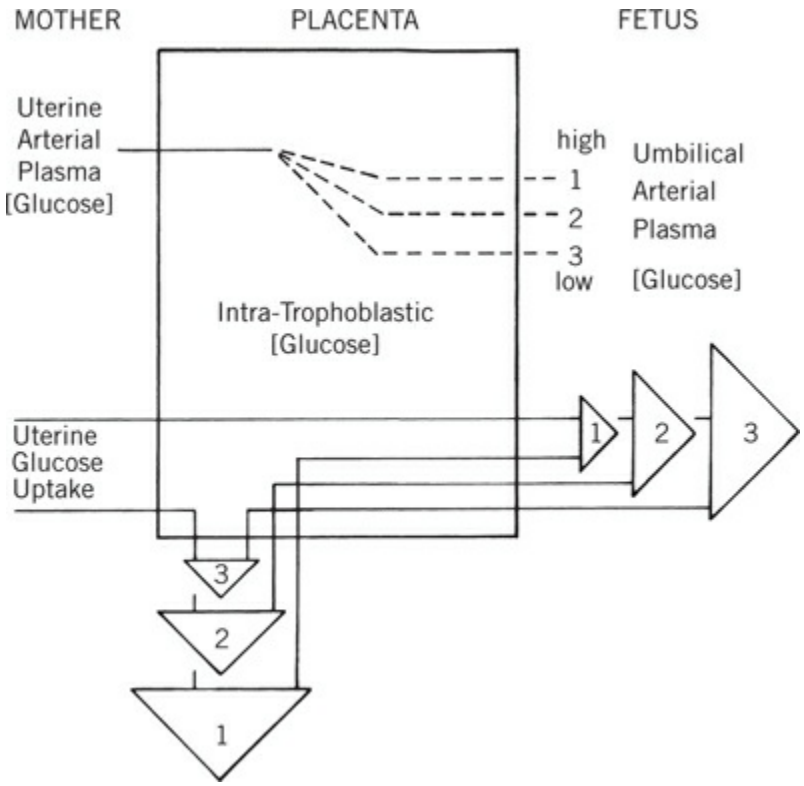


Figure 27-1 Schema of effects of fetal glucose concentration of placental glucose transfer to the fetus and uteroplacental glucose

consumption, adapted from data in sheep. A decrease in fetal glucose concentration relative to maternal glucose concentration promotes maternal-to-fetal glucose transfer at the expense of uteroplacental glucose consumption. These findings demonstrate a reciprocal relationship between placental glucose transfer and uteroplacental glucose consumption that is determined by the fetal glucose concentration. Reproduced with permission of the Nutrition Society.[1]

Placental Glucose Transporters

Glucose uptake and transport by the placenta are mediated by selective glucose transporters. Glut-1 (all species) and Glut-3 (human, rat, sheep) isoforms have been identified at maternal- facing microvillous trophoblast and fetal-facing basal trophoblast membranes. In the human placenta, Glut-1 is the principal transporter at the maternal-facing microvillous membrane, while Glut-3 predominates on the fetal-facing basal membrane or localized in extravillous trophoblast and villous cytotrophoblast cells.[16] However, quantitative distribution has varied among studies and species.[17]

Glut-4 also has been found in the human placenta, probably mostly on fibroblasts, but not consistently on either surface of the trophoblast. There is no evidence, however, that insulin in either the maternal or the fetal circulation increases placental glucose uptake or transfer to the fetus. These observations define the placenta as a nonclassical target organ of insulin regulation of glucose metabolism.

Changes in transporter expression can occur in response to changes in glycemia, particularly when the glycemic changes are chronic. For example, hyperglycemia has been shown to down-regulate the Glut-1 glucose transport system in the placenta in vitro.[18] In contrast, hyperglycemia in diabetic pregnancies that enhances fetal glucose-stimulated insulin and insulin-like growth factor (IGF)-1 production appears also to promote Glut-1 at the fetal-facing basal trophoblast membrane, thereby maintaining an increased placental glucose transport capacity.[19] Hyperglycemia itself also may promote further hyperglycemia in pregnancy. For example, pregnant women with gestational diabetes mellitus release greater amounts of tumor necrosis factor alpha (TNFω) from both the placenta and the maternal adipose tissue in response to high glucose. TNFω specifically interrupts the insulin signal transduction pathway leading to insulin resistance, glucose intolerance, and progression of hyperglycemia.[20] In other species, different patterns have been observed. For example, in the sheep, chronic hyperglycemia was

associated with reduced Glut-1 and Glut-3 concentrations and with increased glucose uptake by the uteroplacenta from the mother. There was no change in fetal glucose uptake, however. In contrast, chronic hypoglycemia decreased placental Glut-1 concentrations but did not appear to change placental Glut-3 concentrations, although uterine, uteroplacental, and fetal glucose uptake rates were decreased.[21] Thus, changes in placental Glut expression are not necessarily directly related to placental glucose transport to the fetus, which appears to exert independent control over its rate of glucose uptake.

Maternal nutritional patterns during gestation also affect the expression of placental glucose transporters. In sheep, for example, early gestational nutrient restriction decreases placental size but does not change placental weight-specific Glut-1 transporter abundance. When such sheep were re-fed later in gestation, both placental mass and Glut-1 abundance increased when fed to sufficiency but neither increased if the ewes were over-fed.[22] Nutrient restriction studies in the pregnant rat have shown no change in placental Glut-1 expression, but did show reduced Glut-3 expression. An additional study compared rodent and ovine placental Glut-1 responses to changes in glucose.[23] In the rat, six days of sustained hyperglycemia caused no change in Glut-1 protein levels, whereas three days of intrauterine growth restriction (IUGR) with fetal hypoglycemia and ischemic hypoxia decreased Glut-1 levels by 50%. In pregnant sheep, maternal and fetal hyperglycemia caused an initial threefold increase in Glut-1 with a persistent decline over the next two to three weeks, whereas maternal and fetal hypoglycemia led to a 30% to 50% decline in placental Glut-1 levels. In a rat placental cell line, high glucose concentrations decreased Glut-1 concentrations, counteracted in part by exposure to low oxygen concentrations.

Clearly, placental glucose transfer to the fetus is regulated by the combined effects of the concentrations of glucose in the maternal and fetal plasma and the kinetic function of the glucose transporter proteins at both the maternal- and fetal-facing trophoblast membranes.[24] There is also considerable need for more research into the regulation of placental glucose transporter expression and related placental glucose uptake, metabolism, and transport to the fetus, since there are demonstrable differences among isoform specificity, time dependency, and species, as well as the impact of changes in selective (glucose, amino acids) nutrient supplies.

Transplacental Glucose Concentration Gradient

Net transport of glucose from mother to fetus across the placenta requires a net maternal-fetal plasma glucose concentration gradient. In all species studied, including humans, the fetal plasma glucose concentration has been observed to be significantly lower than that measured in a simultaneously obtained blood sample from the mother.[25] In addition, a significant positive relationship has been observed between the prevailing glucose concentration in the mother and the fetus.[1] This nearly linear relationship between maternal and fetal glucose concentration covers the physiologic range of glycemia, but experimentally has been seen during acute hyperglycemia induced by glucose infusion to the mother and during hypoglycemia induced by insulin infusion to the mother.

Placental Glucose Consumption

The transplacental glucose concentration gradient is produced by both placental and fetal glucose consumption. The contribution by the placenta is large, as the placenta is a highly metabolically active organ. In pregnant sheep near term, for example, the placenta can consume almost twice as much glucose as the fetus, even though the placental/fetal weight ratio is only about 1:10.[26] Thus, a large part of uterine glucose uptake is consumed by placental glucose metabolism, which contributes significantly to the lower glucose concentrations of the fetus relative to the mother and helps to establish the transplacental glucose concentration gradient; without placental glucose consumption, fetal arterial glucose concentration would markedly increase. Glucose utilization in the placenta also contributes to placental glycogen formation via glycogenin.[27]

Placental Glucose Transport Kinetics

The relationship between the rate of placental glucose transfer to the fetus and the maternal-fetal plasma glucose concentration gradient is complex. Placental and fetal glucose consumption rates vary independently of each other. Furthermore, in vivo studies in pregnant sheep have shown that the glucose transporter proteins at both maternal and fetal placental membranes demonstrate saturation kinetics.[28] (Figure 27-2[28]). In these studies, glucose clamp experiments produced steady-state net fluxes of glucose into and across the placenta and into the fetus at different concentrations of glucose in the maternal and fetal circulations. Uptake and net consumption of glucose by the uteroplacenta demonstrated saturation kinetics with maximal uptake rate (V_{max}) occurring at the upper range of normal ovine maternal plasma glucose concentrations. Similar

results have been measured in a variety of in vitro perfusion experiments in human placentas, although V_{max} in the human placenta studied in vitro is severalfold higher than in pregnant sheep studied under normoglycemic conditions (to date, there are no in vitro, perfused placental studies in the ovine placenta).[29]

Summary of Placental Glucose Uptake, Consumption, and Transport

(Figure 27-3) summarizes the regulation and quantitative aspects of placental glucose uptake and transport. First, uterine, placental, and fetal glucose uptake are directly related to maternal glucose concentration (Figure 27-3), left panel).[29] Second, fetal glucose uptake is separately regulated by fetal glucose concentration by which a relatively lower glucose concentration in the fetus establishes a larger maternal-fetal glucose concentration gradient and thus increased transfer of glucose to the fetus (Figure 27-3, middle panel).[29] In contrast, at any maternal glucose concentration, placental glucose consumption is directly related to fetal arterial plasma glucose concentration (Figure 27-3), right panel).[29] In fact, the fetal side of the placenta has an eightfold greater capacity to take up glucose than the maternal side, so that changes in fetal glucose concentrations have a strong influence on placental glucose flux and metabolism.

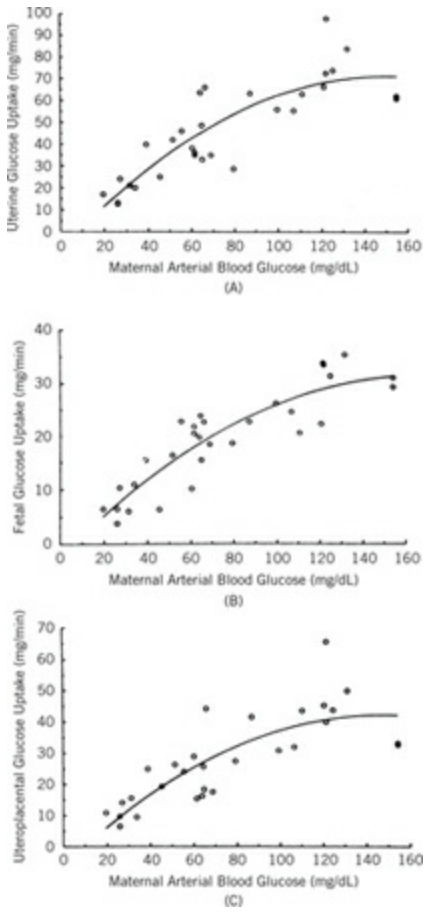


Figure 27-2 Relationship between maternal glucose concentration and the net uptake of glucose by the uterus, fetus, and uteroplacenta. Glucose was infused into pregnant sheep after an overnight fast to produce a large variety of maternal arterial blood glucose concentrations. Net glucose uptake rates by the uterus (A), fetus (B), and uteroplacenta (C) were quantified by the Fick principle. All three relationships show saturation kinetics with an approximate K_m value (a measure of the sensitivity of the transport processes to maternal glucose concentration) in the physiological range of maternal glucose concentration (about 50–60 mg/dL [2.8–3.3 mM]). These results show that maternal glucose concentration determines the entry of glucose into the uterus, uteroplacenta, and fetus. Reproduced with permission of the Society for Experimental Biology and Medicine.[28]

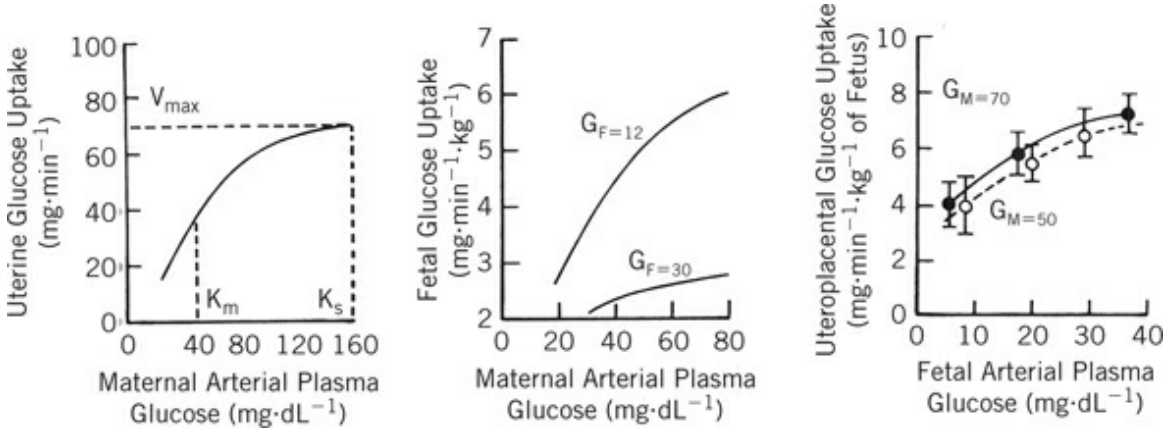


Figure 27-3 Left panel: Net rate of glucose transfer into the uterus from the maternal plasma versus maternal plasma glucose concentration. V_{max} is the maximum rate that glucose can enter the uterus; Km is one-half of V_{max} and a measure of the sensitivity of uterine glucose uptake to maternal glucose concentration; Ks is the maternal glucose concentration at which V_{max} is reached. Middle panel: Net rate of glucose transfer into the fetus from uteroplacenta versus maternal plasma glucose concentration; a lower fetal glucose concentration relative to any maternal glucose concentration produces a greater maternal-fetal glucose concentration gradient and a higher rate of placental to fetal glucose transfer (upper curve versus lower curve). Right panel: Net rate of uteroplacental glucose consumption versus fetal plasma glucose concentration; at any maternal glucose concentration (data for maternal glucose concentrations of 50 and 70 mg/dL are shown), uteroplacental glucose consumption is directly related to fetal glucose concentration. These data show that maternal glucose concentration determines the rate of glucose entry into the uterus (and thus the fetus and placenta), but the rate of uteroplacental glucose consumption is regulated by the fetal glucose concentration. Adapted from Refs. 24 and 29.

Fetal Glucose Utilization

Kinetics of Glucose Utilization in the Fetus

Placental glucose transfer to the fetus is also due to the saturation of the capacity for glucose utilization in the fetus. This capacity itself is variable, as increased fetal glucose supply from the placenta and increased fetal plasma glucose concentration act to increase fetal insulin secretion. In turn, increased fetal insulin concentration augments fetal glucose utilization and

plasma clearance, effectively limiting the increase in fetal glucose concentration as maternal glucose concentration and maternal-to-fetal glucose transfer increase.

Effects of Fetal Plasma Glucose and Insulin on Fetal Glucose Metabolism

The rate of fetal glucose metabolism (total utilization as well as the rate of fetal glucose oxidation) depends directly on the simultaneous interaction of fetal plasma glucose and insulin concentrations. An example in near-term fetal sheep depicting the effect of glucose and insulin concentrations simultaneously on fetal glucose utilization, measured as glucose oxidation to CO2, is shown in (Figure 27-4).[30] Although both glucose and insulin act independently (i.e., additively) to increase glucose utilization and oxidation in the fetus according to saturation kinetics, the relative proportion of glucose oxidized during short-term three- to four-hour studies (about 55% in fetal sheep) does not change significantly over the entire range of glucose utilized. Thus, the principal metabolic effect of insulin in the fetus is to increase the permeability of insulin-sensitive cells to glucose via increased Glut-4 translocation to cell membranes, enhancing glucose uptake and utilization in general, and oxidation of glucose in proportion to uptake and utilization, rather than to differentially affecting intracellular pathways of glucose metabolism. At least this is true on balance for the whole fetus; individual tissues and metabolic pathways may vary significantly in their responsiveness to insulin. Such a permissive role for insulin in fetal sheep might be different from the human fetus, which, like the human adult, has a greater capacity to produce fat in adipose tissue from glucose as well as fatty acids and triglycerides.

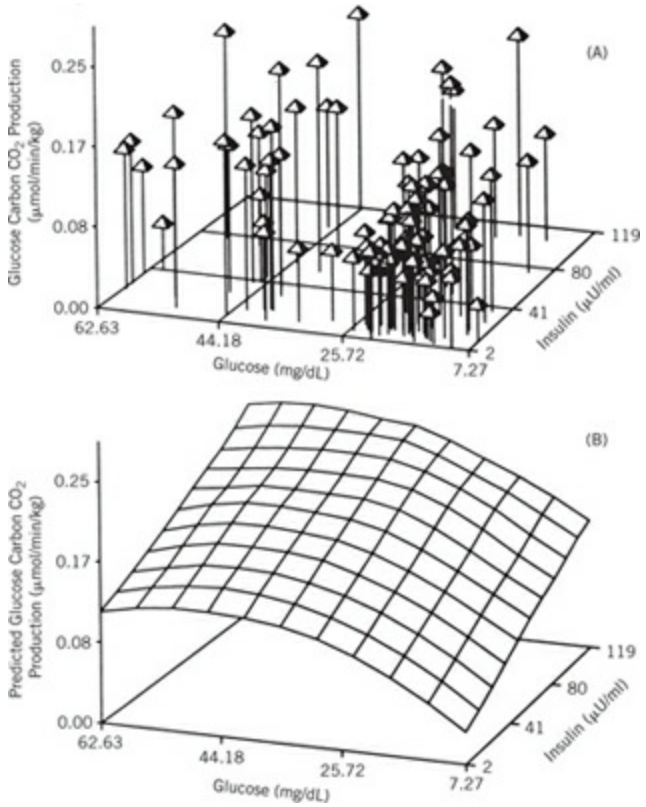


Figure 27-4 Data from fetal sheep showing the additive effects of plasma glucose and insulin concentrations on fetal glucose oxidation rate measured as CO_2 production rate. (A) Three-dimensional plot of individual values of the rate of glucose oxidation to CO_2 at different concentrations of glucose and insulin. (B) Predicted values of the rate of fetal glucose oxidation to CO_2 displayed as a three-dimensional glucose × insulin surface from the values in (A). Reproduced with permission from the American Physiological Society.[30]

Glucose Transporters and Glucose Uptake and Metabolism in the Fetus

Glucose is transported into cells in the fetus, just as in the placenta and newborn and adult, by glucose transporters. Glut-1 is the predominant fetal glucose transporter isoform and is found in abundance in most fetal tissues.[31] In contrast, tissue-specific isoforms, such as the neuronal Glut-3 in brain, Glut-2 in hepatocytes and pancreatic β-cells, and Glut-4 in insulin-responsive tissues such as skeletal muscle, heart, and adipose tissue, are expressed in low amounts in fetal tissues compared with Glut-1.[32,33]

Both acute and chronic changes in fetal glucose and insulin concentrations have been studied in fetal sheep to address the interrelationships among absolute changes in glucose and insulin concentrations, duration of change in glucose and insulin concentrations, expression of glucose transporter concentrations, and glucose utilization rates in the fetus. In chronic studies lasting two-four weeks in late gestation, sustained hyperglycemia was associated with a progressive decrease to normal or subnormal insulin concentrations.[34] Under these conditions there was a transient increase in brain Glut 1 but not Glut-3 concentrations, and a progressive decline in liver and adipose tissue Glut-1 and myocardial and skeletal muscle Glut-1 and Glut-4. Further studies showed that the chronic hyperglycemia and reduction in Glut-4 correlated with the development of insulin resistance.[35] These observations in the fetal sheep mimic results obtained in the rat fetus exposed to a maternal diabetic environment.[36] Chronic hypoglycemia (produced by maternal insulin infusion) produced a decline in brain Glut-3, an increase in brain Glut-1, and a subsequent decline in liver Glut-1, but no significant change in insulin-sensitive myocardium, skeletal muscle, or adipose tissue Glut-1 or Glut-4 concentrations. These time-dependent and tissue- and isoform-specific changes in response to altered circulating glucose and/or insulin concentrations showed that cellular adaptations in Glut-1 and Glut-3 protect the fetus from perturbations in substrate availability, and the adaptations in Glut-4 lead to the development of insulin resistance.

In acute studies, expressions of Glut-1, 3, and 4 were studied in response to increasing duration (1, 2.5, and 24 hours) of selective hyperglycemia or hyperinsulinemia.[37] Hyperglycemia initially increased fetal glucose utilization rate associated with an increase in Glut-1 and 3 isoforms that mediate basal glucose utilization, but no change in Glut-4 that mediates insulin action. Fetal glucose utilization rate subsequently returned to normal values by the end of 24 hours. These results contrast with the tissue-specific effects of selective hyperinsulinemia, which produced a sustained increase in fetal glucose utilization rate associated with a sustained increase in hepatic Glut-1 and a myocardial-specific emergence of mild insulin resistance associated with a down-regulation of Glut-4. In skeletal muscle specifically, there was a lack of sustained temporal association between the increases in transporter proteins and glucose utilization rates,[38] which indicates that subcellular localization and activity of transporter or tissues other than skeletal muscle contribute to net fetal glucose utilization rate.

These observations support an immediate but transient cellular

adaptation of augmented glucose uptake associated with a transient increase in Glut-1 concentration in response to an acute hyperglycemic perturbation in the intrauterine milieu. In contrast, alterations in the hormonal milieu (specifically of insulin) did not demonstrate any acute effects on the mechanisms underlying insulin-induced cellular glucose transport, except for a sustained increase in hepatic Glut-1 concentrations and an increase in skeletal muscle Glut-4 concentrations. Whether chronic hyperinsulinemia in the presence of hyperglycemia or euglycemia leads to adaptations of fetal ovine glucose transporter proteins toward enhancing or limiting intracellular substrate delivery remains unknown. Also unknown are the responsible mechanisms, such as Glut gene transcription or translation and posttranslational protein stability, that underlie both acute and chronic adaptation to changes in circulating glucose and insulin concentrations.

Basal Insulin Concentration

The basal concentration of insulin in the fetus probably has a role in fetal glucose utilization, but direct evidence for this is not as convincing as the effect of hyperinsulinemia. For example, an acute decrease of fetal plasma insulin concentration with somatostatin infusion does not appear to affect fetal glucose concentration or glucose utilization.[39] In contrast, a chronic selective decrease of fetal plasma insulin concentration, for example, by fetal pancreatectomy or streptozotocin injection into the fetus, reduces fetal growth and decreases umbilical glucose uptake in relation to fetal hyperglycemia. The hyperglycemia results from decreased peripheral tissue insulin sensitivity and glucose utilization and from the release of insulin's normal inhibition of hepatic glucose production.[7]

Interaction Between Glucose and Alternative Carbon Substrates for Fetal Oxidative Metabolism

At markedly reduced rates of glucose supply to the fetus, fetal glucose utilization rate decreases proportionally.[29] Under such short-term conditions, fetal oxygen consumption remains near normal,[29] indicating active reciprocal oxidation of other substrates, at least over the short term, such as glucose released from glycogen, lactate, amino acids, fatty acids, and ketoacids. Over longer periods (more than three weeks) of reduced glucose supply, fetal oxygen consumption tends to decrease by 25%–30%; because the rate of fetal growth decreases at the same time and to the same extent as the decrease in oxygen consumption, the reduction in fetal oxygen consumption probably represents the oxidative requirements of the

decreased protein synthetic activity.[40] Furthermore, when exogenous amino acids are infused into the fetal sheep, the glucose/oxygen quotient and glucose uptake rate decrease and leucine oxidation increases, further supporting fetal substrate switching, perhaps coupled with decreased placental glucose transport to the fetus.[41]

Fetal Glucose Utilization Rates

Quantitative aspects of fetal glucose utilization have been made in several large animal models. In most studies, fetal glucose utilization rate is about 5 mg/min/kg fetal weight near term, about half of values in mid-gestation when fetal growth and metabolic rate are also as much as twice as great as they are closer to term.[42] In humans, based on the increase in total maternal glucose production in late gestation, fetoplacental glucose utilization is estimated to be about 5–6 mg/min/kg fetal weight.[11] This rate is similar to that measured in normal term infants.[43] Table 27-2 presents estimated utilization rates of glucose in several fetal organs and the remaining carcass of fetal sheep in late gestation. The fraction of glucose oxidized versus the fraction entering tissue accretion pathways in these organs has not been determined, and the factors that regulate glucose uptake in specific organs remains poorly studied. Most organs appear dependent on plasma glucose concentration for their specific rate of glucose uptake, and certain organs such as skeletal muscle and probably the heart and liver are insulin sensitive (at least in sheep studied in vivo).

TABLE 27-2 Metabolic Rates in the Fetus That Account for Glucose Utilization (Based on Data in Fetal Sheep and Estimates for Human Fetuses for Brain)

	Glucose Utilization Rate (mg/min/kg fetus)	Percent of Total
Whole fetus (sheep, measured)	5.0	100
Whole fetus (human, estimated)	6.0-8.0	100
Brain (sheep, measured)	0.8	16
Brain (human, estimated)	4.0	50–67
Heart (sheep, measured)	0.65	13
Lungs (sheep, estimated)	0.1	2
Liver (sheep, measured)	0.1	2

Red blood cells (human, estimated)	0.1	2
Gut (sheep, estimated)	unknown	unknown
Carcass/skeletal muscle (sheep, estimated)	3.25	65
Total of organs accounted for:		
Sheep	5.0	100
Human	8.2	103–137

Gestational Development of Placental Glucose Transport and Fetal Glucose Utilization

The fetus grows severalfold in size over the second half of gestation, as does its absolute rate of glucose utilization. Placental glucose transfer to the fetus, therefore, must increase to meet the increasing metabolic requirements for glucose of the larger, growing fetus. The increase in placental glucose transport occurs for two reasons: (1) the transplacental glucose concentration gradient increases as the fetal glucose concentration decreases relative to maternal glucose concentration (Figure 27-5, top panel); (2) the placental transport capacity itself increases (Figure 27-5, middle panel).[42]

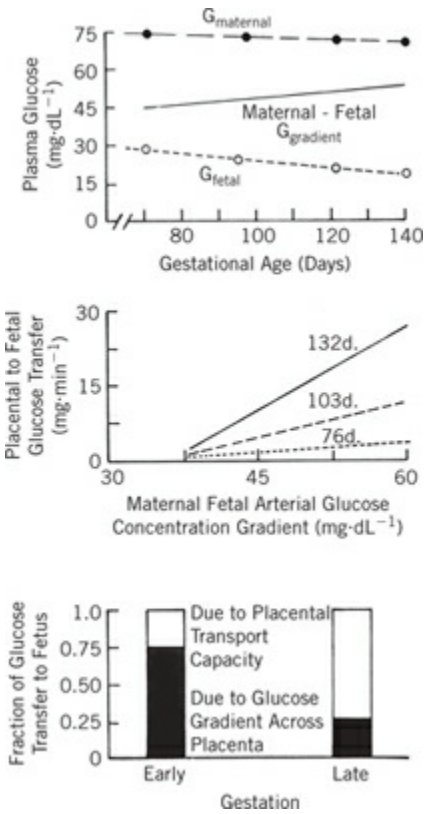


Figure 27-5 Upper panel: Plasma glucose concentration tends to decrease in fetal sheep (–O–) over the second half of gestation relative to the maternal plasma glucose concentration (–•–). This increases the maternal-fetal plasma glucose concentration gradient or the driving force for transplacental glucose transfer (—). Middle panel: Placental-to-fetal glucose transfer (PGT) increases about eightfold over this same period. This increase in transport largely reflects transport capacity, shown by the increasing slope of PGT as gestational age increases. Bottom panel: At mid-gestation, most of PGT is due to the maternal-fetal glucose concentration gradient, whereas by term, the placental transport capacity for glucose accounts for most of PGT. Adapted from Ref. 42.

Placental glucose transport capacity develops primarily by an increase in the total amount rather than the affinity of the glucose transporters.[44] The number of glucose transporter proteins can increase by an increase in the trophoblast membrane surface area, an increase in the concentration of transporters per unit membrane area, or both. There is some evidence from in vitro studies that maternal and fetal glucose concentrations can regulate placental glucose transporter expression, either directly or through IGF-I action.[45] By late gestation, glucose transport capacity by the placenta has a dominant role in the regulation of glucose transport to the fetus (Figure 27-5, bottom panel[42]).

Fetal glucose concentration may decrease relative to maternal glucose concentration over the second half of gestation for several reasons. First, fetal cellularity increases over this period, as does the fraction of body weight that consists of insulin sensitive tissues (e.g., skeletal muscle and adipose tissue).[46] Second, fetal insulin concentration increases, developmentally and in response to glucose and other secretagogues.[47] Third, insulin-sensitive Glut-4 transporters on fetal skeletal muscle and adipose tissue cell membranes also increase over gestation, at least due to the increase in the relative body content of these insulin-sensitive tissues. The result is an increase in the effectiveness of insulin to promote fetal glucose utilization and glucose clearance, thereby reducing the relative fetal plasma glucose concentration. On balance, however, by late gestation, the increasing gradient of glucose concentration across the placenta contributes only limited control over placental transport of glucose to the fetus.

Fetal Insulin Secretion

Many studies have addressed the onset and development of insulin secretion from the fetal pancreas. The fetal pancreas develops in the late first to early second trimester, producing measurable insulin concentrations by mid-gestation. As noted earlier, there is a gradual increase in basal insulin concentration and glucose- and arginine-stimulated insulin secretion toward term.[47] In the rat, very little insulin is secreted in the fetus,[48] while in the human fetus, which normally does not secrete much insulin,[49] sustained hyperglycemia in pregnant diabetics,[50] particularly when the hyperglycemia is pulsatile, produces considerable insulin. Fetal sheep are somewhere in between, showing down-regulation of glucose-induced and basal insulin secretion in the presence of chronic, sustained, marked hyperglycemia as with the rat, but increased insulin secretion with pulsatile hyperglycemia as in humans.[51] Hypoglycemia decreases basal and glucose-induced insulin secretion,[52] perhaps a fundamental aspect of how IUGR with fetal hypoglycemia decreases fetal pancreatic development and insulin secretion capacity. Clearly fetal insulin secretion responds variably to changes in glucose concentration that are dependent on the absolute change in glucose concentration, its magnitude, and its pattern.

Alternate Carbohydrates Produced in the Placenta

The placenta also contributes to fetal carbon supply by metabolizing glucose to lactate (in most or all species) and fructose (in ruminants).[1] Lactate produced in the placenta enters both maternal and fetal circulations at about equal rates under normal conditions, whereas fructose produced in the placenta only enters the fetal circulation. At normal rates of lactate and fructose uptake by the fetus and their oxidation fractions in fetal tissues measured in late-gestation pregnant sheep, these two substrates could supply carbon sufficient to account for another 20%–30% of the carbon requirements for fetal oxygen consumption above that provided by glucose.

Adaptations to Changes in Glucose Supply

Hypoglycemia (Table 27-3)

TABLE 27-3 Fetal Responses to Decreased Glucose

Supply and Hypoglycemia
Acute
Decreased glucose uptake
Decreased insulin production and secretion
Hypoinsulinemia
Decreased glucose utilization
Increase in the rate of placental glucose transfer to the fetus as fetal hypoglycemia tends to increase the maternal–fetal glucose concentration gradient
Decreased placental lactate production
Decreased fetal lactate uptake and utilization
Substitution of other substrates to main oxidative metabolism
Chronic
Initial increase in fetal glucose production, then a return toward the normal condition of little or no fetal glucose production
Initial increase in amino acid oxidation, then a return to normal rates
Decreased insulin sensitivity
Increased ratio of fetal to maternal glucose concentration
Increased ratio of placental glucose consumption to placental glucose transfer

Short-term maternal hypoglycemia has been studied in the late-gestation pregnant sheep.[53] As shown in Figure 27-6,[53] several days of fasting-induced hypoglycemia produce proportional reductions in glucose uptake by maternal nonuterine tissues, the fetus, and the uteroplacenta. Similar changes in glucose utilization by these tissues have been found with acute, several hour insulin-induced hypoglycemia.[29] At the same time, however, uteroplacental oxygen consumption rate does not change. Thus, even with hypoglycemic conditions, other substrates must substitute for glucose to maintain oxidative metabolism. As glucose utilization by the placenta is decreased, however, so is uteroplacental production of lactate (and fructose in sheep). There is little or no change in placental utilization of free fatty acids (FFAs), ketoacids, or amino acids. Thus, uteroplacental oxygen consumption can be maintained by a balance in which a decreased supply of glucose to the fetus also results in a decreased production of exported products of glucose metabolism, allowing glucose carbon to be used preferentially for oxidative metabolism.

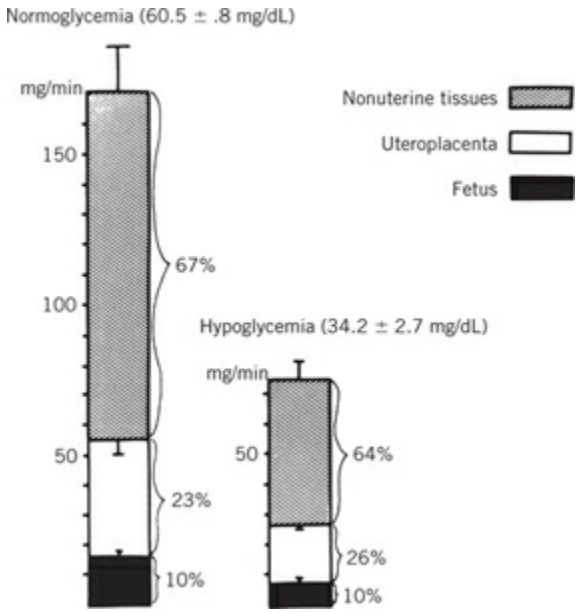


Figure 27-6 Fractional partition of maternal glucose production to nonuterine maternal tissues, uteroplacenta, and fetus in normoglycemic sheep (first column) and hypoglycemic sheep (second column) showing that in short term, there is no change in the relative proportions of glucose used by these three sets of tissues. Thus, fetal and placental glucose needs are dependent on the supply of glucose from the maternal plasma glucose pool; with longer term maternal glucose deficiency, either the fetus must use its own sources of glucose (glycogenolysis or gluconeogenesis) or rely on other substrates such as amino acids for carbon balance. Reproduced with permission from the American Physiological Society.[53]

This pattern is maintained with much longer periods of hypoglycemia, but two other adaptations develop under chronic conditions as well. First, fetal gluconeogenic gene expresion and glucose production develop.[7,41] These increase the fetal glucose concentration relative to that of the mother and shift the balance of glucose uptake by the uterus more to placental glucose consumption and less glucose transfer to the fetus (see (Figure 27-1). Fetal glucose production thus serves two purposes: (1) the maintenance of fetal glucose metabolism, and (2) the diversion of diminished glucose supply to the uterus into placental metabolism. Fetal glucose production, therefore, maintains the glucose metabolic requirements of the fetus and placenta without further drain on the diminished availability of glucose in the mother; this may represent a simple “survival” adaptation. If persistent

in neonatal and postnatal periods, however, this adaptation could have adverse consequences by promoting glucose production in excess of the capacity for glucose utilization, contributing to persistent hyperglycemia, a hallmark of type 2 diabetes.

Gluconeogenesis is regulated such that expression of PEPCK, glucose-6-phosphatase, and fructose-1, 6-bisphosphatase are normally quiescent until just prior to birth when increases in glucagon, cortisol, and catecholamines activate the glycogenolytic and gluconeogenic pathways.[54] Experiments in fetal sheep indicate similar effects of these hormones on glucose metabolism.[6,9,54,55] Insulin is the primary hormone responsible for suppression of hepatic glucose production and gluconeogenic gene expression and activation of glycolysis and lipogenesis. In the fetal sheep exposed to hypoglycemia, concentrations of the counter-regulatory hormones, cortisol, glucagon, and norepinephrine are increased, while insulin concentrations are reduced, producing a hormonal combination (e.g., decreased ratio of insulin to counter-regulatory hormones) favoring gluconeogenesis.[56,57] Importantly, an increase in circulating insulin concentration is able to suppress fetal glucose production and gluconeogenic gene expression that are induced by hypoglycemia. Insulin also increases fetal glucose utilization rates and expression of hepatic glycolytic and lipogenic genes, indicating maintained hepatic insulin action.[57]

The second adaptation to chronic hypoglycemia in late gestation is a reduction in placental and fetal growth rates, resulting in lower fetal and placental weights.[40,58] Because placental glucose transfer capacity does not change at the same time, placental weight reduction, or failure to maintain weight, does not appear to include a reduction in placental membrane surface area or glucose transporter number or affinity. This provides additional evidence that with chronic hypoglycemia and decreased placental glucose supply, placental glucose carbon is shunted to oxidative metabolism. It is interesting to speculate that all these take place to substitute reduced rates of fetal and placental growth for reduced substrate supply, maintaining normal fetal and placental tissue-specific rates of metabolism, and preserving their function and viability without further demands on maternal metabolism.

Intrauterine Growth Restriction

IUGR is a unique example of how fetal glucose metabolism and its regulatory mechanisms adapt to glucose deficiency and hypoglycemia to maintain normal fetal glucose metabolism. Regardless of the model, when

the fetus is deprived of glucose by placental insufficiency or maternal hypoglycemia, fetal weight-specific glucose utilization rate is not very much different from normal rates.[6,7,59] This indicates that one of the mechanisms to enhance glucose uptake by the fetus from the placenta is the production of further hypoglycemia, thereby increasing the maternal-fetal glucose concentration gradient. To do this, there must be an increase in the fetal tissue capacity for glucose uptake and/or utilization. This could come about by increased concentrations and/or activity and/or plasma membrane localization of glucose transporters, increased insulin signal transduction and thereby effectiveness to promote Glut-4 (and perhaps Glut-1) translocation to the cell membrane, or mechanisms of insulin metabolism into oxidative and/or nonoxidative pathways. In both fetal sheep and fetal rats with IUGR, Glut-1 and Glut-4 concentrations in the brain, liver, myocardium, adipose tissue, and skeletal muscle do not decrease with sustained hypoglycemia,[6,31,32,60] perhaps a positive adaptation to maintain glucose utilization despite hypoglycemia. It has not been determined whether this maintenance of glucose transporter concentration is sufficient by itself to maintain the glucose utilization at normal rates. Future studies are necessary to sort out this fascinating and complex adaptation to glucose deficiency among the many tissues in the fetus. Recent studies in late-gestation IUGR fetal sheep show maintained or even increased weight-specific rates of glucose utilization during hyperinsulinemic clamps, indicating increased insulin sensitivity for peripheral glucose disposal.[7,61,62]

Recent data have shown increased fetal hepatic gluconeogenic gene expression and glucose production rates in a variety of conditions that limit nutrient supply to the fetus, including the nutrient-restricted fetal baboon, hypoglycemic fetal sheep, IUGR fetal sheep liver produced from placental insufficiency (PI-IUGR), and IUGR neonatal rodents.[6,7,63–66] Understanding these fetal adaptations is important, as persistence of these fetal phenotypes into postnatal life likely contributes to the increased risk for development of uncontrolled hepatic glucose production, hepatic insulin resistance, and type 2 diabetes.[67]

As mentioned earlier, hepatic insulin action is maintained in the hypoglycemic fetus. This is in contrast to the PI-IUGR fetus that develops hepatic resistance to insulin-mediated suppression of gluconeogenic gene expression and glucose production,[7] indicating that factors other than fetal hypoglycemia in IUGR lead to hepatic insulin resistance. PI-IUGR and hypoglycemic fetuses have increased hepatic phosphorylation of cAMP

response element-binding protein and peroxisome proliferator-activated receptor gamma co-activator (*PGC1γ*) gene expression, supporting the concept that increased counter- regulatory hormone-mediated cAMP activation may drive activation of glucose production.[6,55,66] One of the major differences between the hypoglycemic and PI-IUGR models is fetal oxygenation status. Oxygenation is normal in the hypoglycemic fetus, in contrast to the PI-IUGR fetal sheep, which can have up to a ~50% reduction in fetal arterial blood oxygen content and PO_2. Despite these differences, both hypoglycemic and PI-IUGR fetuses have similar oxygen consumption rates.[65,68–70] Thus, lower oxygen values may have an effect on glucose production and insulin sensitivity in the PI-IUGR fetus rather than on the metabolic rate of oxygen consumption. Future studies are needed to measure the effect of chronic insulin infusion on umbilical venous and arterial oxygenation, umbilical blood flow, and net fetal oxygen consumption rates.

Lactate concentrations and umbilical uptake rates are similar between normal and hypoglycemic fetuses.[68] In contrast, the PI-IUGR fetus has higher lactate concentrations, raising the possibility that lactate might drive glucose production in the PI-IUGR model but not the hypoglycemic model.[7,65,66] Hepatic glycogen content is also similar between normal, hypoglycemic, and PI-IUGR fetuses, supporting gluconeogenesis rather than glycogenolysis as the source of hepatic glucose production when glucose supply is chronically reduced.[7,65,66]

Hyperglycemia (Table 27-4)

TABLE 27-4 Fetal Responses to Increased Glucose Supply and Hyperglycemia

Acute: Mild-moderate
Increased glucose uptake
Increased insulin production and secretion
Hyperinsulinemia
Increased glucose utilization
Increased fetal oxygen consumption
Mild arterial hypoxemia
Respiratory acidosis
Increased placental lactate production
Increased fetal lactate uptake and utilization

Acute: Severe
Arterial hypoxemia
Hypoinsulinemia
Increased erythropoietin
Increased fetal oxygen consumption
Metabolic acidosis
Decreased placental perfusion
Fetal demise
Chronic
Decreased insulin secretion and/or synthesis
Decreased peripheral insulin sensitivity
Increased ratio of placental glucose consumption to placental glucose transfer

Acute maternal hyperglycemia produces an increase in uteroplacental glucose uptake with saturation kinetics that parallel those of the fetus. At the same time, uteroplacental oxygen consumption rate does not change[29]; thus, increasing amounts of glucose consumed by the placenta are shunted into nonoxidative pathways. These include increased rates of lactate and fructose production, as well as increased use of glucose carbon for placental tissue, particularly glycogen.

With chronic hyperglycemia in the mother, produced by glucose infusion or experimental diabetes, similar patterns of glucose metabolism in the placenta have been observed. Fetal adaptations produce other changes as well. For example, chronic fetal hyperglycemia in experimental animals, in response to chronic, relatively constant and marked maternal hyperglycemia, eventually appears to down-regulate fetal pancreatic insulin secretion and causes pancreatic β-cell apoptosis.[71] This affects the basal rate of insulin secretion and also appears to later decrease the secretion of insulin in response to acute hyperglycemia.[72] As a result of hypoinsulinemia in the presence of hyperglycemia, fetal hyperglycemia is exaggerated, shifting the increased rate of uterine glucose uptake more into uteroplacental glucose consumption. Oxidative metabolism, measured as the uteroplacental rate of oxygen consumption, still does not change. Thus, uteroplacental glucose metabolism increasingly enters nonoxidative pathways. The particular metabolic pathways of glucose carbon under these conditions have not been explored. Placental size is not dramatically increased, suggesting that export of placental glucose carbon into lactate and fructose and perhaps other products accounts for the balance of glucose carbon not used for oxidative metabolism. Of course, glucose

carbon could substitute for the oxidation of carbon normally provided by other substrates, including ketoacids and fatty acids, but principally the amino acids. Normally, the glucose oxidation fraction in the placenta of sheep in late gestation, corrected for lactate carbon and fructose carbon export, is still only about 50%. Thus, half of placental oxidative metabolism must be supplied by other substrates. Some amino acids such as leucine and serine may be oxidized within the placenta directly as they are taken up from the maternal plasma and concentrated within the trophoblast cytosol. In contrast, glutamate is produced in the fetal liver from glutamine and then taken up by the placenta where as much as 70%–80% is oxidized.[73] It is certainly possible, therefore, that under hyperglycemic conditions, excess glucose carbon could substitute for carbon that is derived from amino acids, ketoacids, and fatty acids under normoglycemic conditions. Thus, there are clear interrelationships among a variety of carbon substrates that can act as sources of carbon for placental oxidative metabolism, and these interrelationships appear to be regulated by placental glucose supply.

LIPIDS

Placental Lipid Uptake, Metabolism, and Transfer to the Fetus

Less is known about human placental lipid uptake, metabolism, and transfer to the fetus than is known for glucose, principally because most experimental animal models, especially the sheep, demonstrate little capacity for lipid uptake, metabolism, and transport by the placenta. Also, lipid transport by the human placenta has not been studied in vivo, and there is large variation in the data obtained from human placental tissue studied in vitro. Even so, the human placenta appears to have considerable capacity for lipid transfer. Transporters for specific fatty acids, especially the long-chain polyunsaturated fatty acids (LCPUFAs), docosahexaenoic acid (DHA: 22:6 ω-3) important for structural integrity of membranes, and arachidonic acid (AA: 20:4 ω-6) important for neural and retinal development, are primarily involved. More complex pathways include lipoprotein dissociation by placental lipoprotein lipase (LPL) activity (although the exact cellular source of LPL in the placenta remains in question), triglyceride uptake and metabolism (including metabolic pathways of oxidation, chain-lengthening, synthesis, and interconversion pathways), and release into the fetal plasma as FFAs or lipoproteins.

Recent reviews summarize information about placental lipid transport and fetal lipid metabolism and accumulation.[74–76]

Importance of Placental Lipid Metabolism to Fetal Lipid Supply

The amount and type of fatty acid or complex lipid transported by the placenta varies among species-greatest in the hemochorial placenta of the human, guinea pig, and rabbit, and least in the epitheliochorial placenta of the ruminant and the endotheliochorial placenta of the carnivores. Furthermore, it is interesting that fetal fat content at term among species varies reasonably directly with the lipid transport capacity of the placenta.[77] There are many lipid substances in the plasma that are transported across the placenta, which are essential for placental and fetal development even if they do not contribute to nutritional or energy metabolism. Also, brown fat is common to all fetuses; it is essential for postnatal thermogenesis, even if the neonate is not "fat" with white adipose tissue. Furthermore, many lipid substances entering the fetus are qualitatively different from those taken up by the uterus and uteroplacenta, implying active placental metabolism of individual lipid substances. Placental lipid metabolism, therefore, is qualitatively as well as, in some cases, quantitatively important for fetal development and growth, even if the fetus does not get very fat.

Placental Uptake, Synthesis, Metabolism, and Hydrolysis of Fatty Acids

A schema of lipid uptake, metabolism, and transport in the human placenta is shown in Figure 27-7[77] After entering the placenta, fatty acids may be used for triglyceride synthesis, cholesterol esterification, membrane biosynthesis, direct transfer to the fetus, or oxidation. The most important factor that regulates the flux of lipid into the placenta and into the various pathways of transport and metabolism is the concentration of maternal plasma lipids, including FFAs and triglycerides. For example, placental triglyceride content increases in women who are fasting, who deliver preterm infants, or who have diabetes mellitus– all conditions in which maternal plasma FFA concentrations are increased.[78]

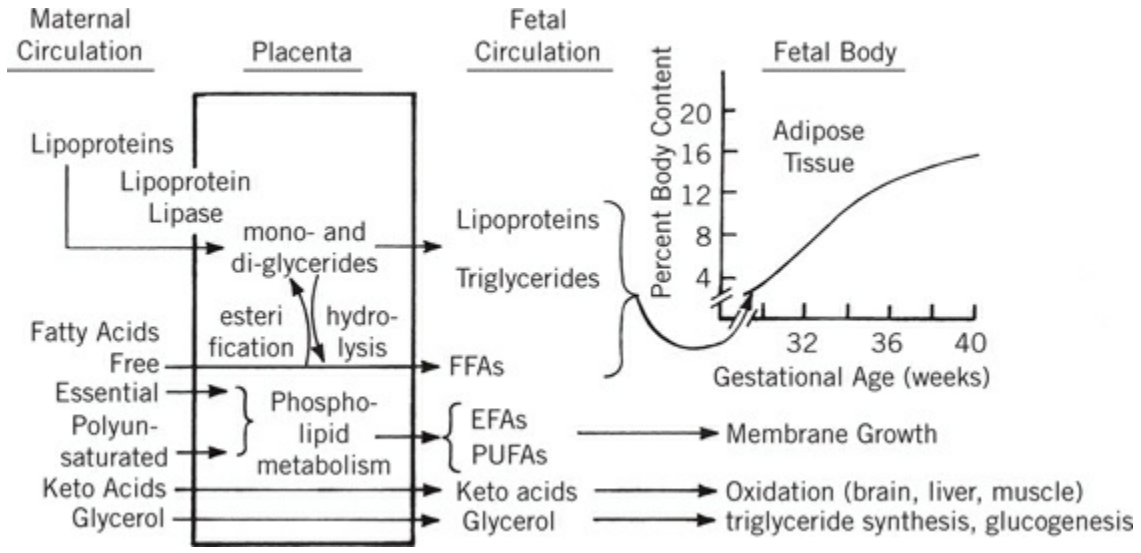


Figure 27-7 Schematic of placental–fetal interrelationships in humans for various aspects of placental lipid metabolism, fetal lipid uptake and metabolism, and fetal lipogenesis into adipose tissue. Adapted from Ref. 77.

Placental tissue from different species has been shown to express LPL activity as well as phospholipase A_2.[79,80] Maternal plasma triglycerides are hydrolyzed by these enzymes, and the released fatty acids are then taken up by the placenta. In the trophoblast cells, the fatty acids are then re-esterified and further hydrolyzed, facilitating their diffusion into the fetal circulation.

All FFAs cross lipid bilayers such as those in the syncytiotrophoblast by diffusion. Partition studies in pure phospholipid bilayers indicate that this diffusion is rapid, on the order of 20–30 milliseconds.[81] In addition, FFA transport across membranes is facilitated by fatty acid–binding proteins (FABPs), which aid in intracellular channeling of FFAs.[82] The main membrane-associated FABPs are the plasma membrane fatty acid–binding protein (FABPpm) and the FFA transfer proteins (FAT/CD36 and FATP). These transfer proteins have been found on both the maternal facing microvillous and the fetal facing basal membranes of the placental trophoblast, while a placenta-specific protein, p-FABPpm, has been found exclusively on the microvillous membrane.[83] p-FABPpm has a unique amino acid composition that enhances the binding affinity and capacity of this protein for AA and DHA compared with linolenic acid (LA) and oleic acids. The syncytiotrophoblast also contains cytoplasmic-binding proteins,

which appear to act by removing FFAs from the inner membrane and channeling them to their respective metabolic fates.[84]

Fatty acid transport into the fetal circulation is largely determined by the transplacental gradient of FFAs relative to available circulating binding protein concentrations and the activity and availability of their binding sites. In humans, fetal patterns of essential fatty acids and structural lipids correlate directly with the fatty acid/lipid composition of the maternal plasma (and, indirectly, the maternal diet).[74-76] The concentration of FFAs in the maternal plasma at term is about three times that in the fetal circulation, but the concentration of albumin, the primary carrier protein for FFAs is actually 10%–20% higher in the fetal than the maternal circulation.[85] Thus, the ratio of plasma FFAs to albumin on the fetal side of the placenta (0.3) is about 25% of that on the maternal side at term (1.3). The higher concentration of FFAs relative to available binding sites on albumin, α-fetoprotein, and other lipoproteins in the maternal compared to the fetal plasma is reflected in the concentration of unbound FFAs, which is greater in the maternal (12 nM) than in the fetal (9 nM) circulation.[86] Human cordocentesis data from 18 to 36 weeks indicates an exponential decrease in the concentration of FFAs in the fetal circulation over this period,[87] presumably because of increasing use of fatty acids in the fetus for rapidly growing structures like the brain and adipose tissue.

Although most fatty acids appear to be transported by diffusion and direct carrier-mediated mechanisms, some studies suggest that chain-altering metabolism in intratrophoblast peroxisomes may lead to a greater transfer of medium-chain fatty acids into the fetal circulation. This might be advantageous to fetal lipid metabolism, which appears to be limited in its capacity for oxidation of long-chain fats, an apparent result of low cytosolic and/or mitochondrial carnitine concentrations.[88] Fatty acids also may be esterified to di- and triglycerides and to phospholipids. These may be stored transiently or for the long term, or hydrolyzed by phospholipases and acylglycerol lipases to release fatty acids into the fetal circulation.

Additional evidence for the hypothesis that transient placental esterification of fatty acids occurs during transport of fatty acid by the placenta comes from measurements of activities of enzymes required for the isolation of glycerol-3-phosphate.[89] Such enzymes are located in the endoplasmic reticulum and have been identified in placental trophoblast cells. Although enzymatic specific activity and total enzyme content do not necessarily reflect metabolic flux, the presence of these enzymes indicates that fatty acid esterification followed by hydrolysis is a

reasonable hypothetical pathway for placental lipid metabolism and transport.

Fatty Acid and Triglyceride Synthesis in the Placenta

Fatty acids are also synthesized in the placenta. Significant fatty acid synthesis has been measured in placental tissue and cultured trophoblast cells from human,[90] sheep,[91] and pig placentas.[92] Primarily long-chain fatty acids (oleic, palmitic, and palmitoleic acids) are formed. Regarding triglycerides, radiolabeled palmitate can be incorporated rapidly into triglycerides in human placental slices, and studies of human trophoblast cells in culture have shown that incubation with albumin-bound ^{14}C-oleate produces ^{14}C-labeled cellular triglycerides. Similar observations have been made in 21-day pregnant rats studied in vivo, showing incorporation of ^{14}C-palmitate into placental triglycerides and phospholipids.[93]

Lipoprotein Synthesis in the Placenta

There is conflicting evidence regarding the synthesis of lipoproteins by placental tissue and their use for transport of fatty acids to the fetal circulation. By day 21 of gestation in the rat, the placenta appears to synthesize apoprotein B, which normally is an integral part of the major plasma lipoprotein carriers of triglycerides.[94] Such observations have not been made in the human placenta, which may secrete lipoproteins only during the early part of gestation, if at all.[95] Small amounts of low-density lipoprotein (LDL) and very-low-density lipoprotein (VLDL) have been identified in human amniotic fluid, but their source has not been determined.

Cholesterol Metabolism and Transfer by the Placenta

LDL cholesterol is taken up by endocytosis into trophoblast cells and degraded by lysozymes. LDL cholesterol appears to be the major precursor for placental production of progesterone and estrogen. high-density lipoprotein cholesterol also contributes to placental progesterone production in cultured human trophoblast cells. Some of this cholesterol is transferred directly to the fetus, although the role of this transport is uncertain, given the large capacity and rate of cholesterol synthesis in the fetal liver.[96]

In the fetal plasma, the fatty acids are bound to albumin and α-fetoprotein and transported to the liver where they are synthesized into triglycerides and released into the fetal circulation.[97] Since maternal

triglyceride concentration determines the activity of placental LPL and fatty acid transport across the placenta is concentration dependent, there is a direct relationship between maternal and fetal plasma triglyceride concentrations, even though there is no direct uptake and transfer of intact triglycerides by the placenta.

The fetal impact of maternal plasma FFA and lipid concentrations is also reflected in fetal lipid content and adipose tissue development.[97] Fatter human fetuses develop in pregnant women who have higher plasma concentrations of fatty acids and other lipids.[98,99] Experimentally, diabetic rats (induced by streptozotocin) have increased FFAs, and their fetuses have carcass and hepatic lipid contents that are increased twofold above normal. Similarly, rabbits fed oil–rich diets show increased neonatal adipose stores. In humans, umbilical venous-arterial fatty acid concentration differences in cord blood samples show that the net flux of FFAs into the fetus from the maternal circulation can account for the fetal requirement of fatty acids during the end of pregnancy. In contrast, the perfused human placenta may transfer fatty acids at a rate that can account for only about 20% of that required for the accumulation of fetal adipose tissue deposited during the third trimester. Other estimates, based on fetal lipid accumulation as well as in vitro transfer experiments, estimate that as much as 50% of fetal fatty acid requirements are transferred across the human placenta[75]; similar estimates have been made in the rat using the incorporation of ^{3}H from $^{3}H_2O$ into fatty acids, indicating that fatty acids are derived about equally from the mother and from fetal fatty acid synthesis. Estimates in the rabbit and the monkey indicate that in these species, placental fatty acid transfer across the placenta could account for all of fetal fat deposition in late gestation. Overall, therefore, it appears that there is a general relationship between the permeability of the placenta to lipids, especially FFAs, and the adiposity of the fetus at term, with human fetuses developing the most fat (15%–18% of body weight at term), guinea pigs second at about 12%, and rabbits third at about 7% (Figure 27-8).[99]

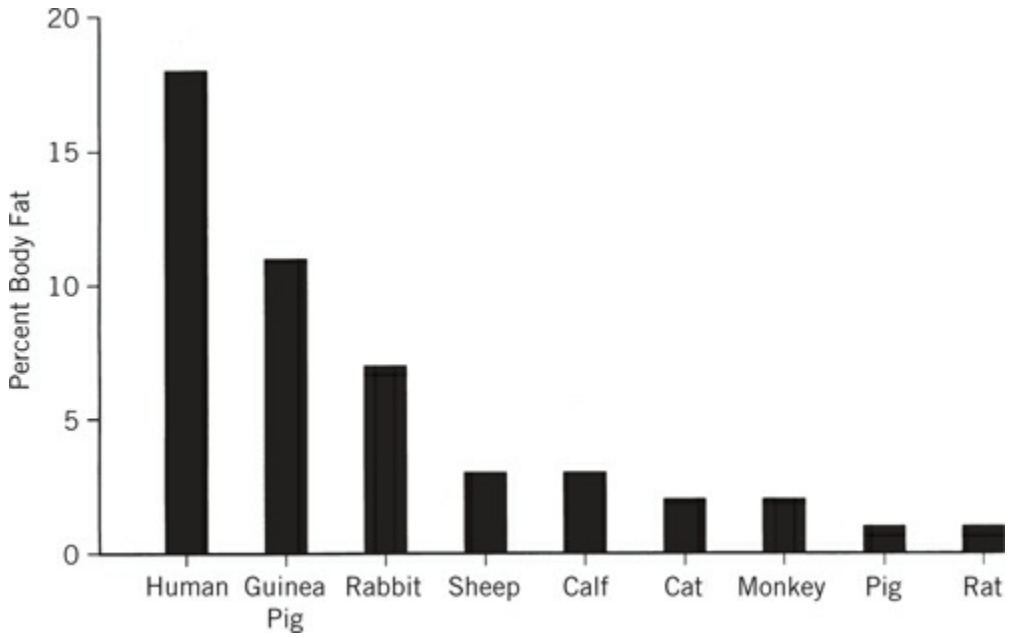


Figure 27-8 Fetal fat content at term as a percent of fetal body weight among species. Adapted from Refs. 1, 3, 77, and 99.

Availability of Essential Fatty Acids to the Fetus

Essential Fatty Acid Metabolism and Transfer by the Placenta

Essential fatty acids are readily transferred to the fetal plasma in all species studied so far. The supply of essential FFAs and especially the LCPUFAs is critical and central to the synthesis of structural lipids and, hence, to normal development of the fetus. LA (18:2, ω-6) and LA (18:3, ω-3) are the only fatty acids known to be essential for complete nutrition and must be supplied in the diet. All of the ω-6 and ω-3 fatty acid structures acquired by the fetus must, therefore, come from the mother via the placenta, either in the form of these two essential fatty acids or their LCPUFA derivatives of which AA (20:4, ω-6) and DHA (22:6, ω-3) are metabolically the most important.[100]

The preference for human placental transfer from the maternal to the fetal circulation has been reported to be docosahexaenoic>α-linolenic>linoleic>oleic>AA.[101,102] However, AA has the highest accumulation of all these fatty acids in the placenta.[103] AA uptake by placental syncytiotrophoblast membranes is highly dependent on ATP and sodium,[104] implying an active transport mechanism for this fatty acid.

Selective LCPUFA placental transfer also is due to trophoblast cellular metabolism; for example, a certain proportion of AA is converted to prostaglandins in the placenta.[101] Concentrations of AA and DHA are three to four times greater in the intervillous space of the placenta than in maternal blood, likely the result of selective LCPUFA lipase activity.[83] Also a selective incorporation of certain fatty acids into phospholipids has been found in the ovine placenta, and selective trophoblast fatty acid oxidation and lipid synthesis have been reported.[81,105,106]

Cultured trophoblast cells incubated with ^{14}C-acetate do not label AA, however, and Δ-5 and Δ-6 desaturase activities in human placental microsomes are absent at least early in gestation,[107] consistent with the absence of measurable conversion of γ-A to AA. This emphasizes the requirement for direct transfer of the essential LCPUFAs to the fetus from the maternal plasma. In fact, higher concentrations of these LCPUFAs are found in the fetus compared with the mother. These results suggest that fetal AA is either transferred directly from the mother or has a higher binding capacity in the fetal plasma, but does not exclude the possibility that some might be synthesized from LA in the fetal liver. Although the formation of AA acid and DHA from essential fatty acid precursors has been shown in term and preterm infants,[108] the degree to which the fetus is capable of fatty acid desaturation and elongation is not clear. Fetal baboons can synthesize both DHA and AA from their precursors, γ-LA and LA, respectively.[109] A low enzymatic activity of ω-5 desaturation has been proposed as one factor limiting AA synthesis,[110] and although high Δ-5 and Δ-6 desaturase activities in the liver of one 18-week and two 22-week fetuses have been reported,[103] which were close to those found in adult liver, human fetal liver desaturase-elongase chain reactions have not been clearly demonstrated in physiological conditions.

Maternal Diet and Essential Fatty Acid Supply to the Fetus

In general, reduced maternal essential fatty acid nutrition has been correlated with reduced neonatal growth and head circumference in humans.[110] Significant linear correlations between the mother and fetus or newborn have been found for both LCPUFA and essential fatty acids in healthy women eating normal, unsupplemented diets.[111] Parallel increases in plasma DHA in the mothers and newborns also have been found after fish oil supplementation during pregnancy.[112] These observations show the importance of maternal dietary fatty acids to regulate the availability of LCPUFA to the fetus and newborn. In fact, because the developing fetus

depends primarily on the maternal supply of essential fatty acids, maternal dietary supplement with LCPUFA-rich oils during the last trimester of pregnancy to increase LCPUFA levels in neonates has been advised. However, the competitive desaturation of the ω-3 and ω-6 series by Δ-6 and Δ-5 desaturases is of major significance because of their controlling role in the desaturating and elongating pathways of the parent essential fatty acids. Thus, excessive dietary intake of LA from vegetable oils, particularly safflower, sunflower, and corn oil, may inhibit Δ-6 desaturase and decrease the formation of DHA from α-LA. Similarly, AA formation is lower when excessive LA is provided, as seen in externally or parentally fed infants receiving corn or safflower oil as the predominant source of fatty.[113] Further, the inhibitory effect of eicosapentaenoic acid on Δ-5 desaturase activity has been considered responsible for the lower plasma AA found when fish oil, high in eicosapentaenoic acid and DHA, is consumed. Also, excessive fish oil intake can inhibit intake of LA from vegetable oils, particularly safflower, sunflower, and corn oil, may inhibit α-6 desaturase activity and decrease AA levels.[114] The consumption of fish oils modifies membrane phospholipid composition, increasing eicosapentaenoic and DHA concentrations at the expense of AA content. Furthermore, adverse effects of low AA concentrations in serum and red blood cell phospholipids on growth during infancy have been observed.[115] AA status in preterm infants also has been correlated with their birth weight.[116] Foods containing lipid peroxides are potentially toxic, and the higher the content of LCPUFAs in the diet, the more likely peroxidation will occur. Thus, excess intake of LCPUFAs may reduce antioxidant capacity, enhancing susceptibility to oxidative damage, a condition that has been shown to be responsible for fetal damage during pregnancy in rats.[117]

Dietary olive oil appears to protect the LCPUFA series better than fish oil.[118] Olive oil also does not affect AA concentrations and is much more resistant to lipid peroxidation. The effect of a diet supplemented with 10% fish oil versus the same amount of olive oil during pregnancy on the fatty acid profile and vitamin E concentration has been studied in the rat. A decrease in AA and α-tocopherol concentrations as well as delayed postnatal development was found in the offspring of rats fed the fish oil–rich diet.[119] This study was extended to determine whether dietary supplementation with either vitamin E or γ-LA as a precursor of AA could ameliorate these changes. AA concentrations and postnatal development indexes, but not α-tocopherol concentrations, were improved when the fish

oil diet was supplemented with γ-LA. In contrast, postnatal development indexes were not improved when the fish oil–rich diet was supplemented with sufficient exogenous vitamin E to normalize άtocopherol levels. Thus, although feeding a fish oil–rich diet during pregnancy and lactation decreased both α-tocopherol and AA concentrations, the latter deficiency rather than the former seemed to be responsible for delayed postnatal development of rat pups. In this same study, another group of pregnant and lactating rats fed the fish oil–rich diet received a supplement with AA instead of ω-LA, and although both treatments restored brain phospholipid content and AA content in the pups, improvement in growth rate and neurodevelopment indexes was more efficient in the γ-LA group. The only difference between the two groups was the absence of LA in brain phospholipids when rats were supplemented with AA, whereas it was present at a normal level in those supplemented with γ-LA.[119] These findings agree with those previously in humans fed diets rich in AA in which the proportion of LA in plasma phospholipids decreased,[120] likely a consequence of replacing LA by AA in tissues.

These observations in humans and experimental animals emphasize the exquisite sensitivity of endogenous LCPUFA metabolism to changes in maternal dietary fatty acid composition during perinatal development and the quite remarkable consequences to fetal and neonatal development. Since benefits and risks of modifying maternal fat intake in pregnancy and lactation are not yet completely established, and the safety of high intakes of LCPUFA during pregnancy is still unclear[121]; further studies are required before recommendations to increase LCPUFA intake in pregnancy can be made.

Fetal Accumulation of Essential Fatty Acids

Intrauterine requirements for LCPUFAs in the human fetus during the last trimester of fetal development through the early weeks of life have been estimated to be 400 and 50 mg/kg/day, respectively. In tissues such as the brain, where lipids constitute about 50% dry weight, almost half of the total lipid content is composed of LCPUFA. Both AA and DHA are readily incorporated into the structural lipids of the developing brain[122] where, besides their role in maintaining membrane fluidity, permeability, and conformation, they play an important functional role. For example, once released from phospholipids by the action of phospholipase A_2, AA is the main precursor for eicosanoids, prostaglandins, and leukotrienes,[74] and it is essential for fetal and neonatal growth, while DHA has a key role

in the development of the retina (both rods and cones, with up to 50% of the fatty acids in the rod outer segment as DHA) and visual function.[123,124] Although the relative rates of desaturation by rat liver and brain differ between those of adult and 10-day-old animals, the degree to which the human fetus is capable of desaturation and elongation is not clear. It is normally believed that the supply of essential fatty acids and LCPUFA is critical and central to the synthesis of structural lipids and hence to normal fetal development.[123,124]

Fetal Lipid Metabolism: Utilization

Physiological changes that develop in the fetus in late gestation and increase nutrient utilization, such as the increase in plasma insulin concentration, act to enhance net maternal-to-fetal fatty acid and lipid transport by increasing fatty acid utilization in the fetus (largely to develop adipose tissue, it appears). Increased utilization of fatty acids by fetal tissues lowers fetal plasma fatty acid concentrations relative to those in the maternal plasma and increases the maternal-to-fetal fatty acid concentration gradients.[99,102] For example, human maternal venous blood concentrations of fatty acids are directly related to the umbilical artery FFA concentrations and the umbilical vein-artery concentration differences of FFAs. Similar observations have been made in the rabbit and the guinea pig, including, in the latter, evidence that experimentally lowering the fetal fatty acid concentration relative to that in the maternal plasma independently increases fatty acid transfer across the placenta (using an in situ perfusion model). Even at the same concentrations, furthermore, naturally occurring or experimentally increased fetal plasma concentrations of albumin directly increase the transfer of fatty acids across the placenta; this observation is based on data from both in vitro perfused human placentas and in situ perfused guinea pig placentas. The effect of albumin seems to be one of simply providing increased esterification capacity in the fetal plasma to bind FFAs transferred directly across the placenta.

When the processes of fatty acid transfer to the fetus by the placenta and fatty acid incorporation into fetal tissues actually begin during gestation, and whether they occur with simultaneous development of placental fatty acid transporters and placental lipid metabolic capacity, remain to be determined, although human fetal fat accretion develops primarily in the third trimester.[3] During late gestation, maternal lipid metabolism changes to a catabolic state, as shown by increased adipose

tissue lipolysis and reduced uptake of circulating triglycerides.[125] The reduced uptake is the result of decreased adipose tissue LPL activity. There is also an overproduction of triglyceride by the liver and enhanced absorption of dietary lipids. Together, these changes in maternal lipid metabolism produce increasing concentrations of nearly all types of circulating plasma lipids as late gestation proceeds, including FFAs, glycerol, and triglyceride-rich VLDL and chylomicron particles. When the pregnant mother is well fed, plasma concentrations of ketoacids (β-hydroxybutyrate and acetoacetate) are not increased, although during fasting, when the concentrations of these substrates do increase, they also may contribute significantly to the supply of nutrient substrates to the uteroplacenta and the fetus.

Fetal Leptin

Leptin mediates a multitude of biological actions in the adult, including regulation of weight by controlling hypothalamic mechanisms involved in food intake and in regulating insulin secretion, insulin action to promote peripheral glucose utilization, and metabolic rate. Emerging evidence supports roles for leptin in the fetus as well, including enhancement of growth.[126] Umbilical cord leptin concentrations correlate with birth weight,[127] being higher in macrosomic infants of diabetic mothers,[128] and lower in infants with IUGR, indicating that the fetal metabolic and hormonal milieu independent of fetal fat content may regulate leptin synthesis and circulating concentrations. In fetal sheep, fetal white adipose tissue leptin mRNA amounts increase with fetal development and gestational age and with basal insulin concentrations.[129] Glucose infusions producing hyperglycemia and hyperinsulinemia and insulin infusions producing hyperinsulinemia alone also increased leptin mRNA, showing that insulin itself can affect fetal leptin production.[130]

Summary of Placental and Fetal Lipid Metabolism

The transport of fatty acids and other lipid substances across the placenta, and the deposition of lipids in adipose tissue, are late-gestation phenomena and highly species specific. The human hemochorial placenta transfers the most lipid and produces the fattest of all land mammals at birth. Essential fatty acid transport, however, occurs in all species and begins early in gestation, allowing membrane lipids, particularly those of neurons, to develop throughout gestation. At preterm birth, however, the human newborn is in a precarious balance with respect to lipid metabolism.

Failure to provide sufficient nonprotein energy will lead to increased rates of lipolysis and fatty acid oxidation. This may or may not result in breakdown of membrane lipids for oxidation, but certainly it produces a metabolic situation in which essential fatty acids will be oxidized along with other fatty acids to produce necessary energy in preference to membrane deposition. This could lead to deleterious reduction in the amount and structure of critical membrane development in the brain, leading potentially to abnormal neurological function and adverse long-term outcome.

ACKNOWLEDGEMENTS

This work was supported in part by grants T32 HD 007186-32 (WWH, PI and Program Director), R01 DK088139 (PJR, PI; WWH, Co-I), KO1 DK090199 (SW, PI), R24 DK090964 (SW, Co-I), Building Interdisciplinary Careers in Women's Health Research Scholar Award K12 HD-057022 (LDB, PI) and R01 HD079404 (LDB, PI) from the National Institutes of Health; Junior Faculty Career Development Awards (SW, PI and LDB, PI) from the UCDenver Center for Women's Health Research; and by Grand Challenges Explorations Grant OPP1061082 (WWH, PI) from the Discovery Program of the Bill & Melinda Gates Foundation.

REFERENCES

1. Hay WW Jr. Energy and substrate requirements of the placenta and fetus. *Proc Nutr Soc*. 1991;50:321–336.
2. Hay WW Jr., Brown LD, Regnault TRH. Fetal requirements and placental transfer of nitrogenous compounds. In: Polin RA, Fox WW, Abman SH, eds. *Fetal and Neonatal Physiology*. 4th ed. Philadelphia, PA: Elsevier (W. B. Saunders Co.); 2011:585–602.
3. Sparks JW, Girard J, Battaglia FC. An estimate of the caloric requirements of the human fetus. *Biol Neonate*. 1980;38:113–119.
4. Kalhan SC, D'Angelo LJ, Savin SM, Adam PAJ. Glucose production in pregnant women at term gestation. Sources of glucose for the human fetus. *J Clin Invest*. 1979;63:388–394.
5. Hay WW Jr, Sparks JW, Quissel BJ, et al. Simultaneous measurements of umbilical glucose uptake, fetal utilization rate, and fetal turnover rate of glucose. *Am J Physiol*. 1981;240:E662–E668.
6. Limesand SW, Rozance PJ, Smith D, Hay WW Jr. Increased insulin sensitivity and maintenance of glucose utilization rates in fetal sheep with placental insufficiency and intrauterine growth restriction. *Am J Physiol Endocrinol Metab*. 2007;293:E1716–E1725.
7. Thorn SR, Brown LD, Rozance PJ, et al. Increased hepatic glucose production in

fetal sheep with intrauterine growth restriction is not suppressed by insulin. *Diabetes*. 2013;62:65–73.
8. Girard J. Gluconeogenesis in late fetal and early neonatal life. *Biol Neonate*. 1986;50:237–258.
9. Fowden AL, Forhead AJ. Adrenal glands are essential for activation of glucogenesis during undernutrition in fetal sheep near term. *Am J Physiol Endocrinol Metab*. 2011;300:E94–E102.
10. Fowden AL, Forhead AJ. Insulin deficiency alters the metabolic and endocrine responses to undernutrition in fetal sheep near term. *Endocrinology*. 2012;153:4008–4018.
11. Shelley HJ, Neligan GA. Neonatal hypoglycemia. *Br Med Bull*. 1966;22:34–39.
12. Greengard O. Enzymatic differentiation of human liver: comparison with the rat model. *Pediatr Res*. 1977;11:669–676.
13. Gilbert M, Pere MC, Baudelin A, Battaglia FC. Role of free fatty acids in hepatic insulin resistance during late pregnancy in conscious rabbits. *Am J Physiol*. 1991;260:E938–E945.
14. Shao J, Yamashita H, Qiao L, et al. Phosphatidylinositol 3-kinase redistribution is associated with skeletal muscle insulin resistance in gestational diabetes mellitus. *Diabetes*. 2002;51:19–29.
15. Barbour LA, Shao J, Qiao L, et al. Human placental growth hormone causes severe insulin resistance in transgenic mice. Am *J Obstet Gynecol*. 2002;186:512–517.
16. Smith CH, Moe AJ, Ganapathy V. Nutrient transport pathways across the epithelium of the placenta. *Ann Rev Nutr*. 1992;12:183–206.
17. Wooding FBP, Fowden AL, Bell AW, et al. Localisation of glucose transport in the ruminant placenta: implications for sequential use of transporter isoforms. *Placenta*. 2005;26:626–640.
18. Hahn T, Hahn D, Blaschitz A, et al. Hyperglycemia-induced subcellular redistribution of GLUT1 glucose transporters in cultured human term placental trophoblast cells. *Diabetologia*. 2000;43:173–180.
19. Illsley NP. Placental glucose transport in diabetic pregnancy. *Clin Obstet Gynecol*. 2000;43:116–126.
20. Kirwan JP, Hauguel-De Mouzon S, Lepercq J, et al. TNF-alpha is a predictor of insulin resistance in human pregnancy. *Diabetes*. 2002;51:2207–2213.
21. Das U, He J, Ehrhardt RA, et al. Time-dependent physiological regulation of ovine placental GLUT-3 glucose transporter protein. *Am J Physiol Regul Integr Comp Physiol*. 2000;279:R2252–R2261.
22. Dandrea J, Wilson V, Gopalakrishnan G, et al. Maternal nutritional manipulation of placental growth and glucose transporter 1 (GLUT-1) abundance in sheep. *Reproduction*. 2001;122:793–800.
23. Das UG, Sadiq HF, Soares MJ, et al. Time-dependent physiological regulation of rodent and ovine placental glucose transporter (GLUT-1) protein. *Am J Physiol Regul Integr Comp Physiol*. 1998;274:R339–R347.
24. Hay WW Jr, Molina RD, DiGiacomo JE, Meschia G. Model of placental glucose consumption and transfer. *Am J Physiol*. 1990;258:R569–R577.
25. Kalhan S, Parimi PS. Metabolism of glucose and methods of investigation in the fetus and newborn. In: Polin RA, Fox WW, Abman SH, eds. *Fetal and Neonatal Physiology*. 4th ed. Philadelphia, PA: Elsevier (W. B. Saunders Co.); 2011:517–533.
26. Meschia G, Battaglia FC, Hay WW Jr, Sparks JW. The utilization of substrates by the ovine placenta in vivo. *Fed Proc*. 1980;9:245–249.

27. Hahn D, Blaschitz A, Korgun ET, et al. From maternal glucose to fetal glycogen: expression of key regulators in the human placenta. *Mol Hum Reprod*. 2011;7:1173–1178.
28. Hay WW Jr, Meznarich HK. Effect of maternal glucose concentration on uteroplacental glucose consumption and transfer in pregnant sheep. *Proc Soc Exp Biol Med*. 1988;190:63–69.
29. Haugel S, Desnaizieres V, Challier JC. Glucose uptake, utilization, and transfer by the human placenta as functions of maternal glucose concentration. *Pediatr Res*. 1986;20:269–273.
30. Hay WW Jr, DiGiacomo JE, Meznarich HK, et al. Effects of glucose and insulin on fetal glucose oxidation and oxygen consumption. *Am J Physiol*. 1989;256:E704–E713.
31. Das UG, Schroeder RE, Hay WW Jr, Devaskar SU. Time-dependent and tissue-specific effects of circulating glucose on fetal ovine glucose transporters. *Am J Physiol Regul Integr Comp Physiol*. 1999;276:R809–R817.
32. Khan JY, Rajakumar RA, McKnight RA, et al. Developmental regulation of genes mediating murine brain glucose uptake. *Am J Physiol Regul Integr Comp Physiol*. 1999;276:R892–R900.
33. Sadiq HF, Das UG, Tracy TF, Devaskar SU. Intrauterine growth restriction differentially regulates perinatal brain and skeletal muscle glucose transporters. *Brain Res*. 1999;823:96–103.
34. Carver TD, Anderson SM, Aldoretta PW, et al. Glucose suppression of insulin secretion in chronically hyperglycemic fetal sheep. *Pediatr Res*. 1995;38:754–762.
35. Schroeder RE, Doria-Medina CL, Das UG, et al. Effect of maternal diabetes upon fetal rat myocardial and skeletal muscle glucose transporters. *Pediatr Res*. 1997;41:11–19.
36. Aldoretta PW, Hay WW Jr. Chronic hyperglycemia induces insulin resistance and glucose intolerance in fetal sheep. *Pediatr Res*. 2001;49(307A):1758.
37. Anderson MS, Flowers-Ziegler J, Das UG, et al. Glucose transporter protein responses to selective hyperglycemia or hyperinsulinemia in fetal sheep. *Am J Physiol Regul Integr Comp Physiol*. 2001;281:R1545–R1552.
38. Anderson MS, He J, Flowers-Ziegler J, et al. Effects of selective hyperglycemia and hyperinsulinemia on glucose transporters in fetal ovine skeletal muscle. *Am J Physiol Regul Integr Comp Physiol*. 2001;281:R1256–R1263.
39. DiGiacomo JE, Hay WW Jr. Effect of hypoinsulinemia and hyperglycemia on fetal glucose utilization and oxidation. *Am J Physiol*. 1990;259:E506–E512.
40. DiGiacomo JE, Hay WW Jr. Fetal glucose metabolism and oxygen consumption during sustained maternal and fetal hypoglycemia. *Metabolism*. 1990;39:193–202.
41. Maliszewski AM, Gadhia MM, O'Meara MC, et al. Prolonged infusion of amino acids increases leucine oxidation in fetal sheep. *Am J Physiol Endocrinol Metab*. 2012;302:E1483–E1492.
42. Molina RD, Meschia G, Battaglia FC, Hay WW Jr. Maturation of placental glucose transfer capacity in the ovine pregnancy. *Am J Physiol*. 1991;261:R697–R704.
43. Kalhan SC, Savin SM, Adam PAJ. Measurement of glucose turnover in the human newborn with glucose-1-^{13}C. *J Clin Endocrinol Metab*. 1976;43:704–707.
44. Ehrhardt RA, McNeill DM, Bell AW. Identification of and developmental increases in glucose transport protein in the sheep placenta. *FASEB J*. 1994;8:A177. Abstract 1024.
45. Bassett NS, Currie MJ, Woodall SM, et al. The effect of maternal IGF-1 treatment on placental GLUT gene expression. Proceeding of Thorburn Symposium,

Hamilton Island, Queensland, Australia, 1994. Program, Combined Meeting of the Australian and New Zealand Perinatal Societies; April 2–5, 1995; Auckland, New Zealand.
46. Bell AW, Battaglia FC, Meschia G. Relation between metabolic rate and body size in the ovine fetus. *J Nutr*. 1987;117:1181–1186.
47. Aldoretta PW, Carver TD, Hay WW Jr. Maturation of glucose-stimulated insulin secretion in fetal sheep. *Biol Neonate*. 1998;73:375–386.
48. Rorsman P, Arkhammar P, Bokvist K, et al. Failure of glucose to elicit a normal secretory response in fetal pancreatic beta cells results from glucose insensitivity of the ATP-regulated K* channels. *Proc Natl Acad Sci USA*. 1989;86:4505–4509.
49. Milner RDG, Hales CN. Effect of intravenous glucose on concentration of insulin in maternal and umbilical-cord plasma. *Br Med J*. 1965;i:284–286.
50. Obenshain SS, Adam PAJ, King KC, et al. Human fetal insulin response to sustained maternal hyperglycemia. *N Engl J Med*. 1970;283:566–570.
51. Carver TD, Anderson SM, Aldoretta PW, Hay WW Jr. Effect of low-level basal plus marked "pulsatile" hyperglycemia on insulin secretion in fetal sheep. *Am J Physiol Endocrinol Metab*. 1996;271:E865–E871.
52. Limesand SW, Hay WW Jr. Adaptation of ovine fetal pancreatic insulin secretion to chronic hypoglycemia and euglycemic conditions. *J Physiol*. 2003;547(pt 1):95–105.
53. Hay WW Jr, Sparks JW, Wilkening RB, et al. Partition of maternal glucose production between conceptus and maternal tissues in sheep. *Am J Physiol*. 1983;245:E347–E350.
54. Fowden AL, Mundy L, Silver, M. Developmental regulation of glucogenesis in the sheep fetus during late gestation. *J Physiol*. 1998;508(pt 3):937–947.
55. Limesand SW, Rozance PJ, Zerbe GO, et al. Attenuated insulin release and storage in fetal sheep pancreatic islets with intrauterine growth restriction. *Endocrinology*. 2006;147:1488–1497.
56. Rozance PJ, Limesand SW, Barry JS, et al. Chronic late-gestation hypoglycemia upregulates hepatic PEPCK associated with increased PGC1alpha mRNA and phosphorylated CREB in fetal sheep. *Am J Physiol Endocrinol Metab*. 2008;*294:*E365–E370.
57. Thorn SR, Sekar SM, Lavezzi JR, et al. A physiological increase in insulin suppresses gluconeogenic gene activation in fetal sheep with sustained hypoglycemia. *Am J Physiol*. 2012;303:R861–R869.
58. Lavezzi JR, Thorn SR, O'Meara MC, et al. Increased fetal insulin concentrations for one week fail to improve insulin secretion or beta-cell mass in fetal sheep with chronically reduced glucose supply. *Am J Physiol*. 2013;304:R50–R58.
59. Wallace JM, Bourke DA, Aitken RP, et al. Blood flows and nutrient uptakes in growth-restricted pregnancies induced by overnourishing adolescent sheep. *Am J Physiol Regul Integr Comp Physiol*. 2002;282:R1027–R1036.
60. Barry JS, Davidsen ML, Limesand SW, et al. Developmental changes in ovine myocardial glucose transporters and insulin signaling following hyperthermia-induced intrauterine fetal growth restriction. *Exp Biolo Med*. 2006;231:566–575.
61. Owens JA, Gatford KL, De Blasio MJ, et al. Restriction of placental growth in sheep impairs insulin secretion but not sensitivity before birth. *J Physiol*. 2007;584:935–949.
62. Wallace JM, Milne JS, Aitken RP, Hay WW Jr. Sensitivity to metabolic signals in late-gestation growth-restricted fetuses from rapidly growing adolescent sheep. *Am J Physiol Endocrinol Metab*. 2007;293:E1233–E1241.
63. Lane RH, MacLennan NK, Hsu JL, et al. Increased hepatic peroxisome

proliferator-activated receptor-gamma coactivator-1 gene expression in a rat model of intrauterine growth retardation and subsequent insulin resistance. *Endocrinology*. 2002;143:2486–2490.
64. Nijland MJ, Mitsuya K, Li C, et al. Epigenetic modification of fetal baboon hepatic phosphoenolpyruvate carboxykinase following exposure to moderately reduced nutrient availability. *J Physiol*. 2010;588:1349–1359.
65. Thorn SR, Brown LD, Rozance PJ, et al. Increased hepatic glucose production in fetal sheep with intrauterine growth restriction is not suppressed by insulin. *Diabetes*. 2013;62:65–73.
66. Thorn SR, Regnault TR, Brown LD, et al. Intrauterine growth restriction increases fetal hepatic gluconeogenic capacity and reduces messenger ribonucleic acid translation initiation and nutrient sensing in fetal liver and skeletal muscle. *Endocrinology*. 2009;150:3021–3030.
67. Thorn SR, Rozance PJ, Brown LD, Hay WW. The intrauterine growth restriction phenotype: fetal adaptations and potential implications for later life insulin resistance and diabetes. *Semin Reprod Med*. 2011;29:225–236.
68. Carver TD, Hay WW Jr. Uteroplacental carbon substrate metabolism and O2 consumption after long-term hypoglycemia in pregnant sheep. *Am J Physiol*. 1995;269:E299–E308.
69. Narkewicz MR, Carver TD, Hay WW Jr. Induction of cytosolic phosphoenolpyruvate carboxykinase in the ovine fetal liver by chronic fetal hypoglycemia and hypoinsulinemia. *Pediatr Res*. 1993;33:493–496.
70. Thureen PJ, Trembler KA, Meschia G, et al. Placental glucose transport in heat-induced fetal growth retardation. *Am J Physiol*. 1992;263:R578–R585.
71. Frost MS, Zehri A, Limesand SW, et al. Differential effects of chronic pulsatile vs. chronic constant maternal hyperglycemia on fetal insulin secretion and pancreatic beta-cell development. *J Pregnancy*. 2012;2012:812094; doi: 10.1155/2012/812094.
72. Hay WW Jr, Aldoretta PW, Carver TD. *Glucose-Specific Suppression of Insulin Secretion and Glucose Utilization in Late-Gestation Fetal Sheep*. Monduzzi Editore. Bologna, Italy: Acta Second World Congress of Perinatal Medicine; 1993:835–839.
73. Moores RR Jr, Vaughn PR, Battaglia FC, et al. Glutamate metabolism in fetus and placenta of late-gestation sheep. *Am J Physiol*. 1994;267:R89–R96.
74. Auestad N. Diverse roles of lipids in neonatal physiology and development. In: Neu J, ed. *Gastroenterology and Nutrition* (Polin RA, Consulting editor: Neonatology Questions and Controversies). Philadelphia, PA: Saunders Elsevier; 2008:253–277.
75. Haggarty P. Effect of placental function on fatty acid requirements during pregnancy. *Eur J Clin Nutr*. 2004;58:1559–1570.
76. Haggarty P. Fatty acid supply to the human fetus. *Annu Rev Nutr*. 2010;30:237–255.
77. Hay WW Jr. Metabolic interrelationships of placenta and fetus. *Placenta*. 1995;16:19–30.
78. Magnusson AL, Waterman IJ, Wennergren M, et al. Triglyceride hydrolase activities and expression of fatty acid binding proteins in the human placenta in pregnancies complicated by intrauterine growth restriction and diabetes. *J Clin Endocrinol Metab*. 2004;89:4607–4614.
79. Rotherwell JE, Elphick MC. Lipoprotein lipase activity in human and guinea pig placenta. *J Dev Physiol*. 1982;4:153–159.
80. Rice GE, Wong MH, Farrugia W, Scott KF. Contribution of type II phospholipase

A2 to in vitro phospholipase A2 enzymatic activity in human term placenta. *J Endocrinol*. 1998;157:25–31.
81. Kamp F, Zakim D, Zhang FL, et al. Fatty-acid flip-flop in phospholipid- bilayers is extremely fast. *Biochemistry*. 1995;34:11928–11937.
82. Veerkamp JH, van Moerkerk HTB, Zimmerman AW. Effect of fatty acid-binding proteins an intermembrane fatty acid transport–studies on different types and mutant proteins. *Eur J Biochem*. 2000;267:5959–5966.
83. Dutta-Roy AK. Transport mechanisms for long-chain polyunsaturated fatty acids in the human placenta. *Am J Clin Nutr*. 2000;71:315S–322S.
84. Veerkamp JH. Fatty-acid transport and fatty acid binding proteins. *Proc Nutr Soc*. 1995;54:23–37.
85. Benassayag C, Mignot TM, Haourigui M, et al. High polyunsaturated fatty acid, thromboxane A2, and alpha-fetoprotein concentrations at the human feto-maternal interface. *J Lipid Res*. 1997;38:276–286.
86. Patel MN, Kleinfeld AM, Richeiri GV, et al. Serum levels of unbound free fatty acids. I: normative data in term newborn infants. *J Am Coll Nutr*. 1997;16:81–84.
87. Economides DL, Crook D, Nicolaides KH. Investigation of hypertriglyceridemia in small for gestational age fetuses. *Fetal Ther*. 1988;3:165–172.
88. Schmidt-Sommerfeld E, Penn D, Sodha RJ, et al. Transfer and metabolism of carnitine and carnitine esters in the in vitro perfused human placenta. *Pediatr Res*. 1985;19:700–706.
89. Szabo AG, De Lellis R, Grimaldi RD. Triglyceride synthesis by the human placenta. I. Incorporation of labeled palmitate into placental triglycerides. *Am J Obstet Gynecol*. 1973;115:257–262.
90. Roux JF, Green R. Lipid metabolism by the human placenta. *Obstet Gynecol*. 1968;29:446.
91. Christie WW, Noble RC. Fatty acid biosynthesis in sheep placenta and maternal and fetal adipose tissue. *Biol Neonate*. 1982;42:79–86.
92. Martin RJ, Housman GJ. Placental development and fatty acid metabolism in pigs fed ad libitum or restricted during gestation. *Proc Soc Exp Biol Med*. 1981;166:472–478.
93. Hummel L, Schwartze A, Schirrmeister W, et al. Maternal plasma triglycerides as a source for fetal fatty acids. *Acta Biol Med Germ*. 1976;35:1635–1641.
94. Knopp RH, Warth MA, Charles D, et al. Lipoprotein metabolism in pregnancy, fat transport to the fetus, and the effects of diabetes. *Biol Neonate*. 1986;50:297–317.
95. Winkel CA, Gilmore J, MacDonald PC, Simpson ER. Uptake and degradation of lipoproteins by human trophoblastic cells in primary culture. *Endocrinology*. 1980;107:1892–1898.
96. Van Aerde JE, Wilke MS, Feldman M, Clandinin MT. Accretion of lipid in the fetus and newborn. In: Polin RA, Fox WW, Abman SH, eds. *Fetal and Neonatal Physiology*. 4th ed. Philadelphia, PA: Elsevier (W. B. Saunders Co.); 2011:454–470.
97. Haggarty P. Placental regulation of fatty acid delivery and its effect on fetal growth: a review. *Placenta*. 2002;23(suppl A):S28–S38.
98. Goldstein R, Levy E, Shafrir E. Increased maternal-fetal transport of fat in diabetes assessed by polyunsaturated fatty acid content in fetal lipids. *Biol Neonate*. 1985;47:343–349.
99. Hay WW Jr. Placental transport of nutrients to the fetus. *Horm Res*. 1994;42:215–222.
100. Herrera E. Implications of dietary fatty acids during pregnancy on placental, fetal and postnatal development–a review. *Trophoblast Res*. 2002;16:S9–S19.

101. Campbell FM, Gordon MJ, Dutta-Roy AK. Preferential uptake of long chain polyunsaturated fatty acids by isolated human placental membranes. *Mol Cell Biochem*. 1996;155:77–83.
102. Haggarty P, Page K, Abramovich DR, et al. Long-chain polyunsaturated fatty acid transport across the perfused human placenta. *Placenta*. 1997;18:635–642.
103. Zimmermann T, Hummel L, Mollr U, Kinzl U. Oxidation and synthesis of fatty acids in human and rat placental and fetal tissues. *Biol Neonate*. 1979;36:109–112.
104. Robertson A, Sprecher H, Karp W. Oxidation of palmitate by human placental tissue slices. *Phys Chem Phys*. 1971;3:293–301.
105. Clandinin MT, Chappell JE, Heim T, et al. Fatty acid utilization in perinatal de novo synthesis of tissues. *Early Hum Dev*. 1981;5:355–366.
106. Chambaz J, Ravel D, Manier MC, et al. Essential fatty acid interconversion in the human fetal liver. *Biol Neonate*. 1985;47:136–140.
107. Hornstra G, Al MDM, V Houwelingen AC, Foreman-van Drongelen MMHP. Essential fatty acids in pregnancy and early human development. *Eur J Obstet Gynecol Reprod Biol*. 1995;61:57–62.
108. Clandinin MT, Chappell JE, Leong S, et al. Intrauterine fatty acid accretion rates in human brain: implications for fatty acid requirements. *Early Hum Dev*. 1980;4:121–129.
109. Su HM, Corso TN, Nathanielsz PW, Brenna JT. Linoleic acid kinetics and conversion to arachidonic acid in the pregnant and fetal baboon. *J Lipid Res*. 1999;1304–1311.
110. Jumpsen J, Van Aerde J, Clandinin MT. Fetal lipid requirements: implications in fetal growth retardation. In: Battaglia FC, ed. *Placental Function and Fetal Nutrition*. Philadelphia, PA: Nesttec Ltd, Vevey/Lippincott-Raven Publ; 1997:157–165.
111. Matorras R, Perteagudo L, Sanjurjo P, Ruiz JI. Intake of long chain ω-3 polyunsaturated fatty acids during pregnancy and the influence of levels in the mother on newborn levels. *Eur J Obstet Gynecol Reprod Biol*. 1999;83:179–184.
112. Van Houwelingen AC, Sorensen JD, Hornstra G, et al. Essential fatty acid status in neonates after fish-oil supplementation during late pregnancy. *Br J Nutr*. 1995;74:723–731.
113. Innis SM. Essential fatty acids in growth and development. *Prog Lipid Res*. 191;30:39–103.
114. Raz A, Kamin-Belsky N, Przedecki F, Obukowicz MG. Fish oil inhibits delta6 desaturase activity in vivo: utility in a dietary paradigm to obtain mice depleted of arachidonic acid. *J Nutr Biochem*. 1997; 8:558–565.
115. Koletzko B, Braun M. Arachidonic acid and early human growth: is there a relation? *Ann Nutr Metab*. 1991;35:128–131.
116. Carlson SE, Werkman SH, Pepples JM. Arachidonic acid status correlates with first year growth in preterm infants. *Proc Natl Acad Sci USA*. 1993;90:1073–1077.
117. Viana M, Herrera E, Bonet B. Teratogenic effects of diabetes mellitus in the rat. Prevention by vitamin E. *Diabetologia*. 1996;39:1041–1046.
118. Navarro MD, Periago JL, Pita ML, Hortelano P. The n-3 polyunsaturated fatty acid levels in rat tissue lipids increase in response to dietary olive oil relative to sunflower oil. *Lipids*. 1994;29:845–849.
119. Amusquivar E, Ruperez FJ, Barbas C, Herrera E. Low arachidonic acid rather than tocopherol is responsible for the delayed postnatal development in offspring of rats fed fish oil instead of olive oil during pregnancy and lactation. *J Nutr*. 2000;130:2855–2865.
120. Sinclair AJ, Mann NJ. Short-term diets rich in arachidonic acid influence plasma

phospholipid polyunsaturated fatty acid levels and prostacyclin and thromboxane production in humans. *J Nutr*. 1996;126:1110S–1114S.
121. Eritsland J. Safety considerations of polyunsaturated fatty acids. *Am J Clin Nutr*. 2000;71:197S–201S.
122. Crawford MA, Hassan AG, Williams G, Whitehouse WL. Essential fatty acids and fetal brain growth. *Lancet*. 1976;1(7957):452–453.
123. Sellmayer A, Koletzko B. Long-chain polyunsaturated fatty acids and eicosanoids in infants–physiological and pathophysiological aspects and open questions. *Lipids*. 1999;34:199–205.
124. Leaf A, Gosbell A, McKenzie L, et al. Long chain polyunsaturated fatty acids and visual function in preterm infants. *Early Hum Dev*. 1996;45:35–53.
125. Herrera E, Gómez-Coronado D, Lascunción MA. Lipid metabolism in pregnancy. *Biol Neonate*. 1987;51:70–77.
126. Hoggard N, Hunter L, Lea RG, et al. Ontogeny of the expression of leptin and its receptor in the murine fetus and placenta. *Br J Nutr*. 2000;83:317–326.
127. Ong KK, Ahmed ML, Sherrif A, et al. Cord blood leptin is associated with size at birth and predicts infancy weight gain in humans. ALSPAC Study Team. Avon Longitudinal Study of Pregnancy and Childhood. *J Clin Edocrinol Metab*. 1999;84:1145–1148.
128. Ehrhardt RA, Bell AW, Boisclair YR. Spatial and developmental regulation of leptin in fetal sheep. *Am J Physiol Regul Integr Comp Physiol*. 2002;282:R1628–R1635.
129. Devaskar SU, Anthony R, Hay WW Jr. Ontogeny and insulin regulation of fetal ovine white adipose tissue leptin expression. *Am J Physiol Regul Integr Comp Physiol*. 2002;282:R431–R438.
130. Devaskar SU, Anthony R, Hay WW Jr. Ontogeny and insulin regulation of fetal ovine white adipose tissue leptin expression. *Am J Physiol Regul Integr Comp Physiol*. 2002;282:R431–R438.

Amino Acid Nutrition in Utero: Placental Function and Metabolism

28

Timothy R.H. Regnault, PhD and Frederick C. Battaglia, MD

This chapter will review placental function and metabolism as it relates to the issue of fetal amino acid nutrition. The review will include information available in human pregnancy, although, of necessity, most of the in vivo information has been collected in various animal species, and this, too, will be reviewed. In all mammalian species, the 20 amino acids of the genetic code are required for net protein accretion. The rate of protein accretion has a wide variation among mammals based primarily on gestation length, which ultimately impacts fetal growth rate. The nutritional supply of amino acids for growth is defined as the net umbilical uptake of amino acids, representing the net transfer from the placenta to the fetus, including back flux from the fetus to the placenta of essential and nonessential amino acids. In the ovine fetus, where these data are available, the quantity taken up by the fetus clearly exceeds requirements for growth[1,2] and is associated with a high rate of fetal amino acid oxidation.[3] In this review, we aim to detail aspects of fetal amino acid requirements, placental transport, and metabolism of amino acids as well as highlight factors

influencing these processes, such as fetal growth restriction (FGR). In addition, we will discuss umbilical uptake, interorgan cycling, and fetal and placental amino acid utilization, including, specifically, fetal hepatic and hindlimb amino acid metabolism. In concluding this chapter, we will deal with the neonatal implications of fetal amino acid metabolism.

OVERVIEW OF PLACENTAL DELIVERY OF FETAL AMINO ACID REQUIREMENTS

The primary source of nitrogen for the growing mammalian fetus is the circulating maternal free amino acid pool. In considering the primary role of the placenta in the delivery of amino acids to the fetus, it is important to consider the multiplicity of factors that may affect these overall delivery rates. Figure 28-1 presents in diagrammatic form the potential fate of an amino acid that has been transported across the plasma membrane of the microvillous maternal surface into the trophoblast cytosol, prior to its entry into the fetal circulation through the basal membrane (BM). Pathway 1 represents the direct transplacental flux of an amino acid from the maternal circulation across the plasma membranes on the maternal and fetal surfaces of the trophoblast and into the fetal circulation. Pathway 2 represents the delivery of an amino acid that has entered the intracellular pool via protein turnover within the trophoblast. Pathway 3 represents the backflux from the fetal circulation into the trophoblast of the same amino acid, and Pathway 4 refers to the transformation of the amino acid to a metabolite that can then enter the fetal circulation. The latter example would be the transamination of leucine to ketoisocaproic acid, which can then be delivered to the fetus. The algebraic sum of these fluxes represents the net umbilical uptake, which is the nutritionally important measurement because it represents the net supply of the amino acid delivered to the fetus. The relative importance of each of the pathways in determining the umbilical uptake differs among amino acids. Examples of amino acids that are utilized extensively within the placenta (such as glutamate) or are synthesized at a relatively high rate within the placenta (such as glycine) will be described in the following sections. There is already ample evidence to show that, for different amino acids, one or another of these pathways may predominate.

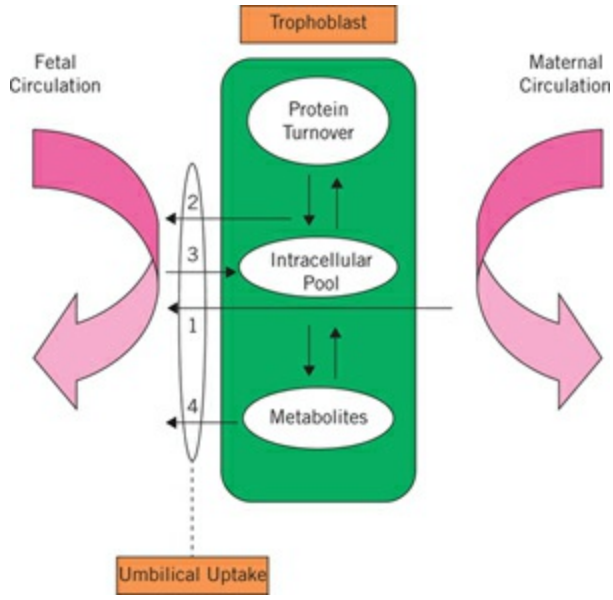


Figure 28-1 A diagrammatic representation of potential pathways for the delivery of an amino acid into the fetal circulation. See text for full description of pathways.

It is apparent that the fetus not only accrues nitrogen in the new tissues formed during growth, but also extensively metabolizes its nitrogen source by synthesizing, oxidizing, and transaminating amino acids.[4] The current estimate of ovine fetal uptake of nitrogen is approximately 0.9 g/kg/d, which is approximately equal to the estimated fetal nitrogen requirement.[5] Therefore, the quantity of amino acids transported is sufficient to meet fetal nitrogen requirements. This estimation includes the fate of amino acids metabolized and excreted as ammonia or urea from the fetoplacental unit.[3,6] Other potential sources of nitrogen, such as intact proteins or peptides, which may be important in embryogenesis do not appear to be required to meet fetal nitrogen needs.

Estimations of fetal protein synthetic rates have been performed in fetal sheep using a variety of methods.[7–9] Leucine was used as a tracer amino acid in chronically catheterized fetal sheep to assess protein synthetic rates at different gestational ages[7,10] and under different physiological conditions.[11,12] These studies demonstrated that the absolute fractional protein synthetic rate (Ks) decreased from mid-gestation to term, and that this decrease was greater than that seen in the fractional protein accretion rate. Figure 28-2 demonstrates the inverse relationship noted

between the fetal Ks and fetal age. Therefore, protein breakdown, defined as the difference between protein accretion and synthesis, decreases in late gestation. The higher protein synthetic rate of the mid-gestation fetus is proportional to the higher metabolic rate at that stage of gestation. Absolute fractional protein synthetic rate declines at a similar rate as fetal O_2 consumption (VO_2), giving a relatively constant ratio of Ks/VO_2.[10] Thus, protein synthesis per millimole of oxygen consumed is quite constant from mid-gestation until term.[10] Fetal metabolic studies, including ones focused upon other metabolic variables such as lactate utilization rate[13,14] and urea production rate,[3,15,16] have advanced our conceptual framework of the key factors that alter metabolic rate per kilogram of body weight during development and, therefore, the developmental changes in the protein synthetic rate.

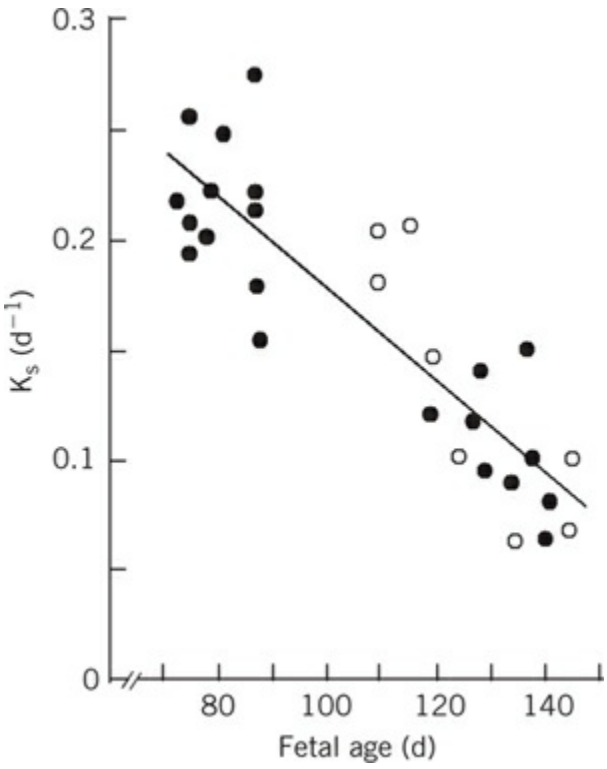


Figure 28-2 In chronically catheterized fetal sheep, the protein synthetic rate (Ks) was noted to decrease with fetal age. Protein synthetic rates were determined using 1^{13} C-leucine (dots) and U^{14}C-lysine (open circles). Regression line (r = –0.87) was drawn according to equation: y = 0.39–0.02Ix. Adapted from Kennaugh et al.[10]

PLACENTAL AMINO ACID TRANSPORT

Both in vivo[5,12,17,18] and in vitro[19–22] studies have brought out the complexity of the placenta as an organ for amino acid transport and metabolism. It is important to emphasize that, unlike an organ such as the liver, the placenta must function primarily as an organ of transport, delivering amino acids from the maternal circulation into the fetal circulation. The complexity of the organ stems not only from the different cell types required to effect such net transport, but also from the capacity of the placenta to metabolize or synthesize various amino acids. The human placenta has two main cell types that are critically important in determining its overall transport characteristics: the trophoblast and the endothelium. In other species, additional cell layers may be present; for example, in the epitheliochorial placenta of sheep and goats, there is an endometrial cell layer as well. However, the syncytium is considered the tissue barrier that is rate limiting to amino acid movement between the maternal and fetal compartments. In any discussion of the human placenta and its tissue layers, it is important to remember that there are enormous changes in the proportion of stem villi to terminal villi at different stages of gestation. Early in gestation, stem villi represent the bulk of the tissue. This means that the interstitial space of the villi and its characteristics for transport and for metabolism cannot be overlooked.

The concentrations of most amino acids, including some of the essentials, are higher in fetal than in maternal plasma, consistent with a process of active transport of amino acids across the placenta. An increased fetal-to-maternal ratio of plasma amino acid concentrations has been documented for humans,[5,23] primates,[24,25] rats,[26,27] guinea pigs,[28,29] sheep,[1,5] and cows.[30,31] The concept of active transport has been corroborated by a variety of experiments. Studies of placental tissue cultured in vitro with nonmetabolizable amino acid analogues, for example, α-aminoisobutyrate (AIB), as well as other amino acids, have shown the intracellular concentration of those substances to be at a level several times greater than the extracellular maternal concentration.[21,32] Placental tissues have also been found to have extremely high levels of the amino acids taurine, glutamate, and aspartate, as well as moderately high levels of alanine, glycine, serine, glutamine, and threonine, when compared to maternal or fetal concentrations.[33] It should be emphasized that measurements of placental tissue amino acid concentrations have not been corrected for extracellular fluid content of the tissue. Therefore, the intracellular tissue concentrations are likely to be considerably higher than

the values in the literature.

The uptake of amino acids occurs through active transport. Mammalian transport systems have been characterized over the years by such general properties as ion dependence, kinetics, substrate specificity, and regulation of activity; and this has allowed the description of multiple transport systems for neutral, anionic, and cationic amino acids, predominantly from human and rat trophoblast studies. Figure 28-3 represents a summary of the major amino acid transport systems described for the maternal microvillous membrane [MVM] and fetal basal membrane (BM) facing membranes of the trophoblast. Each system is distinct but there is extensive overlapping substrate specificity.

System A is a sodium-dependent, unidirectional transporter with a characteristic affinity for *N*-methylated substances, and System A activity has been demonstrated in both MVM and BM.[34] Its activity is increased as an adaptation to cellular depletion of amino acid substrates, similar to that described for System X_{AG}^-[35]; and its mRNA and activity appear to be regulated under a number of factors discussed in more detail in the amino acid protein section.

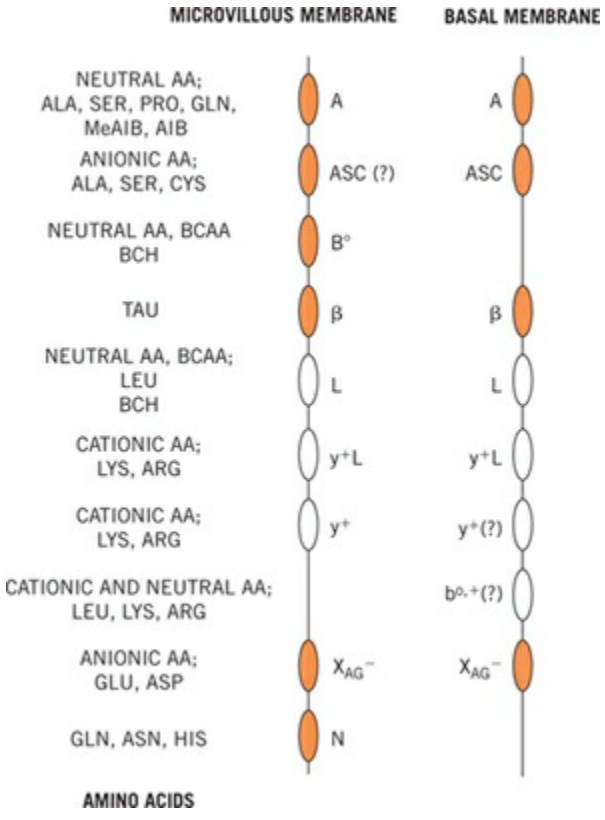


Figure 28-3 Schematic representation of the sodium-dependent (λ) and sodium-independent (γ) amino acid transport systems for both the microvillous (maternal) and basal (fetal) membranes of the trophoblast. Amino acid substrates are listed on the left and references on the right. MeAIB, methylaminoisobutyric acid; AIB, α-aminoisobutyric acid; BCH, 2-aminobicyclo-(2,2,1)-heptane-2-carboxylic acid.

The ASC system has been described for neutral amino acid transport, specifically alanine, serine, and cysteine, as well as for some anionic amino acids.[36] It is a sodium-dependent obligate exchanger localized at the BM, though studies utilizing BeWo cells suggest that System ASC transporters may be present on the MVM as well, but after differentiation and syncytial formation, such activity is lost.[37] A sodium-independent group of transporters (Asc), with similar transport characteristics,[38] has now been reported to exist in placental tissue.[39,40] In addition, the broad-scope, sodium-dependent transporter B^o is responsible for neutral amino acid transport, including branched chain amino acids (BCAAs), and has been localized to the MVM and BM.[41,42] This transporter is highlighted to play an important role in syncytialization, and oxygen tension appears to be a major regulator of its expression.[43] The β-amino acid taurine utilizes the System β transport system. Studies have demonstrated that this β transport system is a sodium-dependent transporter found on both membranes,[44–46] though with a greatly reduced BM activity compared to the MVM.[47] Other studies have now identified this transport system to have a cytoprotective role in syncytiotrophoblasts exposed to hypertonic stress.[48]

System L (or l) transporters are sodium-independent exchange transporters and have been localized to both trophoblast membranes.[49,50] They play a major role in the transport of essential amino acids, having a high affinity for leucine and the other BCAAs.[51–53] The cationic amino acid transport System y^+L is considered a low-capacity, high-affinity system that exchanges cationic amino acids for neutral amino acids and its activity is localized to both the MVM and BM.[54–56] A proposed hypothesis is that this transporter system in the BM is the major supply route of cationic amino acids to the fetus through the uptake of neutral amino acids from fetal circulation, in exchange for cationic amino acids from the placenta.[55,57] The sodium-independent low affinity, high-capacity y^+ transport system is a high-capacity system which transports cationic amino

acids such as arginine, lysine, and ornithine in a sodium-independent manner. It is considered to be the major cationic transport system in placental tissues[5,58,59] and is localized to the MVM[54,55,60] and possibly to the BMs.[61] A role for the related $b^{o,+}$ System in placental tissues has been reported[62,63]; however, further vesicle studies suggest that there is no functional evidence for this system in either term MVM or BM,[55] despite the observation of associated mRNA.[64]

The placenta takes up the anionic amino acids, glutamate and aspartate, from the maternal and fetal circulations, but does not appear to actively transfer these amino acids from mother to fetus. Studies utilizing isolated human and rat placental membrane vesicles and immunohistochemical localization in human placenta have revealed the presence of a high-affinity transport system for aspartate and glutamate via the sodium-dependent X_{AG}^- System within both the microvillous and basal plasma membranes.[34,65,66] This system may mediate the concentrative uptake of anionic amino acids from the maternal and fetal circulations into the placenta. System N transports neutral amino acids, but differs from Systems A and ASC in that it displays a preference for amino acids containing nitrogen-bearing side chains, transporting only glutamine, aspargine, and histidine.[51,67] System N activity is found in the liver and also has been localized in the human placenta,[68] though some studies have failed to report placental mRNA[69] and others have attributed this System N-like transport activity, in the rat placenta, to a combination of System A, possibly System ASC, and $B^{o,+}$ and y^+ L activity.[70]

PLACENTAL AMINO ACID TRANSPORTER PROTEINS

Historically, placental amino acid transport has been described in terms of transport systems based on competitive inhibition studies, though it is now becoming possible to move to a more precise description of the protein chemistry of the transporter system. The ability to clone, sequence, and study the expression of transporters has led to an explosion of data concerning the molecular basis of amino acid transport systems and their potential in vivo function. Thus, the information provided in Table 28-1 and Figure 28-3 should be regarded as an interim report in a rapidly changing field. The identification of individual transporter proteins in the trophoblast will be expanded considerably as sequence data for specific transporters and permit rapid identification of other members of the same

family of transporters; the descriptions in the following section serve to highlight the key points of an expanding field.

TABLE 28-1 Placental Transport Systems and Their Associated Heavy and Light Side Chains

Transporter System	Amino Acid Transporter Type	Protein	Gene
A	Neutral	SNAT-1	SLC38A1
	Sodium dependent	SNAT-2	SLC38A2
		SNAT-4	SLC38A4
ASC	Neutral	ASCT-1	SLC1A4
	Sodium dependent		
Bo	Neutral	ASCT-2	SLC1A5
	Sodium dependent		
β	Neutral	TAUT	SLC6A6
	Sodium dependent		
N	Neutral	SNAT-3	SLC38A3
	Sodium dependent	SNAT-5	SLC38A5
Asc	Neutral	Asc-1/4F2hc[a]	SLC7A10
	Sodium independent	Asc-2/?	SLC7A12
L	Neutral	LAT-1/4F2hc[a]	SLC7A5
	Sodium independent	LAT-2/4F2hc	SLC7A8
		LAT-3	SLC43A1
		LAT-4	SLC43A2
y^+L	Cationic	y^+LAT-1/4F2hc[a]	SLC7A7
	Sodium independent	y^+LAT-2/4F2hc(?)	SLC7A6
y^+	Cationic	CAT-1	SLC7A1
	Sodium independent	CAT-2B	SLC7A2
		CAT-4	SLC7A4
X_{AG}^-	Anionic	EAAT-1	SLC1A3
		EAAT-2	SLC1A2
		EAAT-3	SLC1A1

Refer to text for light chain protein expanded abbreviations.
[a]SLC code for 4F2hc is SLC3A2.

Amino acid transporters exist either as a monomeric protein, while

other transport systems exist as heterodimeric complex.[71,72] Those monomeric transport systems induce amino acid uptake in a similar manner to the amino acid uptake associated with permeases as described in microorganisms,[73,74] though the regulation of placental membrane acquisition of monomeric systems has not yet been studied. Heterodimeric systems involve the grouping together of two proteins to facilitate amino acid transport. In placental tissues, monomeric transport systems are believed to include y^+, ASC, A, and most likely the X_{AG}^- System,[64,75,76] while heterodimeric transport systems include System L, y^+L, and the Asc system.[40,64] Those systems that operate in a heterodimeric manner are generally made up of a common heavy chain, specifically one of the type II membrane glycoproteins, 4F2hc (comparable to the CD98 surface antigen, CD98hc),[76,77] the Neutral and Basic Amino Acid Transport (NBAT), equivalent to rBAT or D2[78–80] protein or as yet undefined proteins,[39] which associate with one of a variety of light chain transport proteins, providing a range of amino acid transport systems.[81,82] There is also evidence that these heavy chains may interact with themselves and each other to form homodimers and/or heterodimers before interacting with a specific light chain to induce activity.[55] A summary of the traditional transport systems and their heavy and light chain associations that have been reported in placental tissues and membranes is presented in Table 28-1.

The 4F2hc mRNA is ubiquitously expressed.[72] While 4F2hc is expressed in placental tissues throughout pregnancy, NBAT expression appears only in first and early second trimester placenta and has not been reported in term tissues.[55,83]In human and rat pregnancy, whole-tissue studies have demonstrated 4F2hc predominantly in the MVM, with the content increasing as gestation advances.[55,77,83] However, 4F2hc has not been detected in the human BM,[55,77] despite reports of 4F2hc mRNA in rat BM preparations[83] and evidence that 4F2hc plays a role in BM amino acid transport in other tissues, including the intestine and kidney.[84]

The glycosylated 4F2hc protein is composed of a single transmembrane region with a large C-terminal extracellular domain.[85] Since mammalian substrate transporters appear to have multiple transmembrane helices, it is unlikely that the 4F2hc protein is capable of inducing amino acid transport alone.[86] It is postulated that 4F2hc is in fact the modulator of heterodimeric transport systems and acts as a “guidance molecule,” translocating the nonglycosylated light chains to the plasma

membrane, after which disulfide linkage between 4F2hc and the light chain occurs.[72,87] The subcellular location of the light chain proteins and their interaction with 4F2hc protein is not well defined. Presently, the model of interaction involves an intracellular pool of light chains that are available for incorporation with the membrane-bound heavy chain.[75,85] The movement is independent of the formation of the disulfide bonds, which suggests a noncovalent steric association.[88] Following membrane localization and, if required, light chain and heavy chain interaction, functional transport may then occur.[72,89,90] The expression of the heavy chain, and not that of the light chains, is best correlated with placental amino acid transport system activity.[75,85]

Neutral Amino Acid Transporter Light Chain Proteins

Currently, three subtypes of the amino acid System A have been cloned, amino acid transporter SNAT1 (SLC38A1), SNAT2 (SLC38A2), and SNAT4 (SLC38A4), and all systems appear to operate in a monomeric manner.[72] The SNAT1 (ATA-1 or GlnT)[91] system has a limited tissue expression, being expressed predominantly in the human placenta and heart, and is responsible for the transport of small short-chain neutral amino acids, such as alanine, serine, methionine, asparagine, and glutamine.[91] The functional characteristics of SNAT2 are similar to those of SNAT1, though found in a wider range of tissues, including the placenta.[91,92] SNAT4 initially appeared to have a unique tissue distribution predominantly in hepatic and skeletal muscle tissues;[93] however, reports also place it in the placenta, in both human MVM and BMs, being expressed from early pregnancy onwards, similar to SNAT1 and SNAT2.[94]

Studies are now identifying the many factors that relate System A expression and activity. These include a number of maternal and fetal influences such as maternal growth hormone (GH),[95] activation of insulin/insulin-like growth factor -I (IGF-I) and mammalian target of rapamycin (mTOR) signaling pathways,[96,97] hypoxia,[98,99] Interleukin 6(IL-6),[100] and leptin concentrations.[101] Other impacts such as amino acid restriction,[102,103] maternal diet,[104] dietary fat and fiber levels,[105] and specifically fatty acids via placental toll-like receptor 4 are also reported to impact System A activity.[106] Maternal age also impacts System A expression, where teenage placentae have been characterized as having reduced System A mRNA, though there appears to be increased activity in

growing teens.[107] Of final note, it appears that synthetic glucocorticoid administration may regulate System A activity, though in a differential manner. In mid-gestation it appears to be associated with decreased System A activity,[108] while near term it is associated with System A activity activation and promotes syncytiotrophoblast differentiation and maturation.[109]

The ASCT-1 (SLC1A4),[110] a component of the ASC transport system, has been localized to human placenta tissue, though its expression appears to be very low.[111] Another component of the ASC system, ASCT-2 has also been cloned,[112] though this is thought to code protein related to the related transport family System Bo. The Bo is a sodium-dependent neutral amino acid transport system for which the related gene ASCT2 (SLC1A5) is proposed to represent the Bo system in the human placental choriocarcinoma cell line, JAR, and isolated human placental Poly(A)$^+$ mRNA.[42] The B title represents this system's broad substrate preference, and the capital status indicates its sodium dependence. The superscript o defines it as accepting zwitterionic or neutral amino acids, which distinguishes it from the Bo,$^+$ system, which also accepts cationic amino acids.[42,113] In early pregnancy, prior to implantation, leucine uptake through System B$^{o,+}$ in blastocysts triggers mTOR signaling believed to result in trophoblast motility and the onset of the penetration stage of blastocyst implantation,[114] and in culture systems altered oxygen tensions appear to play a regulatory role.[43] It has been postulated that suboptimal System Bo,$^+$ regulation of these early pregnancy developmental processes may influence placentation and subsequent embryo nutrition, birth weight and risk of developing metabolic syndrome and obesity.[114] These systems (Boand Bo,$^+$) are differentiated from the bo,$^+$ system that also transports neutral and cationic amino acids, but in a sodium-independent manner.[115] This latter system has been reported in BM preparations,[61] though this localization has been questioned with its activity attributed instead to the y$^+$ L system, as no functional evidence for System bo,$^+$ has been observed in term MVM or BM.[55] The β transporter for taurine has exhibited activity in placenta apical and basal placental preparations,[44,116,117] and this activity is associated with a mRNA encoding TAUT (SLC6A6), in placental cell lines and human placental Poly(A)$^+$ mRNA.[117,118]

The sodium-independent transport of small neutral amino acids is carried out by the Asc system, comprising the proteins, Asc-1 (SLC7A10) and Asc-2 (SLC7A12), both of which have been identified in placental

mRNA.[39,40] Asc-1 forms a heterodimeric complex with 4F2hc (SLC3A2),[40] whereas Asc-2 appears to interact with a presently unknown heavy chain.[39]

Other light chain proteins also associate with 4F2hc to form the functional L transport system,[75,89] catalyzing the uptake of neutral amino acids in a sodium-independent manner. System L-amino acid transporter-1 LAT-1 (SLC7A5), LAT-2 (SLC7A8), LAT-3 (SLC43A1), and, LAT-4 (SLC43A2) have all been reported in placental tissues.[75,77,90,119] The 4F2hc/LAT-1 transport activity displays a narrow specificity towards neutral amino acids, uninfluenced by pH, while 4F2hc/LAT-2 activity has a broad specificity and is stimulated by decreasing pH.[120] Immunological and functional studies suggest that at term, the light chain, LAT-1 is located predominantly in the MVM and the syncytiotrophoblast layer of the villi.[38,77,121] Furthermore, the L transport system phenotype associated with that of the placental MVM is found to occur when the LAT-1 and not the LAT-2 catalytic subunit is co-expressed with the 4F2hc.[121] In a similar manner, human LAT-2 mRNA has been identified in the choriocarcinoma BeWo cell line and placental villous tissues[121,122] and its activity has been localized to the BM[38,121] and not the MVM as is the case for LAT-1.[90,121,123] These studies suggest that the localization of system activity may be further defined through specific light chain location. LAT-1 and LAT-2 function as classic obligatory exchangers, moving substrates in exchange for other amino acids,[124,125] and appear unaffected by hypoxia.[98] Two additional System L light chains, LAT-3 and LAT-4 have recently been reported in placental tissues.[119,126] They appear localized to the BM, are present in the human placental BM, and may play an important role in the net efflux of amino acids to the fetus.[22] While transporting a restricted group of System L substrates (leucine, isoleucine, and phenylalanine) they also appear to differ from the traditional System L transport in that they do not appear to require the co-expression of 4F2hc, and they function as facilitative diffusion transporters.[125,126]

Cationic Transport System Light Chain Proteins

Transport of cationic amino acids occurs predominantly through the y^+L and y^+ systems. Placental y^+L system activity is determined by which light chain, System y^+L-amino acid transporter-1 (y^+LAT-1, SLC7A7) or System y^+L-amino acid transporter-2 (y^+LAT-2, SLC7A6), forms a heterodimer with the membrane-bound 4F2hc. Human placental

preparations contain y^+LAT-1 mRNA,[55,56,127] and when this mRNA is co-expressed with 4F2hc in *Xenopus laevis oocytes*, amino acid uptake characteristics similar to that of System y^+L are displayed.[82] Presently, it is unclear if System y^+LAT light chain proteins display a tissue distribution pattern similar to that displayed by the LAT proteins. However, this system appears to provide an essential role in supplying cationic amino acids to the fetal circulation by exchanging cationic amino acids from within the trophoblast for neutral amino acids such as glutamine and leucine from fetal circulation.[57,127] This mode of operation provides a route of efflux, overcoming the outward positive potential difference across the trophoblast basal plasma membrane.[52]

System y^+ activity is dependent only on the expression of monomeric proteins (or the CAT gene products).[63,128] Currently there are three known mRNAs, which encode for proteins related to this cationic amino acid transport system in placental tissues, namely the cationic amino acid transporters CAT-1 (SLC7A1), CAT-2 (SLC7A2), and CAT-4 (SLC7A4).[55,58] This system's activity has been reported in membrane preparations of the rat and human placenta,[50,62] with a higher uptake in the MVMs compared with the BM.[50,129] However, in the human, contrasting kinetic and substrate inhibition studies and RT-PCR studies localizing CAT-1 and 4 mRNA in human placental tissue suggest that y^+ activity is limited to the MVM and not the BM.[55]

Anionic Transport System Light Chain Proteins

Five cDNA-encoding proteins capable of mediating high-affinity sodium-coupled transport have been reported, and three of these have been cloned from placenta tissue.[65,130] These placental proteins, excitatory amino acid transport (EAAT)-1 (GLAST1), EAAT-2 (GLT1), and EAAT-3 (EAAC1), are known to mediate placental sodium-dependent D-aspartate-inhibitable anionic amino acid transport or System X_{AG}^- activity.[130] All three forms have been detected in human placental tissue[66,131] and in rat placenta tissues.[65,132] The regulation of these proteins also appears to be under the control of GH and IGF family members,[132] and upregulation of the system has been demonstrated through cell density and nutrient deprivation studies in muscle and placental cell lines.[35,65] These latter studies further demonstrated that EAAT-1 and EAAT-3 play a key role in the basal anionic amino acid transfer, while EAAT-2 may be involved in conditions of amino acid depletion.[65]

PLACENTAL AMINO ACID TRANSPORT DURING PREGNANCY

As pregnancy advances, the increasing nutrient demands of the developing conceptus must be met through an appropriate increase in placental nutrient transport. This enhanced performance is facilitated through alterations in placental perfusion and changes in membrane exchange surface area. Recent studies in human pregnancies have shown that total umbilical blood flow increases during gestation in a pattern similar to fetal growth rate. The relationship between blood flow and body size is quite good, although not perfect. As shown in Figure 28-4, when blood flow is expressed per kilogram fetal weight, it falls slightly throughout gestation. This pattern of decline is similar to the changes in fetal heart rate in human pregnancies, which also decreases as gestation advances. Heart rate is closely linked to metabolic rate. Thus, the changes in blood flow and in fetal metabolic rate appear to be reasonably well linked in human pregnancies. The decrease in umbilical blood flow per kilogram fetal weight in human pregnancies does not present a problem in terms of supplying amino acids to the fetus as it grows, as changes in blood flow are compensated for by an increase in placental surface area, and presumably an increase in the number of membrane-bound transporters available for delivery of amino acids. Between the 16th week of pregnancy and term, human fetal weight increases approximately 20-fold, whereas the peripheral villous surface area increases only ninefold.[133,134] It should be emphasized, however, that there are only a few careful ontogenetic studies of how total surface area, including microvillous surface, changes over gestation.[135–137] Furthermore, the most important aspect of human fetal development is the acquisition of huge fat stores, most of which occurs in the last few months of pregnancy.[138] During this time, there is almost no increase in nitrogen content while body fat increases exponentially. Hence, it is carbon that must be acquired, not nitrogen, during these last few months of pregnancy.

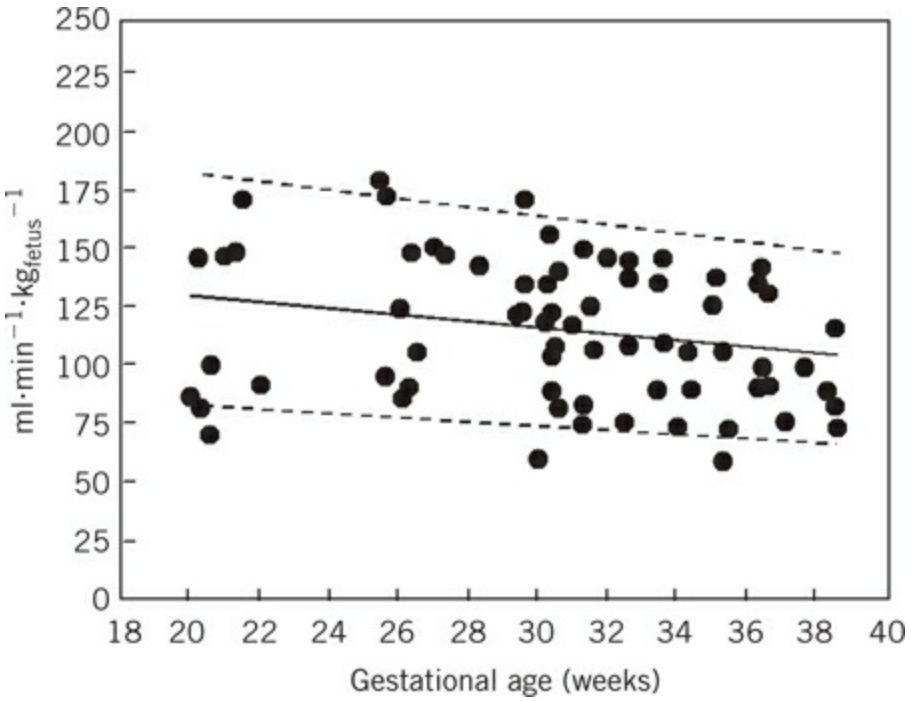


Figure 28-4 Umbilical vein blood flow per kilogram of fetal weight plotted against gestational age for human pregnancies. Slight linear decrease observed was not significant ($y = –1.3636x + 155.9964$). Both 5th and 95th percentiles are shown (dashed lines). Adapted from Barbera et al.[215]

The increases in total surface area of the placenta alone cannot account for the exponential fetal growth occurring over the latter half of gestation. These data suggest the total transport capacity of the trophoblast is likely to be determined by total villous surface area, placental permeability, the concentration of specific amino acid carrier proteins on the cell membrane surfaces, as well as the affinity/activity characteristics of transporter proteins and the circulating amino acid concentrations at both the maternal and fetal surfaces. Indeed, as gestation advances, the efficiency of the placenta as reported as fetal weight per 1 g of placenta increases.

Accompanying the maturational changes in trophoblast surface area, amino acid transport systems are reported to have differing expression and transport parameters through gestation.[55,77,139,140] For example, in term placentae, L-arginine transport in MVM preparation is through both y^+ and y^+L systems, while in the BM transport this may be restricted to the y^+L system.[55] Furthermore, the functional properties of transport systems may

also change as gestation advances. For example, first trimester MVM has increased transport activity compared to term placental vesicles,[140] and 4F2hc protein levels differ between early/mid-pregnancy and term placentae.[55,77] Early in gestation the Km of the MVM high-affinity System y^+L is significantly less than in term preparations.[55] Such studies highlight the complex interactions that occur between developing MVM and BM, within the trophoblast and between the two circulations, to facilitate an increase in nutrient delivery to the growing fetus as gestation advances.

FETOPLACENTAL AMINO ACID METABOLISM

The amino acid requirements of the placenta for energy and for protein accretion during its growth are not well known. The placenta contains enzymes related to carbohydrate and amino acid metabolism that give it the capacity to oxidize a number of amino acids as well as perform glycolysis, glycogenolysis, and glycogen, protein, and fatty acid synthesis.[141,142] Four amino acids (alanine, glutamine, threonine, and serine) that are found in high concentrations in placental tissues and are known gluconeogenic precursors, are transported by the three transport Systems, A, L, and ASC,[38,52,64] which could be a protective mechanism to ensure adequate fetal supply of these important amino acids. So far, amino acid oxidation by human placental mitochondria has been demonstrated for alanine, aspartate, glutamate, and glycine.[143,144] Additionally, given the data on the large uptake of glutamate by the ovine placenta from the fetal circulation, it is possible that glutamate is a predominant amino acid functioning as a metabolic fuel for the placenta[145] where metabolic requirements are very large. In vivo estimates of its oxygen consumption are approximately equal to those for the brain, and oxidation of amino acids is a major metabolic function of the placenta.[146] In vivo, there is a significant production and excretion of ammonia into both uterine and umbilical circulations by the ovine and human placenta.[6,15,16] The absolute rate of uteroplacental ammonia production, approximately 25 μmol/min in the ovine pregnancy at mid-gestation (73–97 days, term 147 ± 3 days), is approximately equal to that of the term placenta. Since in vitro data indicate that there is minimal urea cycle activity within the placenta,[146] this suggests that ammonia, and not urea is the end product of amino acid deamination within the placenta. Some of the ammonia produced by the placenta is consumed by the fetal liver and, in fetal sheep near term, the uptake by the fetal liver is approximately 1.5–1.8 times that taken up by

the umbilical circulation from the placenta.[6,15,146] This indicates that fetal tissues, as well as the placenta, produce ammonia, and uptake of ammonia by the fetal liver undoubtedly contributes to its high rate of fetal urea production.

In part, the placental ammonia production is derived from the transamination of BCAAs.[147] The activity of the branched chain aminotransferases is high in the placenta, and since the activity of α-keto decarboxylases is low, it is clear that the α-ketoacids derived from the BCAA are not oxidized further. These α-ketoacids represent part of a group of compounds that are produced in the placenta and are delivered into both uterine and umbilical circulations. These compounds include ammonia, lactate, and one of the polyols, sorbitol (Figure 28-5). The placental production of glutamate as a consequence of transamination of α-ketoglutarate from BCAA,[145,148,149] together with the placental uptake of fetal plasma glutamate at 4.8 μmol/min/kg fetus,[145] represent the two major influxes of glutamate into the ovine placenta. Although the ovine placenta contains the enzyme glutamine synthetase, only a small fraction of glutamate that is taken up by the placenta from fetal circulation is returned to the fetus as glutamine.[145,150] It is the oxidation of glutamate that appears to be the predominant pathway of placental glutamate disposal.

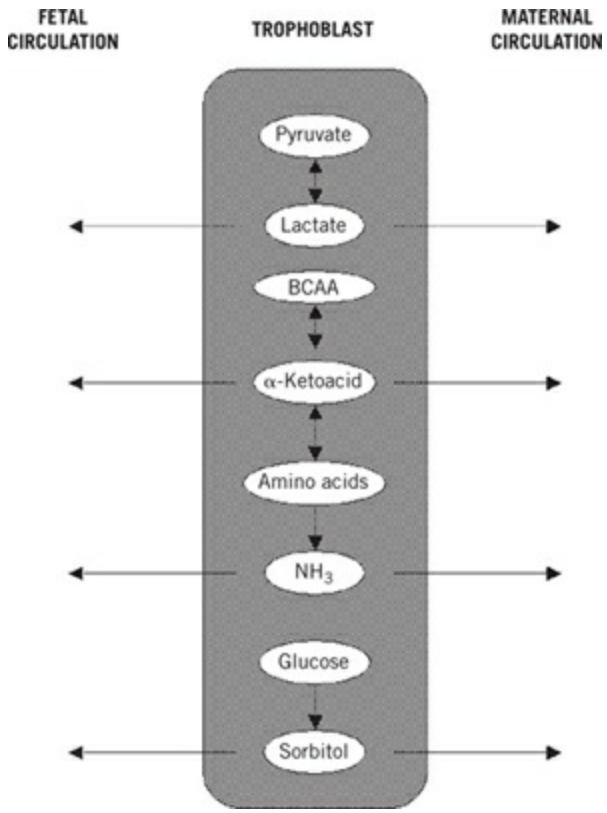


Figure 28-5 A schematic representation of some of the metabolites produced in the placenta that are then delivered into both the maternal and fetal circulations.

It is likely that placental ammonia production is not confined to the epitheliochorial placenta of the sheep. It has been described for the perfused human placenta, and a net efflux of ammonia from the pregnant uterus into the maternal circulation has also been found in rabbits, guinea pigs, and sheep.[148,150–152] It is intriguing to speculate on the possible biologic significance of mammalian fetuses growing and developing in an environment of an elevated ammonia concentration. Finally, ammonia can affect hepatic metabolism and may play a role in glial development and in the development of other tissues rich in glutamine synthetase.

UMBILICAL UPTAKE OF AMINO ACIDS

The umbilical uptake of amino acids (Figure 28-1), is the key step marking the delivery of amino acids into fetal circulation following placental uptake and metabolism. A number of studies involving examination of the

umbilical uptake of amino acids in chronic, unstressed sheep models have revealed that most amino acids (particularly the neutral amino acids) are delivered into the fetal circulation in amounts that exceed their net rates of accretion.[2,5,16,149] The most complete data on the uterine and umbilical uptakes of amino acids are presented in Figure 28-6. 149 A key question, however, is: Upstream of this ultimate delivery to the fetus, how are maternal concentrations of amino acids related to the umbilical uptake? This question is important because it would help define the extent to which changes in maternal amino acid concentrations could impact the fetal delivery of amino acids. The question has been studied in both ovine and human pregnancies by infusions of amino acids into the maternal circulation with measurements of their umbilical uptake. The relationship between maternal concentration and umbilical uptake is a complex one. In ovine studies, the flux from the maternal circulation to the fetal circulation is most rapid for five amino acids and for those five (methionine, phenylalanine, leucine, isoleucine, and valine), differences in their fluxes are directly attributable to differences in their normal maternal plasma concentrations,[17] as shown in Figure 28-7. In sheep and human pregnancies, it is possible to increase the fetal delivery of most amino acids by increasing the maternal concentration in normal pregnancies.[16,153] This appears also to be the case for pregnancies complicated by FGR.[23,154] However, for some amino acids, it has not been possible to increase placental transport when an infusion of many amino acids is given to the mother, presumably due to competitive inhibition by other amino acids in the infusate.[16] Indeed, the description given earlier of the multiple transporter proteins and their amino acid specificity is complex (probably due to difficulties in standardizing techniques for isolating tissue membrane fragments); however, the general principle that emerges is that all transporters from the in vitro and in vivo studies, virtually without exception, are utilized by several amino acids. This simple observation explains the difficulties encountered in trying to increase the delivery of amino acids to the fetus by infusions of amino acids into the maternal circulation. Competitive inhibition has been well demonstrated in vivo leading to a decrease in transport of some amino acids to the fetus despite an increased concentration in the maternal circulation.[16] It also explains the futility of attempting to establish a constant amino acid concentration in the fetus (an amino acid clamp) by a fetal infusion of amino acids. Competitive inhibition will lead to inhibition of the entry of amino acids into the fetal tissues when other amino acids are increased.

Measurements of unidirectional flux are currently available only for the ovine species. A number of studies involving examination of the umbilical uptake of amino acids in chronic, unstressed sheep models have revealed that most amino acids (particularly the neutral amino acids) are delivered into the fetal circulation in amounts that exceed their net rates of accretion.[5,16,149,155,156] The most complete data on the uterine and umbilical uptakes of amino acids are presented in Figure 28-6.

Several studies of the umbilical uptake of amino acids under normal conditions have shown that there is no measurable uptake of aspartate and, in fact, a significant net uptake from the fetal circulation into the placenta of serine and glutamate. Thus, for these three amino acids, it is clear that fetal requirements must be met by production within fetal tissues. Stegink et al.[25,157] infused labeled tracer isotopes of glutamate and aspartate into both fetal and maternal circulations of rhesus monkeys and found essentially no transfer of the compounds across the placenta. Schneider et al., [158] utilizing an in vitro–perfused human placenta preparation, found significant uptake of glutamate from the fluid perfusing the fetal side of the placenta rather than a net transfer of glutamate from the maternal to the fetal side.

It is clear that the umbilical uptake of amino acids cannot be considered solely a reflection of the amino acid supply derived from the maternal plasma pool via transplacental transport, but also may include a contribution derived from production within the placenta or release from placental breakdown.[159] It is important to understand which amino acids are provided to the fetus primarily through placental production versus placental transport, because differing forms of placental pathology may adversely affect the mode of delivery of those amino acids to the fetus.

FETAL AND PLACENTAL OXIDATION

The use of multiple-tracer methodology in animal models has allowed further elucidation of amino acid metabolism within the placenta and the fetus. Fetal and placental metabolism of the essential amino acids, leucine and threonine, have been studied in some detail.[18,160–162] For leucine, Loy et al.[160] were the first to demonstrate that even for an essential amino acid there may be extensive catabolism both within the fetus and within the placenta. They found that 20%–25% of the (1-^{14}C) leucine infused into the fetus could be accounted for as $^{14}CO_2$ produced by the fetus. Unlike glutamate, there was no measurable oxidation of leucine within the

uteroplacental tissues. This indicated an extensive utilization of leucine as a fetal but not placental fuel. From the sheep studies, the extensive fetal catabolism of leucine can be better appreciated by a comparison of its supply to the fetus via the placenta and its rate of oxidation and of accretion in tissue protein. The total umbilical uptake of leucine is approximately 4.9 μmol/min/kg, and net accretion of leucine in tissue proteins accounts for only 2.0 μmol/min/kg in late gestation. On the other hand, the rate of leucine oxidation is 2.8 μmol/min/kg or 57% of its uptake.

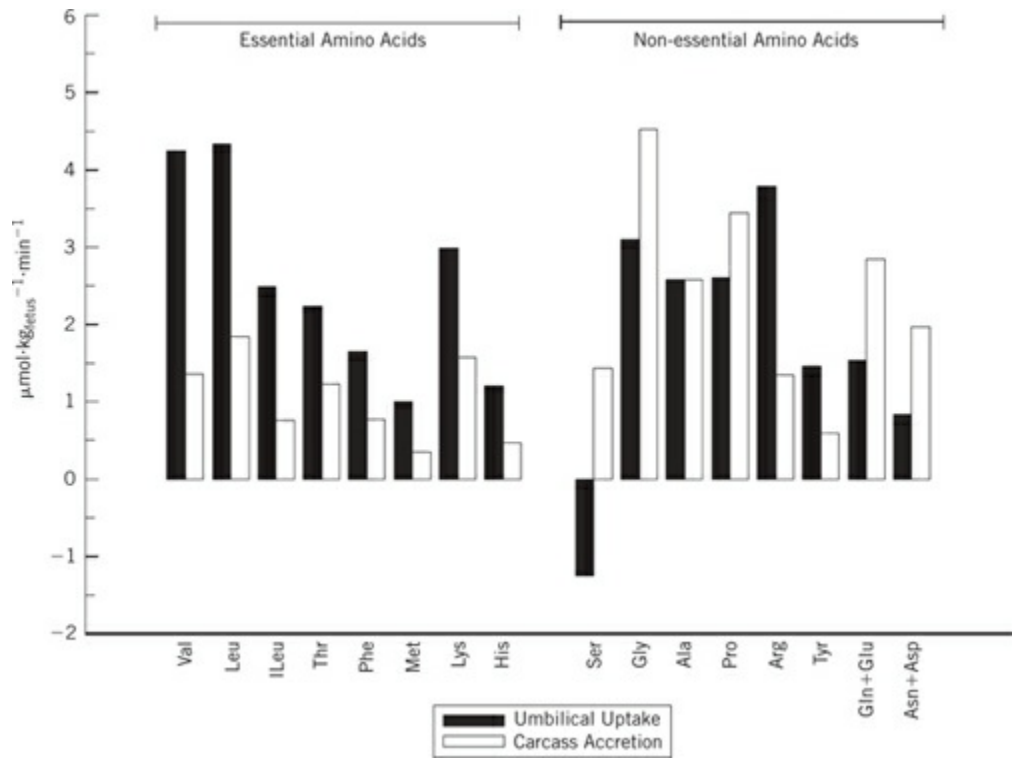


Figure 28-6 Comparison between mean umbilical uptakes and estimated normal fetal accretions of amino acids. For pairs of amino acids (glutamine + glutamate and asparagine + aspartate) individual accretion rates are not known. Adapted from Chung et al.[149]

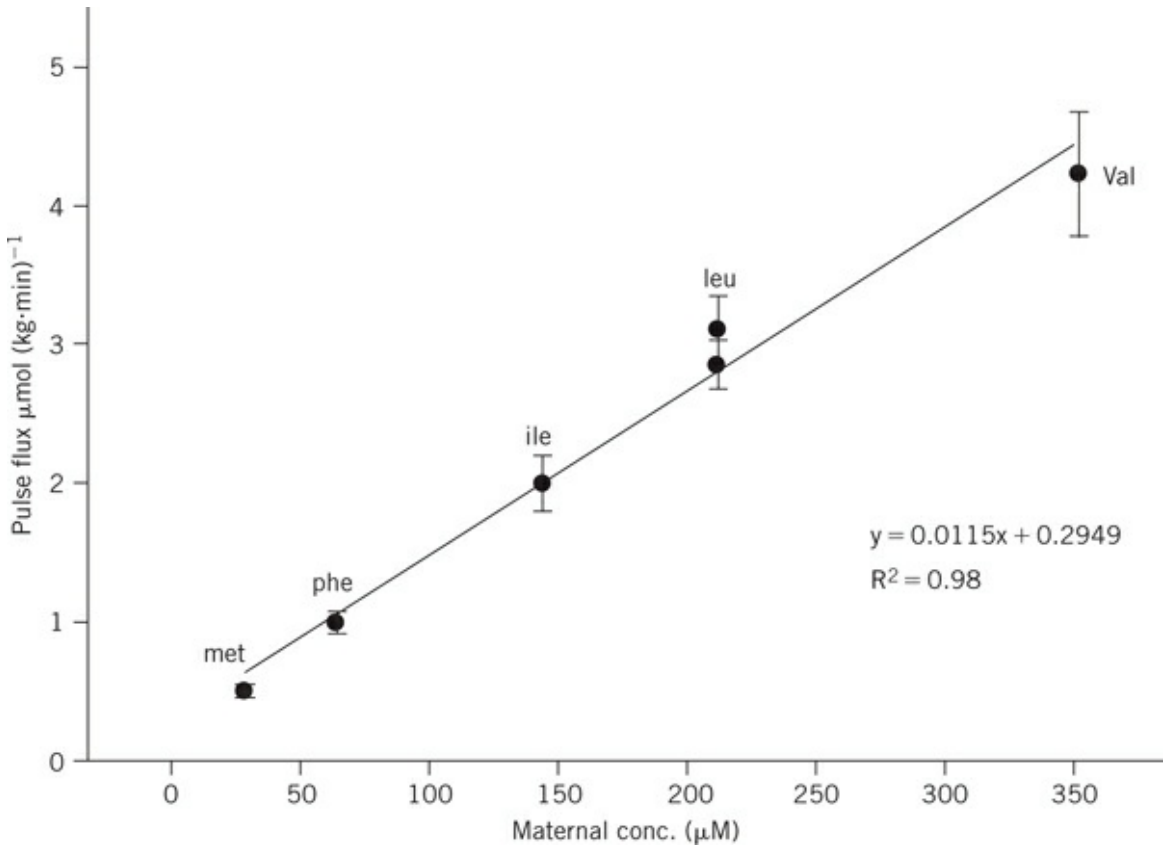


Figure 28-7 Pulse flux calculated over the first 10 minutes after the bolus versus maternal plasma arterial concentration for the five essential amino acids that have high and similar pulse flux clearances. The two leucine data represent two separate studies. Adapted from Paolini et al.[17]

The oxidation rates of amino acids in the fetus and/or placenta of human and predominantly sheep preparations have been measured for leucine, lysine, threonine, alanine, glycine, serine, and glutamate[17,145,160–164]; and it is apparent that fetal oxidation of amino acids is not confined to the nonessential amino acids. In fact, it is an important safety feature for fetal nutrition that amino acids are provided in amounts which exceed their rates of accretion.[3,5] This mandates a fairly high oxidation rate for most amino acids including the essential amino acids. It is not surprising, therefore, that there is a relatively high rate of urea production by the fetus. In the sheep this has been shown by direct measurements of $^{14}CO_2$ production from labeled amino acids and from the high fetal urea production rate in the fetal lamb,[3] and estimated for the

human fetus.[165,166]

FETAL HEPATIC CIRCULATION AND AMINO ACID METABOLISM

The umbilical vein carries the most oxygenated and nutrient-rich blood of the fetus and a large fraction of this blood perfuses the fetal liver. Thus, the fetal liver, in contrast to the postnatal liver, is a site of preferred oxygenation. It is a vitally important defense for the fetus during periods of chronic or acute hypoxia, because it continues to take up lactate from the fetal circulation and releases pyruvate. Other fetal tissues can then metabolize pyruvate. For human pregnancies, the percent of umbilical blood flow shunted away from the fetal liver through the ductus venosus has now been measured.[167] The shunt gradually decreases from 40% to 15% over a gestational age range from 20 weeks to term. However, in some FGR pregnancies, this shunt is significantly increased.[167,168] Since other studies have shown a reduction in umbilical blood flow per kilogram fetal weight in FGR pregnancies,[169] this in itself reduces perfusion of the fetal liver. However, if the reduction in umbilical flow is coupled with an increased ductus venosus shunt, the decrease in hepatic perfusion can be profound. These changes in the fetal circulation must be kept in mind in terms of their potential importance upon fetal hepatic metabolism.

Fetal hepatic uptake of amino acids has been studied in late-gestation fetal lambs, and fetal hepatic uptake and the umbilical uptake of amino acids have been compared.[170] A large hepatic uptake of the entire essential and most of the nonessential amino acids by both lobes of the fetal liver has been reported.[5] During normal fetal development, there is no appreciable rate of fetal gluconeogenesis. This is understandable since fetal hepatic glucose production would be self-defeating for the fetus. It would increase plasma glucose concentration, which would reduce the transplacental glucose gradient leading to a reduced umbilical glucose uptake from the mother. The absence of fetal gluconeogenesis is not accompanied by a reduced amino acid uptake in the fetal circulation from the placenta,[149,171] as shown in Figure 28-8. There is a large uptake of amino acids, including all of the gluconeogenic amino acids, as well as a large fetal uptake of lactate. In fetal life, the carbon is released primarily as glutamate and pyruvate with a smaller contribution occurring with the hepatic release of serine, ornithine, and aspartate. This is in contrast to what occurs in postnatal life, where the carbon from these amino acids is

released from the liver solely as glucose. The hepatic uptake of lactate and release of pyruvate are of special interest because of the importance of these compounds during fetal hypoxia. The uptake of lactate and release of pyruvate continues under conditions of fetal hypoxia providing pyruvate, which can be metabolized by nonhepatic tissues even under hypoxic conditions. The hepatic metabolism of lactate and production of pyruvate help explain the paradox of severe fetal hypoxia associated with a relatively normal pH.[172]

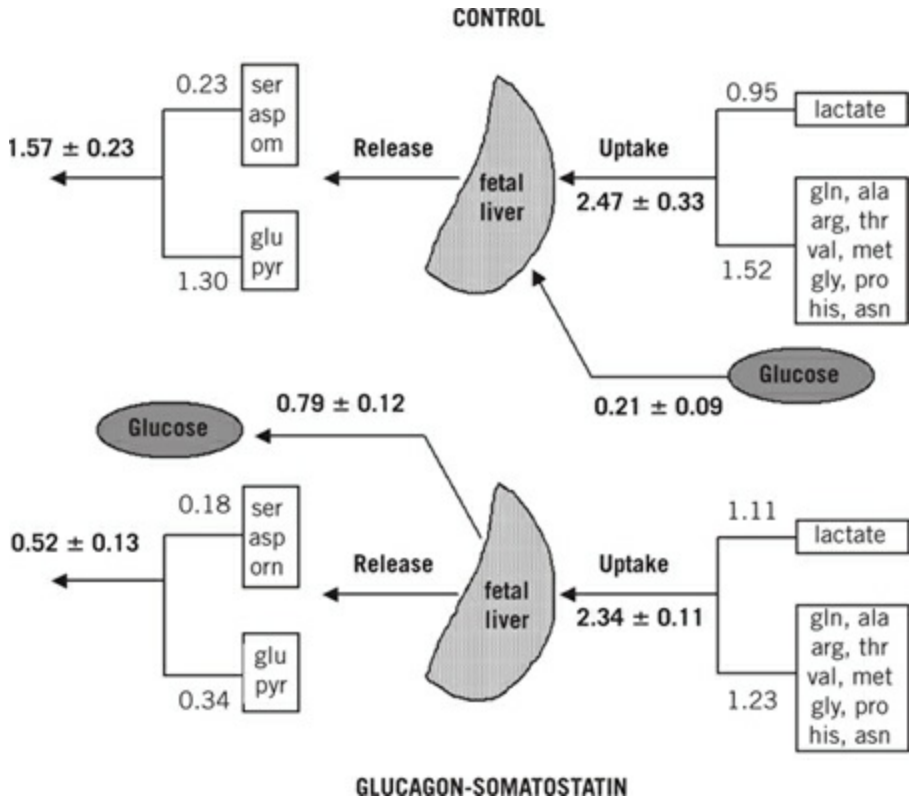


Figure 28-8 Fetal hepatic uptake and output of glucose carbon and glucogenic substrate carbon under normal physiologic conditions (Control) and during a fetal glucagon somatostatin infusion. Each number represents a substrate carbon to oxygen uptake ratio. Adapted from Teng et al.[171]

The interorgan shuttling of the two amino acid pairs, glutamate and glutamine and serine and glycine, play an important role in this overall carbon balance for the fetus. The release of glutamate, serine, and pyruvate effectively replaces the glucose carbon release characteristic of the liver in postnatal life. This is clearly brought out in Figure 28-8, which shows what

happens when gluconeogenesis is triggered in the fetal liver by the infusion of glucagon.[171] With the advent of net glucose released from the liver, the efflux of glutamate, serine, and pyruvate is markedly reduced, and the overall carbon balance is maintained. The exchange of glutamate and glutamine between the fetal liver and placenta does not represent a cycling of carbon between the two organs, as glutamate is largely oxidized in the placenta and in the fetus.[145] The exchange of glutamate and glutamine changes quite markedly during parturition and during glucocorticoid or glucagon administration to the fetus.[171,173] Under these conditions, there is a decrease of fetal hepatic glutamate release that results in a decrease of placental glutamate uptake. What role these metabolic changes in the fetal liver play in parturition is not yet established.

Glycine and serine are a metabolically related pair of amino acids that demonstrate a similar pattern of release and uptake across the ovine fetal liver and placenta to that of glutamate and glutamine,[5,174–176] and there are now studies reporting their metabolism in human pregnancy.[177] The fetal glycine oxidation rate is high, and glycine is almost totally oxidized within the fetal liver. Fetal hepatic glycine oxidation and serine production from glycine are linked in the fetal liver by the enzyme systems glycine oxidase and serine hydroxymethyltransferase (SHMT).[178,179] The combined action of these enzymes is the production of one mole of CO_2 and one mole of serine for every two moles of glycine consumed.[174,180] This is also true in adult tissues,[181] though the adult liver is not normally a site of net serine release.[182]

Infusion of tracer serine into the fetus has demonstrated a rapid bidirectional transport of serine between the fetal blood and placenta, as well as placental conversion of fetal plasma serine to glycine. In contrast to glycine oxidation, the bulk of fetal plasma serine oxidation occurs in extrahepatic tissues.[164,180] Moores et al. infused labeled serine into the maternal circulation of pregnant sheep and found that despite high maternal enrichments of serine, there was no fetal plasma enrichment evident. However, the infusion of maternal tracer serine did lead to the appearance of labeled glycine in the fetal circulation. Thus, placental glycine production utilizes serine derived from both the maternal and fetal circulations.[164] This brings out the complexity of pathways by which an amino acid such as glycine is delivered to the fetus from the placenta. The pathway for glycine production from serine is thought to be via serine SHMT, since ovine placental tissue is known to have moderately high activity for the mitochondrial isoform of this enzyme.[164,179] A byproduct

of placental glycine production via SHMT is methylene tetrahydrofolate, which could be involved in a variety of methylation reactions within the placenta.[179] These examples of placental-fetal interorgan shuttling of amino acids, and particularly between the fetal liver and placenta, are now recognized to be an important and unique aspect of fetal metabolism.

FETAL HINDLIMB METABOLISM OF AMINO ACIDS

Several laboratories have studied the amino acid uptake by the fetal ovine hindlimb as being representative of the fetal carcass/extrahepatic tissues. Its metabolism has been evaluated under normal, maternal fasting, rhIGF infusions, and fetal euglycemic hyperinsulinemic states.[182–185] Under normal physiologic conditions, the hindlimb of a growing fetus shows a net uptake of both essential and nonessential amino acids from the fetal circulation,[184] which is in contrast to adult hindlimb studies that show a large output of alanine and glutamine.[185] It is theorized that the high amino acid uptake by the fetal hindlimb could be explained by the relatively high nitrogen accretion rate of the fetus.[184] There is a net uptake across the hindlimb of two amino acids that are not supplied by the placenta: glutamate and serine. It is estimated that the net fetal hepatic output could account for the combined uptake of glutamate by the placenta and the hindlimb.[184] Given the large uptake of alanine by the fetal liver, it is interesting that the fetal hindlimb shows no net release of alanine.[185]

Liechty et al.[186] reported a relatively high activity for ketoacid carboxylase activity in fetal skeletal muscle. In pregnant sheep, after fasting, there is an approximate fivefold increase in the utilization of glutamine and a twofold increase in the utilization of the BCAAs by the uteroplacental tissues, suggesting a possible interorgan cycle between hindlimb and placenta, with the hindlimb supplying glutamine during the maternal fasted state.[186] Under euglycemic, hyperinsulinemic conditions, fetal lambs were noted to have significantly decreased arterial concentrations of most amino acids, and significantly increased limb uptake of alanine, asparagine, glycine, isoleucine, methionine, and tyrosine, and a decreased uptake of aspartate, but with no significant change in net total nitrogen uptake.[184,186] These studies show that the stimulation of alanine uptake by insulin is opposite to the effect of maternal fasting, suggesting that insulin or IGFs may play a role in regulating an alanine exchange between fetal circulation and nonvisceral

fetal tissues.[184,186]

AMINO ACID TRANSPORT SYSTEMS AND ABNORMAL PREGNANCY OUTCOMES

In several clinical studies, the concentration of amino acids has been observed to be lower in the fetal and maternal plasma of pregnancies that were complicated by FGR,[187–189] suggesting alterations in amino acid delivery. This depression in amino acid concentrations occurs regardless of whether fetal concentrations were determined at the time of delivery or many weeks prior to delivery. Additionally, it has also been found that lower amino acid concentrations are found in FGR pregnancies even if the fetus had normal fetal heart rate and velocimetry measurements,[187,188] suggesting that this may antedate other clinical pathologic findings.[190]

Studies in human FGR demonstrate that System A activity may be impaired.[191,192] An in vitro study utilizing MVM vesicles from the placentae of appropriate for gestational age (AGA) and small for gestational age (SGA) babies demonstrated markedly lower activity (by 63%) of the A system transporters in the SGA compared to the AGA membrane vesicles, suggesting a positive association between fetal growth and System A activity.[192,193]Further, studies with placental villous fragments from pregnancies with SGA infants with and without maternal preeclampsia and from uncomplicated and preeclamptic pregnancies with normal-sized infants have shown there is a reduced uptake of amino acids in SGA pregnancies without preeclampsia.[191]

Inhibition of System A transport in rat pregnancies has been associated with slight decreases in fetal weight, demonstrating a potential role for System A transport in maintaining normal fetal growth.[192] Of particular interest is that imprinted genes control the supply of nutrients to the fetus, and, in the mouse, the System A isoform, SNAT2 (Slc38a4), has been identified as being an imprinted gene. In studies utilizing the placental-specific Igf2 transcript (P0) knockout, placental growth is compromised, though fetal demands are met presumably through an up regulation of transporter genes Slc2a3 (glucose transporter) and the imprinted SNAT during mid-gestation, though intrauterine growth retardation (IUGR) still results near term.[194–196], Interestingly, when fetal demands for nutrients are removed through removal of the fetal Igf2, the up regulation does not occur, providing evidence of regulation of placental transport activity via fetal growth demand genes. Both the Igf2 and Slc38a4 are paternally

expressed, and this interaction is seen as a display of paternally inherited genes maximizing extraction of maternal nutrients.[194–196]

In human FGR vesicle studies the TAUT protein expression is unaltered,[117] though the transport activity of this transporter appears to be reduced.[47] However, it is important to emphasize that vesicle studies have not taken into account the possibility of nonhomogeneity in the placenta for FGR pregnancies. There remains the very real possibility that the proportion of terminal villi to intermediate and stem villi are altered in FGR pregnancies, and, depending on the distribution of any transporter, this maldevelopment could profoundly affect transport in vivo without any changes in transporter expression or activity.

Altered System A, and also Systems X_{AG}^- and y^+ activity, have been reported in maternal protein deprivation induced FGR in rats.[60,197] However, again one must be cautious in extrapolating such studies to humans since protein deprivation in the rat has a profound effect upon maternal plasma amino acid concentrations, a condition not found in human FGR pregnancies. Also, amino acid accretion and utilization of a rat fetus is much greater than the human fetus given the much faster growth rate of the rat fetus, up to 20 times faster near term. With regard to System X_{AG}^- transport, the BM activity of EAAT-1 is reduced in an FGR rat model, which could impact placental glutamate uptake from the fetal circulation,[60] though in human studies, no differences were found in the distribution of the glutamate transporters between control and IUGR placentae.[66]

MVM System y^+ and y^+L activity does not appear to be altered in human FGR,[50] though uterine oxygen tension may impact overall cationic amino acid transport.[127] Additionally, BM transport of lysine is reduced as represented by reduced mediated lysine uptake and a reduced Vmax for the y^+L system,[50] and in pregnancy, array data demonstrate up regulation of System y^+ components.[57] Reductions in the uptake of leucine in both MVM and BM preparations of FGR placentae highlight possible alterations in the L transport system.[50] These changes in BM transport properties could be an important adaptive response by the trophoblast, limiting the back flux of amino acids from the fetal circulation to the placenta. In vivo studies of ovine FGR placental transport have indeed shown a reduced backflux of leucine and threonine from the fetal circulation to the placenta, suggesting that changes in BM function may have occurred.[161] At steady state, the fetal/maternal (F/M) enrichment

ratioof leucine in normal human pregnancies is approximately 0.8, a much higher ratio than the 0.4 ratio in sheep.[23,161,198] However, in both species, this ratio is significantly lower in FGR pregnancies. In the ovine FGR model, the reduction in F/M enrichment ratio was due to a significant reduction in transplacental leucine flux,[161] and that is probably the mechanism in human FGR pregnancies as well. Furthermore, the magnitude of the leucine ratio reduction correlates with a clinical classification of FGR severity based upon a completely different set of clinical data, namely, fetal arterial velocimetry and fetal heart rate data.[190] In addition, recent clinical studies have shown that the leucine and phenylalanine F/M enrichment ratio is significantly lower in human FGR compared to normal pregnancies, and that vesicle transporter activity is also impaired.[23,50,198]

Finally, the observations of differences in amino acid transport activity in the FGR placenta highlight the important point that the activity of transport systems must be considered in conjunction with other contributors to amino acid flux. In studies comparing normal and FGR placentas there are reported reductions in total villous surface area,[135,199] suggesting that morphometric changes contribute to the overall reduction in placental amino acid transport capacity. Thus, at the present time, it appears that both reductions in surface area for exchange and changes in terminal villi/stem villiproportions as well as reductions in specific transporter number and activities are major factors contributing to the reduction in amino acid transport in FGR pregnancies.

While there is a developed knowledge on amino acid transport changes in FGR pregnancy, there is a large deficit in our understanding of how these systems are modulated in pregnancies associated with obesity. Studies show that a maternal high-fat diet in pregnant rats causes up regulation of placental nutrient transport (GLUT1 and SNAT2), resulting in fetal overgrowth.[200] Much of the work concerned with placental function and obesity is currently focused around System A. Regulation of System A activity in obesity is associated withincreased cytokine levels and altered maternal adiponectin levels. Physiological concentrations of the proinflammatory cytokines, IL-6, and tumor necrosis factor-α, in cultured human primary trophoblast cells, are found to stimulate the activity of amino acid transporter System A, but not System L,[100] while adiponectin appears to negatively impact insulin-stimulated System A amino acid transport.[201] Also associated with obesity are elevated circulating fatty acids. Oleic acid in culture is found to significantly

upregulate trophoblast System A activity.[106] Further, studies suggest that obesity associated activation of insulin/IGF-I and mTOR signaling pathway also play a pivotal role in the activation of System A,[97] highlighting that fetal overgrowth associated with obesity is the result of a complex interaction of factors culminating in the increased activity of System A. Many of these studies established that System L was unaltered in these situations,[97] opening the question of how other amino acid transport systems are impacted in obesity.

NEONATAL IMPLICATIONS OF FETAL AMINO ACID SUPPLY AND METABOLISM

One may ask how studies of amino acid nutrition and metabolism during fetal life can contribute to our approach to optimal nutrition for very premature infants in a neonatal intensive care unit. First, it is important to recognize that normal fetal development and postnatal development are vastly different biologic states. Not only are there profound endocrine changes triggered by delivery and its associated separation from the placenta, but also the interorgan cycling of amino acids between the fetal organs and placenta is abruptly terminated. However, certain principles of amino acid metabolism during early development still apply. To summarize, during fetal development, amino acids are taken up in amounts that far exceed their requirements for growth.[7,9,11,18,202] The excess amino acid intake leads to a high rate of amino acid oxidation and a relatively high rate of urea production.[15] A second general characteristic is the absence of gluconeogenesis. The accommodation to this leads to a hepatic production and release of serine and glutamate that reduce the CO_2 production requirements of the fetal liver just as glucose production does in postnatal life. It is important to emphasize that the requirements of amino acids change quite markedly with increasing gestational age, and also depend upon whether there has been normal or abnormal fetal growth. In the normal term newborn, the rate of net protein accretion in utero is quite low, with fat accretion dominating growth. In FGR fetuses with relatively mild FGR, there are reduced plasma amino acid concentrations reflecting the reduced placental supply. In severe FGR, with virtually no fetal growth, plasma concentrations are not affected. There may also be vascular changes, which lead to hepatic injury.

The delivery of optimal nutrition to the small premature infant is a critical factor to the infant's overall care.[203–205] Very premature infants

have been delivered before they have gone through the phase of in utero development associated with rapid accretion of fat depots and glycogen stores. Preterm infants are born at gestational ages associated with rapid accretion of protein in utero. This has led to a neonatal nutritional approach, which emphasizes the early provision of amino acids to very preterm babies.[204] It should be emphasized, however, that this makes an assumption of good liver perfusion and the absence of any prenatal liver injury. Neither of these conditions necessarily applies to an individual newborn in the immediate neonatal period. The best guideline for assessing the nutritional status of the low-birth weight infant is not clear. However, one approach would include achieving a postnatal growth that approximates the in utero growth rate of a normal fetus.[206] By this guideline, the intrauterine accretion rate of amino acids becomes a reference point defining the minimal requirements in the neonatal period. As delineated in the previous discussion, the mammalian fetus utilizes the circulating maternal free amino acid pool as its primary nitrogen source for growth with certain conspicuous exceptions. Since amino acid oxidation is linked to hepatic gluconeogenesis in postnatal life, and since gluconeogenesis is a requirement for postnatal adaptation, one would expect amino acid oxidation to be sustained during postnatal development, significantly increasing amino acid requirements. For the premature infant, the primary sources of nitrogen may be obtained from a variety of sources including that obtained from parenteral nutrition, or human breast milk, or premature infant formulas. Current nutritional management for preterm and FGR infants aims at early provision of amino acids.[204,205]

Recommendations for the quantity of protein delivery include 3.6–3.8 g/kg/d gross protein intake for the stable growing infant at less than 27 weeks gestation and less than 1000 g, and 3.0–3.6 g/kg/d for the 1000- to 1750-g infant at 28- to 34-week gestation. Although the exact quantity is debated (i.e. European guidelines versus American guidelines), it is generally accepted that the stable growing low-birthweight infant requires, on the average, a parenteral amino acid intake of 3.0 g/kg/d to ensure accretion of nitrogen at the intrauterine rate of approximately 300 mg/kg/d. This amino acid intake plus an energy intake of 80–100 kcal/kg/d results in a weight gain that approximates the intrauterine rate of 15 g/kg/d.[207] However, the studies of the fetal circulation in FGR pregnancies makes it clear that the general recommendations should be used with caution in newborns of any weight or gestational age when there is evidence of circulatory compromise in utero. A high carbohydrate and relatively low nitrogen intake is prudent in these circumstances. Certainly, if a high

nitrogen intake is given to such infants, there should be an assessment of ammonia and urea levels as well as measurement of amino acid concentrations to be sure the nitrogen load is not compromising the infant. This caution reflects the fact that there are still no ideal approaches to assess both the quality and quantity of amino acids provided as protein in milk feedings to low-birth-weight infants. The assumption that the accretion of nitrogen should approximate that of a fetus of comparable gestational age has not been adequately tested. Given the radically different environmental conditions of the preterm infant versus the fetus and the marked endocrine changes imposed by delivery of the infant and loss of the placenta, this untested assumption should be viewed with a healthy skepticism. Only long-term follow-up studies of preterm infants fed differently will clarify what nutritional approach is best. Breast milk provides a large array of compounds not included in formula feedings. Some of these compounds such as glycoproteins and peptides contain amino acids. Owing to the instability of glutamine it is currently omitted from parenteral nutrition solutions and infant formulas. Glutamine has recently been found to be an important metabolic fuel for lymphocytes[208,209] and intestinal epithelial cells,[210] and is supplied to the ovine fetus in large amounts throughout gestation.[149]

It has already been demonstrated that formula-fed, very-low-birth-weight infants whose weight gain was comparable to that of a normal fetus retained the same amount of protein (≈ 1.9 g/kg/d), but accumulated fat at approximately three times the rate of the fetus. This indicates that the body composition of the premature infant will differ from that of the comparable placentally nourished fetus.[211] How this will impact on long-term growth and neurodevelopment is unclear and a continuing discussion point.[212,213] It should also be pointed out that the term “amino acids” applies to a group of compounds with vastly different transport properties and metabolic properties. It has already been shown in animal studies that if the concentrations of all amino acids are increased in the maternal circulation, the supply of some amino acids to the fetus is significantly reduced despite the increased maternal concentration through the impact of competitive inhibition.[214]

As the complex interplay among fetus, placenta, and maternal dietary supply of amino acids becomes increasingly clear, it is hoped that this information can be used to understand more completely the remarkable process of fetal growth and maturation. This, in turn, will then be invaluable information in the further modification and design of feeding regimens for premature infants in order to optimize growth and long-term

neurodevelopment.

REFERENCES

1. Young M, Mc Fadyen IR. Placental transfer and fetal uptake of amino acids in the pregnant ewe. *J Perinat Med*. 1973;1(3):174–182.
2. Lemons JA, Schreiner RL. Amino acid metabolism in the ovine fetus. *Am J Physiol*. 1983;244(5):E459–E466.
3. Gresham EL, James EJ, Raye JR, et al. Production and excretion of urea by the fetal lamb. *Pediatrics*. 1972;50(3):372–379.
4. Lemons JA. Fetal-placental nitrogen metabolism. *Semin Perinatol*. 1979;3(2):177–190.
5. Marconi AM, Battaglia FC, Meschia G, Sparks JW. A comparison of amino acid arteriovenous differences across the liver and placenta of the fetal lamb. *Am J Physiol*. 1989;257(6 Pt 1):E909–E915.
6. Holzman IR, Lemons JA, Meschia G, Battaglia FC. Ammonia production by the pregnant uterus. *Proc Soc Exp Biol Med*. 1977;156(1):27–30.
7. van Veen LC, Teng C, Hay WW, et al. Leucine disposal and oxidation rates in the fetal lamb. *Metabolism*. 1987;36(1):48–53.
8. Milley JR. Protein synthesis during hypoxia in fetal lambs. *Am J Physiol*. 1987;252(4 Pt 1):E519–E524.
9. Brown LD, Rozance PJ, Thorn SR, et al. Acute supplementation of amino acids increases net protein accretion in IUGR fetal sheep. *Am J Physiol Endocrinol Metab*. 2012;303(3):E352–E364.
10. Kennaugh JM, Bell AW, Teng C, et al. Ontogenetic changes in the rates of protein synthesis and leucine oxidation during fetal life. *Pediatr Res*. 1987;22(6):688–692.
11. Maliszewski AM, Gadhia MM, O'Meara MC, et al. Prolonged infusion of amino acids increases leucine oxidation in fetal sheep. *Am J Physiol Endocrinol Metab*. 2012;302(12):E1483–E1492.
12. Brown LD, Hay WW. Effect of hyperinsulinemia on amino acid utilization and oxidation independent of glucose metabolism in the ovine fetus. *Am J Physiol Endocrinol Metab*. 2006;291(6):E1333–E1340.
13. Meznarich HK, Hay WW, Sparks JW, et al. Fructose disposal and oxidation rates in the ovine fetus. *Q J Exp Physiol*. 1987;72(4):617–625.
14. Meschia G, Battaglia FC, Hay WW, Sparks JW. Utilization of substrates by the ovine placenta in vivo. *Fed Proc*. 1980;39(2):245–9.
15. Holzman IR, Philipps AF, Battaglia FC. Glucose metabolism, lactate, and ammonia production by the human placenta in vitro. *Pediatr Res*. 1979;13(2):117–120.
16. Jozwik M, Teng C, Battaglia FC, Meschia G. Fetal supply of amino acids and amino nitrogen after maternal infusion of amino acids in pregnant sheep. *Am J Obstet Gynecol*. 1999;180(2 Pt 1):447–453.
17. Paolini CL, Meschia G, Fennessey PV, et al. An in vivo study of ovine placental transport of essential amino acids. *Am J Physiol Endocrinol Metab*. 2001;280(1):E31–E39.
18. van den Akker CH, Schierbeek H, Minderman G, et al. Amino acid metabolism in the human fetus at term: leucine, valine, and methionine kinetics. *Pediatr Res*. 2011;70(6):566–571.
19. Smith CH, Adcock EW, Teasdale F, et al. Placental amino acid uptake: tissue

preparation, kinetics, and preincubation effect. *Am J Physiol*. 1973;224(3):558–564.
20. Boyd CA, Lund EK. L-proline transport by brush border membrane vesicles prepared from human placenta. *J Physiol*. 1981;315:9–19.
21. Schneider H, Proegler M, Sodha R, Dancis J. Asymmetrical transfer of alpha-aminoisobutyric acid (AIB), leucine and lysine across the in vitro perfused human placenta. *Placenta*. 1987;8(2):141–151.
22. Cleal JK, Glazier JD, Ntani G, et al. Facilitated transporters mediate net efflux of amino acids to the fetus across the basal membrane of the placental syncytiotrophoblast. *J Physiol*. 2011;589(Pt 4):987–997.
23. Paolini CL, Marconi AM, Ronzoni S, et al. Placental transport of leucine, phenylalanine, glycine, and proline in intrauterine growth-restricted pregnancies. *J Clin Endocrinol Metab*. 2001;86(11):5427–5432.
24. Kerr GR. The free amino acids of serum during development of macaca mulatta. II. During pregnancy and fetal life. *Pediatr Res*. 1968;2(6):493–500.
25. Stegink LD, Reynolds WA, Filer LJ, et al. Monosodium glutamate metabolism in the neonatal monkey. *Am J Physiol*. 1975;229(1):246–250.
26. Palou A, Arola L, Alemany M. Plasma amino acid concentrations in pregnant rats and in 21-day foetuses. *Biochem J*. 1977;166(1):49–55.
27. Novak DA, Beveridge MJ. Glutamine transport in human and rat placenta. *Placenta*. 1997;18:379–386.
28. Bloxam DL, Tyler CF, Young M. Foetal glutamate as a possible precursor of placental glutamine in the guinea pig. *Biochem J*. 1981;198(2):397–401.
29. Hill PM, Young M. Net placental transfer of free amino acids against varying concentrations. *J Physiol*. 1973;235(2):409–422.
30. Ferrell CL, Ford SP. Blood flow steroid secretion and nutrient uptake of the gravid bovine uterus. *J Anim Sci*. 1980;50(6):1113–1121.
31. Reynolds LP, Millaway DS, Kirsch JD, et al. Growth and in-vitro metabolism of placental tissues of cows from day 100 to day 250 of gestation. *J Reprod Fertil*. 1990;89(1):213–222.
32. Smith CH, Depper R. Placental amino acid uptake. II. Tissue preincubation, fluid distribution, and mechanisms of regulation. *Pediatr Res*. 1974;8(7):697–703.
33. Philipps AF, Holzman IR, Teng C, Battaglia FC. Tissue concentrations of free amino acids in term human placentas. *Am J Obstet Gynecol*. 1978;131(8):881–887.
34. Hoeltzli SD, Kelley LK, Moe AJ, Smith CH. Anionic amino acid transport systems in isolated basal plasma membrane of human placenta. *Am J Physiol*. 1990;259(1 Pt 1):C47–C55.
35. Novak DA, Beveridge MJ, Malandro M, Seo J. Ontogeny of amino acid transport system A in rat placenta. *Placenta*. 1996;17(8):643–651.
36. Johnson LW, Smith CH. Neutral amino acid transport systems of microvillous membrane of human placenta. *AmJPhysiol*. 1988;254(6 Pt 1): C773–C780.
37. Karl PI, Teichberg S, Fisher SE. Na(+)-dependent amino acid uptake by human placental microvillous membrane vesicles: importance of storage conditions and preservation of cytoskeletal elements. *Placenta*. 1991;12(3):239–250.
38. Furesz TC, Smith CH, Moe AJ. ASC system activity is altered by development of cell polarity in trophoblast from human placenta. *Am J Physiol*. 1993;265(1 Pt 1):C212–C217.
39. Kudo Y, Boyd CA. Human placental amino acid transporter genes: expression and function. *Reproduction*. 2002;124:593–600.
40. Chairoungdua A, Kanai Y, Matsuo H, et al. Identification and characterization of

a novel member of the heterodimeric amino acid transporter family presumed to be associated with an unknown heavy chain. *J Biol Chem*. 2001;276(52):49390–49399.
41. Fukasawa Y, Segawa H, Kim JY, et al. Identification and characterization of a na(+)-independent neutral amino acid transporter that associates with the 4F2 heavy chain and exhibits substrate selectivity for small neutral D- and l-amino acids. *JBiolChem*. 2000;275(13):9690–9698.
42. Carbo N, Lopez-Soriano FJ, Argiles JM. Neutral amino acid transport in placental plasma membrane vesicles in the late pregnant rat. Evidence for a b0-like transport system. *Eur J Obstet Gynecol Reprod Biol*. 1997;71(1):85–90.
43. Kudo Y, Boyd CA, Sargent IL, Redman CW. Hypoxia alters expression and function of syncytin and its receptor during trophoblast cell fusion of human placental BeWo cells: implications for impaired trophoblast syncytialisation in pre-eclampsia. *Biochim Biophys Acta*. 2003;1638(1):63–71.
44. Torres-Zamorano V, Leibach FH, Ganapathy V. Sodium-dependent homo- and hetero-exchange of neutral amino acids mediated by the amino acid transporter ATB degree. *Biochem Biophys Res Commun*. 1998;245(3):824–829.
45. Karl PI, Fisher SE. Taurine transport by microvillous membrane vesicles and the perfused cotyledon of the human placenta. *Am J Physiol*. 1990;258(3 Pt 1):C443–C451.
46. Norberg S, Powell TL, Jansson T. Intrauterine growth restriction is associated with a reduced activity of placental taurine transporters. *Pediatr Res*. 1998;44(2):233–238.
47. Moyer MS, Insler N, Dumaswala R. The role of chloride in taurine transport across the human placental brush-border membrane. *Biochim Biophys Acta*. 1992;1109(1):74–80.
48. Nishimura T, Sai Y, Fujii J, et al. Roles of taut and system A in cytoprotection of rat syncytiotrophoblast cell line exposed to hypertonic stress. *Placenta*. 2010;31(11):1003–1009.
49. Ramamoorthy S, Leibach FH, Mahesh VB, Ganapathy V. Modulation of the activity of amino acid transport system L by phorbol esters and calmodulin antagonists in a human placental choriocarcinoma cell line. *Biochim Biophys Acta*. 1992;1136(2):181–188.
50. Jansson T, Scholtbach V, Powell TL. Placental transport of leucine and lysine is reduced in intrauterine growth restriction. *Pediatr Res*. 1998;44(4):532–537.
51. Yudilevich DL, Sweiry JH. Transport of amino acids in the placenta. *Biochim Biophys Acta*. 1985;822(2):169–201.
52. Jansson T. Amino acid transporters in the human placenta. *Pediatr Res*. 2001;49(2):141–147.
53. Battaglia FC, Regnault TR. Placental transport and metabolism of amino acids. *Placenta*. 2001;22:145–61.
54. Eleno N, Deves R, Boyd CA. Membrane potential dependence of the kinetics of cationic amino acid transport systems in human placenta. *J Physiol*. 1994;479(Pt 2):291–300.
55. Ayuk PT, Sibley CP, Donnai P, et al. Development and polarization of cationic amino acid transporters and regulators in the human placenta. *Am J Physiol Cell Physiol*. 2000;278(6):C1162–C1171.
56. Speake PF, Glazier JD, Ayuk PT, et al. L-Arginine transport across the basal plasma membrane of the syncytiotrophoblast of the human placenta from normal and preeclamptic pregnancies. *J Clin Endocrinol Metab*. 2003;88(9):4287–4292.
57. Ayuk PT, Theophanous D, D'Souza SW, et al. L-Arginine transport by the

microvillous plasma membrane of the syncytiotrophoblast from human placenta in relation to nitric oxide production: effects of getation, preeclampsia, and intrauterine growth restriction. *J Clin Endocrinol Metab*. 2002;87(2):747–751.
58. Kamath SG, Furesz TC, Way BA, Smith CH. Identification of three cationic amino acid transporters in placental trophoblast: cloning, expression, and characterization of hcat-1. *J Membr Biol*. 1999;171(1):55–62.
59. Malandro MS, Beveridge MJ, Kilberg MS, Novak DA. Effect of low-protein diet-induced intrauterine growth retardation on rat placental amino acid transport. *Am J Physiol*. 1996;271(1 Pt 1):C295–C303.
60. Malandro MS, Beveridge MJ, Novak DA, Kilberg MS. Rat placental amino acid transport after protein-deprivation-induced intrauterine growth retardation. *Biochem Soc Trans*. 1996;24(3):839–843.
61. Furesz TC, Moe AJ, Smith CH. Two cationic amino acid transport systems in human placental basal plasma membranes. *Am J Physiol*. 1991;261(2 Pt 1):C246–C252.
62. Malandro MS, Beveridge MJ, Kilberg MS, Novak DA. Ontogeny of cationic amino acid transport systems in rat placenta. *Am J Physiol*. 1994;267(3 Pt 1):C804–C811.
63. Furesz TC, Smith CH. Identification of two leucine-sensitive lysine transport activities in human placental basal membrane. *Placenta*. 1997;18(8):649–655.
64. Cariappa R, Heath-Monnig E, Smith CH. Isoforms of amino acid transporters in placental syncytiotrophoblast: plasma membrane localization and potential role in maternal/fetal transport. *Placenta*. 2003;24(7):713–726.
65. Novak D, Quiggle F, Artime C, Beveridge M. Regulation of glutamate transport and transport proteins in a placental cell line. *Am J Physiol Cell Physiol*. 2001;281(3):C1014–C1022.
66. Noorlander CW, de Graan PN, Nikkels PG, et al. Distribution of glutamate transporters in the human placenta. *Placenta*. 2004;25(6):489–495.
67. Kilberg MS, Handlogten ME, Christensen HN. Characteristics of an amino acid transport system in rat liver for glutamine, asparagine, histidine, and closely related analogs. *J Biol Chem*. 1980;255(9):4011–4019.
68. Karl PI, Tkaczevski H, Fisher SE. Characteristics of histidine uptake by human placental microvillous membrane vesicles. *Pediatr Res*. 1989;25(1):19–26.
69. Nakanishi T, Sugawara M, Huang W, et al. Structure, function, and tissue expression pattern of human SN2, a subtype of the amino acid transport system N. *Biochem Biophys Res Commun*. 2001;281(5):1343–1348.
70. Kekuda R, Prasad PD, Fei YJ, et al. Cloning of the sodium-dependent, broad-scope, neutral amino acid transporter bo from a human placental choriocarcinoma cell line. *J Biol Chem*. 1996;271(31):18657–18661.
71. Palacin M, Estevez R, Bertran J, Zorzano A. Molecular biology of mammalian plasma membrane amino acid transporters. *Physiol Rev*. 1998;78(4):969–1054.
72. Estevez R, Camps M, Rojas AM, et al. The amino acid transport system y+L/4f2hc is a heteromultimeric complex. *FASEB J*. 1998;12(13):1319–1329.
73. Horak J. Amino acid transport in eucaryotic microorganisms. *Biochim Biophys Acta*. 1986;864:223–256.
74. Martinez P, Ljungdahl PO. The SHR3 homologue from S. Pombe demonstrates a conserved function of ER packaging chaperones. *J Cell Sci*. 2000;113(Pt 23):4351–4362.
75. Kudo Y, Boyd CA. Heterodimeric amino acid transporters: expression of heavy but not light chains of CD98 correlates with induction of amino acid transport systems in human placental trophoblast. *J Physiol*. 2000;523(Pt 1):13–18.

76. Broer S, Wagner CA. Structure-function relationships of heterodimeric amino acid transporters. *Cell Biochem Biophys*. 2002;36:155–168.
77. Okamoto Y, Sakata M, Ogura K, et al. Expression and regulation of 4f2hc and hlat1 in human trophoblasts. *Am J Physiol Cell Physiol*. 2002;282(1):C196–C204.
78. Markovich D, Stange G, Bertran J, et al. Two mrna transcripts (rbat-1 and rbat-2) are involved in system b0,(+)-related amino acid transport. *J Biol Chem*. 1993;268(2):1362–1367.
79. Peter GJ, Davidson IG, Ahmed A, et al. Multiple components of arginine and phenylalanine transport induced in neutral and basic amino acid transporter-crna-injected xenopus oocytes. *Biochem J*. 1996;318(Pt 3):915–922.
80. Rajan DP, Huang W, Kekuda R, et al. Differential influence of the 4F2 heavy chain and the protein related to b(0,+) amino acid transport on substrate affinity of the heteromeric b(0,+) amino acid transporter. *J Biol Chem*. 2000;275(19):14331–14335.
81. Broer A, Wagner CA, Lang F, Broer S. The heterodimeric amino acid transporter 4f2hc/y+LAT2 mediates arginine efflux in exchange with glutamine. *Biochem J*. 2000;349 (Pt 3):787–795.
82. Torrents D, Estevez R, Pineda M, et al. Identification and characterization of a membrane protein (y+L amino acid transporter-1) that associates with 4f2hc to encode the amino acid transport activity y+L. A candidate gene for lysinuric protein intolerance. *J Biol Chem*. 1998;273(49):32437–3245.
83. Novak DA, Matthews JC, Beveridge MJ, et al. Demonstration of system y+L activity on the basal plasma membrane surface of rat placenta and developmentally regulated expression of 4F2HC mrna. *Placenta*. 1997;18(8):643–648.
84. Yao SY, Muzyka WR, Elliott JF, et al. Cloning and functional expression of a cdna from rat jejunal epithelium encoding a protein (4f2hc) with system y+L amino acid transport activity. *Biochem J*.1998;330 (Pt 2):745–752.
85. Mastroberardino L, Spindler B, Pfeiffer R, et al. Amino-acid transport by heterodimers of 4f2hc/CD98 and members of a permease family. *Nature*. 1998;395(6699):288–291.
86. McGivan JD, Plakidou-Dymock S. Regulation of na(+)-dependent amino acid transport in the bovine renal epithelial cell line NBL-1. *Contrib Nephrol*. 1994;110:103–108.
87. Wells RG, Lee WS, Kanai Y, et al. The 4F2 antigen heavy chain induces uptake of neutral and dibasic amino acids in xenopus oocytes. *J Biol Chem*. 1992;267(22):15285–15288.
88. Nakamura E, Sato M, Yang H, et al. 4F2 (CD98) heavy chain is associated covalently with an amino acid transporter and controls intracellular trafficking and membrane topology of 4F2 heterodimer. *J Biol Chem*. 1999;274(5):3009–3016.
89. Fei YJ, Prasad PD, Leibach FH, Ganapathy V. The amino acid transport system y+L induced in xenopus laevis oocytes by human choriocarcinoma cell (JAR) mrna is functionally related to the heavy chain of the 4F2 cell surface antigen. *Biochemistry*. 1995;34(27):8744–8751.
90. Prasad PD, Wang H, Huang W, et al. Human LAT1, a subunit of system L amino acid transporter: molecular cloning and transport function. *Biochem Biophys Res Commun*. 1999;255(2):283–8.
91. Wang H, Huang W, Sugawara M, et al. Cloning and functional expression of ATA1, a subtype of amino acid transporter A, from human placenta. *Biochem Biophys Res Commun*. 2000;273(3):1175–1179.
92. Hatanaka T, Huang W, Wang H, et al. Primary structure, functional characteristics

and tissue expression pattern of human ATA2, a subtype of amino acid transport system A. *Biochim Biophys Acta*. 2000;1467(1):1–6.
93. Sugawara M, Nakanishi T, Fei YJ, et al. Structure and function of ATA3, a new subtype of amino acid transport system A, primarily expressed in the liver and skeletal muscle. *Biochim Biophys Acta*. 2000;1509:7–13.
94. Desforges M, Lacey HA, Glazier JD, et al. The SNAT4 isoform of the system A amino acid transporter is expressed in human placenta. *Am J Physiol Cell Physiol*. 2006;290(1):C305–312.
95. Tung E, Roberts CT, Heinemann GK, et al. Increased placental nutrient transporter expression at midgestation after maternal growth hormone treatment in pigs: a placental mechanism for increased fetal growth. *Biol Reprod*. 2012;87(5):126.
96. Rosario FJ, Kanai Y, Powell TL, Jansson T. Mammalian target of rapamycin signalling modulates amino acid uptake by regulating transporter cell surface abundance in primary human trophoblast cells. *J Physiol*. 2013;591(Pt 3):609–625.
97. Jansson N, Rosario FJ, Gaccioli F, et al. Activation of placental mtor signaling and amino acid transporters in obese women giving birth to large babies. *J Clin Endocrinol Metab*. 2013;98(1):105–113.
98. Nelson DM, Smith SD, Furesz TC, et al. Hypoxia reduces expression and function of system A amino acid transporters in cultured term human trophoblasts. *Am J Physiol Cell Physiol*. 2003;284(2):C310–C315.
99. Berk JL, Hatch CA, Goldstein RH. Hypoxia inhibits amino acid uptake in human lung fibroblasts. *J Appl Physiol*. 2000;89(4):1425–1431.
100. Jones HN, Jansson T, Powell TL. IL-6 stimulates system A amino acid transporter activity in trophoblast cells through STAT3 and increased expression of SNAT2. *Am J Physiol Cell Physiol*. 2009;297(5):C1228–C1235.
101. von Versen-Höynck F, Rajakumar A, Parrott MS, Powers RW. Leptin affects system A amino acid transport activity in the human placenta: evidence for STAT3 dependent mechanisms. *Placenta*. 2009;30(4):361–367.
102. Taylor PM, Kaur S, Mackenzie B, Peter GJ. Amino-acid-dependent modulation of amino acid transport in xenopus laevis oocytes. *J Exp Biol*. 1996;199(Pt 4):923–931.
103. Jones HN, Ashworth CJ, Page KR, Mcardle HJ. Expression and adaptive regulation of amino acid transport system A in a placental cell line under amino acid restriction. *Reproduction*. 2006;131(5):951–960.
104. Coan PM, Vaughan OR, McCarthy J, et al. Dietary composition programmes placental phenotype in mice. *J Physiol*. 2011;589(Pt 14):3659–3670.
105. Lin Y, Zhuo Y, Fang ZF, et al. Effect of maternal dietary energy types on placenta nutrient transporter gene expressions and intrauterine fetal growth in rats. *Nutrition*. 2012;28(10):1037–1043.
106. Lager S, Gaccioli F, Ramirez VI, et al. Oleic acid stimulates system A amino acid transport in primary human trophoblast cells mediated by toll-like receptor 4. *J Lipid Res*. 2013;54(3):725–733.
107. Hayward CE, Greenwood SL, Sibley CP, et al. Effect of maternal age and growth on placental nutrient transport: potential mechanisms for teenagers' predisposition to small-for-gestational-age birth? *Am J Physiol Endocrinol Metab*. 2012;302(2):E233–E242.
108. Audette MC, Challis JR, Jones RL, et al. Antenatal dexamethasone treatment in midgestation reduces system a-mediated transport in the late-gestation murine placenta. *Endocrinology*. 2011;152(9):3561–3570.

109. Audette MC, Greenwood SL, Sibley CP, et al. Dexamethasone stimulates placental system A transport and trophoblast differentiation in term villous explants. *Placenta*. 2010;31(2):97–105.
110. McGivan JD, Pastor-Anglada M. Regulatory and molecular aspects of mammalian amino acid transport. *Biochem J*. 1994;299(Pt 2):321–334.
111. Shafqat S, Tamarappoo BK, Kilberg MS, et al. Cloning and expression of a novel na(+)-dependent neutral amino acid transporter structurally related to mammalian na+/glutamate cotransporters. *J Biol Chem*. 1993;268(21):15351–15355.
112. Utsunomiya-Tate N, Endou H, Kanai Y. Cloning and functional characterization of a system asc-like na+-dependent neutral amino acid transporter. *J Biol Chem*. 1996;271(25):14883–14890.
113. Van Winkle LJ, Christensen HN, Campione, AL. Na+-dependent transport of basic, zwitterionic, and bicyclic amino acids by a broad-scope system in mouse blastocysts. *J Biol Chem*. 1985;260(22):12118–12123.
114. Van Winkle LJ, Tesch JK, Shah A, Campione AL. System B0,+ amino acid transport regulates the penetration stage of blastocyst implantation with possible long-term developmental consequences through adulthood. *Hum Reprod Update*. 2006;12(2):145–157.
115. Chillaron J, Estevez R, Mora C, et al. Obligatory amino acid exchange via systems bo,+-like and y+l-like. A tertiary active transport mechanism for renal reabsorption of cystine and dibasic amino acids. *J Biol Chem*. 1996;271(30):17761–17770.
116. Kulanthaivel P, Cool DR, Ramamoorthy S, et al. Transport of taurine and its regulation by protein kinase C in the JAR human placental choriocarcinoma cell line. *Biochem J*. 1991;277(Pt 1):53–58.
117. Roos S, Powell TL, Jansson T. The human placental taurine transporter in uncomplicated and IUGR pregnancies: cellular localization, protein expression and regulation. *Am J Physiol Regul Integr Comp Physiol*. 2004;287(4):R886–R893.
118. Ramamoorthy S, Leibach FH, Mahesh VB, et al. Functional characterization and chromosomal localization of a cloned taurine transporter from human placenta. *Biochem J*. 1994;300(Pt 3):893–900.
119. Babu E, Kanai Y, Chairoungdua A, et al. Identification of a novel system L amino acid transporter structurally distinct from heterodimeric amino acid transporters. *J Biol Chem*. 2003;278(44):43838–43845.
120. Rajan DP, Kekuda R, Huang W, et al. Cloning and expression of a b(0,+)-like amino acid transporter functioning as a heterodimer with 4f2hc instead of rbat. A new candidate gene for cystinuria. *J Biol Chem*. 1999;274(41):29005–29010.
121. Kudo Y, Boyd CA. Characterisation of l-tryptophan transporters in human placenta: a comparison of brush border and basal membrane vesicles. *J Physiol*. 2001;531(Pt 2):405–416.
122. Pineda M, Fernandez E, Torrents D, et al. Identification of a membrane protein, LAT-2, that co-expresses with 4F2 heavy chain, an l-type amino acid transport activity with broad specificity for small and large zwitterionic amino acids. *J Biol Chem*. 1999;274(28):19738–19744.
123. Deves R, Boyd CA. Surface antigen CD98(4F2): not a single membrane protein, but a family of proteins with multiple functions. *J Membr Biol*. 2000;173(3):165–177.
124. Broer S. Adaptation of plasma membrane amino acid transport mechanisms to physiological demands. *Pflugers Arch*. 2002;444(4):457–466.
125. Cleal JK, Brownbill P, Godfrey KM, et al. Modification of fetal plasma amino

acid composition by placental amino acid exchangers in vitro. *J Physiol*. 2007;582(Pt 2):871–882.
126. Bodoy S, Martin L, Zorzano A, et al. Identification of LAT4, a novel amino acid transporter with system L activity. *J Biol Chem*. 2005;280(12):12002–12011.
127. Radaelli T, Cetin I, Ayuk PT, et al. Cationic amino acid transporter activity in the syncytiotrophoblast microvillous plasma membrane and oxygenation of the uteroplacental unit. *Placenta*. 2002;23(suppl A):S69–S74.
128. Verrey F, Closs EI, Wagner CA, et al. CATs and hats: the SLC7 family of amino acid transporters. *Pflugers Arch*. 2004;447(5):532–542.
129. Closs EI, Graf P, Habermeier A, et al. Human cationic amino acid transporters hcat-1, hcat-2a, and hcat-2b: three related carriers with distinct transport properties. *Biochemistry*. 1997;36(21):6462–6468.
130. Matthews JC, Beveridge MJ, Malandro MS, et al. Activity and protein localization of multiple glutamate transporters in gestation day 14 vs. Day 20 rat placenta. *Am J Physiol*. 1998;274(3 Pt 1):C603–C614.
131. Nakayama T, Kawakami H, Tanaka K, Nakamura S. Expression of three glutamate transporter subtype mrnas in human brain regions and peripheral tissues. *Brain Res Mol Brain Res*. 1996;36(1):189–192.
132. Matthews JC, Beveridge MJ, Dialynas E, et al. Placental anionic and cationic amino acid transporter expression in growth hormone overexpressing and null IGF-II or null IGF-I receptor mice. *Placenta*. 1999;20(8):639–650.
133. Baur R. Morphometry of the placental exchange area. *Adv Anat Embryol Cell Biol*. 1977;53(1):3–65.
134. Teasdale F. Gestational changes in the functional structure of the human placenta in relation to fetal growth: a morphometric study. *Am J Obstet Gynecol* 1980;137(5):560–568.
135. Mayhew TM, Manwani R, Ohadike C, et al. The placenta in pre-eclampsia and intrauterine growth restriction: studies on exchange surface areas, diffusion distances and villous membrane diffusive conductances. *Placenta*. 2007;28:233–238.
136. Teasdale F, Jean-Jacques G. Morphometric evaluation of the microvillous surface enlargement factor in the human placenta from mid-gestation to term. *Placenta*. 1985;6(5):375–381.
137. Woods DL, Rip MR. Placental villous surface area of light-for-dates infants at term. *Early Hum Dev*. 1987;15(2):113–117.
138. Sparks JW, Girard JR, Battaglia FC. An estimate of the caloric requirements of the human fetus. *Biol Neonate*. 1980;38(3–4):113–119.
139. Hoeltzli SD, Smith CH. Alanine transport systems in isolated basal plasma membrane of human placenta. *Am J Physiol*.1989;256(3 Pt 1): C630–C637.
140. Mahendran D, Donnai P, Glazier JD, et al. Amino acid (system A) transporter activity in microvillous membrane vesicles from the placentas of appropriate and small for gestational age babies. *Pediatr Res*. 1993;34(5):661–665.
141. Gustke HH, Kowalewski S. Glycolytic enzymes in the normal human term placenta. *Enzyme*. 1975;19(3):154–164.
142. Diamant YZ, Mayorek N, Neumann S, Shafrir E. Enzymes of glucose and fatty acid metabolism in early and term human placenta. *Am J Obstet Gynecol*. 1975;121(1):58–61.
143. Robertson AF, Karp WB. Placental transport of nutrients. *South Med J*. 1976;69(10):1358–1362.
144. Tran N, Laplante M, Brady N, Lebel E. Oxidation of dl-3-phenylalanine-1- 14 C, dl-leucine-1- 14 C, and d-glucose-1- 14 c-6-phosphate to 14 CO 2 in human

placenta. *J Nucl Med*. 1972;13(1):41–44.
145. Moores RR, Vaughn PR, Battaglia FC, et al. Glutamate metabolism in fetus and placenta of late-gestation sheep. *Am J Physiol*. 1994;267(1 Pt 2): R89–R96.
146. Meschia G, Battaglia FC, Hay WW, Sparks JW. Utilization of substrates by the ovine placenta in vivo. *Fed Proc*. 1980;39(2):245–249.
147. Jozwik M, Teng C, Meschia G, Battaglia FC. Contribution of branched-chain amino acids to uteroplacental ammonia production in sheep. *Biol Reprod*. 1999;61(3):792–796.
148. Jozwik M, Teng C, Wilkening RB, et al. Effects of branched-chain amino acids on placental amino acid transfer and insulin and glucagon release in the ovine fetus. *Am J Obstet Gynecol*. 2001;185(2):487–495.
149. Chung M, Teng C, Timmerman M, et al. Production and utilization of amino acids by ovine placenta in vivo. *Am J Physiol*. 1998;274(1 Pt 1):E13–22.
150. Pell JM, Jeacock MK, Shepherd MK. Glutamate and glutamine metabolism in the ovine placenta. *J Agric Sci*.1982;101:275–281.
151. Johnson RL, Gilbert M, Block SM, Battaglia FC. Uterine metabolism of the pregnant rabbit under chronic steady-state conditions. *Am J Obstet Gynecol*. 1986;154(5):1146–1151.
152. Carstensen MH, Leichtweiss HP, Schröder H. The metabolism of the isolated artificially perfused guinea pig placenta. I. Excretion of hydrogen ions, ammonia, carbon dioxide and lactate, and the consumption of oxygen and glucose. *J Perinat Med*. 1982;10(3):147–153.
153. Ronzoni S, Marconi AM, Cetin I, et al. Umbilical amino acid uptake at increasing maternal amino acid concentrations: effect of a maternal amino acid infusate. *Am J Obstet Gynecol*. 1999;181(2):477–483.
154. Ronzoni S, Marconi AM, Paolini CL, et al. The effect of a maternal infusion of amino acids on umbilical uptake in pregnancies complicated by intrauterine growth restriction. *Am J Obstet Gynecol*. 2002;187(3):741–746.
155. Lemons JA, Adcock EW, Jones MDJ, et al. Umbilical uptake of amino acids in the unstressed fetal lamb. *J Clin Invest*. 1976;58(6):1428–1434.
156. Bell AW, Kennaugh JM, Battaglia FC, Meschia G. Uptake of amino acids and ammonia at mid-gestation by the fetal lamb. *Q J Exp Physiol*. 1989;74(5):635–643.
157. Stegink LD, Pitkin RM, Reynolds WA, et al. Placental transfer of glutamate and its metabolites in the primate. *Am J Obstet Gynecol*. 1975;122(1):70–78.
158. Schneider H, Möhlen KH, Dancis J. Transfer of amino acids across the in vitro perfused human placenta. *Pediatr Res*. 1979;13(4 Pt 1):236–240.
159. Regnault TR, de Vrijer B, Galan HL, et al. Umbilical uptakes and transplacental concentration ratios of amino acids in severe fetal growth restriction. *Pediatr Res*. 2013;73(5):602–611.
160. Loy GL, Quick AN, Battaglia FC, et al. Measurement of leucine and alpha-ketoisocaproic acid fluxes in the fetal/placental unit. *J Chromatogr*. 1991;562:169–174.
161. Ross JC, Fennessey PV, Wilkening RB, et al. Placental transport and fetal utilization of leucine in a model of fetal growth retardation. *Am J Physiol*. 1996;270(3 Pt 1):E491–E503.
162. Anderson AH, Fennessey PV, Meschia G, et al. Placental transport of threonine and its utilization in the normal and growth-restricted fetus. *Am J Physiol*. 1997;272(5 Pt 1):E892–E900.
163. Timmerman M, Chung M, Wilkening RB, et al. Relationship of fetal alanine uptake and placental alanine metabolism to maternal plasma alanine

concentration. *Am J Physiol*. 1998;275(6 Pt 1):E942–E950.
164. Moores RR, Rietberg CC, Battaglia FC, et al. Metabolism and transport of maternal serine by the ovine placenta: glycine production and absence of serine transport into the fetus. *Pediatr Res*. 1993;33(6):590–594.
165. Jóźwik M, Jóźwik M, Pietrzycki B, et al. Maternal and fetal blood ammonia concentrations in normal term human pregnancies. *Biol Neonate*. 2005;87(1):38–43.
166. Gresham EL, Simons PS, Battaglia FC. Maternal-fetal urea concentration difference in man: metabolic significance. *J Pediatr*. 1971;79(5):809–811.
167. Bellotti M, Pennati G, De Gasperi C, et al. Role of ductus venosus in distribution of umbilical blood flow in human fetuses during second half of pregnancy. *Am J Physiol Heart Circ Physiol*. 2000;279(3):H1256–H1263.
168. Baschat AA. Ductus venosus Doppler for fetal surveillance in high-risk pregnancies. *Clin Obstet Gynecol*. 2010;53(4):858–868.
169. Ferrazzi E, Rigano S, Bozzo M, et al. Umbilical vein blood flow in growth-restricted fetuses. *Ultrasound Obstet Gynecol*. 2000;16(5):432–438.
170. Timmerman M, Teng C, Wilkening RB, et al. Net amino acid flux across the fetal liver and placenta during spontaneous ovine parturition. *Biol Neonate*. 2001;79(1):54–60.
171. Teng C, Battaglia FC, Meschia G, et al. Fetal hepatic and umbilical uptakes of glucogenic substrates during a glucagon-somatostatin infusion. *Am J Physiol Endocrinol Metab*. 2002;282(3):E542–E550.
172. Wilkening RB, Boyle DW, Meschia G. Fetal ph improvement after 24 h of severe, nonlethal hypoxia. *Biol Neonate*. 1993;63(2):129–132.
173. Timmerman M, Teng C, Wilkening RB, et al. Effect of dexamethasone on fetal hepatic glutamine-glutamate exchange. *Am J Physiol Endocrinol Metab*. 2000;278(5):E839–E845.
174. Cetin I, Fennessey PV, Quick AN, et al. Glycine turnover and oxidation and hepatic serine synthesis from glycine in fetal lambs. *Am J Physiol*. 1991;260(3 Pt 1):E371–E378.
175. Thureen PJ, Narkewicz MR, Battaglia FC, et al. Pathways of serine and glycine metabolism in primary culture of ovine fetal hepatocytes. *Pediatr Res*. 1995;38(5):775–782.
176. Narkewicz MR, Thureen PJ, Sauls SD, et al. Serine and glycine metabolism in hepatocytes from mid gestation fetal lambs. *Pediatr Res*. 1996;39(6):1085–1090.
177. Kalhan SC, Gruca LL, Parimi PS, et al. Serine metabolism in human pregnancy. *Am J Physiol Endocrinol Metab*. 2003;284(4):E733–E740.
178. Moores RR, Carter BS, Meschia G, et al. Placental and fetal serine fluxes at midgestation in the fetal lamb. *Am J Physiol*. 1994;267(1 Pt 1): E150–E155.
179. Narkewicz MR, Moores RR,, Battaglia FC, Frerman FF. Ontogeny of serine hydroxymethyltransferase isoenzymes in fetal sheep liver, kidney, and placenta. *Mol Genet Metab*. 1999;68(4):473–80.
180. Cetin I, Fennessey PV, Sparks JW, et al. Fetal serine fluxes across fetal liver, hindlimb, and placenta in late gestation. *Am J Physiol*. 1992;263(4 Pt 1): E786–E793.
181. Jois M, Ewart HS, Brosnan JT. Regulation of glycine catabolism in rat liver mitochondria. *Biochem J*. 1992;283(Pt 2):435–439.
182. Ewart HS, Jois M, Brosnan JT. Rapid stimulation of the hepatic glycine-cleavage system in rats fed on a single high-protein meal. *Biochem J*. 1992;283 (Pt 2):441–447.
183. Boyle DW, Denne SC, Moorehead H, et al. Effect of rhigf-i infusion on whole

fetal and fetal skeletal muscle protein metabolism in sheep. *Am J Physiol*. 1998;275(6 Pt 1):E1082–E1091.
184. Lemons JA, Liechty EA. Nitrogen flux across ovine maternal and fetal hindquarters during fasting. *J Dev Physiol*. 19879(2):151–158.
185. Wilkening RB, Boyle DW, Teng C, et al. Amino acid uptake by the fetal ovine hindlimb under normal and euglycemic hyperinsulinemic states. *Am J Physiol*. 1994;266(1 Pt 1):E72–E78.
186. Liechty EA, Polak MJ, Lemons JA. Branched-chain amino acid carbon and nitrogen arteriovenous concentration differences across the ovine fetal hindlimb. *Pediatr Res*. 1987;21(1):44–48.
187. Cetin I, Marconi AM, Bozzetti P, et al. Umbilical amino acid concentrations in appropriate and small for gestational age infants: a biochemical difference present in utero. *Am J Obstet Gynecol* 1988;158(1):120–126.
188. Cetin I, Corbetta C, Sereni LP, et al. Umbilical amino acid concentrations in normal and growth- retarded fetuses sampled in utero by cordocentesis. *Am J Obstet Gynecol*. 1990;162(1):253–261.
189. Economides DL, Nicolaides KH, Gahl WA, et al. Plasma amino acids in appropriate- and small-for-gestational-age fetuses. *Am J Obstet Gynecol*. 1989;161(5):1219–1227.
190. Pardi G, Cetin I, Marconi AM, et al. Diagnostic value of blood sampling in fetuses with growth retardation. *N Engl J Med*. 1993;328(10):692–696.
191. Shibata E, Hubel CA, Powers RW, et al. Placental system A amino acid transport is reduced in pregnancies with small for gestational age (SGA) infants but not in preeclampsia with SGA infants. *Placenta*. 2008;29(10):879–882.
192. Glazier JD, Cetin I, Perugino G, et al. Association between the activity of the system A amino acid transporter in the microvillous plasma membrane of the human placenta and severity of fetal compromise in intrauterine growth restriction. *Pediatr Res*. 1997;42(4):514–519.
193. Dicke JM, Henderson GI. Placental amino acid uptake in normal and complicated pregnancies. *Am J Med Sci*. 1988;295(3):223–227.
194. Sibley CP, Coan PM, Ferguson-Smith AC, et al. Placental-specific insulin-like growth factor 2 (igf2) regulates the diffusional exchange characteristics of the mouse placenta. *Proc Natl Acad Sci U S A*. 2004;101(21):8204–8208.
195. Constancia M, Angiolini E, Sandovici I, et al. Adaptation of nutrient supply to fetal demand in the mouse involves interaction between the igf2 gene and placental transporter systems. *Proc Natl Acad Sci U S A*. 2005;102(52):19219–192124.
196. Constancia M, Hemberger M, Hughes J, et al. Placental-specific IGF-II is a major modulator of placental and fetal growth. *Nature*. 2002;417(6892):945–948.
197. Malandro MS, Kilberg MS. Molecular biology of mammalian amino acid transporters. *Annu Rev Biochem*. 1996;65:305–336.
198. Marconi AM, Paolini CL, Stramare L, et al. Steady state maternal-fetal leucine enrichments in normal and intrauterine growth-restricted pregnancies. *Pediatr Res*. 1999;46(1):114–119.
199. Teasdale F, Jean-Jacques G. Intrauterine growth retardation: morphometry of the microvillous membrane of the human placenta. *Placenta*. 1988;9(1):47–55.
200. Jones HN, Woollett LA, Barbour N, et al. High-fat diet before and during pregnancy causes marked up-regulation of placental nutrient transport and fetal overgrowth in C57/BL6 mice. *FASEB J*. 2009;23(1):271–278.
201. Jones HN, Jansson T, Powell TL. Full-length adiponectin attenuates insulin signaling and inhibits insulin-stimulated amino acid transport in human primary

trophoblast cells. *Diabetes*. 2010;59(5):1161–1170.
202. van Lingen RA, van Goudoever JB, Luijendijk IH, et al. Effects of early amino acid administration during total parenteral nutrition on protein metabolism in preterm infants. *Clin Sci (Lond)*. 1992;82(2):199–203.
203. Poindexter BB, Langer JC, Dusick AM, Ehrenkranz RA, National Institute of Child Health and Human Development Neonatal Research Network. Early provision of parenteral amino acids in extremely low birth weight infants: relation to growth and neurodevelopmental outcome. *J Pediatr*. 2006;148(3):300–305.
204. Thureen PJ. The neonatologist's dilemma: catch-up growth or beneficial undernutrition in very low birth weight infants-what are optimal growth rates? *J Pediatr Gastroenterol Nutr*. 2007;45(suppl 3):S152–S154.
205. Hay WW, Thureen P. Protein for preterm infants: how much is needed? How much is enough? How much is too much? *Pediatr Neonatol*. 2010;51(4):198–207.
206. American academy of pediatrics committee on nutrition: nutritional needs of low-birth-weight infants. *Pediatrics*. 1985;75(5):976–986.
207. Zlotkin SH, Bryan MH, Anderson GH. Intravenous nitrogen and energy intakes required to duplicate in utero nitrogen accretion in prematurely born human infants. *J Pediatr*. 1981;99(1):115–120.
208. Parry-Billings M, Evans J, Calder PC, Newsholme EA. Does glutamine contribute to immunosuppression after major burns? *Lancet*. 1990;336(8714):523–525.
209. Newsholme EA, Crabtree B, Ardawi MS. Glutamine metabolism in lymphocytes: its biochemical, physiological and clinical importance. *Q J Exp Physiol*. 1985;70(4):473–489.
210. Souba WW, Klimberg VS, Hautamaki RD, et al. Oral glutamine reduces bacterial translocation following abdominal radiation. *J Surg Res*. 1990;48(1):1–5.
211. Reichman B, Chessex P, Putet G, et al. Diet, fat accretion, and growth in premature infants. *N Engl J Med*. 1981;305(25):1495–1500.
212. Sydsjö G. Long-term consequences of non-optimal birth characteristics. *Am J Reprod Immunol*. 2011;66(suppl 1):81–87.
213. Barker DJ, Gluckman PD, Godfrey KM, et al. Fetal nutrition and cardiovascular disease in adult life. *Lancet*. 1993;341(8850):938–941.
214. Jozwik M, Teng C, Wilkening RB, et al. Reciprocal inhibition of umbilical uptake within groups of amino acids. *Am J Physiol Endocrinol Metab*. 2004;286(3):E376–E383.
215. Barbera A, Galan HL, Ferrazzi E, et al. Relationship of umbilical vein blood flow to growth parameters in the human fetus. *AmJObstetGynecol*. 1999;181(1):174–179.

Human Milk: Nutritional Properties

29

Marcia H. Monaco, PhD Sharon M. Donovan, PhD, RD

HEALTH BENEFITS OF HUMAN MILK FOR THE INFANT

Human milk provides nutrients with high bioavailability and in sufficient qualities to support the needs of the rapidly growing human infant, and is defined as the "normative standard of infant feeding" by the American Academy of Pediatrics (AAP).[1] As such, the recommended dietary intakes for infants in the first six months of life are based on the nutrient analysis of human milk and average volumes of milk consumption by healthy, full-term, exclusively breast-fed infants.[2] The specific nutrient requirements of the term infant are discussed further in Chapter 36. Human milk also is recommended for the preterm, low-birth-weight (LBW) infant due to the associations between human milk feeding and reduced incidence of necrotizing enterocolitis[3] and improved IQ scores.[4] However, the nutrient content of either term or preterm human milk is insufficient to meet the needs of infants less than 1500 g and should be supplemented with a human milk fortifier for optimal nutrition.[1,5] The specific nutrient requirements of the LBW infant are presented in Chapters 32 and 33.

In addition, human milk contains bioactive factors whose functions extend beyond providing essential nutrients and energy and appear to facilitate the optimal transition of the infant from intra- to extrauterine life.[6] Components in human milk actively protect the infant from pathogenic infection and facilitate the establishment of the microbiota, the latter of which is required to activate the mucosal immune system.[7]

Indeed, clinical and epidemiological data provide strong evidence that human milk feeding decreases the incidence and/or severity of diarrhea, respiratory and urinary tract infections, otitis media, and necrotizing enterocolitis.[1] This protection likely is mediated through the immuno protective proteins, lipids, and carbohydrates (oligosaccharides) in human milk that exert antibacterial and antiviral activities[7] (see Chapter 30 for additional discussion of protective properties of human milk).

In addition, there is evidence that breastfeeding confers longer term risk reduction for autoimmune diseases. Exposure to beneficial components in human milk that promote the development of the immune system and/or a lack of exposure to stimuli that would trigger an inappropriate immunologic response in genetically predisposed individuals have been proposed as mechanisms underlying the reduced risk of developing autoimmune diseases,[8,9] although the relative contributions of these two effects are unknown. Breastfeeding is associated with lower risk of celiac disease[10] when the introduction of gluten occurs concomitantly with breastfeeding and insulin-dependent diabetes mellitus.[9] Evidence for prevention of atopic eczema,[11] asthma,[12] wheezing,[13] and inflammatory bowel diseases[8] is less conclusive and further rigorous research is needed to elucidate the interaction between the infant's genetic predisposition and breastfeeding in modulating the risk of these diseases. As dietary antigens can cross the placenta, as well as pass into human milk, pediatricians have recommended maternal antigen avoidance during pregnancy and/or lactation as a strategy to prevent or treat atopic diseases in children at high risk for developing atopic diseases. A systematic review of the randomized or quasi- randomized trials found that antigen avoidance during pregnancy or lactation did not substantially reduce the risk of the child developing atopy and may adversely affect maternal and fetal/infant nutritional status and, thus, is not recommended.[14] The benefits of antigen avoidance diets during lactation on the development and severity of atopic eczema are equivocal at best, and trials with large sample sizes are needed in this area.

Strong evidence indicates the association of breastfeeding with higher cognitive development compared to formula feeding.[4,15] Breastfeeding for more than four months was associated with a 4.3 points augmentation in the Bayley mental developmental index of 2-year-old, even after adjusting for confounding factors (socioeconomic, parental IQ, etc.).[15] The greatest benefit was gained by preterm versus term infants, particularly those who received human milk for at least six months.[16] In one study, duration of breastfeeding, the infant's gestational week at birth, and maternal milk

docosahexaenoic acid to arachidonic acid ratio (DHA-to-AA) yielded a three-factor model that explained 76% of the variation in total IQ ($P < 0.001$).[17]

Recent systematic meta-analysis and reviews support an inverse relationship between breastfeeding and the risk of pediatric overweight, though the extent of this relationship tend to decrease when adjustments for confounding factors, such as maternal education, maternal body mass index (BMI), socioeconomic status, and maternal smoking, among others, are taken into consideration.[18] Breastfeeding for greater than four months is associated with an approximately 20%–50% reduction in the risk of pediatric overweight[19]–[21] and each month of prolonged exclusive breastfeeding was associated with a weight gain reduction of approximately 100 g after adjustments for infant and maternal factors.[22] Moreover, breastfeeding for greater than 25 days was associated with a 14% decrease in fat mass measured by dual-energy X-ray absorptiometry (DXA) in 16-year-old adolescents.[23] An additive interaction was observed between maternal pre-pregnancy BMI and breastfeeding, in that 2- to 14-year-old children of obese mothers (BMI ≥ 30) who were never breast-fed had a 6.1-fold greater risk (95% confidence interval 2.8, 13.1) of being overweight (BMI ≥ 95th percentile) than children of normal-weight mothers who were breast-fed.[19] Even in women who were overweight (BMI ≥ 25) or had diabetes, breastfeeding was protected against the development of overweight in their 9- to 14-year-olds.[20] Taken together, these latter two studies provide compelling justification to promote breastfeeding in all mothers, particularly those with high BMI. However, several recent studies suggest that overweight women experienced delayed initiation of lactation and may require additional measures of lactation support.[24] Additional information on breastfeeding can be found in Chapter 31. Reducing the prevalence of childhood overweight by breastfeeding may exert long-term protection against the development of type-2 diabetes. A recent study analyzing data on over 10,000 subjects from five prospective studies conducted between 1969 and 1990 in low- and middle-income countries failed to find any association between duration of breastfeeding and development of diabetes adult in adulthood.[25] It is clear that more studies are needed to define the impact of breastfeeding on the onset of adult type 2 diabetes.

POLICY RECOMMENDATIONS AND

BREASTFEEDING TRENDS

Breastfeeding is the preferred form of nutrition for the human infant and is recommended by national and international health organizations and pediatric societies. AAP recommend exclusive breastfeeding for the first six months of life,[1] but strongly supports the continuation of breastfeeding for at least the first year of life and beyond, as long as mutually desired by the mother and child, with the addition of complementary foods at 6 months of age.[1] The timing and composition of complementary feeding are discussed further in Chapter 35.

Breastfeeding rates have risen slowly but steadily over the past decade and, in 2009, approximately 76% of infants received some human milk in the early postpartum period.[26] The breastfeeding rates decrease to 47% and 25% at 6 and 12 months postpartum, respectively. Breastfeeding initiation rates (approximately 76.9%)[26] are near the Surgeon General's Healthy People 2020 goal of 81.9%[27] and it was estimated that if breastfeeding were to continue to increase at the current rate (approximately 2%/y), inhospital breastfeeding in the United States should meet or exceed the Healthy People 2020 goal. However, breastfeeding rates at 6 and 12 months postpartum are still lower than the Healthy People 2020 goals of 60% and 34%, respectively,[27] and these goals will likely not be met in all subgroups in the population if current trends persist. Maternal age, education, race/ethnicity, income, geographic location, and participation in the Special Supplemental Nutrition Program for Women, Infants, and Children (WIC) affected breastfeeding rates, with rates being highest for white non-Hispanic, older, college-educated mothers.[28] Using data collected in the 2007 National Immunization Survey, breastfeeding initiation among WIC participants was nearly 17% lower than nonWIC participants, and breastfeeding duration differed significantly between these two groups (6.7 vs. 8.2 months, respectively).[29] Thus, for Healthy People 2020 goals to be achieved, effective strategies must be developed to promote breastfeeding in young, less educated mothers and those participating in WIC.[30] In 2011, the Surgeon General issued a Call to Action to support breastfeeding that described specific steps people can take to participate in a society-wide approach to support breastfeeding mothers and babies. This approach will increase the public health impact of everyone's efforts, reduce inequities in the quality of health care that mothers and babies receive, and improve the support that families receive in employment and community settings.[30]

ASSESSING THE GROWTH OF BREAST-FED INFANTS

It has long been recognized that the patterns of linear growth, weight gain, and body composition during the first year of life differ between breast-fed and formula-fed infants[31] and even between infants who are breast-fed for short duration versus longer term exclusive breastfeeding.[32] A recently published systematic review (15 studies) and meta-analysis (11 studies) examined the pattern of changes in body composition of healthy breast-fed and formula-fed infants.[31] Fat-free mass, fat mass, and % fat mass were determined by a variety of methods (i.e., total body potassium determination, total body electrical conductivity, DXA, MRI, air displacement plethysmography, and isotope dilution). Formula-fed infants had higher fat-free mass at 3–4 months (mean difference + 130 g), 8–9 months (+290 g) and 12 months (+300 g), where as fat mass was lower at 3–4 months (−90 g) and 6 months (−180 g) than breast-fed infants. Conversely, at 12 months, formula-fed had higher fat mass (+290 g) than breast-fed. This particular review did not include infants older than 12 months, however, others have reported that any difference in growth trajectory between formula-fed and breast-fed infants detected in the first year of life disappeared after 24 months postpartum.[33] Importantly, head circumference does not differ between breast-fed and formula-fed infants.[34]

Assessment of growth in children is important for monitoring health status. There are several growth charts available for assessing the adequacy of children's growth, but it is recommended that the WHO child growth charts, which were generated from anthropometric data collected from approximately 8500 children in Brazil, Ghana, India, Norway, Oman, and the United States through the WHO Multicenter Growth Reference Study be used for children under 2 years of age.[35] The growth standards are available on the Internet (http://www.who.int/childgrowth/standards/en). Importantly, the WHO standards demonstrated that breast-fed infants worldwide, who are raised in healthy environments, exhibit similar patterns of growth.[35] More recently, WHO released growth velocity standards for weight, length, and head circumference. The standards cover from birth to 2 years of age, and are based on another multi country study involving breast-fed infants and young children from six separate geographical regions.[36] Growth velocity curves are considered a superior quantitative measure of growth compared to attained size (weight or length) for age. Furthermore, monitoring growth

velocity supports early detection of abnormal growth patterns and allows for timely clinical intervention.[36]

In summary, the WHO international growth standards are based on exclusively breast-fed infants and represent the growth patterns of infants following current AAP and WHO recommendations.[1] Whichever growth charts a practitioner chooses to use, it should be kept in mind that growth charts are tools that contribute to forming an overall clinical impression of the child and should not be used as a sole diagnostic instrument.

PHYSIOLOGY OF LACTATION AND MILK PRODUCTION

Initiation of Lactation

The lactation cycle consists of four phases: mammogenesis, lactogenesis, galactopoiesis, and involution. Mammogenesis is a long-term process that is hormonally stimulated during adolescence, with further development during pregnancy and terminal differentiation during lactation.[37] Lactogenesis is associated with the expression of lactation-specific genes in mammary secretory epithelial cells and is divided into two stages.[37] Lactogenesis I occurs during mid-pregnancy and represents the initiation of the unique synthesis capacity of the mammary gland. Lactogenesis II occurs just before or at the time of parturition and is associated with a drop in circulating progesterone and estrogen concentrations, sustained prolactin secretion, the initiation of copious milk secretion, and marked changes in milk composition.[37] Colostrum, the first milk produced following delivery, is characterized by high concentrations of total protein, immunoglobulin, sodium and chloride, and low concentrations of lactose, potassium, glucose, and citrate.[37] The increase in lactose and decrease in sodium and chloride concentrations signal closure of paracellular pathways at day 1–2 postpartum.[37] From days 2–4 postpartum, increased milk citrate, free phosphate, glucose and calcium concentrations, and decreased milk pH are observed. Changes in one or more of these milk constituents are used to identify the onset of lactogenesis II, which typically occurs between 30 and 40 hours postpartum in term mothers. Premature delivery,[38] cesarean delivery,[39] and insulin- dependent diabetes mellitus[40] have been reported to suppress or delay lactogenesis II and detrimentally impact the successful establishment of lactation. Women at risk for delayed initiation of lactation often are advised to use breast pumps to

stimulate milk production. For example, in a recent study, mothers of very low-birth-weight infants born prematurely (<32 weeks of gestation) were randomized to initiate milk expression either within 60 minutes of birth or one to six hours following delivery. Women who started milk expression within one hour of birth had increased milk volume and experienced decreased time to onset of lactogenesis stage II.[41] This study suggests the timing for milk expression may be critical for milk production and prolonged lactation. Chapman and colleagues[42] conducted a randomized clinical trial to assess the effectiveness of pumping on early milk transfer and duration of breastfeeding among primiparous women after cesarean delivery at term. Unexpectedly, breast pumping between 24 and 72 hours after delivery did not improve milk transfer in the first 72 hours postpartum and appeared to negatively affect overall breastfeeding duration.

Even among highly educated, motivated women who delivered at term, nearly 50% of mothers experienced suboptimal breastfeeding behaviors (SIBB) on the day of delivery, which declined to 22% and 14% by day 3 and 7 postpartum, respectively.[43] Primiparity, cesarean delivery, prolonged stage II labor (>1 hour), birth medications, and infant status at birth (low APGAR score) were associated with SIBB, suggesting that birth-related events can detrimentally impact the initiation of lactation.[42] Flat or inverted nipples, the use of nonhuman milk fluids within the first 48 hours postpartum and pacifier use were also risk factors for SIBB.[42] Thus, lactation success can be improved by proactively addressing modifiable factors during delivery and the early postpartum period and all breastfeeding mothers should be evaluated for SIBB at 72–96 hours postpartum.[43] Dewey and colleagues[43] also identified high maternal BMI (>27) as a risk factor for SIBB. Subsequent studies confirmed that pre-pregnancy BMI was negatively associated with the onset of lactogenesis II[24] and overweight or obese mothers breast-fed their infants for a shorter duration than normal-weight women.[24,44] There may be a physiological basis for the lower lactation success in that overweight and obese women had a reduced prolactin response to suckling in the first week postpartum.[44]

Milk Production

The volume of human milk produced can be assessed by “test weighing” a breastfeeding infant before and after each feeding for a 12 or 24 hours period, or by the deuterium oxide dilution method. A systematic review of

22 studies found that mean human milk intake of exclusively breast-fed infants increased approximately 20–30 g/mo from 699 g/d at 1 month, to 731, 751, 780, 796, and 854 g/d at months 2–6, respectively.[33] The greatest changes in milk volume occur in the first week after birth. A recent meta-analysis on 25 published papers indicated changes in milk consumption from 22 ± 4.2 ml/d on day 1 postpartum to 100 ± 22, 285 ± 40, 384 ± 17, 457 ± 21, and 495 ± 33 mL/d on days 2–7. By day 14, milk intake was approximately 1.4-fold higher than day 7.[45] A cross-sectional study by Kent and colleagues[46] monitored 775 breastfeeding episodes of infants aged 1–6 months and assessed milk production by test weighing and the extent of breast emptying by milk fat content. In that study, infants nursed on average 11 ± 3 times/d (range: 6–18), with an inter-feeding interval of 138 ± 43 minutes (range 4–658 minutes) and 64% of infants breast-fed 1–3 times per night.[46] There were no differences in breastfeeding frequency with age or between girls and boys; however, the volume per feeding increased with age and was greater in male than female infants.[46] Infants consumed 76.0 ± 12.6 g milk (range 0–240 g) per feeding and overall 24-hour milk production was 788 ± 169 g/d (range 440–1365 g/d).[46] Interestingly, they found that milk production differed between breasts, with the right breast producing on average nearly 90 g/d more than the left breast (443 vs. 356 g/d).[46] Interestingly, for 53% of the breastfeeding episodes, the contents of one breast were sufficient to satisfy the infant for at least one hour and, on average, only 67.3 ± 7.8% (range 0–100%) of the available milk in the breast was removed at each feeding. These data suggested that both breasts do not need to be offered at each feeding episode, nor does a breast need to be fully drained at every feeding to maintain adequate milk production.[46] The authors concluded that breastfeeding mothers should be encouraged to feed their infants on demand rather than conform to average recommendations that may not be appropriate for that "mother–infant dyad."[46]

DYNAMIC NATURE OF HUMAN MILK COMPOSITION

The nutrient composition of human milk is dynamic and changes in response to mammary gland physiology (lactogenesis, involution) and maternal diet.[47] Milk composition also differs between mothers who deliver prematurely versus at term, which, in part, may reflect the degree of mammary gland maturation at delivery.[48] How stage of lactation and

maternal diet impact the content and composition of individual classes of nutrients will be discussed in depth in subsequent sections of this chapter.

In general, the composition of human milk varies throughout the course of lactation.[47,48] Colostrum, the first secretions produced by the mammary gland, persists from birth to five days postpartum. Colostrum is a low-fat, high-protein fluid that is rich in immune protective components. The milk produced over the next nine days is termed "transitional milk" and is most variable in its composition as women transition at different rates from lactogenesis II to galactopoiesis.[47] Typically, by 15 days postpartum, "mature" human milk is produced and its composition is relatively stable until weaning.[47,49] At weaning, minor changes in milk protein, lactose, chloride, and sodium concentrations were observed when milk volume declined to less than 400 mL/d. Although considerable information is available regarding changes in human milk composition in early lactation[47,49] or at weaning, less is known about the effect of sustained extended lactation (>6–12 months) on the nutrient content of human milk, particularly for micronutrients.[50] Karra and colleagues[50] reported that, zinc, calcium, and vitamins B_6 and C tended to decrease between 7 and 25 months postpartum, where as folate concentrations remained constant and magnesium levels only decreased after 18 months of lactation. Lactose, protein, and sodium increased slightly, but significantly after 10 months of lactation. These decreases in milk nutrient content between 7 and 25 months of lactation may have little nutritional significance for the infant as the mean number of breast feedings declined from 6 to 2 to 3 per day over this period and infants at this age are consuming other foods in their diets.

In addition, milk composition changes markedly within a single feeding. The first milk (foremilk) released by the mammary gland is low in fat and the fat content increases throughout the feeding and is highest in the "hindmilk."[49] Comparing paired fore- and hind milk samples collected from the same mother, protein and lactose contents were similar, where as the fat content of hind milk was two- to threefold higher and provided approximately 25–35 kcal/100 mL more energy than foremilk.[49] These variations in human milk composition within a feeding must be taken into account when obtaining samples for compositional analysis. The most accurate determination will be obtained when the complete contents of the breast are expressed and mixed prior to taking a sample for analysis.

BIOCHEMICAL COMPOSITION

Nutrients in human milk are contained in discreet compartments that reflect their chemical characteristics as well as their processing and packaging during milk synthesis. Most water-soluble nutrients are present in the aqueous phase (whey), which represents the largest compartment of human milk.[47] Casein proteins are present in milk as a colloidal dispersion and fats and fat-soluble components are present in an emulsion of milk-fat globules surrounded by a milk-fat globule membrane.[47] The physical compartmentalization of nutrients in human milk differs from infant formulas and likely contributes to the dissimilarities in nutrient bioavailability between the two fluids.

Mature human milk is approximately 88% water with a specific gravity of 1.030. The osmolarity of human milk is approximately 286 mOsm/L compared to 185–374 mOsm/L in infant formulas.[51] There is no evidence that healthy appropriate birth weight[52] or LBW[53] infants require additional water, even in hot climates. The total solids in human milk are approximately 124 g/L, of which 70 g/L is carbohydrate, 40 g/L is fat, 10 g/L is protein, and 2 g/L is mineral ash.[47,54] In comparison, bovine milk contains three-times more protein (35 g/L) and mineral ash (7 g/L), less lactose (9 g/L) and similar amounts of total solids (128 g/L), fat (37 g/L), and water (87%).[47,54]

Energy Content

The energy content of human milk can be directly measured by bomb calorimetry or approximated from the protein, carbohydrate, and fat content of milk multiplied by their Atwater values.[55] Typically, mature human milk contains between 2730 and 2940 kJ/L (650–700 kcals/L) (Table 29-1).[45,47,55] The energy content of colostrum (days 2–7 postpartum; 571 ± 80 kcals/L) and transitional (days 8–20 postpartum; 629 ± 76 kcals/L) human milk is lower than mature milk (≥21 days postpartum; 642 ± 36 kcals/L).[55] Milk caloric density, calculated as the sum of the energy provided by each macronutrient, was 6%–10% higher than that obtained by bomb calorimetry of the same milk samples, suggesting that the former method may overestimate the energy content of human milk.[55] In addition, it is important to keep in mind that some human milk proteins[56] and oligosaccharides[57] are resistant to digestion in the neonatal gastrointestinal tract. Therefore, bomb calorimetry may also overestimate the bio available energy content of human milk.

Carbohydrate

The predominant macronutrient present in human milk is carbohydrate in the form of lactose and oligosaccharides.[47,54] Lactose, the disaccharide of glucose and galactose, is readily digested by nearly all infants, where as oligosaccharides are non digestible and play a number of functional roles in the infant.[54]

Lactose and Glucose

After water, lactose is the most prevalent nutrient in human milk, with an average concentration of 70 g/L (67–76 g/L) in mature human milk (Table 29-1).[45,47] The lactose content of colostrum is lower than mature milk,[45] although colostrum is rich in oligosaccharides.[58] All oligosaccharides contain lactose at their reducing end,[58] therefore, the amount of free lactose in human milk may be overestimated if the analytical method used to quantify lactose cleaves the lactose from oligosaccharides. Lactose is the major osmol in human milk and its concentration is positively correlated with milk yield. The concentrations of lactose and alpha-lactalbumin (α-LA) in milk also are positively correlated, because lactose is synthesized by the actions of α-LA and galactosyltransferase. Lactose is one of the most stable constituents in human milk and varies little after lactation is established.[49,50] Human milk contains very low levels of free glucose[59] (Table 29-1) and other glucose metabolites,[59] which are likely of little nutritional significance to the infant. Glucose concentrations were not higher in the milk of insulin- dependent diabetic (IDDM) mothers.[59] Glucose normally rises in milk with the onset of lactogenesis II[59]; however, this rise was delayed by 1–4 days in IDDM mothers, associated with compromised initiation of lactation.[59]

TABLE 29-1 Energy and Macronutrient Composition of Colostrum and Mature Human Milk

Constituent (per L)	Colostrum	Mature Milk
Energy (kJ)[a]	2244	2729
Total carbohydrate (g)	26–76	50–83
Lactose (g)	30–50	67–70
Oligosaccharides (g)	22–24	5–15
Glucose (g)	0.2–1.0	0.2–0.3
Total lipid (g)	10–27	28–49
Triglyceride (g)	14.5–19.5	34–47

Fatty acids (g)	13–17	30–42
Cholesterol (g)	0.2–0.3	0.1–0.2
Cholesteryl esters (g)	0.05	0.01
Phospholipids & sphingolipids (g)	N.R.[b]	0.25–0.30
Total nitrogen (g)	3.0	1.9
Nonprotein nitrogen (g)	0.5	0.45
Protein nitrogen (g)	2.5	1.45
Total protein (g)	14–65	8–21
Casein (g)	3.8	3–5
β-casein (g)	2.6	3–4
κ-casein (g)	1.2	1–2
Whey proteins (g)	11–15	5–6
α-lactalbumin (g)	3.6	2–3
IgM (g)	0.02	0.01
IgG (g)	0.01	0.05
Lactoferrin (g)	3.5	1–3
Lysozyme (g)	0.1–0.2	0.1
Secretory IgA (g)	2.0	0.5–1.0
Serum albumin (g)	0.4	0.3

Mean values adapted from References 45,47,54,58,75.

[a]1 kJ = 0.2389 kcal.

[b]Data not reported

Oligosaccharides

Human milk is unique in that it contains a diverse array of highly complex oligosaccharides (HMOs), which comprise the third largest component in human milk.[54,60] Oligosaccharide concentrations in human colostrum and mature milk are 22–24 g/L and 12–14 g/L, respectively[54] (Table 29-1). Over 200 separate forms of acidic or neutral HMO have been described in human milk and the composition is influenced by maternal genetics.[58,60] These oligosaccharides are formed by the elongation of lactose with galactose in (Gal) β-1,3 or β-1,4 linkages, or *N*-acetyl glucosamine (GlcNAc) in a β-1,3 linkage to form the two core neutral oligosaccharides, lacto-*N*-tetraose (LNT, Galβ-1, 3GlcNAcβ-1, 3Galβ-1, 4Glc) and lacto-*N*-neotetraose (LNnT, Galβ-1, 4 GlcNAcβ-1, 3Galβ-1, 4Glc), respectively.[60] More complex HMOs then are formed by the addition of fucose and sialic acids (SA),[60] the latter of which are a subset of the nine-carbon sugar, neuraminic acid. The *N*-acetylated form, *N*-acetylneuraminic acid (Neu5Ac; NANA), is the most predominant form and the only one

synthesized in humans.[61] The SA residue is always found at the non reducing end of the oligosaccharide and is attached either by α-2, 3 or α-2,6 linkages to galactose or *N*-acetylgalactosamine (GalNAc).[61]

Many neutral HMOs contain fucose (Fuc) on the non reducing end.[54] These HMOs contain antigenic determinants of the Lewis blood group and/or secretor systems[62] and are synthesized by one of three fucosyltransferases (FTs) that are not uniformly distributed in the population.[58,60] The "secretor" or "nonsecretor" status of a mother is characterized by the presence of FT II activity; nonsecretor mothers have two inactive copies of the gene.[62] FT II catalyzes synthesis of α-1-2 fucosylated oligosaccharides, such as 2'-fucosyllactose (2'-FL, Fuc α-1, 2Galβ-1, 4Glc) and lacto-*N*-fucopentaose (LNF-I, Fucα-1, 2Galβ-1, 3GlcNAcβ-1, 3Galβ-1,4Glc).[62] The other fucosylated HMOs include the 3'-FL derivatives (Gal β-1, 4[Fucα-1, 3]Glc), which are synthesized by several FTs, and difucosyllactose (DFL), which contains both α-1, 2 and α-1, 3 linages (Fucα-1, 2 Gal β-1, 4[Fucα -1,3]Glc).[62] Analysis of the neutral oligosaccharide composition of human milk samples collected from over 400 women living in 10 countries demonstrated the presence of structures based on LNnT and LNT in all samples, where as fucosyloligosaccharides were not uniformly expressed in all women.[62] For example, 100% of the milk samples from women in Mexico contained 2-FL, where as only 46% of the samples from the Philippines contained 2-FL.[62]

HMOs are resistant to hydrolysis in the neonatal small intestine.[57] As a result, structurally intact HMO are absorbed and excreted in the urine and feces of preterm[63] and term[64–66] infants. Obermeier and colleagues[64] orally administered ^{13}C-galactose to lactating women to assess the degree of utilization of dietary carbohydrates for lactose and oligosaccharide synthesis. Approximately 7% of the oral dose (2 g) of ^{13}C-galactose was incorporated in lactose and neutral oligosaccharides within 8 hours and isotopically labeled lactose, 2'-FL, LNT, fucosyl-LNT, and DFL[64,65] and other slightly modified metabolites[65] were detectable in the urine of their nursing infants. Oligosaccharides excreted in urine have the potential to act as analogous receptors for viral and bacterial adhesion molecules and prevent urinary tract infections.[67] Within the blood, acidic oligosaccharides inhibit platelet–neutrophil complexes and neutrophil activation.[63] A more complete discussion of the carbohydrate components present in human milk is presented in Chapter 5.

Oligosaccharides are absorbed to a limited extent, with most reaching the large intestine in an intact form, where in they have the potential to exert widespread beneficial effects on neonatal intestinal health and host defense.[58,60] Oligosaccharides are capable of serving as receptors or receptor analogs for enteric pathogens, including viruses, bacteria, and parasites.[60] In addition, they serve as a pre biotic for growth of bifidogenic bacteria[68] and for the generation of short-chain fatty acids (SCFAs).[68] A recent study utilizing pyrosequencing of 16s rDNA determined that the stool of breast-fed infants contained higher amounts of bacteria from the phyla Actinobacteria (e.g., *Bifidobacteria)* and Bacteroidetes, where as formula-fed infants had higher proportions of *bacteria* from the phyla Firmicutes and Proteobacteria.[69] In another study, stool of breast-fed infants contained primarily acetic and lactic acids, with smaller amounts of butyric or propionic acids, suggesting increased amounts of *bifidobacteria*[70]. In contrast, feces of formula-fed infants contained mostly acetic and propionic acids, the latter of which is produced primarily by *Bacteroides* and *Clostridia*. The SCFAs reduce intestinal pH and, subsequently, decrease potentially harmful gut microbiota.[70]

In contrast to human milk, oligosaccharides are negligible in cow milk and cow milk-based infant formulas.[71] Thus, formula-fed human infants are deprived of these unique bioactive components and their addition to infant formulas has been endorsed as a means to decrease the incidence of viral and microbial infections.[58,60,72] Because HMOs are not yet available in sufficient quantities or approved for addition to infant formula, nonhuman milk oligosaccharides, namely galactooligosaccharides (GOS), polydextrose (PDX), and fructooligosaccharides (FOS) are currently added to infant formula.[60] GOS, PDX, and FOS differ structurally from HMO, and are neither fucosylated nor sialylated.[60] Many clinical trials have investigated the use of the these alternative forms of oligosaccharides and, although the results have been promising,[73] expert committees recommend additional research be conducted to define the optimal quantity and type of oligosaccharide, duration of administration and short- and long-term outcomes before recommendations can be made for the use of oligosaccharides in infant diets.[74]

Fat

Fat is second most prevalent class of macronutrients, averaging 15–20 g/L in colostrum and 35–48 g/L in mature milk and representing approximately 45%–55% of the energy content of human milk.[47,75,76]

Triglyceride accounts for nearly all of the fat in human milk (97.5%–98%), with small amounts of phospholipids and sphingolipids (0.7%) and cholesterol (0.5%)[47,75] (Table 29-1). Triglycerides are synthesized within mammary epithelial cells from glycerol and fatty acids, which are either synthesized within the mammary gland or extracted from the maternal circulation through the action of lipoprotein lipase.[76] Maternal plasma lipids are the major source of longer chain (>16 carbon) fatty acids in human milk, where as saturated fatty acids (10–14 carbons) are synthesized de novo within the mammary gland and their proportion increases in women consuming high-carbohydrate or low-fat diets.[76] Maternal diet contributes approximately 30% of the long-chain fatty acids (16:0, 18:0, 18:1*n*9, and 18:2), where as maternal adipose tissue likely contributes the majority of the remaining 70%, including the long-chain polyunsaturated fatty acids (LCPUFAs).[76]

Within the mammary epithelial cell, milk fat is packaged with other lipophilic compounds and secreted into milk as a milk fat globule, which range in size from 0.6 to 46, 0.6 to 11, and 0.7 to 17 μm in colostrum, transitional, and mature milk.[77] Each globule is surrounded by a bipolar milk fat globule membrane (MFGM) that is derived from the mammary apical plasma and mature Golgi vesicle membranes.[77] The MFGM emulsifies the milk fat and contains a number of bioactive lipids, proteins, and carbohydrates, including lactadherin and MUC1, which may play a role in host defense.[77]

Fatty Acid Composition

Fatty acids esterified to glycerol as triglycerides account for 88% of the fat; very little nonesterified fatty acids are present in human milk.[47,75,76] Approximately 43% of fatty acids are saturated, 35% are cis-monounsaturated, 1%–7% are trans-monounsaturated, and 20% are polyunsaturated fatty acids (19% *n*-6 and 1% *n*-3),[78] although these values can be markedly influenced by maternal diet[47,75] and, to a lesser degree, by stage of lactation.[75,78] For example, the percentage linoleic acid can vary from 1% to 45% of total fatty acids in milk.[47,75] Although 160 different fatty acids have been identified in human milk, the predominant fatty acids are palmitic ($C_{16:0}$; 22%), stearic ($C_{18:0}$; 7%), oleic ($C_{18:1n-9}$; 34%), and linoleic ($C_{18:2}$; 8%) acids (Table 29-2).[47,75,78–80] Digestion and absorption of fat in human milk is more efficient than fat in infant formula. One mechanism may be the presence of bile salt-stimulated lipase (BSSL)

in human milk, which is complementary to pancreatic lipase.[79] Another potential explanation is that human milk triglycerides contain palmitic acid primarily in the sn-2 position, where as infant formula fat blends contain palmitic acid primarily in the sn-1 and sn-3 positions.[79] Pancreatic lipase selectively hydrolyzes triglycerides in the sn-1 and sn-3 positions, producing two free fatty acids and a monoglyceride. Human infants efficiently absorb the 2-monopalmitin, produced during human milk fat digestion, where as free palmitate may form soaps with calcium and be lost in the feces.[79]

TABLE 29-2 Major Fatty Acids in Colostrum and Mature Human Milk

Fatty Acids (% by wt of total fatty acids)	Colostrum	Mature Milk
Saturated	40	**45 (32–52)**
C8:0	0.10–0.30	0.10–0.24
C10:0	0.50–0.75	0.9–1.3
C12:0	2.0–4.4	5.0–8.2
C14:0	5.2–8.0	6.7–8.9
C16:0	20–27	15–29
C18:0	7.0–7.4	6.5–7.5
C20:0	0.20–0.30	0.22–0.44
C22:0	0.10–0.40	0.10–0.40
C24:0	0.10–0.40	0.10–0.40
***Cis*-Monounsaturated**		**40 (30–50)**
C14:1*n*-5		0.3
C16:1*n*-9		0.4
C16:1*n*-7		2.2–3.5
C18:1*n*-9		30–34
C18:1*n*-7		1.9
C20:1*n*-11		0.15
C20:1*n*-9	0.45–0.62	0.16–0.39
C22:1*n*-9	0.04	0.02–0.06
C24:1*n*-9	0.10–0.40	0.10–0.40
***Trans*-Monounsaturated**		**7.0 (2.5–13.8)**
C16:1*n*-9t	N.R.	0.24–0.53
C18:1*n*-5t	N.R.	0.0–0.04
C18:1*n*-6t	N.R.	0.0–0.06

C18:1*n*-7+8t	N.R.	0.10–1.3
C18:1*n*-9t	1.41.	0.17–1.7
C18:1*n*-10t	N.R.	0.31–3.3
C18:1*n*-11t	N.R.	0.28–3.0
C18:1*n*-12t	N.R.	0.27–5.7
C18:1*n*-13t	N.R.	0.0–2.7
C18:1*n*-14t	N.R.	0.0–0.32
C18:1*n*-15t	N.R.	0.0–0.15
C18:1*n*-16t	N.R.	0.02–0.76
Total polyunsaturated	12.0–19.5	**14.0 (11–27)**
n-3 series	**0.85–2.0**	***0.90–1.8***
C18:3	0.5–0.7	0.7–1.2
C20:4	0.16	0.12
C22:5	0.16–0.27	0.10–0.13
C22:6	0.09–0.5	0.03–0.3
n-6 series	***9.0–18.6***	***11.0–18.5***
C18:2	7.0–17.8	8.0–18
C18:3	0.06	0.10
C20:2	0.75–1.09	0.20–0.55
C20:3	0.57–0.69	0.25–0.48
C20.4	0.7–1.0	0.34–0.64
C22.4	0.28–0.61	0.04–0.16
C22.5	0.05–0.16	0.02–0.07
CLA isomers	***0.40–0.50***	***0.45–0.55***
C18:2*n*-9c,11t	0.06–0.25	0.06–0.43
C18:2*n*-9c,12t	0.05	0.06
C18:2*n*-10t,12c	0.02	0.01
C18:2*n*-11c,13t	0.04	0.05
C18:2*n*-11c,13c	0.02	0.03

N.R., data not reported.
Mean values (range) adapted from References 78–82,84–86.

Human milk contains relatively low concentrations of SCFAs (C_4-C_8), but significant amounts of the *n*-3 and *n*-6 LCPUFA AAIC20:4*n*6) and DHA (C22:6*n*3) (Table 29-2). The AA content of human milk averages 0.3–0.6 wt% and is relatively constant worldwide. In contrast, the DHA content of human milk can vary sixfold (< 0.1–1.0 wt%) and reflects maternal DHA intake.[81] Studies indicate that the LCPUFA content of milk produced by mothers delivering prematurely to be greater than that in milk

produced by mothers delivering at term.[82] The LCPUFA levels in neonatal plasma, erythrocyte membranes, neural, and retinal tissues are determined primarily by dietary intake.[81] Beneficial effects of dietary AA and DHA intake on visual and cognitive development[17] of preterm and, to a lesser degree, term infants have been demonstrated and LCPUFAs are now routinely supplemented to preterm and most term infant formulas.[81]

The trans-fatty acid content of human milk also is influenced by material diet.[83,84] The main source of trans fatty acids in the diet is partially hydrogenated fats; trans fatty acids can comprise up to 60% of the fatty acids in shortenings and food sources made with shortenings. Trans fatty acids represented approximately 7.7% of the fatty acid intake (2.5% of energy) of lactating women and the primary dietary sources were bakery products and breads (32%), snacks (15%), fast foods (11%), and margarines and shortenings (11%).[85] Human milk trans fatty acid content varies worldwide, ranging from 0.2% to 0.8% in milk samples from China and Africa to 7% in milk of North American mothers.[85,86] High concentrations of trans fatty acids in human milk are of potential concern as they may adversely affect the growth and development of infants by interfering with *n*-3 and *n*-6 fatty acid metabolism or by altering membrane fatty acid composition.[83,85] Indeed, milk trans fatty acids are inversely associated with milk linoleic and α-linolenic acids.[83,85] Furthermore, trans fatty acid, linoleic acid, α-linolenic acid, AA, and DHA concentrations in milk were directly related to their concentrations in the plasma of breast-fed infants.[85] The inclusion of trans fatty acid content on food labels was associated with a decline in milk trans fatty acid content in Canadian mothers and a reduction in exposure of infants from 2.0 mg/kg/d in 1998 to 1.3 mg/kg/d in 2005, suggesting that increased consumer awareness of food sources of trans fatty acids has reduced their intake and subsequent secretion into milk.[85]

Dairy products and the meat of ruminant animals also are sources of trans fatty acids produced by bio hydrogenation of fatty acids in the rumen.[84,86] Dairy products typically contain 2%–5% trans fatty acids, primarily as conjugated isomers of linoleic acid.[86] Conjugated linoleic acids (CLAs) constitute a group of structurally related fatty acids with 18 carbons and two conjugated double bonds. The most abundant naturally occurring CLA in the human diet and human milk is rumenic acid ($18:2_{c9t11}$) (Table 29-2).[86] Rumenic acid is produced in the bovine and human mammary gland via the desaturation of vaccenic acid ($18:2_{t11}$) by

delta 9-desaturase (Δ9-desaturase).[86] Other forms of CLAs, such as $18:2_{t10c12}$, are found in dietary supplements and in low levels in human milk (Table 26-2).[86] Moreover, milk fat content was reduced by 25% in lactating women after consuming a CLA supplement for five days.[87] Thus, nursing mothers should be cautioned against consuming CLA supplements.[87]

Sterols

Milk sterols are composed of 90% cholesterol and 8.6% desmosterol with negligible quantities of phytosterols.[47,76] The cholesterol content of human milk declines from approximately 300–350 mg/L in colostrum to 100–200 mg/L in mature milk and concentrations are correlated with milk fat content.[47,76] Approximately 10%–15% of the cholesterol is present as cholesteryl esters, which, similarly, decline from 50 to 10 mg/L from colostrum to mature human milk.[88] Interestingly, unsaturated fatty acids are preferentially esterified to cholesterol (73 wt% of the fatty acids in cholesteryl esters).[88] Linoleic acid represented 31% of the fatty acids esterified to cholesterol versus 13% of the total fatty acids in milk.[88] Breast-fed infants consume approximately fourfold more cholesterol per day than adults (4.3 vs. 25 mg/kg body weight) and have higher circulating cholesterol concentrations than infants fed formula devoid of cholesterol.[81]

Phospholipids and Sphingolipids

Phospholipids are dipolar lipids that are present as a component of the MFGM. Their concentration in milk is approximately 250 mg/L or 0.6 g/100 g lipid.[47,76] The principal classes of phospholipids are sphingomyelin, phosphatidylcholine, phosphatidyl-ethanolamine, phosphatidylserine, and phosphatidylinositol.[47,76] Choline, which can be endogenously synthesized from phosphatidylcholine and sphingomyelin, is a major constituent of cell membranes in neural tissues and is essential for the formation of acetylcholine.[89,90] Recent studies have established a key role for prenatal and early postnatal choline intake for optimal hippocampal development and memory.[90] Human infants excrete in urine large amounts of betaine, a product of choline oxidation, regardless of choline intake, suggesting an obligatory choline requirement.[89] Infant formulas contain choline; however, the concentration may not be sufficient to meet the increased demand of the human infant.[89] In contrast, human milk choline concentration doubles between colostrum and mature milk

and appears to be sufficient to account for betaine excretion in full-term infants.[89] In addition, human milk contains several classes of sphingomyelin (100–200 μM), neutral glycosylceramides (1–20 mg/L), and acidic glycolipids (1–20 mg/L), including gangliosides.[47,76] Gangliosides may play a role in host defense by binding cholera and other toxins,[91] and in neural development.[92] Analysis of neural tissue collected postmortem from infants who had died of sudden infant death syndrome demonstrated greater ganglioside and glycoprotein SA content in the brain frontal cortex of breast-fed than formula-fed infants, suggesting increased synaptogenesis and differences in neurodevelopment.[92]

Protein and Nonprotein Nitrogen

Human milk nitrogen content declines from colostrum (3.0 g/L) to mature milk (1.9 g/L) (Table 29-1). The nitrogen component of human milk is composed of whey and casein proteins and over 200 nonprotein nitrogen (NPN) compounds.[47,93,94] Approximately 25% of the nitrogen in human milk is NPN; thus, human milk protein content is most accurately determined as (total nitrogen – NPN) × 6.25. Using this approach, the protein content of human colostrum and mature milk averages approximately 15–20 g/L and 9–10 g/L, respectively.[47,93,94] The protein content of mature human milk is markedly lower than cow's milk (35 g/L) and cow's milk- (13–15 g/L) or soy protein-based infant formulas (18–20 g/L).[93] Formulas not only contain more total protein than human but also contain a different pattern of amino acids.[93] Infants fed cow's milk-based formulas typically have higher urea N and amino acids, except taurine and cystine, than breast-fed infants.[93]

Protein

Human milk contains a heterogeneous mixture of proteins (casein and whey) and peptides. The protein content and composition of human milk undergoes dramatic changes during the course of lactation, which are matched to the developmental needs of the infant.[47,93,94] For example, colostrum is whey-predominant (90:10 whey to casein ratio) and contains higher concentrations of growth factors[94] and immune protective proteins, including sIgA, lactoferrin, and lysozyme, than mature milk.[94] As lactation progresses, the concentrations of growth factors and antimicrobial proteins decrease and casein increases, resulting in a whey to casein ratio of approximately 60:40 in mature human milk.[47,93,94] Milk produced by mothers delivering preterm is higher in total protein and some differences

in the casein and whey protein content compared to term human milk have been reported.[49,93]

Casein

There are four major families of casein proteins: α-casein, β-casein, γ-casein, and κ-casein.[47] Human milk is relatively low in casein (3–5 g/L) compared to bovine milk (26 g/L) and the forms of casein differ between the two species (Table 29-1).[93] The main caseins in human milk are β-casein (3–5 g/L) and κ-casein (1–3 g/L).[93,94] Alpha-casein, the predominant casein in bovine milk, is absent from human milk.[47,93] Casein proteins are high in proline and low in cystine and undergo extensive posttranslational phosphorylation and glycosylation.[93] Human milk κ-casein is more highly glycosylated (40% carbohydrate) than bovine κ-casein (10% carbohydrate).[93] Casein proteins precipitate at pH 4.6 and will form a "curd" in the infant's stomach. Human milk caseins tend to form looser micelles and softer curds, making them more easily digestible by carboxypeptidase. A number of peptides are formed during the proteolytic degradation of casein, which has been proposed to regulate intestinal motility (opioid-like compounds) or enhance calcium absorption (phosphopeptides).[95]

Whey

Whey is the fluid remaining following precipitation of casein micelles. In human milk, whey proteins account for 60%–70% of the total proteins versus 20% in bovine milk.[93] Human milk whey contains a variety of proteins, some of which are milk-specific and synthesized by the mammary gland (e.g., α-LA) and others of which are extracted from the maternal circulation (e.g., serum albumin) (Table 29-1). Alpha-lactalbumin is the predominant whey protein in human milk (28% of total human milk protein and 40% of whey protein), where as β-lactoglobulin is the primary whey protein in bovine milk and infant formulas and is absent from human milk.[94,96] A number of beneficial physiologic functions for both the mother and the infant have been ascribed to α-LA.[94,96] Within the mammary gland, α-LA participates in lactose synthesis and, as the major osmol in milk, lactose facilitates milk production and secretion.[96] For the infant, α-LA provides a well-balanced supply of essential amino acids and may enhance mineral and trace element absorption.[94,96] During α-LA digestion, peptides that have immunostimulatory and antibacterial

properties appear to be transiently formed.[96] Furthermore, a novel folding variant of multimeric α-LA, termed HAMLET (human α-LA made lethal to tumor cells), has been discovered that specifically enhances apoptosis of tumor, but not normal, cells.[96] The formation of HAMLET requires partial unfolding of the protein and its antitumor activity requires binding of oleic acid (C18:1) to the protein as a cofactor.[96] HAMLET has been proposed potentially to impact intestinal mucosal cell turnover, however, there is no evidence to date that it is formed in the gastrointestinal tract of human infants.[96] Although α-LA only composes 2%–3% of the protein in bovine milk, bovine α-LA-enriched whey sources are commercially available and currently are being used to produce infant formulas with higher α-LA content that have high protein quality.[96]

Functional Roles of Milk Proteins

In addition to serving as a source of amino acids, broader physiologic and immunologic roles of human milk proteins have been identified that provide nutritional and developmental benefits to the neonate.[96] For example, human milk proteins bind essential nutrients present in milk, which may enhance their bioavailability either through improved solubility or via direct interaction with receptors lining the gastrointestinal tract. Examples include haptocorrin for vitamin B_{12} and folate-binding protein, and the binding of minerals and trace elements to β-casein, α-LA, lactoferrin, and β-glycomacropeptide.[96]

As previously discussed, another key function of human milk proteins involves host defense, which occurs through antimicrobial protection, stimulation of immuno competence, and promotion of the establishment of a beneficial microbiota.[7] Roles for sIgA, lactoferrin, lactoperoxidase, and lysozyme in neonatal host defense have long been appreciated.[94] In addition, other milk proteins have been added to the cadre of immuno protective components, including haptocorrin[97] and κ-casein.[98] Another example of a multifunctional milk protein with a key role in the development and maintenance of the immune system is osteopontin (OPN).[99] OPN has been shown to regulate Th1-mediated immunity and to enhance host resistance to infections.[99] In addition, proteolytic products of milk proteins have been shown to exert bioactivities distinct from the intact proteins, contributing to the complexity of the potential interactions between milk components and the infant. For example, antimicrobial peptides were produced following an in vitro digestion of lactoferrin[100] and α-LA,[101] and bifidogenic peptides were produced from lactoferrin and

secretory component.[102] The multifunctional roles of proteins such as α-LA and lactoferrin and the potential for their proteolytic products to exert bioactive functions within the neonatal gastrointestinal tract illustrate the complexity of human milk protein bioactivity.[96]

Hormones and Growth Factors

Hormones and growth factors constitute another class of bioactive proteins and peptides in milk.[103] Many hormones have been reported in human milk, including parathyroid hormone,[104] insulin,[105] leptin,[106] ghrelin,[107] apelin,[107] nesfatin-1,[107] obestatin,[106,108] and adiponectin,[105,109,110] among others. The function of these hormones in milk and in the infant is still incompletely understood, but it is hypothesized that they are involved in the regulation of growth of various tissues and may be associated with the neonate metabolism and overall growth[110,111] On the other hand, growth factors such as insulin-like growth factor-I (IGF-I), epidermal growth factor, transforming growth factor-α and factor-β are among the most well studied.[103,112] Cell culture and animal models show that these growth factors can initiate cellular growth and expression of differentiated function and that they may be important in repair following intestinal injury.[103,112] There is evidence that orally administered recombinant growth factors survive digestion and exert biological effects within the neonatal gastrointestinal tract.[113] However, most of the published research investigating growth factors has applied supra physiological doses and there is little clinical data to support a direct role of any growth factor at the concentrations normally present in human milk on neonatal intestinal development.

Recent studies have identified neurotrophic growth factors in human milk, such as brain-derived neurotrophic factor (BNDF), glial cell-line-derived neurotrophic factor (GNDF), ciliary neurotrophic factor, and S100B protein.[114,115] Neurotrophic factors have been proposed to be important for the development of the postnatal enteric nervous system, peristalsis, and synaptic communication,[116] in addition to promote learning and memory processing.[117] BNDF and GNDF content in milk tends to decrease as lactation progresses,[114] where as S100B concentration in mature milk is higher than colostrum.[115]

Enzymes

A variety of enzymes are present in human milk that influence the

digestion of other milk components including plasmin, prothrombin, cathepsins, and kallikrein.[118] Plasmin and cathepsin are responsible for the cleavage of milk OPN into biologically active fragments that have a role in cell adhesion.[119] BSSL improves the digestion and absorption of lipids and lipid-soluble vitamins,[120] where as protease inhibitors, such as α_1-antitrypsin and antichymotrypsin, limit the digestion of some milk proteins.[121] This latter function may be critical to preserving the physiologic functions of the nutrient-binding and immunologic proteins within the neonatal gastrointestinal tract. Indeed, intact human proteins are present in the feces of both term and preterm infants.[56]

NPN

Approximately 20%–25% of the nitrogen in human milk is present as NPN, which is considerably higher than the proportion of total N present at NPN in bovine milk (5%).[47,93,122] The NPN content of infant formulas is variable and is influenced by whether the whey protein concentrate was prepared by electro dialysis or ultra filtration, the latter of which removes more NPN components.[122] The NPN components are less than 10,000 mol wt nitrogen-containing compounds. The NPN in human milk comprises a heterogeneous group of more than 200 compounds including urea, free amino acids, such as taurine and glutamine, small peptides, including many growth factors, amino sugars, nucleic acids, polyamines, nucleotides, and carnitine.[47] A number of these compounds have physiological roles in the infant and their concentrations are lower in the plasma of formula-fed versus breast-fed infants. This observation led to the addition to infant formulas of taurine and carnitine in the 1980s and, most recently, nucleotides. Urea N accounts for approximately 50% of the NPN in human milk[122] and appears to be utilized to synthesize nonessential amino acids. Heine and colleagues[123] estimated that 2%–3.6% of serum proteins are synthesized from urea N, and 10%–40% of the ^{15}N from an oral dose of ^{15}N, ^{15}N-urea was retained by preterm infants.[122] Thus, while NPN is not included in the protein fraction of human milk, it may contribute to the pool of available nitrogen for protein synthesis by the infant.

Vitamins

Human milk contains all the essential water-soluble and fat-soluble vitamins and their content can be influenced by maternal status diet.[124–127]

Healthy breast-fed infants of well-nourished mothers are at low risk for vitamin deficiencies and the need for vitamin supplementation is rare, with a few exceptions where milk concentrations may be inadequate. Examples include vitamin B_{12} in milk of vegan mothers, vitamin D in milk produced by mothers with insufficient dietary intake or sun exposure, and vitamin K in most mothers.[128] Vitamin D and K deficiencies have been described in breast-fed infants and these are the only two vitamins for which supplementation for the breast-fed infant is recommended by the AAP.[129,130] These will be discussed in greater detail below.

Fat-Soluble Vitamins

Most fat-soluble vitamins are delivered to the infant within the MFGM and their absorption is enhanced by BSSL in milk.[120] An exception is vitamin D, which is found in the whey bound to vitamin D-binding proteins.

Retinoids and Carotenoids

Retinoids and carotenoids comprise a group of closely related compounds that have vitamin A activity (e.g., retinol), can be converted to vitamin A (e.g., β-carotene, α-carotene, β-cryptoxanthin), or do not have provitamin A activity, but have other physiological functions in the body (e.g., lutein, zeaxanthin, lycopene).[131,132] For example, lutein and zeaxanthin are the only dietary carotenoids present in macular pigment and may protect photoreceptors against light-initiated oxidative damage.[131] Colostrum and breast milk are rich in retinoids, principally as retinol esters (Table 29-3).[130–134] Retinol ester is the principal form of vitamin A in human milk. Its concentration is influenced by maternal diet and varied approximately twofold across eight countries.[134] Retinol concentration in term human milk declined as lactation progressed; however, this trend was not observed in preterm human milk and the absolute concentrations were higher in transitional and mature preterm than term milk.[133] Human milk contains approximately 1.8 μmol/L total carotenoids, with the provitamin A carotenoids (α-carotene, β-carotene, β-cryptoxanthine) accounting for 50% of the carotenoids.[132] In general, the carotenoid content declines from colostrum to mature milk, but the magnitude of decrease was influenced by the polarity of the carotenoids, with the most polar carotenoids exhibiting the smallest decrease.[134] Thus, colostrum had a higher proportion of less polar carotenoids (e.g. β-carotene), where as mature milk contained more lutein and zeaxanthin.[133] Lietz and colleagues[131] demonstrated that human milk α- and β-carotene content could be

increased by dietary supplementation, without decreasing milk lutein or zeaxanthin content. They also showed that the concentrations of hydrocarbon carotenoids (α- and β-carotene) were greater in maternal plasma than milk, where as the opposite was true for xanthophylls (lutein or zeaxanthin), suggesting regulated uptake and transport of polar carotenoids into milk.[131]

Vitamin D

Unlike the other fat-soluble vitamins, vitamin D is present in whey-attached vitamin D-binding proteins.[47] The principal form of vitamin D is 25-OH vitamin D and its content is influenced by maternal diet and sun exposure.[47,130,135] Endogenous vitamin D synthesis is detrimentally impacted by darker skin pigmentation and other factors that reduce skin UVB exposure, including the winter season, environmental pollution, use of sunscreens, and lifestyle or cultural practices that limit sunlight exposure.[136] Greer and colleagues showed that milk vitamin D content could be increased 10-fold by 1.5 MED (minimal erythemal dose of total body UVB exposure), which is approximately 90 seconds of irradiation in Caucasians.[137]

Exclusively breast-fed infants can obtain vitamin D from human milk, by endogenous synthesis from sunlight exposure or by mobilizing vitamin D stored during gestation. However, vitamin D stores of infants are rapidly depleted, even in infants born to mothers with normal vitamin D status and after 2 months of age, infants are dependent upon exogenous sources of vitamin D.[130] Human milk typically contains 1 μg or 40 IU/L or less of vitamin D (Table 29-3), which is insufficient to meet the current vitamin D recommendation for infants of 400 IU/d.[130] Furthermore, the AAP recommends that infants less than 6 months of age be kept out of direct sunlight and that protective clothing, as well as sunscreen, should be used for infants 6 months and older.[130] To reduce the risk of rickets, the AAP currently recommends a daily supplement of 400 IU for all breast-fed infants and for non breast-fed infants receiving less than 1000 mL/d of vitamin D-supplemented formula for the first two months of life.[130]

TABLE 29-3 Vitamin Composition of Human Milk

Constituent (per L)	Colostrum	Mature Milk
Fat-soluble vitamins		
Vitamin A and carotenoids		

Retinol esters (μmol)	1.45–2.0	1.22
Total Carotenoids (μmol)	5.35	1.8–3.0
α-Carotene (μmol)	0.17	0.02–0.03
β-Carotene (μmol)	0.42	0.04–0.07
β-Cryptoxanthin (μmol)	0.23	0.03–0.06
Lutein/Zeaxanthin (μmol)	0.2	0.04–0.10
Lycopene (μmol)	0.51	0.02–0.06
Vitamin D3 (cholecalciferol) (μg)	0.1–0.3	0.1–1.0
Vitamin E		
α-tocopherol (mg)	15	3-5
γ-tocopherol (mg)	1.5	1.0
Vitamin K		
Phylloquinone, K1 (μg)	0.64–3.7	0.86–3.9
Menaquinone, K2 (μg)	1.8–2.6	1.2–2.0
Water-soluble vitamins		
Ascorbic Acid (mg)	80–100	40–100
Biotin (μg)	4.5	5–9
Vitamin B_{12} (μg)	1.2–1.5	0.5–0.8
Folate (μg)	5–10	50–100
Niacin (mg)	0.5	1.5–2.0
Pantothenic Acid (mg)	2.5	2.0–2.5
Pyridoxine, B_6 (μg)	20–40	90–300
Riboflavin, B_2 (μg)	500	300–400
Thiamin, B_1 (μg)	20	160–210

Mean values adapted from References.[47,128,132,133,145,152,153,177]

Vitamin E and Coenzyme Q10

Analogous to vitamin A, the term “vitamin E” describes a family of closely related compounds (α, β- and γ-tocopherol) that function as antioxidants in cell membranes by scavenging free radicals. Human milk contains α- and γ-tocopherol, but insignificant levels of β-tocopherol.[134] The α- and γ-tocopherol content of colostrum is greater than of mature milk (Table 29-3).[134] In addition, the α- and γ-tocopherol content of preterm human milk is greater than milk from mothers delivering at term.[134] Infants appear to differentially utilize natural and synthetic forms of vitamin E,[138] but both human milk and formula contain sufficient vitamin E to meet requirements.

Recently, coenzyme Q10, a lipophilic antioxidant, has been reported in

term and preterm human milk[139,140] and the coenzyme Q10, α- and γ-tocopherol concentrations were directly correlated with the antioxidant capacity of the milk.[140] Compared to term human milk, the coenzyme Q10 content was lower in colostrum (0.50 vs. 0.81 μmol/L) and transitional (0.45 vs. 0.75 μmol/L) milk from mothers delivering preterm.[140] In term human milk, coenzyme Q10 content declined from colostrum (0.80 μmol/L) to transitional (0.57 μmol/L) and mature milk (0.44 μmol/L) samples, which was more evident when expressed on the basis of milk cholesterol content, 137, 86, and 67 μmol/mol, respectively.[139]

Vitamin K

Vitamin K (phylloquinone) is a necessary cofactor for the vitamin K-dependent carboxylase, which catalyzes the formation of γ-carboxyglutamic acid (Gla). The formation of Gla residues creates calcium-binding sites that are necessary for the function of coagulation factors (II, VII, IX, and X), protein C, protein S, protein Z, osteocalcin, Prolin rich Gla proteins (one and two), and Conantokin (G and T).[141] Vitamin K is not readily transported across the placenta; thus, vitamin K deficiency can cause unexpected bleeding in the first week of life.[129] The AAP currently recommends that a single, intramuscular dose of vitamin K (0.5–1.0 mg) be given to all newborns to prevent vitamin K deficiency bleeding.[129] In addition, it has been suggested that exclusively breast-fed infants receive oral supplementation of 1 mg of Vitamin K weekly until 12 weeks postpartum.[141]

Human milk contains both forms of vitamin K, phylloquinone (K_1) and menaquinones (K_2),[142,143] with milk menaquinone-4 (MK-4) being synthesized in the body from dietary phylloquinone.[144] Overall, a weak relationship has been noted between maternal dietary phylloquinone intake and milk phylloquinone content, however, the vitamin K content and composition can be impacted by maternal diet and supplement use.[126,142,144] Maternal supplementation with a pharmacological dose of 5 mg phylloquinone per day increased its concentration in milk and in the serum of breastfeeding infants.[126] Another form of menaquinome (MK-7) has been detected in breast milk of Japanese women.[145] MK-7 is synthesized by bacteria and is associated with the intake of a regional fermented soybean product. Prophylactic vitamin K administration is particularly important for exclusively breast-fed infants because human milk vitamin K (phylloquinone) concentrations (typically 1–3 μg/L) (Table

29-3) are insufficient to meet the recommended intake for infants of 2 µg/d.[129] In addition, *bifidobacteria*, which are more predominant in breast- than formula-fed infants, do not synthesize vitamin K and the vitamin K content in the stool of breast-fed infants was insignificant compared to formula-fed infants.[146]

Water-Soluble Vitamins

The concentrations of water-soluble vitamins in human milk are influenced by maternal nutritional intake and vitamin supplementation.[127,147] The thiamin and riboflavin contents of human milk typically are sufficient to meet the needs of the exclusively breast-fed infant, but vitamin B_6, vitamin B_{12}, and folate may be low in milk produced by mothers from lower socioeconomic groups.[127] Although supplementation may be beneficial for women with poor dietary intake, only niacin and vitamin B_{12} can be increased further in milk when the mother's vitamin status is adequate.[47,147]

Vitamin C

The concentration of ascorbic acid in human milk can range from 40 to 100 mg/L in well-nourished women and within an individual mother the concentration decreases from colostrum to mature milk[47] (Table 29-3). Low maternal intake reduces milk ascorbic acid content. For example, mean human milk ascorbic acid was 50% lower in the milk of African (31 mg/L) versus European (63 mg/L) women.[148] Furthermore, ascorbic acid supplementation (1000 mg/d for 10 days) modestly increased milk ascorbic acid content in European women (60–70 mg/L) versus (19–60 mg/L) in African women.[148] In African women, one serving of orange juice per week had no significant effect, where as three or five servings/week (100 mg ascorbic acid/serving) for six weeks doubled mean human milk ascorbic acid content from 16 to 32 mg/L.[148] Human milk ascorbic acid is sufficient to meet the needs of exclusively breast-fed infants and no supplementation is recommended.[128]

Thiamine

Thiamine is present in both free (30%) and phosphorylated (70%) forms in human milk.[124] The thiamine content of colostrum is low (20 µg/L), but increases to 160–200 µg/L in mature milk and this amount can be further increased with maternal supplementation (Table 29-3). The thiamine

intake of breast-fed infants is lower than the recommended dietary allowance (RDA), however, the intake appears adequate to meet their needs and infantile beriberi is rare in the United States[128] Other than ensuring adequate maternal thiamine intake, supplementation of the infant is not recommended.[128]

Riboflavin

Riboflavin is present in human milk as both riboflavin and flavin adenine dinucleotide.[124] The riboflavin content of colostrum is high and decreases to 400–600 µg/L in mature human milk (Table 29-3). Riboflavin deficiency in women is associated with low concentration of riboflavin in milk.[124] However, in well-nourished women, riboflavin is only increased by pharmacological intakes (>3 times the RDA).[47] The riboflavin intake of breast-fed infants is adequate and no supplementation of infants is needed.[128]

Niacin

The niacin content of colostrum is approximately 0.5 mg/L and 1.5–2.0 mg/L in mature milk (Table 29-3). The content in milk can be increased to as high as 6.0 mg/L by maternal intake.[47,147] In addition, niacin can be synthesized from excess dietary tryptophan, which contributes to the milk niacin content.[47,147] Niacin supplementation is not recommended for exclusively breast-fed infants.[128]

Vitamin B_6

The vitamin B_6 (pyridoxal, pyridoxal phosphate, pyrodoxamine, and pyridoxine) content of colostrum is low and increases to 90–310 µg/L in mature milk (Table 29-3).[47,127] Vitamin B_6 in human milk of well-nourished women is adequate for the exclusively breast-fed infant[128]; however, low maternal intake can reduce milk B_6 content.[127,149]

Folate and Vitamin B_{12}

The folate (5-methyltetrahydrofolate) content of human milk increases from 5 to 10 µg/L in colostrum to 50–100 µg/L in mature milk[127] and its concentrations remain constant with maternal deficiency or supplementation.[124] It was estimated that one-third of pregnant and lactating women are not meeting their folate requirement by diet alone,

even after the mandatory supplementation of folate to enriched flour, pasta, and grains in North America in 1998.[150] However, folate deficiency has not been reported in breast-fed infants and supplements are not recommended for the infant.[128]

The content of vitamin B_{12} in human milk decreases from colostrum (approximately 1.2 μg/L) to mature milk (approximately 0.5 μg/L).[128,147] Infants of well-nourished mothers with adequate vitamin B_{12} intake do not require supplementation. However, the vitamin B_{12} content of milk of vegan mothers can be insufficient to meet the needs of the exclusively breast-fed infant and deficiencies have been reported.[151] Thus, in some clinical situations, supplements of vitamin B_{12} are warranted for the mother or infant.[128]

Biotin and Pantothenic Acid

The biotin content of colostrum is approximately 4.5 μg/L and its concentration increases to 5–9 μg/L in mature milk (Table 29-3).[152] However, mature milk biotin does not reach a stable plateau and varies substantially between mothers (0–27 μg/L)[152] (Table 29-3). The concentration of pantothenic acid in human milk is 2.0– 2.5 mg/L[153] (Table 29-3). There were no differences in milk pantothenic acid concentrations between two weeks and four months postpartum, but its concentration was responsive to maternal dietary intake.[153] Pantothenic acid was also higher in preterm (3.5 mg/L) versus term (2.5 mg/L) human milk.[153]

In summary, in well-nourished women, human milk contains all essential vitamins at adequate concentrations to meet the needs of the exclusively breast-fed infant. Two exceptions are vitamins D and K, but deficiencies can be avoided by adequate maternal dietary and maternal sun exposure, in the case of vitamin D, and prophylactic vitamin K injection at birth. Mothers following restricted diets or those with previous gastric bypass surgery are at risk for low milk vitamin B_{12} content and should be encouraged to take supplemental vitamin B_{12}. As the content of all fat- and water-soluble vitamins in human milk are influenced by maternal intake, it may be prudent to encourage all lactating mothers to consider a daily multivitamin supplement to ensure adequate milk nutrition content.[124]

Macro- and Microminerals

Over 20 macro- and microminerals have been identified in human milk and, in general, their content changes over the course of lactation, with most declining from colostrum to mature milk.[154–156] In direct contrast to vitamins, mineral concentrations in milk generally do not correlate to maternal serum or typical dietary intake[157] and do not respond to supplementation.[158] Mineral transport into milk is tightly regulated by the mammary gland.[159] Specific micromineral (Cu, Zn, and Fe) transporters have been identified and their molecular and hormonal regulation is under study.[159] The absolute concentrations of most macro- and microminerals are lower in human milk than bovine milk or infant formulas;[155,156] however, they are characterized by enhanced bioavailability.[160] Thus, the mineral requirements of exclusively breast-fed term infants can be met for the first six months of life.[47,160]

Macrominerals

The major macrominerals in milk include calcium, magnesium, phosphorus, sodium, potassium, and chloride[47,94,155] (Table 29-4). Electrolytes, such as sodium and potassium, are totally soluble in water and found primarily in whey. In contrast, calcium forms complexes with phosphate and casein; 85% is found in the whey and 15% associated with casein micelles in calcium–phosphate linkages.[161]

TABLE 29-4 Macromineral and Micromineral Composition of Human Milk

Constituent (per L)	Colostrum	Mature Milk
Macrominerals		
Calcium (mg)	250	200–250
Chloride (mg)	600–800	400–450
Magnesium (mg)	30–35	30–35
Phosphorus (mg)	120–160	120–140
Potassium (mg)	600–700	400–550
Sodium (mg)	300–400	150–250
Microminerals		
Boron (μg)	30	28
Chromium (μg)	17	25–75
Copper (mg)	0.5–0.8	0.1–0.3

Fluoride (μg)	5–20	4–15
Iodine (μg)	40–50	140–150
Iron (mg)	0.5–1.0	0.3–0.75
Manganese (μg)	5–12	3–4
Selenium (μg)	25–32	10–25
Zinc (mg)	5–12	1–3

Mean values adapted from References.[47,154–156,161,162,165,166]

Calcium, Magnesium, and Phosphorus

Calcium content of colostrum (300–350 mg/L) is greater than human mature milk (200–250 mg/L)[154,155] (Table 29-4), but both are markedly lower than that present in bovine milk (1200–1350 mg/L).[154] Once lactation is established, the calcium and magnesium content of human milk is relatively stable between (3–26 weeks postpartum), where as human milk phosphorus content declines approximately 40% during that time frame.[162] Calcium bioavailability is high from human milk, due in part to the calcium to phosphorus ratio of 2:1, and the levels of calcium, phosphorus, and magnesium in human milk are adequate to meet the needs of the term human infant.[47] For preterm infants, preterm or donor human milk should be fortified with a human milk fortifier to prevent rickets and hyocalcemic tetany.[163]

Sodium, Potassium, and Chloride

The concentrations of electrolytes in milk are influenced by an electrochemical gradient in the mammary secretory cells, which is established by the passage of water.[47] In early lactation, changes in the concentrations of lactose, sodium, and chloride signal closure of paracellular pathways between the mammary epithelial cells at one to two days postpartum.[37] Overall, the concentrations of sodium, potassium, and chloride are lower in mature milk than colostrum, but the magnitude of difference is greater for potassium and chloride (60%–70% lower) than sodium (30%–35% lower).

Microminerals

The predominant trace elements in human milk include iron, copper, and zinc, as well as, manganese, selenium, chromium, fluoride, iodine, and boron[47,154,155] (Table 29-4). The concentrations of trace elements in human milk are low; however, their bioavailability is much higher than from bovine milk or infant formula.[160] The presence of lactoferrin in

human milk has been proposed to facilitate iron and manganese uptake via an intestinal receptor for this protein.[160] Furthermore, high concentrations of ascorbic acid and citrate also may facilitate the uptake of trace elements from human milk, where as a high concentration of casein in cow's milk and cow's milk formulas may limit trace element absorption from these diets.[160] The physical distribution of trace elements differs between human and cow milk, which also may influence their bioavailability.[161] For example, in bovine milk 24% of the iron, 44% of the copper, and 84% of the zinc are bound to casein, where as comparable values for human milk are 9%, 7%, and 8%, respectively, and most of these trace elements are found in the human milk whey.[160] A significant proportion of the iron in both bovine (14%) and human (33%) milk is associated with the outer MFGM.[161]

Iron, Zinc, Copper, and Manganese

The iron concentration in human milk averages 0.5–1.0 mg/L in colostrum and 0.35–0.70 mg/L in mature milk[154,162] (Table 29-4). The bioavailability of iron from human milk is high (20%–50%) relative to infant formulas (4%–7%).[160] Exclusively breast-fed infants generally do not need iron supplements prior to 6 months of age, at which time the AAP recommends the gradual introduction of iron-enriched solid foods, such as iron supplemented rice cereal.[164] Infants who have low iron stores may require iron supplementation prior to 6 months of age to avoid iron-deficiency anemia.[164] However, trace elements that share absorptive pathways compete for uptake, and excessive iron supplementation may create imbalances in the ratios between trace elements (Fe/Zn, Zn/Cu, Fe/Mn) and impair trace element absorption.[160] The zinc concentration of human milk rapidly declines from 5 to 12 mg/L in colostrum to 1–3 mg/L in mature milk,[154,162] and does not seem to be influenced by maternal dietary intake.[156] Similarly to iron, the bioavailability of zinc from human milk (60%) is greater than from formula (approximately 30%).[160] Following a similar trend to iron and zinc, the copper content of human colostrum and milk declines from colostrum (0.5–0.8 mg/L) to mature milk (0.1–0.3 mg/L), although the extent of the decline is less than those observed with iron and zinc.[156,165] Copper absorption is similar from human milk (25%) and cow's milk-based infant formula (23%); however, the copper concentrations in formula are double that of human milk, suggesting the potential for excess copper intake in formula-fed infants.[165] Similar to the

other trace elements, the manganese content of human milk (3–4 μg/L) is lower than colostrum (5–12 μg/L)[154,162] and its bioavailability from human milk is greater than from infant formula.[160]

Selenium, Iodine, and Fluoride

The selenium, fluoride, and iodine concentrations inhuman milk vary worldwide. Mean selenium concentrations in colostrum and milk are 33–40 and 8–12 μg/L, respectively[154,162] (Table 29-4). Maternal selenium status has a strong influence on milk selenium content and a multi country comparison showed that levels in milk from the Philippines was much higher than milk from Swedish or Hungarian mothers.[154] Likewise, milk fluoride was reported to vary from 7 to 118 μg/L in samples collected worldwide, but does not appear to be affected by stage of lactation.[154] In many countries, fluoride is supplemented to the water supply. Esala and colleagues[166] compared milk samples collected from mothers living in two areas having an average of 0.2 and 1.7 mg fluoride/L in the drinking water. The fluoride content of breast milk in the high-fluoride areas (10.9 μg/L; range 4.5–50.7 μg/L) was significantly higher than the low-fluoride areas (7.0 μg/L; range 4.3–14.0 μg/L), respectively.[166] To avoid fluorosis, the AAP recommends that fluoride not be directly administered to infants during the first six months after birth, whether they are breast- or formula-fed.[1] During the period from 6 months to 3 years of age, breast-fed (and formula-fed) infants require fluoride supplementation, only if the water supply is severely deficient in fluoride (<0.3 mg/L).[1] Lastly, milk iodine content varies widely by geographic location and maternal intake and iodized salt and seafood are primary dietary sources.[167] A recent systematic review of human milk iodine content showed a range of 12–155 μg/L, with the lowest concentration found in areas with high prevalence of goiter.[167] In Germany, the mean iodine content of human milk increased sixfold from 1982 to 1996 due to maternal iodine supplementation.[167]

In summary, milk mineral content, with the exception of selenium, iodine, and fluoride, is not significantly influenced by maternal diet. In well-nourished women, human milk contains all essential macro- and microminerals at adequate concentrations to meet the needs of the exclusively breast-fed infant for the first 6 months of life and supplements are not recommended under normal circumstances.

DRUGS, ENVIRONMENTAL CONTAMINANTS,

AND VIRUSES IN HUMAN MILK

The presence in breast milk of environmental chemicals that may disrupt the nervous, endocrine, and reproductive systems of humans has gained increased attention from regulatory agencies and women's and children's health advocates.[168–171] Environmental chemicals in milk encompass substances that are introduced from food, pharmaceuticals, or exposure to environmental contaminants.[168–171] Although scientific evidence to date indicates that the advantages of breastfeeding outweigh any risks from contaminants, it is important to identify contaminant trends, locate disproportionately exposed populations, and take public health measures to decrease and eliminate chemical pollutants from mother's milk.[169] This is particularly important, as human milk often serves as the sole source of nutrition for the breast-fed infant and owing to the rapid mental and physical development that occurs in the early postnatal period, infants can be more susceptible to adverse effects resulting from chemical exposures than adults.[169,170]

Pharmaceuticals and Drugs

Drugs may enter into milk through medical treatment or recreational drug use.[168,171] Factors affecting the transfer of compounds into milk include their ionization (pKa of the compound), lipid solubility, maternal plasma protein binding, and molecular weight.[168] In general, small (<200 Da), ionized, lipid-soluble compounds will appear in milk within one hour after maternal ingestion and the plasma to milk ratio of the drug will be close to one.[168] For most drugs, the percentage of the maternal dose that is transferred to milk is between 0.5% and 1.0%, but there are some striking exceptions including certain beta blockers, where more than 5% of the maternal dose has been identified as passing into milk.[168] There have been very small numbers of reports of adverse reactions in the infant as a result of maternal drug ingestion.[169] Although the use of antidepressant medication during lactation is controversial, Davanzo[172] concluded after a recent review of the literature that selective serotonin reuptake inhibitors and nortriptyline were safe during lactation, but tricyclic doxepin and nefazodone should be avoided. Specific recommendations for the clinical management of breastfeeding mothers when medical treatment is necessary, including the use of radioisotopes, can be found in the AAP statement[171] and in the LactMed database, under the Toxicology data network of the U.S. National Library of Medicine

(http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?LACT).

Environmental Contaminants

Decades of inadequately controlled pollution of the environment by "persistent, bioaccumulative and toxic" (PBT) chemicals has resulted in the contamination of human milk by polychlorinated biphenyls (PCBs), dichlorodiphenyltrichloroethane and its metabolites, dioxins, dibenzofurans, polybrominated diphenyl ethers, and heavy metals.[168] Women may be exposed to lipophilic chemicals through air, food, water, and occupational and household environments. These compounds are encountered to differing extents among women in industrially developed versus developing countries.[168] Some of the highest levels of milk contaminants are seen in developing countries among women living in agricultural areas, which are extensively treated with pesticides, and in the Canadian Inuit, who eat species high on the marine food chain that accumulate heavy burdens of PBT chemicals.[170]

Lipophilic chemicals are stored and accumulated over time in body fat and are mobilized during lactation. Generally, chemicals enter breast milk by passive transfer from plasma, and their concentration in milk is proportional to their solubility and lipophilicity. Twenty percent or more of maternal body burden of some persistent pollutants, such as PCBs, can be transferred during six months of lactation.[168] Although the levels of PBTs in human milk increase with the age of the mother,[169] the pattern of contaminant concentrations in milk over the course of lactation is still debatable. LaKind and colleagues reported that lipid-adjusted concentration of certain organic chemicals in milk and serum of lactating women did not decreased consistently over time. The study was conducted in samples collected from 10 women, thus additional studies with larger cohorts are needed to corroborate the findings.[173] However, studies of infants exposed to environmental chemicals in human milk have, for the most part, not shown adverse health effects for the nursing infant.[169] Thus, while more research is needed, current scientific evidence does not support altering the global public health recommendation of exclusive breastfeeding for six months followed by continued breastfeeding for the first year or beyond.[1,33,169,170]

Human Immunodeficiency Virus Type 1

Transmission of human immunodeficiency virus type 1 (HIV-1) through breastfeeding has been conclusively demonstrated.[174] Therefore, to avoid

mother-to-child transmission, the AAP and CDC recommend complete avoidance of breastfeeding by HIV-1-infected mothers in the United States.[175] However, in low- to mid-income countries, where infant morbidity and mortality rates are high, breastfeeding may outweigh the risk of HIV infection. Infants who received mixed feeding (breastfeeding and solid foods or formula) were at a higher risk of acquiring HIV infection.[176] For HIV endemic areas, the recommendation is for exclusive breastfeeding during the first months of life followed by extended antiretroviral therapy to the infant.[1,176]

OVERALL SUMMARY AND CONCLUSIONS

Breastfeeding is the optimal form of nutrition for the human infant and has numerous nutritional advantages over infant formula. In addition, human milk confers short- and long-term health protection against infection and autoimmune disease and may have lifelong consequences reducing chronic disease incidence. All mothers should be strongly encouraged by health care providers to breastfeed their infants, and medical, governmental, and societal programs should be established to support exclusive breastfeeding for the first six months of life, including ensuring that all hospitals become "Baby Friendly."[30]

REFERENCES

1. Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129:e827–e841.
2. Yates AA. Dietary reference intakes: concepts and approaches underlying protein and energy requirements. *Nestle Nutr Workshop Ser Pediatr Program*. 2006;58:79–90.
3. Sullivan S, Schanler RJ, Kim JH, et al. An exclusively human milk-based diet is associated with a lower rate of necrotizing enterocolitis than a diet of human milk and bovine milk-based products. *J Pediatr*. 2010;156:562–567.
4. Isaacs EB, Fischl BR, Quinn BT, et al. Impact of breast milk on intelligence quotient, brain size, and white matter development. *Pediatr Res*. 2010;67:357–362.
5. Hanson C, Sundermeier J, Dugick L, et al. Implementation, process, and outcomes of nutrition best practices for infants <1500 g. *Nutr Clin Pract*. 2011;26:614–624.
6. Rautava S, Walker WA. Academy of breastfeeding medicine founder's lecture 2008: breastfeeding–an extrauterine link between mother and child. *Breastfeed Med*. 2009;4:3–10.
7. Walker A. Breast milk as the gold standard for protective nutrients. *J Pediatr*. 2010;156(suppl 2):S3–S7.
8. Barclay AR, Russell RK, Wilson ML, et al. Systematic review: the role of breastfeeding in the development of pediatric inflammatory bowel disease. *J*

Pediatr. 2009;155:421–426.
9. Patelarou E, Girvalaki C, Brokalaki H, et al. Current evidence on the associations of breastfeeding, infant formula, and cow's milk introduction with type 1 diabetes mellitus: a systematic review. *Nutr Rev*. 2012;70:509–519.
10. Szajewska H, Chmielewska A, Piescik-Lech M, et al. Systematic review: early infant feeding and the prevention of coeliac disease. *Aliment Pharmacol Ther*. 2012;36:607–618.
11. Foisy M, Boyle RJ, Chalmers JR, et al. Overview of reviews the prevention of eczema in infants and children: an overview of cochrane and non-cochrane reviews. *Evid Based Child Health*. 2011;6:1322–1339.
12. Oddy WH. Longer duration of exclusive breastfeeding associated with reduced risk of childhood asthma up to age six. *Evid Based Nurs*. 2013;16:18–19.
13. Brew BK, Allen CW, Toelle BG, Marks GB. Systematic review and meta-analysis investigating breast feeding and childhood wheezing illness. *Paediatr Perinat Epidemiol*. 2011;25:507–518.
14. Kramer MS, Kakuma R. Maternal dietary antigen avoidance during pregnancy or lactation, or both, for preventing or treating atopic disease in the child. *Cochrane Database Syst Rev*. 2012;9:CD000133.
15. Gomez-Sanchiz M, Canete R, Rodero I, et al. Influence of breast-feeding and parental intelligence on cognitive development in the 24-month-old child. *Clin Pediatr (Phila)*. 2004;43:753–761.
16. Rao MR, Hediger ML, Levine RJ, et al. Effect of breastfeeding on cognitive development of infants born small for gestational age. *Acta Paediatr*. 2002;91:267–274.
17. Gustafsson PA, Duchen K, Birberg U, Karlsson T. Breastfeeding, very long polyunsaturated fatty acids (PUFA) and IQ at 6 1/2 years of age. *Acta Paediatr*. 2004;93:1280–1287.
18. Arenz S, Von Kries R. Protective effect of breast-feeding against obesity in childhood: can a meta-analysis of published observational studies help to validate the hypothesis? *Adv Exp Med Biol*. 2009;639:145–152.
19. Li C, Kaur H, Choi WS, et al. Additive interactions of maternal prepregnancy BMI and breast-feeding on childhood overweight. *Obes Res*. 2005;13:362–371.
20. Mayer-Davis EJ, Rifas-Shiman SL, Zhou L, et al. Breast-feeding and risk for childhood obesity: Does maternal diabetes or obesity status matter? *Diabetes Care*. 2006;29:2231–2237.
21. McCrory C, Layte R. Breastfeeding and risk of overweight and obesity at nine-years of age. *Soc Sci Med*. 2012;75:323–330.
22. Chu L, Retnakaran R, Zinman B, et al. Impact of maternal physical activity and infant feeding practices on infant weight gain and adiposity. *Int J Endocrinol*. 2012;2012:293821.
23. Yin J, Quinn S, Dwyer T, et al. Maternal diet, breastfeeding and adolescent body composition: a 16-year prospective study. *Eur J Clin Nutr*. 2012;66:1329–1334.
24. Wojcicki JM. Maternal prepregnancy body mass index and initiation and duration of breastfeeding: a review of the literature. *J Womens Health*. 2011;20:341–347.
25. Fall CH, Borja JB, Osmond C, et al. Infant-feeding patterns and cardiovascular risk factors in young adulthood: data from five cohorts in low- and middle-income countries. *Int J Epidemiol*. 2011;40:47–62.
26. Breastfeeding: Data: NIS | DNPAO | CDC. Breastfeeding among U.S. children born 2000–2009, CDC National Immunization Survey. http://www.cdc.gov/breastfeeding/data/NIS_data/index.htm. Updated 2012.
27. United States Breastfeeding Committee (USBC). Healthy people 2020:

Breastfeeding objectives. http://www.usbreastfeeding.org/LegislationPolicy/FederalPolicies/HealthyPeople2 Updated 2010.
28. Belanoff CM, McManus BM, Carle AC, et al. Racial/ethnic variation in breastfeeding across the US: a multilevel analysis from the national survey of children's health, 2007. *Matern Child Health J*. 2012;16(suppl 1):S14–S26.
29. Jensen E. Participation in the supplemental nutrition program for women, infants and children (WIC) and breastfeeding: national, regional, and state level analyses. *Matern Child Health J*. 2012;16:624–631.
30. U.S. Department of Health and Human Services. *The Surgeon General's Call to Action to Support Breastfeeding*. Washington, DC: U.S. Department of Health and Human Services, Office of the Surgeon General; 2011.
31. Gale C, Logan KM, Santhakumaran S, et al. Effect of breastfeeding compared with formula feeding on infant body composition: a systematic review and meta-analysis. *Am J Clin Nutr*. 2012;95:656–669.
32. Betoko A, Charles MA, Hankard R, et al. Determinants of infant formula use and relation with growth in the first 4 months. *Matern Child Nutr*. 2014;10(2):267–279.
33. Kramer MS, Kakuma R. Optimal duration of exclusive breastfeeding. *Cochrane Database Syst Rev*. 2012;8:CD003517.
34. Andres A, Cleves MA, Bellando JB, et al. Developmental status of 1-year-old infants fed breast milk, Cow's milk formula, or soy formula. *Pediatrics*. 2012;129:1134–1140.
35. de Onis M, Onyango AW, Borghi E, et al. Comparison of the world health organization (WHO) child growth standards and the national center for health Statistics/WHO international growth reference: implications for child health programmes. *Public Health Nutr*. 2006;9:942–947.
36. de Onis M, Siyam A, Borghi E, et al. Comparison of the world health organization growth velocity standards with existing US reference data. *Pediatrics*. 2011;128:e18–e26.
37. Neville MC, Morton J. Physiology and endocrine changes underlying human lactogenesis II. *J Nutr*. 2001;131:3005S–3008S.
38. Henderson JJ, Hartmann PE, Newnham JP, Simmer K. Effect of preterm birth and antenatal corticosteroid treatment on lactogenesis II in women. *Pediatrics*. 2008;121:e92–e100.
39. Matias SL, Nommsen-Rivers LA, Creed-Kanashiro H, Dewey KG. Risk factors for early lactation problems among Peruvian primiparous mothers. *Matern Child Nutr*. 2010;6:120–133.
40. Hartmann P, Cregan M. Lactogenesis and the effects of insulin-dependent diabetes mellitus and prematurity. *J Nutr*. 2001;131:3016S–3020S.
41. Parker LA, Sullivan S, Krueger C, et al. Effect of early breast milk expression on milk volume and timing of lactogenesis stage II among mothers of very low birth weight infants: a pilot study. *J Perinatol*. 2012;32:205–209.
42. Chapman DJ, Young S, Ferris AM, Pérez-Escamilla R. Impact of breast pumping on lactogenesis stage II after cesarean delivery: a randomized clinical trial. *Pediatrics*. 2001;107:1–7.
43. Dewey KG, Nommsen-Rivers LA, Heinig MJ, Cohen RJ. Risk factors for suboptimal infant breastfeeding behavior, delayed onset of lactation, and excess neonatal weight loss. *Pediatrics*. 2003;112:607–619.
44. Rasmussen KM, Kjolhede CL. Prepregnant overweight and obesity diminish the prolactin response to suckling in the first week postpartum. *Pediatrics*.

2004;113:e465–e471.
45. Hester SN, Hustead DS, Mackey AD, et al. Is the macronutrient intake of formula-fed infants greater than breast-fed infants in early infancy? *J Nutr Metab*. 2012;2012:891201.
46. Kent JC, Mitoulas LR, Cregan MD, et al. Volume and frequency of breast feedings and fat content of breast milk throughout the day. *Pediatrics*. 2006;117:e387–e395.
47. Picciano MF. Nutrient composition of human milk. *Pediatr Clin North Am*. 2001;48:53–67.
48. Molto-Puigmarti C, Castellote AI, Carbonell-Estrany X, Lopez-Sabater MC. Differences in fat content and fatty acid proportions among colostrum, transitional, and mature milk from women delivering very preterm, preterm, and term infants. *Clin Nutr*. 2011;30:116–123.
49. Saarela T, Kokkonen J, Koivisto M. Macronutrient and energy contents of human milk fractions during the first six months of lactation. *Acta Paediatr*. 2005;94:1176–1181.
50. Karra MV, Udipi SA, Kirksey A, Roepke JL. Changes in specific nutrients in breast milk during extended lactation. *Am J Clin Nutr*. 1986;43:495–503.
51. Paxson CL, Jr, Adcock EW,3rd, Morriss FH, Jr. Osmolalities of infant formulas. *Am J Dis Child*. 1977;131:139–141.
52. Ashraf RN, Jalil F, Aperia A, Lindblad BS. Additional water is not needed for healthy breast-fed babies in a hot climate. *Acta Paediatr*. 1993;82:1007–1011.
53. Cohen R, Brown K, Rivera L, Dewey K. Exclusively breastfed, low birthweight term infants do not need supplemental water. *Acta Paediatr*. 2000;89:550–552.
54. Kunz C, Rodriguez-Palmero M, Koletzko B, Jensen R. Nutritional and biochemical properties of human milk, part I: general aspects, proteins, and carbohydrates. *Clin Perinatol*. 1999;26:307–333.
55. Hosoi S, Honma K, Daimatsu T, et al. Lower energy content of human milk than calculated using conversion factors. *Pediatr Int*. 2005;47:7–9.
56. Davidson LA, Donovan SM, Lönnerdal B, Atkinson SA. Excretion of human milk proteins by term and premature infants. In: Atkinson SA, Lonnerdal B, eds. *Protein and Non-Protein Nitrogen in Human Milk*. Boca Raton, FL: CRC Press; 1989:117–136.
57. Gnoth MJ, Kunz C, Kinne-Saffran E, Rudloff S. Human milk oligosaccharides are minimally digested in vitro. *J Nutr*. 2000;130:3014–3020.
58. Kunz C, Rudloff S, Baier W, et al. Oligosaccharides in human milk: Structural, functional, and metabolic aspects. *Annu Rev Nutr*. 2000;20:699–722.
59. Arthur PG, Kent JC, Hartmann PE. Metabolites of lactose synthesis in milk from diabetic and nondiabetic women during lactogenesis II. *J Pediatr Gastroenterol Nutr*. 1994;19:100–108.
60. Bode L, Jantscher-Krenn E. Structure-function relationships of human milk oligosaccharides. *Adv Nutr*. 2012;3:383S–391S.
61. Sprenger N, Duncan PI. Sialic acid utilization. *Adv Nutr*. 2012;3:392S–397S.
62. Erney RM, Malone WT, Skelding MB, et al. Variability of human milk neutral oligosaccharides in a diverse population. *J Pediatr Gastroenterol Nutr*. 2000;30:181–192.
63. Rudloff S, Pohlentz G, Diekmann L, et al. Urinary excretion of lactose and oligosaccharides in preterm infants fed human milk or infant formula. *Acta Paediatr*. 1996;85:598–603.
64. Obermeier S, Rudloff S, Pohlentz G, et al. Secretion of ^{13}C-labelled oligosaccharides into human milk and infant's urine after an oral ^{13}Cgalactose

load. *Isotopes Environ Health Stud*. 1999;35:119–125.
65. Rudloff S, Pohlentz G, Borsch C, et al. Urinary excretion of in vivo ^{13}C-labelled milk oligosaccharides in breastfed infants. *Br J Nutr*. 2012;107:957–963.
66. Albrecht S, Schols HA, van Zoeren D, et al. Oligosaccharides in feces of breast- and formula-fed babies. *Carbohydr Res*. 2011;346:2173–2181.
67. Bode L, Rudloff S, Kunz C, et al. Human milk oligosaccharides reduce platelet-neutrophil complex formation leading to a decrease in neutrophil β 2 integrin expression. *J Leukoc Biol*. 2004;76:820–826.
68. Li M, Bauer LL, Chen X, et al. Microbial composition and in vitro fermentation patterns of human milk oligosaccharides and prebiotics differ between formula-fed and sow-reared piglets. *J Nutr*. 2012;142:681–689.
69. Schwartz S, Friedberg I, Ivanov IV, et al. A metagenomic study of diet-dependent interaction between gut microbiota and host in infants reveals differences in immune response. *Genome Biol*. 2012;13:r32.
70. Harmsen HJ, Wildeboer-Veloo AC, Raangs GC, et al. Analysis of intestinal flora development in breast-fed and formula-fed infants by using molecular identification and detection methods. *J Pediatr Gastroenterol Nutr*. 2000;30:61–67.
71. Martín-Sosa S, Martín M, García-Pardo L, Hueso P. Sialyloligosaccharides in human and bovine milk and in infant formulas: variations with the progression of lactation. *J Dairy Sci*. 2003;86:52–59.
72. Newburg DS. Neonatal protection by an innate immune system of human milk consisting of oligosaccharides and glycans. *J Anim Sci*. 2009;87(suppl 13):S26–S34.
73. Rinne MM, Gueimonde M, Kalliomaki M, et al. Similar bifidogenic effects of prebiotic-supplemented partially hydrolyzed infant formula and breastfeeding on infant gut microbiota. *FEMS Immunol Med Microbiol*. 2005;43:59–65.
74. Thomas DW, Greer FR, Committee on Nutrition, Section on Gastroenterology, Hepatology, and Nutrition. Probiotics and prebiotics in pediatrics. *Pediatrics* 2010;126:1217–1231.
75. Agostoni C, Marangoni F, Lammardo AM, et al. Long-chain polyunsaturated fatty acid concentrations in human hind milk are constant throughout twelve months of lactation. *Adv Exp Med Biol*. 2001;501:157–161.
76. Koletzko B, Agostoni C, Bergmann R, et al. Physiological aspects of human milk lipids and implications for infant feeding: a workshop report. *Acta Paediatr*. 2011;100:1405–1415.
77. Zou XQ, Guo Z, Huang JH, et al. Human milk fat globules from different stages of lactation: a lipid composition analysis and microstructure characterization. *J Agric Food Chem*. 2012;60:7158–7167.
78. Minda H, Kovacs A, Funke S, et al. Changes of fatty acid composition of human milk during the first month of lactation: a day-to-day approach in the first week. *Ann Nutr Metab*. 2004;48:202–209.
79. Straarup EM, Lauritzen L, Faerk J, et al. The stereospecific triacylglycerol structures and fatty acid profiles of human milk and infant formulas. *J Pediatr Gastroenterol Nutr*. 2006;42:293–299.
80. Berenhauser AC, Pinheiro do Prado AC, da Silva RC, et al. Fatty acid composition in preterm and term breast milk. *Int J Food Sci Nutr*. 2012;63:318–325.
81. Decsi T, Fekete M, Koletzko B. Plasma lipid and apolipoprotein concentrations in full term infants fed formula supplemented with long-chain polyunsaturated fatty acids and cholesterol. *Eur J Pediatr*. 1997;156:397–400.

82. Kovacs A, Funke S, Marosvolgyi T, et al. Fatty acids in early human milk after preterm and full-term delivery. *J Pediatr Gastroenterol Nutr*. 2005;41:454–459.
83. Decsi T. Nutritional relevance of trans isomeric fatty acids in human milk. *Acta Paediatr*. 2003;92:1369–1371.
84. Mosley EE, Wright AL, McGuire MK, McGuire MA. Trans fatty acids in milk produced by women in the united states. *Am J Clin Nutr*. 2005;82:1292–1297.
85. Innis SM, King DJ. Trans fatty acids in human milk are inversely associated with concentrations of essential all-cis n-6 and n-3 fatty acids and determine trans, but not n-6 and n-3, fatty acids in plasma lipids of breast-fed infants. *Am J Clin Nutr*. 1999;70:383–390.
86. Mosley EE, McGuire MK, Williams JE, McGuire MA. Cis-9, trans-11 conjugated linoleic acid is synthesized from vaccenic acid in lactating women. *J Nutr*. 2006;136:2297–2301.
87. Masters N, McGuire MA, Beerman KA, et al. Maternal supplementation with CLA decreases milk fat in humans. *Lipids*. 2002;37:133–138.
88. Bitman J, Wood DL, Mehta NR, et al. Comparison of the cholesteryl ester composition of human milk from preterm and term mothers. *J Pediatr Gastroenterol Nutr*. 1986;5:780–786.
89. Holmes HC, Snodgrass GJ, Iles RA. Changes in the choline content of human breast milk in the first 3 weeks after birth. *Eur J Pediatr*. 2000;159:198–204.
90. Zeisel SH. The fetal origins of memory: the role of dietary choline in optimal brain development. *J Pediatr*. 2006;149(suppl 5):S131–S136.
91. Laegreid A, Otnaess AB, Fuglesang J. Human and bovine milk: comparison of ganglioside composition and enterotoxin-inhibitory activity. *Pediatr Res*. 1986;20:416–421.
92. Wang B, McVeagh P, Petocz P, Brand-Miller J. Brain ganglioside and glycoprotein sialic acid in breastfed compared with formula-fed infants. *Am J Clin Nutr*. 2003;78:1024–1029.
93. Rudloff S, Kunz C. Protein and nonprotein nitrogen components in human milk, bovine milk, and infant formula: quantitative and qualitative aspects in infant nutrition. *J Pediatr Gastroenterol Nutr*. 1997;24:328–344.
94. Darragh A, Lönnerdal B. Milk | human milk. In: Editor-in-Chief: Fuquay JW, ed. *Encyclopedia of Dairy Sciences*. 2nd ed. San Diego, CA: Academic Press; 2011: pp. 581–590.
95. Ferranti P, Traisci MV, Picariello G, et al. Casein proteolysis in human milk: tracing the pattern of casein breakdown and the formation of potential bioactive peptides. *J Dairy Res*. 2004;71:74–87.
96. Lonnerdal B, Lien EL. Nutritional and physiologic significance of alpha-lactalbumin in infants. *Nutr Rev*. 2003;61:295–305.
97. Adkins Y, Lonnerdal B. Potential host-defense role of a human milk vitamin B-12-binding protein, haptocorrin, in the gastrointestinal tract of breastfed infants, as assessed with porcine haptocorrin in vitro. *Am J Clin Nutr*. 2003;77:1234–1240.
98. Stromqvist M, Falk P, Bergstrom S, et al. Human milk kappa-casein and inhibition of helicobacter pylori adhesion to human gastric mucosa. *J Pediatr Gastroenterol Nutr*. 1995;21:288–296.
99. Schack L, Lange A, Kelsen J, et al. Considerable variation in the concentration of osteopontin in human milk, bovine milk, and infant formulas. *J Dairy Sci*. 2009;92:5378–5385.
100. Tomita M, Bellamy W, Takase M, et al. Potent antibacterial peptides generated by pepsin digestion of bovine lactoferrin. *J Dairy Sci*. 1991;74:4137–4142.
101. Pellegrini A, Thomas U, Bramaz N, et al. Isolation and identification of three

bactericidal domains in the bovine alpha-lactalbumin molecule. *Biochim Biophys Acta*. 1999;1426:439–448.
102. Liepke C, Adermann K, Raida M, et al. Human milk provides peptides highly stimulating the growth of bifidobacteria. *Eur J Biochem*. 2002;269:712–718.
103. Donovan SM. Role of human milk components in gastrointestinal development: current knowledge and future NEEDS. *J Pediatr*. 2006;149(suppl 5):S49–S61.
104. Lubetzky R, Weisman Y, Dollberg S, et al. Parathyroid hormone-related protein in preterm human milk. *Breastfeed Med*. 2010;5:67–69.
105. Ley SH, Hanley AJ, Sermer M, et al. Associations of prenatal metabolic abnormalities with insulin and adiponectin concentrations in human milk. *Am J Clin Nutr*. 2012;95:867–874.
106. Aydin S, Ozkan Y, Erman F, et al. Presence of obestatin in breast milk: relationship among obestatin, ghrelin, and leptin in lactating women. *Nutrition*. 2008;24:689–693.
107. Aydin S. The presence of the peptides apelin, ghrelin and nesfatin-1 in the human breast milk, and the lowering of their levels in patients with gestational diabetes mellitus. *Peptides*. 2010;31:2236–2240.
108. Lacquaniti A, Donato V, Chirico V, et al. Obestatin: an interesting but controversial gut hormone. *Ann Nutr Metab*. 2011;59:193–199.
109. Woo JG, Guerrero ML, Altaye M, et al. Human milk adiponectin is associated with infant growth in two independent cohorts. *Breastfeed Med*. 2009;4:101–109.
110. Newburg DS, Woo JG, Morrow AL. Characteristics and potential functions of human milk adiponectin. *J Pediatr*. 2010;156(suppl 2):S41–S46.
111. Savino F, Liguori SA, Sorrenti M, et al. Breast milk hormones and regulation of glucose homeostasis. *Int J Pediatr*. 2011;2011:803985.
112. Howarth GS, Shoubridge CA. Enhancement of intestinal growth and repair by growth factors. *Curr Opin Pharmacol*. 2001;1:568–574.
113. Houle VM, Park YK, Laswell SC, et al. Investigation of three doses of oral insulin-like growth factor-I on jejunal lactase phlorizin hydrolase activity and gene expression and enterocyte proliferation and migration in piglets. *Pediatr Res*. 2000;48:497–503.
114. Li R, Xia W, Zhang Z, Wu K. S100B protein, brain-derived neurotrophic factor, and glial cell line-derived neurotrophic factor in human milk. *PLoS One*. 2011;6:e21663.
115. Gazzolo D, Bruschettini M, Lituania M, et al. Levels of S100B protein are higher in mature human milk than in colostrum and milk-formulae milks. *Clin Nutr*. 2004;23:23–26.
116. Boesmans W, Gomes P, Janssens J, et al. Brain-derived neurotrophic factor amplifies neurotransmitter responses and promotes synaptic communication in the enteric nervous system. *Gut*. 2008;57:314–322.
117. Bekinschtein P, Cammarota M, Katche C, et al. BDNF is essential to promote persistence of long-term memory storage. *Proc Natl Acad Sci USA*. 2008;105:2711–2716.
118. Palmer DJ, Kelly VC, Smit A, et al. Human colostrum: Identification of minor proteins in the aqueous phase by proteomics. *Proteomics*. 2006;6:2208–2216.
119. Christensen B, Schack L, Kläning E, Sørensen ES. Osteopontin is cleaved at multiple sites close to its integrin-binding motifs in milk and is a novel substrate for plasmin and cathepsin D. *J Biol Chem*. 2010;285:7929–7937.
120. Hernell O, Blackberg L. Digestion of human milk lipids: physiologic significance of sn-2 monoacylglycerol hydrolysis by bile salt-stimulated lipase. *Pediatr Res*. 1982;16:882–885.

121. Lindberg T, Ohlsson K, Westrom B. Protease inhibitors and their relation to protease activity in human milk. *Pediatr Res*. 1982;16:479–483.
122. Donovan SM, Lonnerdal B. Non-protein nitrogen and true protein in infant formulas. *Acta Paediatr Scand*. 1989;78:497–504.
123. Heine W, Tiess M, Wutzke KD. ^{15}N tracer investigations of the physiological availability of urea nitrogen in mother's milk. *Acta Paediatr Scand*. 1986;75:439–443.
124. Allen LH. B vitamins in breast milk: relative importance of maternal status and intake, and effects on infant status and function. *Adv Nutr*. 2012;3:362–369.
125. Olafsdottir AS, Wagner KH, Thorsdottir I, Elmadfa I. Fat-soluble vitamins in the maternal diet, influence of cod liver oil supplementation and impact of the maternal diet on human milk composition. *Ann Nutr Metab*. 2001;45:265–272.
126. Greer FR, Marshall SP, Foley AL, Suttie JW. Improving the vitamin K status of breastfeeding infants with maternal vitamin K supplements. *Pediatrics*. 1997;99:88–92.
127. Sneed SM, Zane C, Thomas MR. The effects of ascorbic acid, vitamin B6, vitamin B12, and folic acid supplementation on the breast milk and maternal nutritional status of low socioeconomic lactating women. *Am J Clin Nutr*. 1981;34:1338–1346.
128. Greer FR. Do breastfed infants need supplemental vitamins? *Pediatr Clin North Am*. 2001;48:415–423.
129. Committee on Fetus and Newborn. Controversies concerning vitamin K and the newborn. *Pediatrics*. 2003;112:191–192.
130. Perrine CG, Sharma AJ, Jefferds MED, et al. Adherence to vitamin D recommendations among US infants. *Pediatrics*. 2010;125:627–632.
131. Lietz G, Mulokozi G, Henry JC, Tomkins AM. Xanthophyll and hydrocarbon carotenoid patterns differ in plasma and breast milk of women supplemented with red palm oil during pregnancy and lactation. *J Nutr*. 2006;136:1821–1827.
132. Canfield LM, Clandinin MT, Davies DP, et al. Multinational study of major breast milk carotenoids of healthy mothers. *Eur J Nutr*. 2003;42:133–141.
133. Macias C, Schweigert FJ. Changes in the concentration of carotenoids, vitamin A, alpha-tocopherol and total lipids in human milk throughout early lactation. *Ann Nutr Metab*. 2001;45:82–85.
134. Chappell JE, Francis T, Clandinin MT. Vitamin A and E content of human milk at early stages of lactation. *Early Hum Dev*. 1985;11:157–167.
135. Dawodu A, Tsang RC. Maternal vitamin D status: effect on milk vitamin D content and vitamin D status of breastfeeding infants. *Adv Nutr*. 2012;3:353–361.
136. Brannon PM, Picciano MF. Vitamin D in pregnancy and lactation in humans. *Annu Rev Nutr*. 2011; 31:89–115.
137. Greer FR, Hollis BW, Cripps DJ, Tsang RC. Effects of maternal ultraviolet B irradiation on vitamin D content of human milk. *J Pediatr*. 1984;105:431–433.
138. Stone WL, LeClair I, Ponder T, et al. Infants discriminate between natural and synthetic vitamin E. *Am J Clin Nutr*. 2003;77:899–906.
139. Niklowitz P, Menke T, Giffei J, Andler W. Coenzyme Q10 in maternal plasma and milk throughout early lactation. *Biofactors*. 2005;25:67–72.
140. Quiles JL, Ochoa JJ, Ramirez-Tortosa M, et al. Coenzyme Q concentration and total antioxidant capacity of human milk at different stages of lactation in mothers of preterm and full-term infants. *Free Radic Res*. 2006;40:199–206.
141. Van Winckel M, De Bruyne R, Van De Velde S, Van Biervliet S. Vitamin K, an update for the paediatrician. *Eur J Pediatr*. 2009;168:127–134.
142. Kojima T, Asoh M, Yamawaki N, et al. Vitamin K concentrations in the maternal

milk of Japanese women. *Acta Paediatr*. 2004;93:457–463.
143. Indyk HE, Woollard DC. Vitamin K in milk and infant formulas: determination and distribution of phylloquinone and menaquinone-4. *Analyst*. 1997;122:465–469.
144. Thijssen HH, Drittij MJ, Vermeer C, Schoffelen E. Menaquinone-4 in breast milk is derived from dietary phylloquinone. *Br J Nutr*. 2002;87:219–226.
145. Kamao M, Tsugawa N, Suhara Y, et al. Quantification of fat-soluble vitamins in human breast milk by liquid chromatography–tandem mass spectrometry. *J Chromatogr B Analyt Technol Biomed Life Sci*. 2007;859:192–200.
146. Greer FR. Vitamin K the basics—What's new? *Early Hum Dev*. 2010;86(suppl 1):S43–S47.
147. Picciano MF. Vitamins in milk. Water-soluble vitamins in human milk. In: Jensen RG, ed. *Handbook of Milk Composition*. San Diego, CA: Academic Press; 1995: pp. 675–688.
148. Daneel-Otterbech S, Davidsson L, Hurrell R. Ascorbic acid supplementation and regular consumption of fresh orange juice increase the ascorbic acid content of human milk: studies in European and African lactating women. *Am J Clin Nutr*. 2005;81:1088–1093.
149. West KD, Kirksey A. Influence of vitamin B6 intake on the content of the vitamin in human milk. *Am J Clin Nutr*. 1976;29:961–969.
150. Sherwood KL, Houghton LA, Tarasuk V, O'Connor DL. One-third of pregnant and lactating women may not be meeting their folate requirements from diet alone based on mandated levels of folic acid fortification. *J Nutr*. 2006;136:2820–2826.
151. Weiss R, Fogelman Y, Bennett M. Severe vitamin B12 deficiency in an infant associated with a maternal deficiency and a strict vegetarian diet. *J Pediatr Hematol Oncol*. 2004;26:270–271.
152. Mock DM, Mock NI, Dankle JA. Secretory patterns of biotin in human milk. *J Nutr*. 1992;122:546–552.
153. Song WO, Chan GM, Wyse BW, Hansen RG. Effect of pantothenic acid status on the content of the vitamin in human milk. *Am J Clin Nutr*. 1984;40:317–324.
154. Parr RM, DeMaeyer EM, Iyengar VG, et al. Minor and trace elements in human milk from Guatemala, Hungary, Nigeria, Philippines, Sweden, and Zaire. Results from a WHO/IAEA joint project. *Biol Trace Elem Res*. 1991;29:51–75.
155. Atkinson S, Alston-Mills B, Lönnerdal B, Neville MC. Major minerals and ionic constituents of human and bovine milks. In: Jensen RG, ed. *Handbook of Milk Composition*. San Diego, CA: Academic Press; 1995: pp. 593–622.
156. Casey CE, Smith A, Zhang PC. Microminerals in human and animal milks. In: Jensen RG, ed. *Handbook of Milk Composition*. San Diego, CA: Academic Press; 1995: pp. 622–674.
157. Domellof M, Lonnerdal B, Dewey KG, et al. Iron, zinc, and copper concentrations in breast milk are independent of maternal mineral status. *Am J Clin Nutr*. 2004;79:111–115.
158. Chierici R, Saccomandi D, Vigi V. Dietary supplements for the lactating mother: influence on the trace element content of milk. *Acta Paediatr Suppl*. 1999;88:7–13.
159. Kelleher SL, Lonnerdal B. Nutrient transfer: mammary gland regulation. *Adv Exp Med Biol*. 2009;639:15–27.
160. Lonnerdal B. Trace element absorption in infants as a foundation to setting upper limits for trace elements in infant formulas. *J Nutr*. 1989;119(suppl 12):1839–1845.
161. Fransson GB, Lonnerdal B. Distribution of trace elements and minerals in human

and cow's milk. *Pediatr Res*. 1983;17:912–915.
162. Krachler M, Li FS, Rossipal E, Irgolic KJ. Changes in the concentrations of trace elements in human milk during lactation. *J Trace Elem Med Biol*. 1998;12:159–176.
163. Cohen RS, McCallie KR. Feeding premature infants: why, when, and what to add to human milk. *JPEN J Parenter Enteral Nutr*. 2012;36(suppl 1):20S–24S.
164. Baker RD, Greer FR, The Committee on Nutrition. Diagnosis and prevention of iron deficiency and iron-deficiency anemia in infants and young children (0–3 years of age). *Pediatrics*. 2010;126:1040–1050.
165. Lonnerdal B. Copper nutrition during infancy and childhood. *Am J Clin Nutr*. 1998;67(suppl 5):1046S–1053S.
166. Esala S, Vuori E, Helle A. Effect of maternal fluorine intake on breast milk fluorine content. *Br J Nutr*. 1982;48:201–204.
167. Azizi F, Smyth P. Breastfeeding and maternal and infant iodine nutrition. *Clin Endocrinol (Oxf)*. 2009;70:803–809.
168. LaKind JS, Amina Wilkins A, Berlin CM, Jr. Environmental chemicals in human milk: a review of levels, infant exposures and health, and guidance for future research. *Toxicol Appl Pharmacol*. 2004;198: 184–208.
169. Landrigan PJ, Sonawane B, Mattison D, et al. Chemical contaminants in breast milk and their impacts on children's health: an overview. *Environ Health Perspect*. 2002;110:A313–A315.
170. Pronczuk J, Akre J, Moy G, Vallenas C. Global perspectives in breast milk contamination: infectious and toxic hazards. *Environ Health Perspect*. 2002;110:A349–A351.
171. Committee on Drugs. The transfer of drugs and other chemicals into human milk. *Pediatrics*. 2001;108:776–789.
172. Davanzo R, Copertino M, De Cunto A, et al. Antidepressant drugs and breastfeeding: a review of the literature. *Breastfeed Med*. 2011;6:89–98.
173. LaKind JS, Berlin CM, Jr, Sjodin A, et al. Do human milk concentrations of persistent organic chemicals really decline during lactation? Chemical concentrations during lactation and milk/serum partitioning. *Environ Health Perspect*. 2009;117:1625–1631.
174. Read JS, Committee on Pediatric AIDS. Human milk, breastfeeding, and transmission of human immunodeficiency virus type 1 in the United States. *Pediatrics*. 2003;112:1196–1205.
175. Little KM, Hu DJ, Dominguez KL. HIV-1 and breastfeeding in the United States. *Adv Exp Med Biol*. 2012;743:261–270.
176. Horvath T, Madi BC, Iuppa IM, et al. Interventions for preventing late postnatal mother-to-child transmission of HIV. *Cochrane Database Syst Rev*. 2009;1:CD006734.
177. Korchazhkina O, Jones E, Czauderna M, Spencer SA. Effects of exclusive formula or breast milk feeding on oxidative stress in healthy preterm infants. *Arch Dis Child*. 2006;91:327–329.

30 Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut

Anne Donnet-Hughes, PhD, Eduardo J. Schiffrin, MD, PhD, and W. Allan Walker, MD

Human breast milk is an undeniably unique, naturally adapted nutritional source for the human infant. It contains macronutrients, micronutrients and biologically active compounds.[1] However, in addition to the nutritive value it imparts, breast milk and breastfeeding are well recognized to protect against gastrointestinal infections, diarrheal diseases,[2] and respiratory infections.[3] Such protective effects have been variously ascribed to the presence in milk of maternal leukocytes,[4] immunoglobulins,[5] growth factors, oligosaccharides, and other immune factors.[6] While some of these components interact directly with bacteria, virus, and other pathogens, others mediate their effects by promoting proliferation, maturation, and activation of neonatal immune cells and by ensuring homeostasis. The net result is an enhanced protection of the immature intestine against colonization and invasion by pathogenic

bacteria.

An emerging concept is that breast milk can influence the neonatal immune system's perception of "danger" or "infectious non-self". To do so, some milk molecules[7] may actually facilitate the intestinal response to specific microbial motifs by activating intracellular signaling pathways such as that of nuclear factor (NF)-κB, a major regulator of the inflammatory response. At first, this might appear to contradict the recognized anti-inflammatory effect of breast milk. However, controlled innate activation is responsible for triggering homeostatic and defensive functions.[8]

The present chapter will elaborate further on how breast milk factors may modulate the neonate's perception of danger signals while avoiding exaggerated inflammation and tissue damage. It will address factors that promote a beneficial microbial flora either directly or indirectly via education of neonatal cells and challenge the theory that milk is strictly anti-inflammatory by proposing novel mechanisms by which the mother already educates her infant in the immediate postnatal period.

THE INNATE MUCOSAL IMMUNE RESPONSE IN THE MATURE INTESTINE

The intestine is the largest immune compartment in the body and, as such, the immunological events taking place in its tissues have a major impact on the overall immune status of the host. A prerequisite for a healthy intestine is an immune system that can detect harmful luminal antigens and pathogens and initiate a protective response, while remaining tolerant to innocuous microbes and dietary antigens as well as endogenous host cells and molecules.[9,10]

A single layer of epithelial cells forms an effective physical barrier that limits interaction between the contents of the intestinal lumen and the lamina propria and lymphoid follicles of the underlying tissue (Figure 30-1). Reinforcement of this barrier is provided by a thick layer of mucin (MUC) glycoproteins that are secreted by goblet cells and cover the epithelium surface. Together with other secretory molecules such as secretory IgA (sIgA) and antimicrobial peptides (AMP), the MUC sensure that the bulk of the gut microbiota is retained in the lumen.[11]

Virtually all human tissues and cells exposed to microbes are able to produce AMP. These molecules have broad-spectrum antibiotic activity against bacteria, viruses, yeast, and fungi and as such, they can shape the

composition of the commensal microbiota and confer protection against environmental and pathogenic organisms.[12] The gut AMP comprise multiple families, including defensins, cathelicidins, and C-type lectins that are secreted by epithelial cells and are retained within the mucus layer.[12] Only low concentrations reach the lumen. Moreover, there is a clear partitioning of the AMP within the mucus layer[8] with C-type lectin products such as RegIIIγ, being concentrated in the poorly colonized inner mucus layer, and defensins being retained in the outer layer.

Most AMP belong to the two structurally distinct classes of defensins and the C-type lectins.[13] α-Defensins are constitutively expressed in human neutrophils and Paneth cells of the small intestinal crypts and do not require bacterial signals for expression. β-defensins, produced predominantly in intestinal epithelial cells (IECs), can be both constitutive and inducible, while the inducible C-type lectin RegIIIγ is up-regulated in the small intestine and colon in response to bacteria.[13] Another third type of AMP, cathelicidin (LL-37/hCAP18), is expressed by neutrophils and mast cells, as well as by differentiated epithelial cells in the colon and stomach and in Brunner's glands of the duodenum.[13]

Paneth cells are considered particularly important in antibacterial defense as they secrete other AMP such as lysozyme and secretory phospholipase A2.[14] The functional significance of these cells and their expression of AMP have been demonstrated in animals which have increased bacterial translocation and susceptibility to infection[15] following ablation of the Paneth cell population.

The intestinal epithelium is not an inert barrier. It monitors changes in the lumen and discriminates the diverse luminal antigens. When needed, it collaborates with professional immune cells in the intraepithelial compartment and in the lamina propria, to mount rapid immune responses to protect the host against harm.[16] Any delay in response to real threat could seriously compromise the host thus, an alert is triggered by all microorganisms. However, the magnitude and precise nature of the response are determined by the characteristics of the microbe.

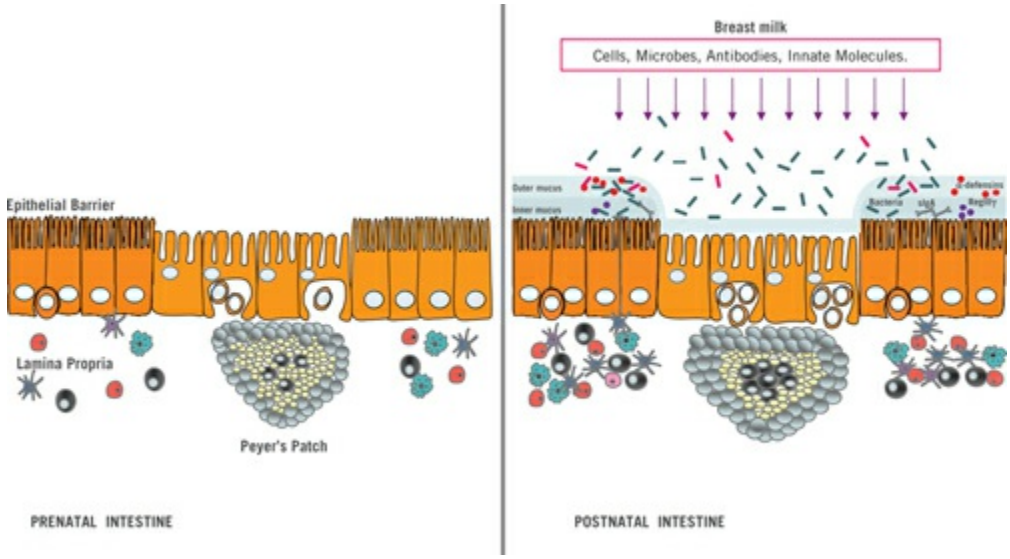


Figure 30-1 The prenatal intestine has all the main elements of the intestinal immune system including the epithelial barrier, immune cells in the lamina propria and organized lymphoid tissues such as Peyer's patches. In the postnatal period, breast milk compensates for deficits in endogenous immune factors and, together with colonizing bacteria, promotes functional maturation of this mucosal immune system. Maturational changes include increased production of mucus and antimicrobial peptides (e.g., defensins, RegIIIγ) into the lumen; increased number of lymphocytes in the intraepithelial compartment; larger and more numerous Peyer's patches with more developed germinal centers and increased numbers of immunoglobulin-producing cells; and increased cellularity in the lamina propria.

Broadly speaking, non-pathogens induce limited responses with a transient innate component[17] that might explain the low grade, physiological inflammation observed in the intestine. In contrast, pathogens induce rapid and more vigorous responses that are accompanied by some tissue damage. Thus, the host reaction can be considered as a stratified response whose primary tier is triggered by most bacteria and is pro-inflammatory, but whose second level of activation involves an assembly of gene products that are defined by the virulence traits of the organism.[18]

The initial innate response depends on the expression of pattern recognition receptors (PRRs) such as toll-like receptors (TLR), Nucleotide-binding oligomerization domain (NOD)-like receptors, and C-

type lectinreceptors.[19] These PRRs empower host cells to recognize and respond appropriately to a number of microbial components and endogenous host proteins.

The transmembrane TLR family of PRR molecules is perhaps the best-studied. Their expression can be constitutive or induced by different signals, and is dependent on the cell type and intestinal location.[19] TLR1, 2, 4, 5, and 6 are localized on the plasma membrane whereas TLR 3, 7, 8, and 9 are localized on the endosome membrane and interact with internalized ligands.[20] In enterocytes and colonocytes, TLR expression is polarized with low expression of TLR2 and 4 on the apical membrane and expression of TLR5 on the basolateral membrane. Activation of specific, spatially regulated TLR may provide a measure of microbial virulence[20] and permit some level of specificity in the primary immune response.

Upon ligand binding to TLRs, their cytoplasmic domain activates a cascade of adapter signaling molecules that include myeloid differentiation marker (MyD88), IL-1 receptor-activated kinase (IRAK), and tumor necrosis factor receptor-associated factor (TRAF)-6. Their sequential recruitment leads to activation of NF-κB and mitogen-activated protein kinases (MAPK) and induction of an inflammatory response.[21]

NOD1 and NOD2 are a structurally distinct family of intracellular, cytosolic PRRs that are expressed in IECs, Paneth cells,[22] in macrophages, T and B cells, and dendritic cells (DCs).[23] As such, these innate immune receptors detect internalized microbial ligands including the Gram-negative peptidoglycan (PGN) derivative γ-C-glutamyl-meso-diaminopimelic acid and muramyldipeptide[24] which bind to NOD1 and NOD2, respectively.

LINKING INNATE AND ADAPTIVE IMMUNITY

Besides alerting the immune system of danger signals in the lumen, the innate system activates and coordinates adaptive immunity, which in turn, controls the composition and localization of components of the commensal microbiota. The secretory immune system, in particular sIgA, works in concert with innate components of the mucosal epithelium to limit colonization of the inner mucus layer (Figure 30-1)[8,11]. The production of sIgA depends on the intervention of innate immune cells and sometimes T helper cells.

Certain types of mucosal immune cells, like macrophages, are able to sample bacteria from the lumen of the gut and clear the bacteria which

have breached the epithelium layer.[25] The TLR-expressing DCs, are environmentally conditioned andare at the heart of the decision-making processes determining the outcome of the primary response in the intestine.[26] These mucosal DCs phagocytose a limited bacterial load from the lumen of the gut, migrate to the mesenteric lymph nodes and promote B and T cell[27] responses that result in the production of sIgA. Thedimeric IgA produced by plasma cells is then transported by the polymeric immunoglobulin receptor across the epithelium, out to the lumen of the gut.[28] In addition, DC supregulate their expression of major histocompatibility complex and costimulatory molecules following TLR engagement and in so doing, acquire the capacity to prime naïve T cells, which then develop into CD4+ T helper 1 (Th1), Th2, Th17, or T regulatory (Treg) effector cells.[26] The CD4+ lineage induced must allow effective defense, of appropriate magnitude, against any pathogen initiating the response yet keep autoimmune mechanisms under control.[29]

The CD4+ Th1/Th2 paradigm has dominated cellular immunity for several years. Th1 cells produce interferon (IFN)-γ, lymphotoxin and tumor necrosis factor(TNF)-α and protect against viruses and intracellular pathogens[29] whereas the Th2 subset secrete the signature cytokines IL-4 IL-5 and Il-13 and promote B cell antibody isotype switching to IgG1, IgA, and IgE. A number of observations suggest that exposure to IL-4 is essential for the priming of Th2 effector cells and that exposure to IL-12 is essential for the priming of Th1cells.[29]

The latest CD4+ T helper lineage to be identified, Th17, produces IL-17A, IL-17F, and IL-22. Th17 effector cells are critical mediators of immunity to bacterial and fungal infections at epithelial barriers and are present early in life.[30] However, Th17 effector cells can also be mediators of inflammatory processes.[31] Peripherally induced Treg development depends on extrinsic tissularsignals.[32] These cells mediate their effects via secretion of IL-10 or transforming growth factor (TGF)-β, and are thought to prevent autoimmune responses by specifically suppressing activation and proliferation of CD4 and CD8 effector T cells. The development of CD4 + effector and regulatory lineages is determined by the transcription factors the cells express and the genes they transcribe.[29] Clearly, DC play an important role in the choice of CD4+ lineage commitment.

Interestingly, recent studies have shown that not all commensals are equal in their capacity to interact with immune cells and to control the development and activation of regulatory or effector immune responses.[33] Segmented filamentous bacteria stimulate Th17 while Clostridia and the

polysaccharide A of *Bacteroides fragilis* stimulate Treg. However, the mechanisms behind these differential responses are, as yet, unknown.[34]

COMPONENTS OF THE NEONATAL INTESTINAL BARRIER

For successful colonization, healthy immune status and a beneficial relationship with the microbiota throughout life, the most elementary immunoregulatory mechanisms must already exist in the neonatal intestine at birth (Figure 30-1). Certainly, some immune instruction already begins prenatally and depends on the thymus for the generation of self-tolerant effector and regulatory T cells. Immature T cells interact with thymic epithelial cells in a vital process that ensures the maturation, differentiation, and selection processes of the mature T cell pool. These prenatal events explain the central features of adaptive immunity and self-tolerance already present at birth.[35] However, while it is important to restrict antigen exposure and actively down-regulate immune reactions *in utero* for fetal survival, this leads to deficits in innate and adaptive immunity in the newborn period. It takes several years before the immune system fully develops but in the immediate to early postnatal period, gestational age, and environmental factors have a profound impact on the maturation process. Indeed, there is clinical evidence that preterm infants are at great risk of infections associated with disorders of the gut ecology[36] and at higher risk of systemic infections due to intracellular pathogens.[37]

There is compelling evidence from animal studies that the symbiotic relationship between the host and the microbiota is critical to the normal development and maturation of the mucosae and their associated lymphoid tissues after birth.[38] Compared to specific pathogen-free animals, germ-free animals have less developed Peyer's patches and mesenteric lymph nodes; reduced numbers of intraepithelial lymphocytes, IgA-secreting cells and Paneth cells[39,40]; limited production of AMP as well as an attenuated mucus layer, in spite of a normal number of goblet cells.[41]

Secondary lymphoid tissues such as Peyer's patches and mesenteric lymph nodes are present prenatally in the developing intestine[40], but postnatal bacterial colonization is thoughtto initiate multiple events that promote the development and functional maturation of the gut associated lymphoid tissue (Figure 30-1).[20,40] For example, the recognition of PGNby NOD1 elicits the production of CCL20 and β-defensins that promote B cell recruitment to the mucosal lymphoid organs and induce expression of

sIgA.[42]

Both the numbers of Paneth cells and the expression of AMP are also developmentally regulated in humans and can already be detected in fetal intestine, at 24 weeks ofgestation.[41,43] However, the newborn intestine has a fewer number of Paneth cells and lower expression of AMP than adult intestine both of which most likely render the infant more susceptible to disease. The observation that Paneth cells of both preterm and term infants with necrotizing enterocolitis (NEC)do not express lysozyme, supports this hypothesis.[44]

Although the innate immune response is not considered to require previous immunological experience to be activated, the homeostatic mechanisms operating in the adult intestine are not expected of the naïve neonatal immune system, particularly if one considers that the same host receptors recognize both pathogens and non-pathogens. Indeed, the increased susceptibility of neonates to infection and certain allergens suggests that the capacity to handle antigenic challenge is not inherent but develops over time and/or through education and experience. In the immediate postnatal period, the cellular composition of the microbiota is highly variable as many organisms vie to colonize the mucosa. This physiological process can trigger an inflammatory response not only in the gut, but also systemically owing to the high permeability of the neonatal intestine. Remarkably, detrimental inflammatory reactions do not normally occur and immune homeostasis prevails.

Certainly, the neonatal intestine has the molecular machinery to recognize microbial conserved molecules. Postnatalintestinal exposure to bacterial product sresults in transcriptional activation in the epithelium and a transient inflammatory status.[41] As in the mature intestine, this is achieved through TLR mediated signal transduction. Effective control of the inflammatory process in the neonatal mucosa is critical for survival and involves different cell types and mechanisms such as low expression of TLR2 and TLR4 on IECs[20] and lamina propria cells and/or limited cellular signaling following engagement with non-pathogens.[24] However, the innate immune response determines the subsequent adaptive response —the choices made during CD4+ Th cell differentiation being of particular importance in the immature neonatal period. In the human newborn, they result in a strong bias toward Th2 and Th17 cell differentiation and a defect in Th1-polarizing cytokine production.[45] The net consequence of which is an increased vulnerability to intracellular pathogens but adequate defense against extracellular organisms, possibly to cope with the

transition from the sterile intrauterine environment to a contaminated extrauterine environment.

When non-pathogens come in contact with innate sentinel cells, transient IEC activation is followed by a tolerogenic, functional hyporesponsiveness. Several mechanisms seem to be involved in the development of this innate immune tolerance[41] which is characterized by down-regulation of TLR2 and TLR4 expression on IECs, and repression of the critical TLR signal transduction molecule, IRAK-1.

DEVELOPMENT OF THE INTESTINAL MICROBIOTA

The mature microbiota is composed of up to 40,000 bacterial species distributed in several phyla, the two most dominant being *Firmicutes* and *Bacteroidetes*. The gastrointestinal immune system has an influence on systemic immune function. Bacterial challenge is a major driving force for immune responses due to the broad diversity of bacterial haplotypes and derivedmolecules.[8,46] Approximately 80% of the body's immune cells are located in the intestine.

Humans start life in the aseptic intrauterine environment but at birth all epithelial surfaces are suddenly exposed to, and then colonized by, environmental microbes. Indeed, microbial colonization of newborn feces can be detected only a few hours after birth.[38] Neonatal intestinal colonization is a potentially hazardous process and yet, it is probably the most important factor influencing the development and maturation of the intestinal immune system and if successful, is fundamental for health throughout life. As discussed above, several simultaneously acting processes protect the neonatal mucosa from a massive inflammatory reaction during this process.[47]

The number and type of bacteria competing for, and colonizing the intestinal habitat, depends on host and environmental factors. During the first days of life, the intestinal microbiota undergoes major compositional changes until a steady state is reached at approximately 1 year of age.[38,48] The mode of delivery, the gestational age, exposure to antibiotics, hospitalization, and length of hospital stay, as well as the type of nutrition have all some bearing on the composition of the neonatal microbiota.

Intestinal colonization seems to be different between pre-term and full-term babies and may play a role in a variety of immune and inflammatory neonatal conditions such as NEC and gut-derived sepsis.[49] Indeed, an

inappropriately energetic inflammatory response towards intestinal microbes seems to be one of the pathogenic mechanisms at play in NEC.

The presence of oxygen and the positive redox potential of the neonatal intestine seem to favor the initial colonization by facultative anaerobes, Staphylococcus, Enterobacteriaceae, and Streptococcus being the most commonly isolated genera in newborn fecal flora soon after birth. Thereafter, strict anaerobes such as Bifidobacterium species and Clostridium species appear,[38] followed by other strict anaerobes such as Bacteroides and additional Clostridium species.[50] A recent study using state-of-the-art molecular methods has challenged this generally accepted view of microbial succession and has shown that total anaerobes already outnumber total facultative bacteria during the first week of life in breast-fed infants.[51] Further studies involving a higher number of infants will be required to clarify this controversial point.

The interplay between milk factors, the microbiota, and the neonatal immune system is complex and difficult to decipher but there is increasing evidence that the events taking place during the early stages of life impact immune function and health throughout the lifespan. The hygiene hypothesis, proposed to explain the rise in atopic diseases, suggested that early in life, microbial antigens interacting with the innate immune system tip adaptive immune responses toward a Th1, and away from a Th2 response.[52] A recent adaptation of this hypothesis proposes that instead of priming immune activation, such microbial stimulation primes immunoregulation via Treg cells and regulatory antigen presenting cells.[46] However, the capacity of the microbiota to promote genetic regulation of immune homeostasis via diverse mechanisms are only now being elucidated.[53]

Molecular studies have also enabled a deeper understanding of how environmental factors such as breast milk, influence neonatal intestinal colonization, and as a consequence, immune development. A recent study showed greater phylogenetic heterogeneity in the microbiota of healthy breast-fed infants compared to that of formula-fed babies.[54] Furthermore, while there is an undeniable preponderance of Bifidobacterium in the stools of breast-fed infants, Enterobacteria, coliforms, Clostridia, and Klebsiella are present at equivalent levels to Bifidobacterium in the stools of formula-fed babies.[55,56] Global intestinal epithelial gene activation also differs between the two populations.[54] It appears that virulence related genes of the microbiome trigger divergent genetic activation of immune defenses depending on whether infants are breast-fed or formula-fed. More

specifically, host genes that mediatebacterially induced reactive oxygen species signaling and epithelial homeostasis were up-regulated in breast-fed infants whereas genes that prime inflammatory mucosal responses were up- regulated in formula-fed babies. The greater incidence of infections, inflammation and allergy in formula-fed infants compared to breast-fed infants suggests that breast milk is a source of protective regulatory factors. Molecular studies will deepen our appreciation of not only the manner in which the microbiota shapes host immunity but the importance of breast milk and specific milk factors in determining mucosal gene activation during the colonization process.

BREAST MILK FACTORS MODULATING THE RESPONSE TO MICROBES AND SHAPING THE INTESTINAL MICROBIOTA

As there is a delay in stabilization of the intestinal microbiota and in the complete maturation of the immune system, much of the immunologic benefits of breast milk have been attributed to the passive protection that it provides.[57] It is apparent that although breast milk supplies a combination of protective factors, functional redundancy is not rare. Not only do several factors exhibit the same biological function, a single molecule can have a range of biological activities. This complexity and the change in the concentration of specific components as lactation proceeds are testimony to a dynamic biological fluid, the complete function of which has still to be fully appreciated.

Despite a normal, integrated luminal barrier, some components of the intestinal microbiota and some pathogens circumvent host defense mechanisms and enterinto contact with epithelial cells. Such encounters are likely to occur more frequently in an immature intestine, thus additional protective mechanisms are required to ensure epithelial barrier integrity and preservation of mucosal homeostasis. To compensate for developmental delays and poor endogenous production of protective molecules in the neonatal intestine, soluble factors in breast milk prevent microbial antigens, pathogenic or not, from reaching the cells in host tissues. Moreover, some factors through their bacteriostatic or bactericidal actions can define the nature of microbial antigens that succeed in interacting with the intestinal immune system (Table 30-1). Examples are milk oligosaccharides, glycans, MUCs[58] and AMP including defensins, cathelicidins, lysozyme, and lactoferrin and its peptides.[59]

TABLE 30-1 Examples of Human Milk Factors That Define the Intestinal Microbiota	
Oligosaccharides and glycoconjugates	Receptor analogues that inhibit binding of pathogens and their toxins to epithelial cells Bifidogenic effect
Mucin 1	Inhibits binding of S-fimbriated *E. coli* and rotavirus to epithelial cells
Lactadherin	Inhibits binding of rotavirus to epithelial cells
Defensins and cathelicidins	Bacterial killing via membrane disruption
Lactoferrin	Bacterial killing by lactoferricin peptide Bacteriostatic activity by binding ferric iron. Bifidogenic effect
Lysozyme	Bacterial killing via membrane disruption
Secretory IgA	Inhibits binding of pathogens to the intestinal mucosa. Facilitates biofilm formation by the normal flora. Bifidogenic effect of secretory component.
α-Lactalbumin	Anti-bacterial peptides
Fatty acids	Anti-bacterial activity

Trefoil factors (TFF), secretory products of goblet cells, also contribute to epithelial surface integrity and improve repair of damaged epithelium through a process called epithelial restitution.[60] It is not known if the neonatal intestine is able to produce TFF in sufficient quantities, but TFF are present in human milk[61] and while their contribution to neonatal intestinal development and protection against disease remain unknown, they have been shown experimentally to promote β-defensin production and down regulate the inflammatory response in human IECs.[62]

Closer scrutiny of the interplay among specific milk molecules, microbes, and the mucosal immune system is beginning to unravel some of the intricacies of breast milk protection. It is becoming apparent that exogenous factors in milk play a regulatory, or even an educational, role in the underlying immune processes. Simplistically, soluble factors in breast milk can interact directly with microbial cells to limit subsequent contact with the intestinal epithelium. However, the most recent research now suggests that breast milk permits, and indeed encourages, interaction between particular microbial motifs and the intestinal immune system[7] but curbs any energetic immunereaction.[63] In so doing, breast milk promotes a symbiotic relationship between the neonatal immune system and the various bacterial communities that constitute the permanent microbiota of

the gut.

Prebiotic and Antimicrobial Activities of Human Milk Oligosaccharides and Glycoconjugates

Human milk oligosaccharides (HMOs), a heterogeneous group of unconjugated compounds, are the third most abundant solidcomponent in breast milk after fat and lactose. Colostrum levels of HMOs can reach about 20 g/L but their concentration decreases as lactation proceeds.[64]

HMOs are synthesized by specific mammary gland glycosyltransferases, which sequentially add fucose, galactose, *N*-acetylglucosamine, and sialic acid to lactose.[65] HMOs are not nutrients for the neonate since infants lack the enzymes necessary for their digestion. Instead, they can pass undigested into the colon where they not only create an acidic environment that is antagonistic for pathogens, they stimulate the growth and metabolism of symbiotic bacteria, such as *Bifidobacterium*, that then compete with pathogens for occupation of intestinal niches.[66]

HMOs provide further protection through molecular mimicry. Pathogens such as *Streptococcus pneumoniae*, *Escherichia coli*, C*ampylobacter jejuni*, *Listeria monocytogenes*, *Shigella*, *Salmonella*, and *Vibrio cholera*, initiate the infectious process by adhering to oligosaccharides expressed on epithelial cell membranes.[67] In vitro, epidemiological and clinical studies suggest that by resembling structures on the mucosal cells, HMOs serve as decoy receptors for pathogen adhesion factors and thereby prevent pathogen binding to mucosal surfaces.[68] HMOs may also mediate their protective effects by modifying the composition of the glycocalyx or the expression of glycoproteins on the epithelial surface,[69] as well as modifying bacterial genes involved in host-microbe interactions.[70] Certainly, the important concentrations of 1, 2-linked fucosylated glycans in human milk have been shown to protect breast-fed infants against diarrhea caused by campylobacter, caliciviruses, and the stable toxin of enterotoxigenic *E. coli*, as well as against other etiologies of moderate-to-severe diarrhea.[71]

While HMOs are bifidogenic, it is not known if they have a more global influence on the composition of the neonatal microbiota. Nevertheless, the observation that breast-fed infants harbor less P-fimbriated *E. coli* than type 1 fimbriated *E. coli* and are less likely to suffer from urinary infections,[72] would suggest that a more global effect on the microbiota is indeed plausible.

There is an increasing body of evidence that HMOs can also modify

the activity of epithelial and immune cells and as such, can exert immunomodulatory properties at the mucosal or systemic levels.[67] Many of the observations have been made *in vitro* and the clinical relevance remains speculative, nevertheless, HMOs promote *Bifidobacterium*-mediated expression of tight junction proteins and anti-inflammatory cytokines by IECsas well asencourage lymphocyte maturation and activation.[73] Furthermore, by interacting with immunomodulatory lectins such as galectins, which influence T helper cell development and inflammation[74] and with selectins, which regulate leukocyte migration and the inflammatory response,[67] HMOs may exert additional, currently unknown, indirect effects on the immune status of the neonate. Irrespective of the mechanism, studies in animal models support the contention that HMOs are important breast milk factors in the prevention of NEC.[75]

Milk also contains several glycoconjugates which confer protection on the suckling neonate. The milk fat globule membrane glycoproteins, MUC-1 and lactadherin, inhibit rotavirus replication and prevent experimental gastroenteritis through their sialic acid residues.[76] Several other glycoconjugates of human milk are now known and include the ganglioside GM1, which binds to cholera toxin and labile toxins of *E. coli* and *C. jejuni*; the neutral glycolipids globotriaosylceramide, which binds to toxins of *Shigella* and *E. coli*; a mannosylated glycoprotein that inhibits binding of some strains of *E.coli* to human intestinal epithelium; and finally, glycosaminoglycans, which inhibit binding of the outer membrane protein of the AIDS virus to CD4 T cells.[77]

Antimicrobial Proteins and Peptides

The presence of an exogenous source of AMP in human breast milk may compensate for the deficit in Paneth cells and AMP observed in the immature neonatal intestine. The presence of multiple molecules with antimicrobial activity has been reported in human milk including lysozyme,[78] defensins[79] and cathelicidin.[80] They are produced by mammary gland epithelial cells and milk cells,[81] are present in milk in significant concentrations throughout lactation, and have demonstrable antimicrobial activity against a broad range of microorganisms.

Lactoferrin is a major protein of breast milk. It is present throughout lactation and has multiple biological effects.[65] By virtue of its iron binding properties, lactoferrin has been proposed to play a role in iron uptake by the intestinal mucosa and to act as a bacteriostatic agent by withholding iron from iron-requiring bacteria.[82] An antimicrobial effect of lactoferrin,

independent of the degree of iron saturation, has also been described and may be due to the generation of the bactericidal peptide, lactoferricin, during digestion.[83] However, lactoferrin is relatively stable in the neonatal intestine and can be found intact or partially hydrolyzed in the feces of suckling but not formula-fed infants.[84] Interestingly, lactoferrin[85] and its peptides have been shown to promote growth of lactic acid bacteria such as *Bifidobacterium* and *Lactobacillus*. There have been some promising clinical studies which have also shown a beneficial effect of recombinant human lactoferrin in the prevention of NEC.[86] Moreover the use of bovine milk lactoferrinseems to prevent late-onset sepsis and NEC in preterm infants when given in combination with a probiotic microorganism.[87]

Immunoglobulins

Although intestinal lymphoid structures are present at birth, immunoglobulin-producing B cells are not fully operational for several weeks.[40] This delay in B cell activation and maturation into antibody–producing plasma cells causes a transient deficit in secretory IgA (sIgA) production and reduced protection of the neonatal intestinal mucosa.[88] Microbial colonization of neonatal tissues leads to B cell activation by 1 month postpartum. The B lymphoblaststhen mature into plasma cells that produce polymeric IgM and, most frequently, dimeric IgA, which are transported across the intestinal epithelium by polymeric immunoglobulin receptors.[89]

In the early newborn period, breast milk-derived antibodies compensate for the deficit in neonatal production. These milk antibodies are produced in the mammary gland by the B lymphoblasts that have trafficked from maternal gastrointestinal and bronchial lymphoid tissues to the mammary gland.[90] The repertoire of milk antibody specificity is governed by environmental antigens, including the infectious agents to which the mother has been exposed,[91] and can be directed against both enteric and respiratory pathogens, including viruses. Concentrations of sIgA are higher in the feces of breast-fed infants than in those of formula-fed infants[92] and may reflect the capacity of milk sIgA to resist gastrointestinal digestion.

The dominant immunoglobulin in human milk is sIgA, which protects neonatal mucosal surfaces by a mechanism called “immune exclusion” This involves agglutination and immobilization of different pathogens,[93] including viruses, bacteria, and parasites as well as neutralization of endotoxin and other bacterial products. In so doing, sIgA prevents their

interaction with the intestinal epithelium and facilitates their elimination by peristalsis, without triggering inflammatory tissue damage.[28,91,93] Although, many bacteria in the intestinal microbiota are coated with sIgA, they are not permanently eliminated from the intestine.[94] The reasons for this apparent inconsistency are not known but may be related to the type and specificity of the antibody.

Paradoxically, sIgA binds to M cells of the intestinal Peyer's patches and then to underlying DC and, as a consequence, facilitates transport of microbes and antigens to the mucosal immune system.[95] It is tempting to speculate that milk sIgA uses this mechanism to educate the neonatal immune system in the management of commensal microbiota and pathogens. Certainly, physiological transport of microbesfrom the gut lumen to the mesenteric nodes has been described as an important process in immune homeostasis.[27]

EDUCATION OF THE MUCOSAL IMMUNE SYSTEM BY FACTORS IN BREAST MILK

Protection and education of offspring are integral parts of evolutionary success, and breast milk composition has adapted to meet these requirements. However, while the passive protection afforded by breast milk is well recognized, the manner by which it educates the neonatal immune system is less obvious. There is certainly accumulating evidence to suggest that the effects of breast milk extend beyond the neonatal period and that there is a more intimate association between maternally derived milk factors and the neonatal immune system, beyond that required to prevent pathogen colonization and infection. The mechanisms underlying the long-lasting benefits of breast milk are likely to be multifarious. At least two different and partially overlapping processes are probably involved:

1. First, breast milk factors, in shaping the composition of the neonatal microbiota, indirectly influence immune system development and response.[51,96] Breast milk contains a myriad of factors that, qualitatively or quantitatively, may modulate how neonatal cells perceive microbes and/or react to microbial contact. Examples are immunoglobulins, oligosaccharides, glycoproteins and glycolipids, which interfere with bacterial binding to epithelial cells, and AMP, which limit bacterial challenge and encourage the preponderance of particular

microbial species. In addition, breast milk-derived bacteria may be a preferential inoculum to build up the neonatal microbiota.[96,97]

2. Second, breast milk directly "educates" the neonatal immune system to react with appropriate innate or adaptive immune responses upon microbial or antigenic challenge. It can modulate the homeostatic balance between tolerogenic and effector immune responses and influence the development of pathologic diseases later in life.[5]

A crucial role for breast milk in the induction of immunological tolerance to exogenous and endogenous antigens and in the prevention of pathological conditions later in life, is gaining credence. The tolerogeniccapacity of breast milk is mediated by different mechanisms including (a) the transmission of antigens from the maternal environment to the suckling newborn via breast milk and, (b) the presence of immunomodulatory factors in breast milk.[63] Indeed, simultaneous exposure to antigens potentially involved in human pathological processes, together with immunomodulatory factors in the milk,[6,63] may be explain the protection breast milk provides against immune-mediated diseases such as allergy, Type 1 diabetes mellitus and celiac disease.[63,98] Furthermore, it is feasible that exogenous and maternal self- antigens in breast milk may prevent such diseases through antigen-specific induction of immunological tolerance. Certainly, maternal exposure to environmental antigens leads to the formation of immune complexes which are transferred to the infant via the milk and then educate the neonatal immune system to remain tolerant to the antigen.[99]

IMMUNE EDUCATION BY BREAST MILK DURING NEONATAL INTESTINAL COLONIZATION

Through their immunomodulatory activity, milk growth factors and cytokines may affect neonatal immune development, but arguably, the finest education is achieved through experience. To this end, milk limits, but does not completely veto, bacterial interaction with the mucosal surface and the intestinal immune system. Indeed, such interactions are important for postnatal maturation of intestinal immunity[33] and any delay to this maturation, such as that caused by reduced microbial stimulation,

may increase susceptibility to allergic disease.[46] The following sections discuss how milk may modulate microbial interactions with receptors on host cells, thereby encouraging cellular activation to particular microbial motifs but prevent crude, exaggerated inflammatory responses that may occur more readily in immature cells.

PRR–Bacteria Interactions in the Presence of Breast Milk

Lipopolysaccharide of gram-negative bacteria is a potent stimulator of the innate immune response, but it requires several host humoral and cell-surface proteins to mediate its effects. These include the acute-phase lipopolysaccharide (LPS)-binding protein (LBP), and the glycoproteins CD14, TLR4, and myeloid differentiation protein (MD)–2.[100] Cellular activation begins when LPS binds to LBP, which subsequently accelerates LPS binding to CD14 on the cell surface. LPS is then transferred from the CD14–LBP complex to a cell surface TLR4–MD-2 complex, which transduces intracellular signaling. In contrast, cellular activation by the lipoteichoic acid (LTA) of gram-positive bacteria involves TLR2-mediated signals.[101]

TABLE 30-2 Examples of Human Milk Factors That Modulate Responses to Bacteria

Soluble CD14	Transfers lipopolysaccharide and lipoteichoic acid to TLR complexes or to lipoproteins Modulates TLR signaling. Pro-inflammatory
Soluble TLR2	Modulates TLR signaling. Anti-inflammatory
Lactoferrin	Modulates TLR signaling Anti-inflammatory
Interleukin-10	Increases expression of CD14 Anti-inflammatory
Transforming Growth Factor-β	Decreases expression of CD14 and TLR4 Anti-inflammatory
Osteoprotegerin	Anti-inflammatory
β-Defensin 2	Activates dendritic cells and induces Th1 immune responses via TLR4

Human breast milk contains high levels of soluble forms of CD14 (sCD14)[7,102] and TLR2[103] which, like lactoferrin, may modulate TLR

signaling (Table 30-2). Depending on its concentration and environment, sCD14 can activate or inhibit responses to LPS by respectively transferring LPS to, or diverting LPS away from,[104] membrane CD14 (mCD14). Furthermore, sCD14 can transfer LPS directly to the TLR4–MD2 receptor complex in cells that lack mCD14, such as endothelial and epithelial cells[105].

It is feasible, therefore, that the ambivalent nature of sCD14 allows physiological activation of cells for homeostasis is but prevents exaggerated inflammation during infection or sepsis. Further assurance may be provided by additional milk proteins that bind sCD14 and lessen sCD14–LPS or sCD14-bacteriallipoprotein(BLP)-mediated cell activation. An example of the former is the AMP lactoferr in that binds specifically, and with high affinity, to the lipid A region of LPS[106] as well as to sCD14 and the sCD14–LPS complex.[107] The more recently discovered sTLR2 also decreases in concentration over the lactation period and, by binding to sCD14 in milk, has been postulated to moderate responses to bacterial lipopeptide and gram-positive bacteria.[103]

Thus, some receptors in cell-surface TLR complexes exist as soluble forms in milk and act as decoy receptors for microbial motifs (Table 30-2). Still other proteins found in milk, such as IL-10 and TGF-β, at constant levels throughout the first months of lactation, may modify TLR-mediated responses by regulating the expression of the various proteins in the TLR receptor complex. In this respect, IL-10 increases the expression of CD14 on monocytes but does not alter their expression of TLR-4.[108] In contrast, TGF-β down regulates the expression of CD14 on macrophages.[109] Importantly, TGF-β null-mice have increased TLR4 expression and inflammatory lesions in multiple tissues.[110]

Cellular Reactivity to Bacteria in the Presence of Breast Milk

The predominant production of Th2 cytokines in fetal and neonatal life is a key element in depressing innate immune responses in the newborn.[52] This deficiency in response results in impaired LPS-induced production of the pro-inflammatory, Th1-polarizing cytokines TNF-α, IFN-γ,[111] and IL-12 by neonatal mononuclear cells. An increased ratio of IL-6/TNF-α further polarizes toward a Th2 response.[112] It follows that low expression of TLR2 and TLR4 in the neonatal intestinal epithelium together with no expression of MD2[113] may explain hyporeactivity to components of the

microbiota.[20] In contrast to neonatal mononuclear cells and adult IECs, human fetal enterocytes express TLR2 and TLR4 and are hyper-responsive to LPS.[114] This hyper-reactivity may also depend on a lower expression of the inhibitory intracellular molecule I-κBα, which binds NF-κB and retains it in the cytosol.[115]

The interaction between luminal microbes and highly expressed TLR4 on neonatal IECs may represent a risk factor for the development of NEC.[114] Thus, increased TLR expression on human fetal enterocytes could explain the increased susceptibility of premature infants to NEC. Nevertheless, a transient activation of IECs, with IκB-α phosphorylation and subsequent NF-κB nuclear translocation, may be required for subsequent postnatal tolerance toendotoxin.[116]. Indeed, continuous activation in the presence of bacterial ligands is then thought to down-regulate TLR expression on IECs,[113] perhaps via shedding of the receptors.[103]

Remarkably, the presence of soluble PRRs and their partners in breast milk may participate in both the transient activation of innate immunity and in its subsequent down regulation. Certainly, milk sCD14 mediates the production of pro-inflammatory cytokines IL-8 and TNF-α, and of the chemokine ENA-78, by IECs exposed to LPS,[7] as well as the production of IL-8 in monocytes and DCs exposed to LPS.[117] At first glance, it is difficult to reconcile why high levels of sCD14 in breast milk, in the presence of an important bacterial inoculum, do not lead to excessive inflammation in the intestine. One possible explanation is that strong activation leads to shedding of the TLR involved in the response.[103] However, it is also possible that other proteins in milk refine the response by interacting with sCD14 or sCD14 complexes as described earlier. An example is lactoferrin, which in addition to downregulating LPS-induced production of pro-inflammatory cytokines via NF-κB, also inhibits sCD14–LPS-induced expression of IL-8 by human endothelial cells as well as their increased expression of adhesion molecules, E-selectin and intercellular adhesion molecule (ICAM)-1.[118]

It is also noteworthy that the TLR2 ligand LTA does not stimulate pro-inflammatory cytokine production by IECs in the presence of milk, but rather inhibits sCD14-mediated LPS effects.[119] This observation suggests that gram-positive bacteria, via their LTA, may moderate cellular reactivity to LPS. However, more recent work has shown that sTLR2 in human milk can bind sCD14 and that sTLR2 can inhibit monocyte production of IL-8 and TNF-α in response to synthetic bacterial

lipopeptide.[103] It is now known that multiple rounds of NF-κB activation are necessary in a first instance for immune defense and thereafter, for resolution of inflammation and tissue repair, and that the latter is achieved through IKK-α inhibition of NF-κB.[120] It is becoming apparent that breast milk has evolved to provide molecules that act at several levels of this process. First, it permits the neonate to mount a limited response to the microbial "danger" signal by promoting the production of pro-inflammatory cytokines. Thereafter, it may activate a second set of genes that are be part of a negative feedback mechanism.[117]

Perhaps as a precautionary measure, breast milk already provides anti-inflammatory factors and AMP whose expression in the neonatal mucosa would otherwise require additional NF-κB activation (Table 30-2).[79] In agreement with this, a certain ambivalence is seen with some milk factors such as TGF-β,[121] sCD14[7] and the antimicrobial factors lactoferrin and lysozyme, which have the capacity to activate or inhibit NF-κB activation depending on the concentration and the cellular environment.[82]

The TRAF6 signal transducer of the TLR/IL-1R superfamily is also important for signaling of the receptor activator of NF-κB (RANK) and its ligand (RANKL),[122] both of which are key regulators of lymph node formation and T cell–DC crosstalk. Osteoprotegerin (OPG), a natural, soluble decoy receptor for RANKL, is expressed by human mammary gland and breast milk cells, is present in human breast milk for at least the first six months postpartum and can be detected in the serum of neonatal animals after gavage. OPG is also expressed by IECs and is upregulated in the tissues of inflammatory bowel disease patients.[123]

Maternal milk also inhibits IL-1β–induced NF-κB signaling in human IECs in vitro, perhaps due to the presence of the IL-1 receptor antagonist (IL-1ra) whose levels persist throughoutlactation.[124] Certainly, both human milk and an infant formula supplemented with humanIL-1ra, have anti-inflammatory effects on chemically induced colitis in rats.[125] Soluble TNF-α receptor[124] and IL-10[126] in milk may behave in a similar manner.

As well as being a fuel for bacteria, HMO may also keep the reactivity of the neonatal immune response in check. They can interfere in protein–carbohydrate interactions, such as those mediated by selectins, and can inhibit formation of platelet–neutrophil complexes and the subsequent transmigration of neutrophils through activated endothelium at sites of inflammation.[67]

Education of the Neonatal Immune System via Microbes

in Milk

A variety of viable leukocytes are found in human milk but their concentration declines as lactation proceeds.[127] These leukocytes are cells that have migrated from the maternal gut- and bronchial-associated lymphoid tissue to the lactating mammary gland via the lymphatics and the circulation.[128] The cell populations include granulocytes, macrophages, and lymphocytes, predominantly T cells. A significant proportion of the latter being $CD8^+$ effector cells that may be important in the control of viral passage from mother to infant.[4]

Ingested milk leukocytes may survive gastrointestinal passage and a small number may be taken up by the neonatal intestinal barrier and migrate to the mesenteric lymph nodes.[127] Macrophages and granulocytes are considered to be the predominant cell types. However, work demonstrating that $CD14^+$ milk mononuclear cells express HLA-DR, CD86, CD83, and DC-specific ICAM-3–grabbing non integrin suggests that these cells may, in fact, be partially differentiated DCs.[129] These cells are probably the source of some of the soluble factors in milk such as sCD14 and OPG, as well as several cytokines and chemokines.[130] Furthermore, they contain ingested IgA, which they may release upon encountering bacteria in the gut.[128]

In addition to maternal eukaryotic cells, there is accumulating evidence that bacteria of maternal origin are also transmitted to the infant via colostrum and breast milk.[1,96,97] Previously, most studies on the microbiology of human milk addressed the transmission of pathogens or contaminating commensals in samples meant for milk banks.[131] Recently, it has been suggested that breast milk is not sterile, even when collected aseptically. Rather, it is a natural source of diverse microorganisms that may be beneficial to the health of the suckling infant. Indeed, *Staphylococcus*, *Streptococcus*, and *Lactobacillus* species are readily identified. However, culture-independent techniques reveal a rich biodiversity of microbes also comprising *Enterococcus*, *Bifidobacterium*, *Corynebacterium*, *Propionibacterium*, *Peptostreptococcus*, and *Escherichia* species, amongst others.[96] The majority of milk organisms arise from the mother's skin or the infant's mouth. However, it has been postulated that cells migrating from the maternal gut-associated lymphoid tissue to the lactating breast, capture components of the luminal microbiota prior to their departure and transfer them to the suckling infant via breast milk.[132]

The relevance of the milk microbiome to infant health is not, as yet,

known but it may represent an inoculum for the development of the neonatal microbiota. Certainly, some species are common to a mother's breast milk and her infant feces.[96,133] Moreover, the milk microbiome may educate the neonatal immune system and confer protection. Indeed, there have been studies to suggest that *Bifidobacterium* strains isolated from human milk have probiotic properties such as antimicrobial and anti-inflammatory activities, anti-allergic effects and immunomodulatory activity and that they can improve gastrointestinal function.[134]

Together, these studies indicate that milk bacteria are not contaminants and suggest that the milk microbiome is influenced by several factors that significantly skew its composition. As breast milk bacteria are among the first microbes entering the human body, current data emphasize the necessity to understand the biological role that the milk microbiome may play in human health.[135]

Immune Education by Milk TGF-β

TGF-β is a multifunctional cytokine that inhibits the development of exaggerated responses to self or non-harmful antigens without compromising immune responses to pathogens.[121] It controls the initiation and resolution of inflammation through the regulation and survival of multiple cell types and together with IL-2, induces the differentiation of peripheral Treg from naïve CD4+ T cells. In addition, TGF-β regulates inflammation, oral tolerance, induction of IgA production and barrier function in the intestine.[121,136] There are several studies to suggest that exogenous TGF-β in milk may also have these effects in the host.

There are three isoforms of TGF-β: TGF-β1, TGF-β2, and TGF-β3 but TGF-β2 is the most abundant isoform in breast milk. It has been demonstrated that the intestine of premature human infants, particularly those experiencing NEC, express higher levels of pro-inflammatory cytokines and lower expression of TGF-β2 when compared to the intestine of full-term infants.[137] The same group also showed that orally administered recombinant TGF-β2 provides protection against inflammatory mucosal injury in an animal model of NEC. Together, these results support the hypothesis that an exogenous source of TGF-β in breast milk may be important for intestinal homeostasis in the neonate and may offer protection against immune-mediated disease. Further support for this is provided by a recent review that has evaluated animal studies addressing the capacity of milk TGF-β to influence immune responses in various experimental models.[138] Various studies suggest that milk TGF-β survives

passage through the digestive system and can be taken up by the neonatal intestine to exert effects in extraintestinal tissues. It can protect against endotoxic shock and exaggerated inflammatory responses. Furthermore, it induces oral tolerance in the host. It has been shown that airborne allergens are transferred from the mother to the suckling neonate[139] and that simultaneous exposure of neonatal animals to the allergen and TGF-β in the milk leads to oral tolerance induction and antigen-specific protection from allergic airway disease. This protective effect is mediated by regulatory CD4+ T lymphocytes and is dependent on TGF-β1 signaling in T cells. Together, the various studies suggest a role for milk TGF-β in influencing neonatal immune responses. Indeed, a correlation has been observed between TGF-β levels in breast milk and serum IgA levels in human newborn infants.[140]

Breast Milk Effects on Adaptive Immunity

As mentioned above neonates are susceptible to microbial infections and mount poor adaptive immune responses. The reasons for this are multifactorial and may include a lack of memory B and T cells and reduced T cell responses,[141] reduced numbers and maturation of DCs,[142] a high proportion of Treg cells but with naïve phenotype, and a bias toward Th2 immune responses.[143] However, specific TLR stimulation can overcome some of these defects[142] and may inhibit the developmentof Th2 responses.[144]

Breast-fed infants are less prone to develop asthma, atopic dermatitis, eczema, and allergy than formula-fed infants. Some studies have reported a correlation between the development of these diseases and the concentration of milk molecules sCD14, TGF-β, and sIgA.[145] Mechanistic studies certainly support a regulatory role for milk molecules in adaptive immunity and in correcting imbalances in Th17, Th1, Th2 and Treg responses. For example, sCD14 mediates LPS-induced TNF-α, IL-6, IL-8, and IL-12 release by DCs, as well as their expression of costimulatorymolecules.[146] Furthermore, it acts directly on activated human T cells to inhibit IL-2 production and cell proliferation, as well as the production of another Th1-like cytokine, IFN-β, and a Th2-like cytokine, IL-4.[147] Direct interaction of sCD14 with B cells, resulting in higher levels of IgG1 and marked inhibition of IgE production, has also been observed.[148] Consistent with these observations, administration of milk sCD14 to neonatal mice enhances antibody secretion.[102]

Several *in vitro* and *in vivo* studies have now demonstrated a role for

lactoferrin in T cell differentiation and the capacity of orally administered lactoferrin to correct the Th1/Th2 cytokine imbalance in both Th1 and Th2 models of disease.[149]

Studies in OPG null mice have shown that OPG regulates B cell maturation and the development ofefficient antibody responses.[150] It is also note worthy that Peyer's patch DCs express RANK and that treatment with antagonists of RANKL enhances the induction of tolerogenic immuneresponses.[151] It is feasible therefore that the high levels of OPG in milk may mediate similar effects.

HMO, specifically the Lewis X component, can interact with DCs via the cell-surface molecule dendriticcell-specific intercellular adhesion moleculegrabbingnon-integrin (DC-SIGN).[152] DC-SIGN is implicated in various functions, mainly related to pathogen recognition and antigen processing and presentation to CD4+lymphocytes, and can function both as an adhesion receptor and as a phagocyticpathogen-recognition receptor, similar to theTLRs.[153] Although likely, more studies are required to determine whether HMO interference with the interaction of DC-CD4+ cell interactions render the newborn host tolerant to components of the microbiota.

Other Protective Factors in Milk

Additional immunomodulatory factors in milk may regulate innate and/or adaptive immune responses. These include the antimicrobial β-defensin which induces cytokines and chemokines, is chemotactic for DCs, and uses TLR4 to activate DCs and induce Th1 immune responses.[154] Numerous cytokines and chemokines, such as IL-1β, IL-6, and TNF-α, monocyte colony stimulating factor, monocyte chemotactic protein 1, IL-8, and RANTES, as well as fatty acids, have also been described.[124,126] α-Lactalbumin is a major milk protein in all mammals. Its digestion provides a source of biologically important amino acids, AMP, and an immunomodulatory peptide that enhances macrophage phagocytic activity. Milk is also a rich source of hormones and growth factors that may aid nutrient transport and the growth and differentiation of neonatal tissues.[6]

CONCLUSION

Breast milk provides a well-recognized passive protection against pathogens in the neonatal period. However, recently discovered functions suggest that protection extends beyond the newborn period. The cocktail

of biologically active factors that milk provides, targets neonatal innate and adaptive immune responses at multiple levels. Milk's primordial task is to alert the innate arm of potential danger. To do so, it supplies molecules that actively promote a pro-inflammatory primary response upon exposure to antigens or microbial components. However, as a pivotal link between innate and adaptive immunity, it ultimately instructs the neonatal immune system to remain tolerant to nonthreatening antigens and components of the commensal microbiota, but to eliminate potential pathogens. To achieve this, it already provides a variety of anti-inflammatory factors that would otherwise imply further activation of neonatal cells. These additional factors fine-tune the type and level of reactivity. Differences between breast-fed and formula-fed infants suggest that in some infant formulas, certain elements needed for the control of these processes are lacking and the infant is more susceptible to antigenic challenge.

REFERENCES

1. Tackoen M. Breast milk: its nutritional composition and functional properties. *Rev Med Brux*. 2012;33:309–317.
2. Gribble, KD. Mechanisms behind breastmilk's protection against, and artificial baby milk's facilitation of, diarrhoeal illness. *Breastfeed Rev*. 2011;19:19–26.
3. Tarrant M, Kwok MK, Lam TH, et al. Breast-feeding and childhood hospitalizations for infections. *Epidemiology*. 2010;21:847–854.
4. Peroni DG, Chirumbolo S, Veneri D, et al. Colostrum-derived B and T cells as an extra-lymphoid compartment of effector cell populations in humans. *J Matern Fetal Neonatal Med*. 2013;26:137–142.
5. Brandtzaeg P. The gut as communicator between environment and host: immunological consequences. *Eur J Pharmacol*. 2011;668(suppl 1):S16–S32.
6. Rautava S, Lu L, Nanthakumar NN, et al. TGF-β2 induces maturation of immature human intestinal epithelial cells and inhibits inflammatory cytokine responses induced via the NF-κB pathway. *J Ped Gastroenterol Nutr*. 2012;54:630–638.
7. Labéta M O, Rey Nores JE, Mauricio A, et al. Innate recognition of bacteria in human milk is mediated by a milk-derived highly expressed pattern recognition receptor, soluble CD14. *J Exp Med*. 2000;191:1807–1812.
8. Hooper LV, Littman DR, Macpherson AJ. Interactions between the microbiota and the immune system. *Science*. 2012;336:1268–1273.
9. Pabst O, Mowat AM. Oral tolerance to food protein. *Mucosal Immunol*. 2012;5:232–239.
10. Rescigno M, Lopatin U, Chieppa M. Interactions among dendritic cells, macrophages, and epithelial cells in the gut: implications for immune tolerance. *Curr Opin Immunol*. 2008;20:669–675.
11. Hooper LV, Macpherson AJ. Immune adaptations that maintain homeostasis with the intestinal microbiota. *Nat Rev Immunol*. 2010;10:159–169.
12. Wiesner J, Vilcinskas A. Antimicrobial peptides: the ancient arm of the human

immune system. *Virulence*. 2010;1:440–464.
13. Muniz LR, Knosp C, Yeretssian G. Intestinal antimicrobial peptides during homeostasis, infection and disease. *Front Immunol*. 2012;3:1–13.
14. Ouellette AJ. Paneth cells and innate immunity in the crypt microenvironment. *Gastroenterology*. 1997;113:1779–1784.
15. Sherman MP, Bennett SH, Hwang FFY, et al. Paneth cells and antibacterial host defense in neonatal small intestine. *Infect Immun*. 2005;73:6143–6146.
16. Miron N, Cristea V. Enterocytes: active cells in tolerance to food and microbial antigens in the gut. *Clin Exp Immunol*. 2012;167:405–412.
17. Haller D, Bode C, Hammes WP, et al. Non-pathogenic bacteria elicit a differential cytokine response by intestinal epithelial cell/leucocyte co-cultures. *Gut*. 2000;47:79–87.
18. Jenner RG, Young RA. Insights into host responses against pathogens from transcriptional profiling. *Nat Rev Micro*. 2005;3:281–294.
19. Takeuchi O, Akira S. Pattern recognition receptors and inflammation. *Cell*. 2010;140:805–820.
20. Abreu MT. Toll-like receptor signalling in the intestinal epithelium: how bacterial recognition shapes intestinal function. *Nat Rev Immunol*. 2010;10:131–144.
21. O'Neill LA, Dunne A, Edjeback M, et al. Mal and MyD88: adapter proteins involved in signal transduction by toll-like receptors. *J Endotoxin Res*. 2003;9:55–59.
22. Lala S, Ogura Y, Osborne C, et al. Crohn's disease and the NOD2 gene: a role for paneth cells. *Gastroenterology*. 2003;125:47–57.
23. Pauleau AL, Murray PJ. Role of Nod2 in the response of macrophages to Toll-like receptor agonists. *Mol Cell Biol*. 2003;23:7531–7539.
24. Cario E. Bacterial interactions with cells of the intestinal mucosa: Toll-like receptors and NOD 2. *Gut*. 2005;54:1182–1193.
25. Niess JH, Brand S, Gu X, et al. CX3CR1–mediated dendritic cell access to the intestinal lumen and bacterial clearance. *Science*. 2005;307:254–258.
26. Coombes JL, Powrie F. Dendritic cells in intestinal immune regulation. *Nat Rev Immunol*. 2008;8:435–446.
27. Macpherson AJ, Uhr T. Induction of protective IgA by intestinal dendritic cells carrying commensal bacteria. *Science*. 2004;303:1662–1665.
28. Brandtzaeg P. Induction of secretory immunity and memory at mucosal surfaces. *Vaccine*. 2007;25:5467–5484.
29. Zhu J, Yamane H, Paul WE. Differentiation of effector CD4 T cell populations. *Annu Rev Immunol*. 2010;28:445–489.
30. Black A, BhaumikS, Kirkman RL, et al. Developmental regulation of Th17–cell capacity in human neonates. *EurJ Immunol*. 2012;42:311–319.
31. Aggarwal S, Ghilardi N, Xie MH, et al. Interleukin-23 promotes a distinct CD4 T cell activation state characterized by the production of interleukin-17. *J Biol Chem*. 2003;278:1910–1914.
32. Josefowicz SZ, Lu LF, Rudensky AY. Regulatory T cells: mechanisms of differentiation and function. *Annu Rev Immunol*. 2012;30:531–564.
33. Molloy MJ, Bouladoux N, Belkaid Y. Intestinal microbiota: shaping local and systemic immune responses. *Semin Immunol*. 2012;24:58–66.
34. Lee YK, Mazmanian SK. Has the microbiota played a critical role in the evolution of the adaptive immune system? *Science*. 2010;330:1768–1773.
35. Stritesky GL, Jameson SC, Hogquist KA. Selection of self-reactive T cells in the thymus. *Annu Rev Immunol*. 2012;30:95–114.
36. Manzoni P, Stolfi I, Messner H, et al. Bovine lactoferrin prevents invasive fungal

infections in very low birth weight infants: a randomized controlled trial. *Pediatrics*. 2012;129:116–123.
37. Prendergast AJ, Klenerman P, Goulder PJR. The impact of differential antiviral immunity in children and adults. *Nat Rev Immunol*. 2012;12:636–648.
38. Kaplan JL, Shi HN, Walker WA. The role of microbes in developmental immunologic programming. *Pediatr Res*. 2011;69:465–472.
39. Ibnou-Zekri N, Blum S, Schiffrin EJ, et al. Divergent patterns of colonization and immune response elicited from two intestinal *Lactobacillus* strains that display similar properties in vitro. *Infect Immun*. 2003;71:428–436.
40. Maynard CL, Elson CO, Hatton RD, et al. Reciprocal interactions of the intestinal microbiota and immune system. *Nature*. 2012;489:231–241.
41. Duerr CU, Hornef MW. The mammalian intestinal epithelium as integral player in the establishment and maintenance of host–microbial homeostasis. *Semin Immunol*. 2012;24:25–35.
42. Bouskra D, Brézillon C, Bérard M, et al. Lymphoid tissue genesis induced by commensals through NOD1 regulates intestinal homeostasis. *Nature*. 2008;456:507–510.
43. Mallow EB, Harris A, Salzma N, et al. Human enteric defensins: gene structure and developmental expression. *J Biol Chem*.1996;271:4038–4045.
44. Coutinho HB, da Mota HC, Coutinho VB, et al. Absence of lysozyme (muramidase) in the intestinal Paneth cells of newborn infants with necrotising enterocolitis. *J Clin Pathol*. 1998;51:512–514.
45. Kollmann TR, Levy O, Montgomery RR, et al. Innate immune function by Toll-like receptors: distinct responses in newborns and the elderly. *Immunity*. 2012;37:771–783.
46. Round JL, Mazmanian SK. Inducible Foxp3+ regulatory T-cell development by a commensal bacterium of the intestinal microbiota. *Proc Nat Acad Sci*. 2010;107:12204–12209.
47. Chassin C, Hempel C, Stockinger S, et al. MicroRNA-146a-mediated downregulation of IRAK1 protects mouse and human small intestine against ischemia/reperfusion injury. *EMBO Mol Med*. 2012;4:1308–1319.
48. Bäckhed F, Ley RE, Sonnenburg JL, et al. Host-bacterial mutualism in the human intestine. *Science*. 2005;307:1915–1920.
49. Morowitz MJ, Denef VJ, Costello EK, et al. Strain-resolved community genomic analysis of gut microbial colonization in a premature infant. *Proc Nat Acad Sci*. 2011;108:1128–1133.
50. Rotimi VO, Duerden BI. The development of the bacterial flora in normal neonates. *J Med Microbiol*. 1981;14:51–62.
51. Jost T, Lacroix C, Braegger CP, et al. New insights in gut microbiota establishment in healthy breast fed neonates. *PLoS One*. 2012;7:e44595.
52. Lau S, Matricardi PM. Worms, asthma, and the hygiene hypothesis. *Lancet*. 2006;367:1556–1558.
53. Xue X, Feng T, Yao S, et al. Microbiota down regulates dendritic cell expression of miR-10a, which targets IL-12/IL-23p40. *J Immunol*. 2011;187:5879–5886.
54. Schwartz S, Friedberg I, Ivanov I, et al. A metagenomic study of diet-dependent interaction between gut microbiota and host in infants reveals differences in immune response. *Genome Biol*. 2012;13:32.
55. Penders J, Thijs C, Vink C, et al. Factors influencing the composition of the intestinal microbiota in early infancy. *Pediatrics*. 2006;118:511–521.
56. Madan JC, Farzan SF, Hibberd PL, et al. Normal neonatal microbiome variation in relation to environmental factors, infection and allergy. *Curr Opin Pediatr*.

2012;24:753–759
57. Hanson L. Breastfeeding provides passive and likely long-lasting active immunity. *Ann Allergy Asthma Immunol*. 1998;81:523–537.
58. Newburg DS. Neonatal protection by an innate immune system of human milk consisting of oligosaccharides and glycans. *J Anim Sci*. 2009;87:26–34.
59. Håversen L, Kondori N, Baltzer L, et al. Structure-microbicidal activity relationship of synthetic fragments derived from the antibacterial α-helix of human lactoferrin. *Antimicrob Agents Chemother*. 2010;54:418–425.
60. Kjellev S. The trefoil factor family–small peptides with multiple functionalities. *Cell Mol Life Sci*. 2009;66:1350–1369.
61. Vestergaard EM, Nexo E, Wendt A, et al. Trefoil factors in human milk. *Early Hum Dev*. 2008;84:631–635.
62. Gonzalez GJ, Sanchez G, Gonzalez JE. Trefoil factor 3 isolated from human breast milk downregulates cytokines (IL8 and IL6) and promotes human beta defensin (hBD2 and hBD4) expression in intestinal epithelial cells HT-29. *Bosn J Basic Med Sci*. 2012;12:256–264.
63. Verhasselt V. Neonatal tolerance under breastfeeding influence. *Curr Opin Immunol*. 2010;22:623–630.
64. Newburg D, Shen Z, Warren C. Quantitative analysis of human milk oligosaccharides by capillary electrophoresis. *Adv Exp Med Biol*. 2000;478:381–382.
65. Ballard O, Morrow AL. Human milk composition: nutrients and bioactive factors. *Pediatr Clin North Am*. 2013;60:49–74.
66. Yu ZT, Chen C, Kling DE, et al. The principal fucosylated oligosaccharides of human milk exhibit prebiotic properties on cultured infant microbiota. *Glycobiology*. 2013;23:169–177.
67. Bode L. Human milk oligosaccharides: every baby needs a sugar mama. *Glycobiology*. 2012;22:1147–1162.
68. Newburg DS, Ruiz-Palacios GM, Morrow AL. Human milk glycans protect infants against enteric pathogens. *Annu Rev Nutr*. 2005;25:37–58.
69. Angeloni S, Ridet JL, Kusy N, et al. Glycoprofiling with micro-arrays of glycoconjugates and lectins. *Glycobiology*. 2005;15:31–41.
70. Garrido D, Kim JH, German JB, et al. Oligosaccharide binding proteins from *Bifidobacterium longum* subsp. *infantis* reveal a preference for host glycans. *PLoS One*. 2011;6:e17315.
71. Morrow AL, Ruiz-Palacios GM, Jiang X, et al. Human-milk glycans that inhibit pathogen binding protect breast-feeding infants against infectious diarrhea. *J Nutr*. 2005;135:1304–1307.
72. Wold AE, Adlerberth I. Breast feeding and the intestinal microflora of the infant —implications for protection against infectious diseases. *Adv Exp Med Biol*. 2000;478:77–93.
73. Eiwegger T, Stahl B, Schmitt J, et al. Human milk-derived oligosaccharides and plant-derived oligosaccharides stimulate cytokine production of cord blood T-cells in vitro. *Pediatr Res*. 2004;56:536–540.
74. Cedeno-Laurent F, Opperman M, Barthel SR, et al. Galectin-1 triggers an immunoregulatory signature in Th cells functionally defined by IL-10 expression. *J Immunol*. 2012;188:3127–3137.
75. Jantscher-Krenn E, Zherebtsov M, Nissan C, et al. The human milk oligosaccharide disialyllacto-N-tetraose prevents necrotising enterocolitis in neonatal rats. *Gut*. 2012;61:1417–1425.
76. Yolken RH, Peterson JA, Vonderfecht SL, et al. Human milk mucin inhibits

rotavirus replication and prevents experimental gastroenteritis. *J Clin Invest*. 1992;90:1984–1991.
77. Dai D, Nanthkumar NN, Newburg DS, et al. Role of oligosaccharides and glycoconjugates in intestinal host defense. *J Pediatr Gastroenterol Nutr*. 2000;30:S23–S33.
78. Ibrahim HR, Imazato K, Ono H. Human lysozyme possesses novel antimicrobial peptides within its N-terminal domain that target bacterial respiration. *J Agric Food Chem*. 2011;59:10336–10345.
79. Jia HP, Starner T, Ackermann M, et al. Abundant human β-defensin-1 expression in milk and mammary gland epithelium. *J Pediatr*. 2001;138:109–112.
80. Murakami M, Dorschner RA, Stern LJ, et al. Expression and secretion of cathelicidin antimicrobial peptides in murine mammary glands and human milk. *Pediatr Res*. 2005;57:10–15.
81. Armogida SA, Yannaras NM, Melton AL, et al. Identification and quantification of innate immune system mediators in human breast milk. *Allergy Asthma Proc*. 2004;25:297–304.
82. Lönnerdal B, Iyer S. Lactoferrin: molecular structure and biological function. *Annu Rev Nutr*. 1995;15:93–110.
83. Longhi C, Conte MP, Penta M, et al. Lactoferricin influences early events of *Listeria monocytogenes* infection in THP-1 human macrophages. *J Med Microbiol*. 2004;53:87–91.
84. Goldman AS, Garza C, Schanler RJ, et al. Molecular forms of lactoferrin in stool and urine from infants fed human milk. *Pediatr Res*. 1990;27:252–255.
85. Coppa GV, Zampini L, Galeazzi T, et al. Prebiotics in human milk: a review. *Dig Liver Dis*. 2006;38(suppl 2):S291–S294.
86. Adamkin DH. Mother's milk, feeding strategies, and lactoferrin to prevent necrotizing enterocolitis. *J Parenter Enteral Nutr*. 2012; 36:25S-29S.
87. Manzoni P, Inaldi M, Catani S, et al. Bovine lactoferrin supplementation for prevention of late-onset sepsis in very low-birth-weight neonates: a randomized trial. *JAMA*. 2009;302:1421–1428.
88. Brandtzeag P, Nilssen DE, Rogum TO, et al. Ontogeny of the mucosal immune system and IgA deficiency. *Gastroenterol Clin North Am*. 1991;20:397–439.
89. Mostov KE, Friedlander M, Blobel G. The receptor for transepithelial transport of IgA and IgM contains multiple immunoglobulin-like domains. *Nature*. 1984;308:37–43.
90. Bourges D, Meurens F, Berri M, et al. New insights into the dual recruitment of IgA+ B cells in the developing mammary gland. *Mol Immunol*. 2008;45:3354–3362.
91. Brandtzaeg P. Role of secretory antibodies in the defence against infections. *Int J Med Microbiol*. 2003;293:3–15.
92. Schanler RJ, Goldblum RM, Garza C, et al. Enhanced fecal excretion of selected immune factors in very low birth weight infants fed fortified human milk. *Pediatr Res*. 1986;20:711–715.
93. Corthesy B. Role of secretory immunoglobulin A and secretory component in the protection of mucosal surfaces. *Future Microbiol*. 2010;5:817–829.
94. van der Waaij LA, Limburg PC, Mesander G, et al. In vivo IgA coating of anaerobic bacteria in human faeces. *Gut*. 1996;38:348–354.
95. Kadaoui KA, Corthésy B. Secretory IgA mediates bacterial translocation to dendritic cells in mouse Peyer's patches with restriction to mucosal compartment. *J Immunol*. 2007;179:7751–7757.
96. Perez PF, Doré J, Leclerc M, et al. Bacterial imprinting of the neonatal immune

system: lessons from maternal cells? *Pediatrics*. 2007;119:e724–e732.
97. Martín R, Langa S, Reviriego C, et al. Human milk is a source of lactic acid bacteria for the infant gut. *J Pediatr*. 2003;143:754–758.
98. Ludvigsson JF, Fasano A. Timing of introduction of gluten and celiac disease risk. *Ann Nutr Metab*. 2012;60:22–29.
99. Mosconi E, Rekima A, Seitz-Polski B, et al. Breast milk immune complexes are potent inducers of oral tolerance in neonates and prevent asthma development. *Mucosal Immunol*. 2010;3:461–474.
100. Aderem A, Ulevitch RJ. Toll-like receptors in the induction of the innate immune response. *Nature*. 2000;406:782–787.
101. Schröder NWJ, Morath S, Alexander C, et al. Lipoteichoic acid (LTA) of *Streptococcus pneumoniae* and *Staphylococcus aureus* activates immune cells via Toll-like receptor (TLR)-2, lipopolysaccharide-binding protein (LBP), and CD14, whereas TLR-4 and MD-2 are not involved. *J Biol Chem*. 2003;278:15587–15594.
102. Filipp D, Alizadeh-Khiavi K, Richardson C, et al. Soluble CD14 enriched in colostrum and milk induces B cell growth and differentiation. *Proc Nat Acad Sci*. 2001;98:603–608.
103. LeBouder E, Rey-Nores JE, Rushmere NK, et al. Soluble forms of Toll-like receptor (TLR)2 capable of modulating TLR2 signaling are present in human plasma and breast milk. *J Immunol*. 2003;171:6680–6689.
104. Hailman E, Vasselon T, Kelley M, et al. Stimulation of macrophages and neutrophils by complexes of lipopolysaccharide and soluble CD14. *J Immunol*. 1996;156:4384–4390.
105. Pugin J, Schurer-Maly CC, Leturq D, et al. Lipopolysaccharide activation of human endothelial and epithelial cells is mediated by lipopolysaccharide-binding protein and soluble CD14. *Proc Natl Acad Sci USA*. 1993;90:2744–2748.
106. Appelmelk BJ, An YQ, Geerts M, et al. Lactoferrin is a lipid A-binding protein. *Infect Imm*. 1994;62:2628–2632.
107. Baveye S, Elass E, Fernig DG, et al. Human lactoferrin interacts with soluble CD14 and inhibits expression of endothelial adhesion molecules, E-selectin and ICAM-1, induced by the CD14–lipopolysaccharide complex. *Infect Imm*. 2000;68:6519–6525.
108. Moreno C, Merino J, Vázquez B, et al. Anti-inflammatory cytokines induce lipopolysaccharide tolerance in human monocytes without modifying toll-like receptor 4 membrane expression. *Scand J Immunol*. 2004;59:553–558.
109. Imai K, Takeshita A, Hanazawa S. Transforming growth factor-β inhibits lipopolysaccharide-stimulated expression of inflammatory cytokines in mouse macrophages through downregulation of activation protein 1 and CD14 receptor expression. *Infect Imm*. 2000;68:2418–2423.
110. Kulkarni AB, Huh CG, Becker D, et al. Transforming growth factor 181 null mutation in mice causes excessive inflammatory response and early death. *Proc Nati Acad Sci USA*.1993;90:770–774.
111. Levy O. Innate immunity of the newborn: basic mechanisms and clinical correlates. *Nat Rev Immunol*. 2007;7:379–390.
112. Angelone DF, Wessels MR, Coughlin M, et al. Innate immunity of the human newborn is polarized toward a high ratio of IL-6/TNF-[alpha] production in vitro and in vivo. *Pediatr Res*. 2006;60:205–209.
113. Abreu MT, Vora P, Faure E, et al. Decreased expression of Toll-like receptor-4 and MD-2 correlates with intestinal epithelial cell protection against dysregulated proinflammatory gene expression in response to bacterial lipopolysaccharide. *J Immunol*. 2001;167:1609–1616.

114. Nanthakumar N, Meng D, Goldstein AM, et al. The mechanism of excessive intestinal inflammation in necrotizing enterocolitis: an immature innate immune response. *PLoS One*. 2011;6:e17776.
115. Claud EC, Lu L, Anton PM, et al. Developmentally regulated IκB expression in intestinal epithelium and susceptibility to flagellin-induced inflammation. *Proc Natl Acad Sci USA*. 2004;101:7404–7408.
116. Lotz M, Gütle D, Walther S, et al. Postnatal acquisition of endotoxin tolerance in intestinal epithelial cells. *J Exp Med*. 2006;203:973–984.
117. LeBouder E, Rey-Nores JE, Raby AC, et al. Modulation of neonatal microbial recognition: TLR-mediated innate immune responses are specifically and differentially modulated by human milk. *J Immunol*. 2006;176:3742–3752.
118. Baveye S, Elass E, Fernig DG, et al. Human lactoferrin interacts with soluble CD14 and inhibits expression of endothelial adhesion molecules, E-selectin and ICAM-1, induced by the CD14–lipopolysaccharide complex. *Infect Imm*. 2000;68:6519–6525.
119. Vidal K, Donnet-hughes A, Granato D. Lipoteichoic acids from *Lactobacillus johnsonii* strain La1 and *Lactobacillus acidophilus* strain La10 antagonize the responsiveness of human intestinal epithelial HT29 cells to lipopolysaccharide and gram-negative bacteria. *Infect Immun*. 2002;70:2057–2064.
120. Lawrence T, Bebien M, Liu GY, et al. IKK[alpha] limits macrophage NF-[kappa]B activation and contributes to the resolution of inflammation. *Nature*. 2005;434:1138–1143.
121. Li MO, Wan YY, Sanjabi S, et al. Transforming growth factors-β regulation of immune responses. *Annu Rev Immunol*. 2006;24:99–146.
122. Wong BR, Besser D, Kim N, et al. TRANCE, a TNF family member, activates Akt/PKB through a signaling complex involving TRAF6 and c-Src. *Mol Cell*. 1999;4:1041–1049.
123. Vidal K, Serrant P, Schlosser B, et al. Osteoprotegerin production by human intestinal epithelial cells: a potential regulator of mucosal immune responses. *Am J Physiol Gastrointest Liver Physiol*. 2004;287:G836–G844.
124. Buescher ES, Malinowska I. Soluble receptors and cytokine antagonists in human milk. *Pediatr Res*. 1996;40:839–844.
125. Grazioso CF, Werner AL, Alling DW, et al. Antiinflammatory effects of human milk on chemically induced colitis in rats. *Pediatr Res*. 1997;42:639–643.
126. Fituch CC, Palkowetz KH, Goldman AS, et al. Concentrations of IL-10 in preterm human milk and in milk from mothers of infants with necrotizing enterocolitis. *Acta Pædiatrica*. 2004;93:1496–1500.
127. Goldman AS, Goldblum R M. Transfer of maternal leukocytes to the infant by human milk. *Curr Top Microbiol Immunol*. 1997;222: 205–213.
128. Brandtzaeg P. The mucosal immune system and its integration with the mammary glands. *J Pediatr*. 2010;156:S8–S15.
129. Ichikawa M, Sugita M, Takahashi M, et al. Breast milk macrophages spontaneously produce granulocyte–macrophage colony-stimulating factor and differentiate into dendritic cells in the presence of exogenous interleukin-4 alone. *Immunology*. 2003;108:189–195.
130. Garofalo R. Cytokines in human milk. *J Pediatr*. 2010;156:S36–S40.
131. Glenn W, Whitaker N, Lawson J. High risk human papillomavirus and Epstein Barr virus in human breast milk. *BMC Res Notes*. 2012;5:477.
132. Donnet-Hughes A, Perez PF, Doré J, et al. Potential role of the intestinal microbiota of the mother in neonatal immune education. *Proc Nutr Soc*. 2010;69:407–415.

133. Martín V, Maldonado-Barragán A, Moles L, et al. Sharing of bacterial strains between breast milk and infant feces. *J Hum Lact*. 2012;28:36–44.
134. Maldonado J, Cañabate F, Sempere L, et al. Human milk probiotic *Lactobacillus fermentum* CECT5716 reduces the incidence of gastrointestinal and upper respiratory tract infections in infants. *J Pediatr Gastroenterol Nutr*. 2012;54:55–61.
135. Cabrera-Rubio R, Collado MC, Laitinen K, et al. The human milk microbiome changes over lactation and is shaped by maternal weight and mode of delivery. *Am J Clin Nutr*. 2012;96:544–551.
136. Planchon SM, Martins CA, Guerrant RL, et al. Regulation of intestinal epithelial barrier function by TGF-beta 1. Evidence for its role in abrogating the effect of a T cell cytokine. *J Immunol*. 1994;153:5730–5739.
137. Maheshwari A, Kelly DR, Nicola T, et al. TGF-β2 suppresses macrophage cytokine production and mucosal inflammatory responses in the developing intestine. *Gastroenterology*. 2011;140:242–253.
138. Oddy WH, McMahon RJ. Milk-derived or recombinant transforming growth factor-beta has effects on immunological outcomes: a review of evidence from animal experimental studies. *Clin Exp Allergy*. 2011;41:783–793.
139. Verhasselt V, Milcent V, Cazareth J, et al. Breast milk-mediated transfer of an antigen induces tolerance and protection from allergic asthma. *Nat Med*. 2008;14:170–175.
140. Ogawa J, Sasahara A, Yoshida T, et al. Role of transforming growth factor-β in breast milk for initiation of IgA production in newborn infants. *Early Hum Dev*. 2004;77:67–75.
141. Siegrist CA. The challenges of vaccine responses in early life: selected examples. *J Comp Pathol*. 2007;137(suppl 1):S4–S9.
142. Gold MC, Donnelly E, Cook MS, et al. Purified neonatal plasmacytoid dendritic cells overcome intrinsic maturation defect with TLR agonist stimulation. *Pediatr Res*. 2006;60:34–37.
143. Maródi L. Innate cellular immune responses in newborns. *Clin Immunol*. 2006;118:137–144.
144. Andersson AC, Seppälä U, Rudin A. Activation of human neonatal monocyte-derived dendritic cells by lipopolysaccharide down-regulates birch allergen-induced Th2 differentiation. *Eur J Immnol*. 2004;34:3516–3524.
145. Siltanen M, Kajosaari M, Savilahti EM, et al. IgG and IgA antibody levels to cow's milk are low at age 10 years in children born preterm. *J Allergy Clin Immunol*. 2002;110:658–663.
146. Verhasselt V, Buelens C, Willems F, et al. Bacterial lipopolysaccharide stimulates the production of cytokines and the expression of costimulatory molecules by human peripheral blood dendritic cells: evidence for a soluble CD14–dependent pathway. *J Immunol*. 1997;158:2919–2925.
147. Rey-Nores JE, Bensussan A, Vita N, et al. Soluble CD14 acts as a negative regulator of human T–cell activation and function. *Eur J Immunol*. 1999;29:265–276.
148. Arias MA, Rey Nores JE, Vita N, et al. Cutting edge: human B cell function is regulated by interaction with soluble CD14: opposite effects on IgG1 and IgE production. *J Immunol*. 2000;164:3480–3486.
149. Fischer R, Debbabi H, Dubarry M, et al. Regulation of physiological and pathological Th1 and Th2 responses by lactoferrin. *Biochem Cell Biol*. 2006;84:303–311.
150. Yun TJ, Tallquist MD, Aicher A, et al. Osteoprotegerin, a crucial regulator of

bone metabolism, also regulates B cell development and function. *J Immunol*. 2001;166:1482–1491.
151. Williamson E, Bilsborough JM, Viney JL. Regulation of mucosal dendritic cell function by receptor activator of NF-κB (RANK)/RANK ligand interactions: impact on tolerance induction. *J Immnunol*. 2002;169:3606–3612.
152. Naarding MA, Ludwig IS, Groot F, et al. Lewis X component in human milk binds DC-SIGN and inhibits HIV-1 transfer to CD4+ T lymphocytes. *J Clin Invest*. 2005;115:3256–3264.
153. Neumann AK, Thompson NL, Jacobson K. Distribution and lateral mobility of DC-SIGN on immature dendritic cells–implications for pathogen uptake. *J Cell Sci*. 2008;121:634–643.
154. Chu H, Pazgier M, Jung G, et al. Human α-defensin 6 promotes mucosal innate immunity through self-assembled peptide nanonets. *Science*. 2012;337:477–481.

Approach to Breastfeeding

31

Robert M. Lawrence, MD, FAAP, FABM and Ruth A. Lawrence, MD, FAAP, FABM, FAAC

There are various memorable phrases and taglines associated with initiatives and campaigns to promote breastfeeding; “Babies were born to be breast-fed,” “Mother’s milk is never forgotten,” “Good health begins with breastfeeding,” or “Breastfeeding is more than just good nutrition.” The Fourth Annual Summit on Breastfeeding held in Washington, DC in June of 2012 was titled “First Food: The Essential Role of Breastfeeding.”[1–5] This chapter and others within this text provides knowledge and understanding to support these statements.

The “Breastfeeding Report Card” for the United States presented by the Centers for Disease Control and Prevention (CDCP) in 2012 reports that the average national rates for breastfeeding initiation reached 76.9%, breastfeeding at 6 months of age was at 47.2% and at 12 months at 25.5% and exclusive breastfeeding averages reached 36% at 3 months and 16.3% at 6 months in 2009.[6] The health goals of our nation for 2020 include a statement regarding breastfeeding. The goals particularly address high-risk women, those from minority, low-income, and undereducated groups.[2] The objectives focus on increasing the proportion of infants who are breastfed, for all infants, with targets of 81.9% for ever breastfeeding, 60.6% breastfeeding at 6 months and 34.1% at 12 months and exclusive breastfeeding through three months of 46.2% and through 6 months of 25.5%.[7]

The Institute of Medicine issued a report on nutrition during lactation as part of a review of nutrition in the perinatal period that stated that breastfeeding was ideal for all infants under ordinary circumstances.[8] It further stated that even women without perfect diets could produce good

milk and nourish their young well. Professional medical associations such as the American Academy of Pediatrics,[9] the American College of Obstetrics and Gynecology, and the Academy of Family Practice have developed policies encouraging universal breastfeeding. The World Health Organization and United Nations International Children's Emergency Fund (UNICEF) have taken very strong positions in support of worldwide breastfeeding, including the development of the Baby Friendly Hospital Initiative.[10,11]

Human milk is specifically designed for the needs of the human infant. Its nutritional advantages have been noted to be especially important for brain growth.[12–14] In the first year of life, the brain of the human infant doubles in size.[15] The myelinization of nerves is equally as important and occurs extensively in the first year of life. Taurine, cholesterol, and omega fatty acids are essential to brain growth and are uniquely present in human milk.[16]

Data from evidence-based systematic review and meta-analyses demonstrate clear benefits for breastfeeding infants and mothers in developed countries.[17,18] The benefits for the infants include a reduced risk of acute otitis media, severe lower respiratory tract infections, non-specific gastroenteritis, atopic dermatitis, asthma in young children, obesity, type 1 and 2 diabetes, childhood leukemia, and necrotizing enterocolitis. A history of lactation for women showed a reduced risk of type 2 diabetes and breast and ovarian cancer.[17,18]

The presence of dozens of active enzymes, the immunologic properties, infection protection properties, and allergy protection are some of the compelling reasons breastfeeding is superior for human infants.[19–22]

The number of women who elect to breastfeed has continued to increase, and the renaissance of breastfeeding is well established.[6,23] It is important for the clinician to be knowledgeable about the value of human milk, the advantages of breastfeeding, the clinical management of lactation, and the diagnosis and treatment of problems.[24] The current scientific literature provides a large resource of information on these topics, which will be summarized here.

ANATOMY AND PHYSIOLOGY

Lactation is the completion of the normal reproductive cycle. It is a physiologic process triggered by the termination of pregnancy, but anticipated both anatomically and physiologically from early

development.[16]

The breast bud is present at birth in both sexes, but remains dormant until early pubescence, when growth is stimulated by the increase in estrogen and progesterone in the female.[25,26] The ductal system proliferates and the breast matures. This maturation continues with stimulus from each menstrual cycle until age 25. When growth stabilizes, further proliferation does not occur until pregnancy intervenes (Figure 31-1).

Changes in circulating hormones result in profound changes in the ductular-lobular-alveolar growth during pregnancy.[27] There is marked increase in ductular sprouting, branching, and lobular formation evoked by luteal and placental hormones (Figure 31-2). Placental lactogen, prolactin, and chorionic gonadotropin have been identified as contributors to the accelerated growth.

From the third month of gestation, secretory material resembling colostrum appears in the alveoli. By the second trimester, placental lactogen begins to stimulate the production of colostrum so that a women delivering immaturely as early as 16 weeks may secrete colostrum although her baby is not viable. Until delivery, the production of milk is suppressed by prolactin-inhibiting hormone (PIH) produced by the placenta. Progesterone produced by the placenta has been recognized as important in blocking milk production in pregnancy. At delivery, the withdrawal of placental and luteal sex hormones and the infant’s sucking result in the loss of the inhibiting hormones and the stimulation of prolactin releasing factors.[28]

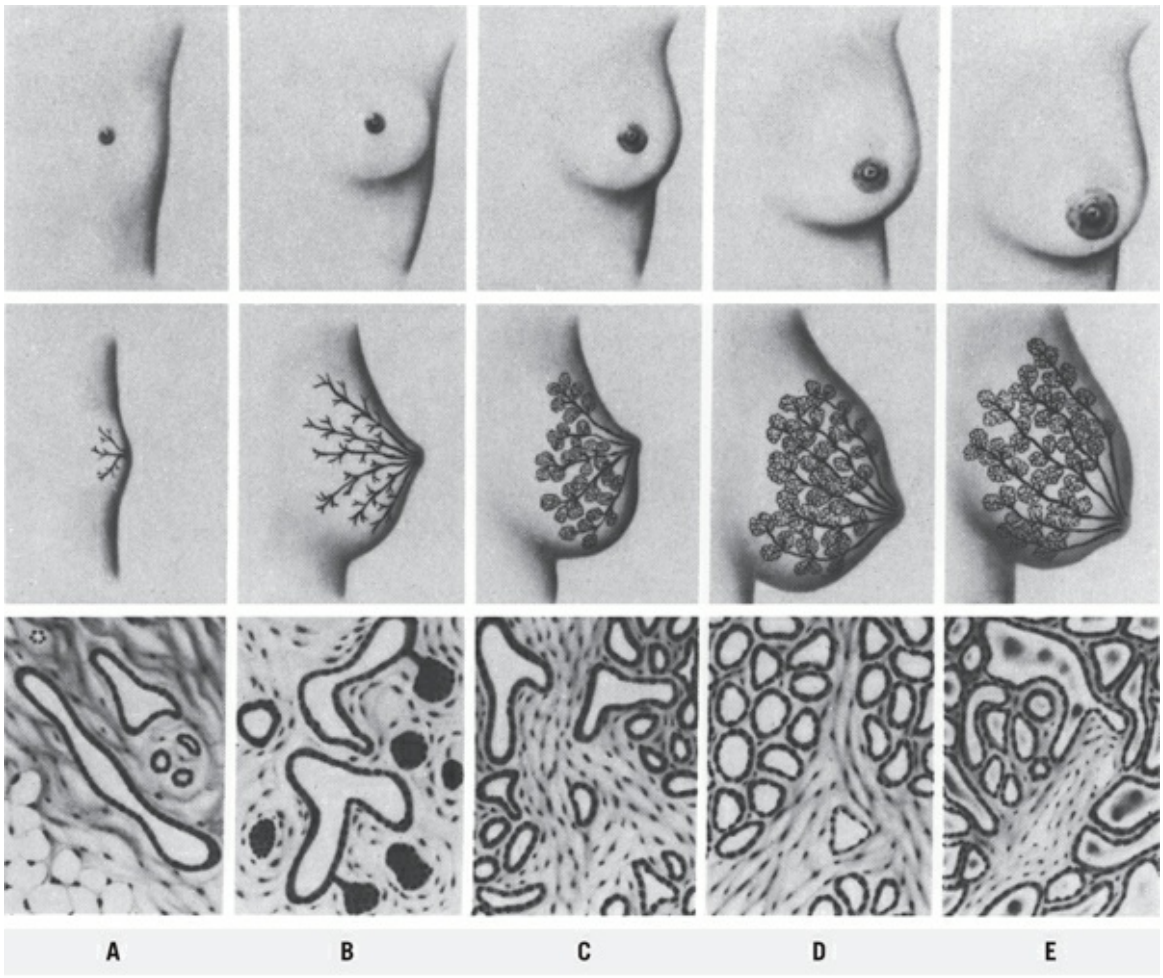


Figure 31-1 Female breast from infancy to lactation with corresponding cross-section and duct structure. *A, B, C,* Gradual development of well-differentiated ductular and peripheral lobular-alveolar system. *D*, Ductular sprouting and intensified peripheral lobular-alveolar development in pregnancy Glandular luminal cells begin actively synthesizing milk fat and proteins near term. Only small amounts are released into lumen *E*, With postpartum withdrawal of luteal and placental sex steroids and placental lactogen, prolactin is able to induce full secretory activity of alveolar cells and release of milk into alveoli and smaller ducts. Reproduced with permission from Lawrence and Lawrence.[10]

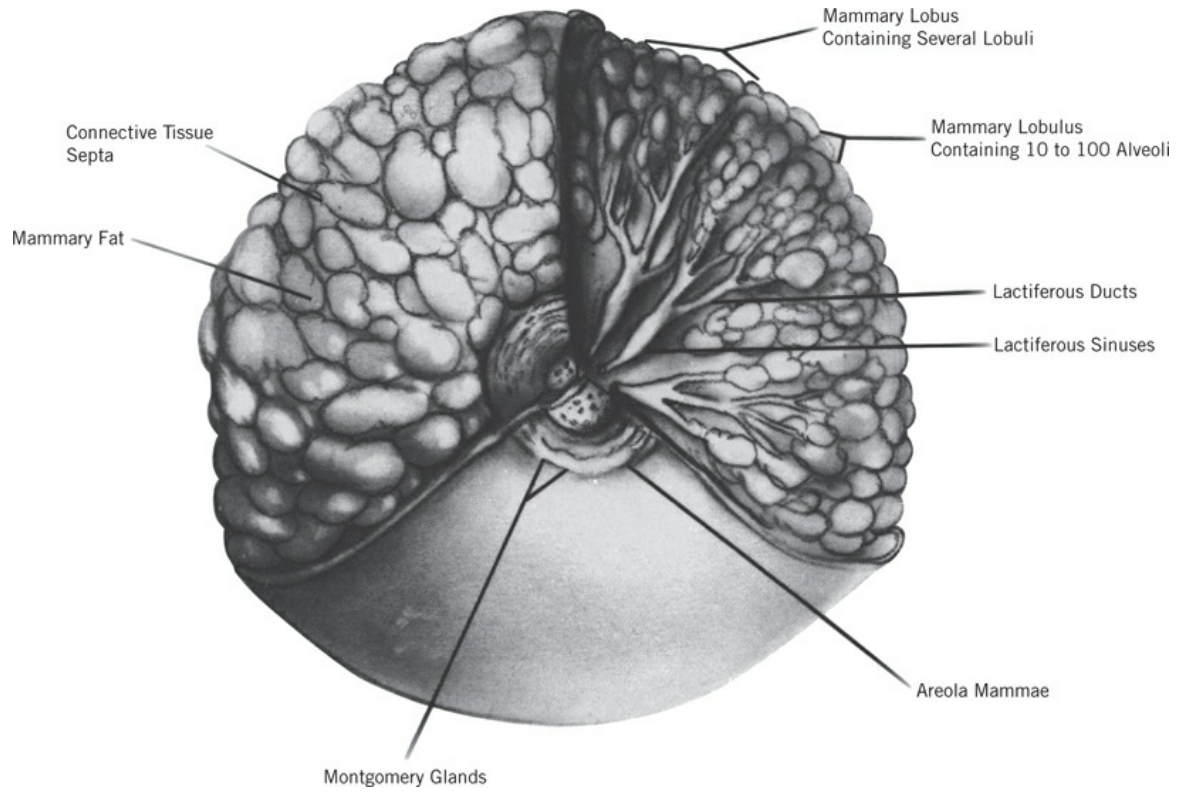


Figure 31-2 Morphology of mature breast with dissection to reveal mammary fat and duct system. Reproduced with permission from Lawrence and Lawrence.[10]

The initiation of milk secretion at delivery and the continued production of milk occur because the breast has developed extensively throughout pregnancy.[29] The ductal system has arborized to form an extended network of collecting ducts. The alveoli are richly lined with epithelial cells varying from flat to low columnar in shape, all capable of producing milk. Some cells protrude into the lumen of the alveoli; others are short and smooth. The lumen of the alveolus is crowded with fine granular material and lipid droplets (Figure 31-3). The division and differentiation of the mammary epithelial cells and presecretory alveolar cells into secretory milk-releasing alveolar cells completes the preparation for milk production. The biosynthesis of milk involves this cellular site, where the metabolic processes occur. There are stem cells and highly differentiated secretory alveolar cells at the terminal ducts. The stem cells are stimulated by growth hormone and insulin, which is synergized by prolactin to stimulate the cells to secretory activity. The breast acts in response to the interactions of the pituitary, thyroid, pancreatic, adrenal,

and ovarian hormones (Figure 31-4).

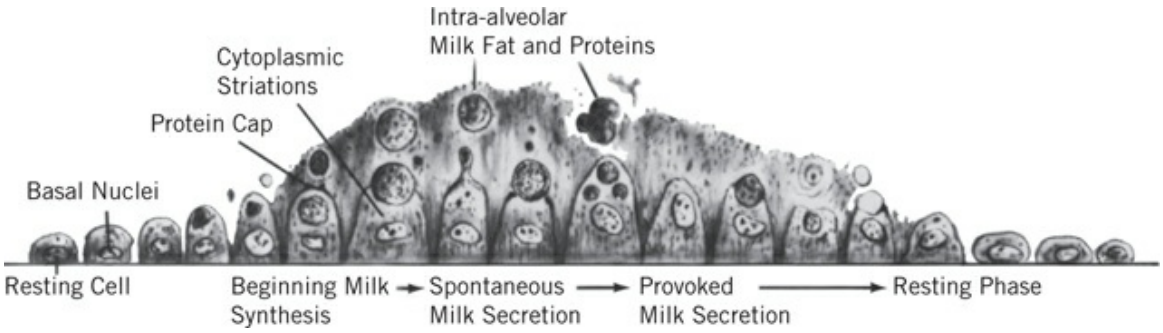


Figure 31-3 Cycle of secretory cells from resting stage to secretion and return to resting stage. Reproduced with permission from Lawrence RA and Lawrence RM.[10]

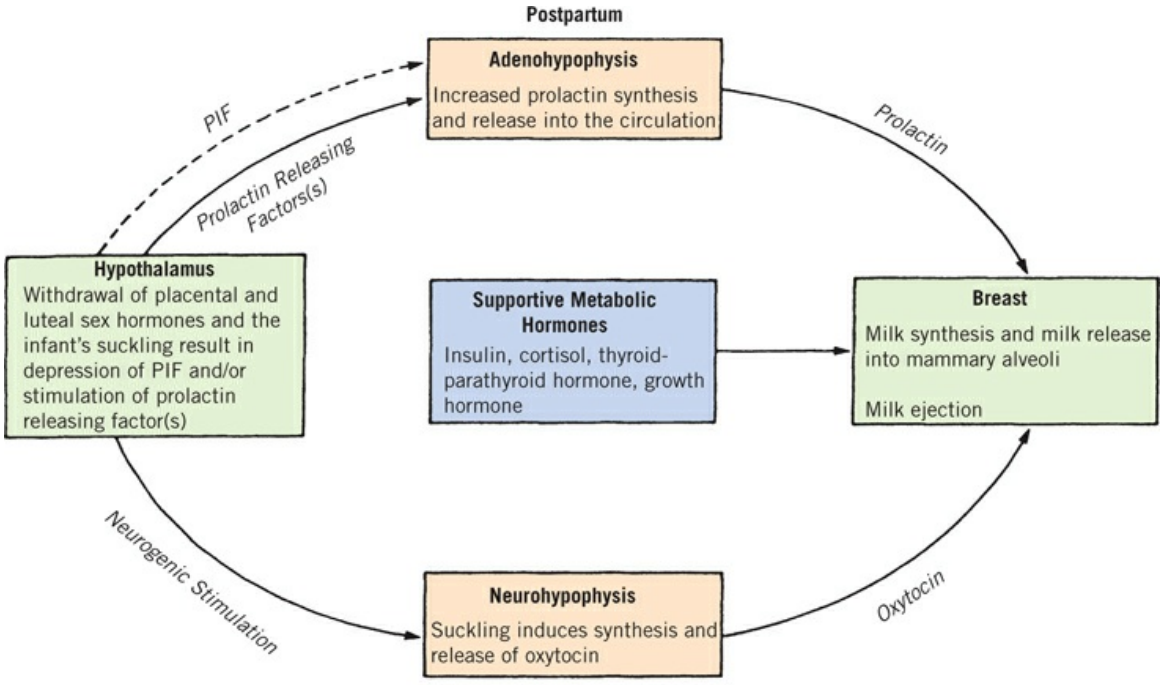


Figure 31-4 Hormonal preparation of breast postpartum for lactation. Reproduced with permission from Lawrence and Lawrence.[10]

The process of milk synthesis involves apocrine secretion for the de novo production of fat and protein and the merocrine secretion of lactose

synthesized from glucose.[25] Ions diffuse across the membrane and, in some cases, are actively transported. The primary alveolar milk is then diluted within the lumen to be isotonic with plasma by water that diffuses from extracellular fluid.[28,29] The pathways for milk synthesis and secretion into the mammary alveolus include[28]: (1) exocytosis of protein and lactose; (2) formation of the milk fat globule; (3) secretion of ions and water; (4) pinocytosis-exocytosis of immunoglobulins; and (5) the paracellular pathway (Figure 31-5).

Because lactation is anticipated, the body prepares the breast during pregnancy and also develops additional nutritional maternal stores that will be needed during lactation, in the form of 6–8 pounds of body weight apart from the uterus and its contents. When lactation begins, there is a redistribution of blood supply from the uterus to the breast, where there is an increased demand for nutrients and an increased metabolic rate to accommodate the demands of milk production. The mammary gland may have to produce milk at the expense of other organs if stores are inadequate. There are cardiovascular adjustments as mammary blood flow increases. The mammary blood flow, cardiac output, and milk secretion are suckling dependent. In addition, suckling induces the release of anterior pituitary hormones, prolactin and oxytocin, which act directly on the breast tissue and on the uterus.[28]

In addition to glandular preparation, the nipple and areola are also preparing for lactation. There is an increase in vascularization. The Montgomery glands, which are sebaceous glands on the areolar circling the nipple, become enlarged and begin to secrete a substance that lubricates and protects the areola and nipple during pregnancy and lactation.[16]

The use of ultrasound imagery[30–34] to reexamine the working of the human breast has replaced some beliefs about the anatomy which were originally derived from the dissection of formalin prepared specimens in 1840.[35] Imaging the actively secreting breast has revealed that only about 9–12 (range 4–18) ducts are at the base of the nipple not the original number of 15–25. The duct were measured to be 1.9 ± 0.6 mm (1.0–4.4 mm) in diameter. The number of ducts and their diameter did not correlate with nipple size or radius of the areolar or actual milk production. The amount of glandular tissue in the lactating breast was about 64% (range 45%–83%) of the breast tissue and the fatty tissue only accounted for 38% (16%–51%). There was no correlation between milk production and the amount of glandular tissue as measured by ultrasound.[30,31] The nipple has

many sensory nerve fibers, but the areola does not, an important fact in terms of comfort for the mother while nursing.[32] The response to tactile sensation of the nipple increases dramatically at delivery as an adaptation for lactation that enhances the nervous response to suckling by the infant (Figure 31-6).

INITIATION OF MILK SECRETION

Withdrawal of placental and luteal sex hormones and stimulation of prolactin-releasing factor result in the increased prolactin synthesis by the adenohypophysis, which stimulates milk synthesis in the mammary alveoli. The release of milk from the alveolar collecting ductules depends on the ejection or let-down reflex (Figure 31-6). The let-down reflex is a simple arc that is initiated by the suckling of the infant.[36] This suckling stimulates the mechanoreceptors in the nipple and areola that send stimuli along nerve pathways to the hypothalamus, which stimulates the posterior pituitary to release oxytocin.[37,38] Oxytocin is carried via the bloodstream to the breast and uterus.[39,40] Oxytocin stimulates the myoepithelial cells, which envelop the secretory alveoli and the collecting ductules in the breast, to contract, ejecting milk through the ductule (Figure 31-4) The oxytocin also stimulates the myoepithelial cells in the uterus to contract, causing the “after pains” a mother associates with lactation. Physiologically, this uterine contraction enhances the uterine postpartum involution, so that the uterus of the lactating woman returns to normal more quickly postpartum. Oxytocin release can also be stimulated by seeing or hearing the infant; thus a woman notices that her milk begins to drip when she sees her infant.[40] Prolactin, however, is only released when the breast is stimulated by suckling or pumping.

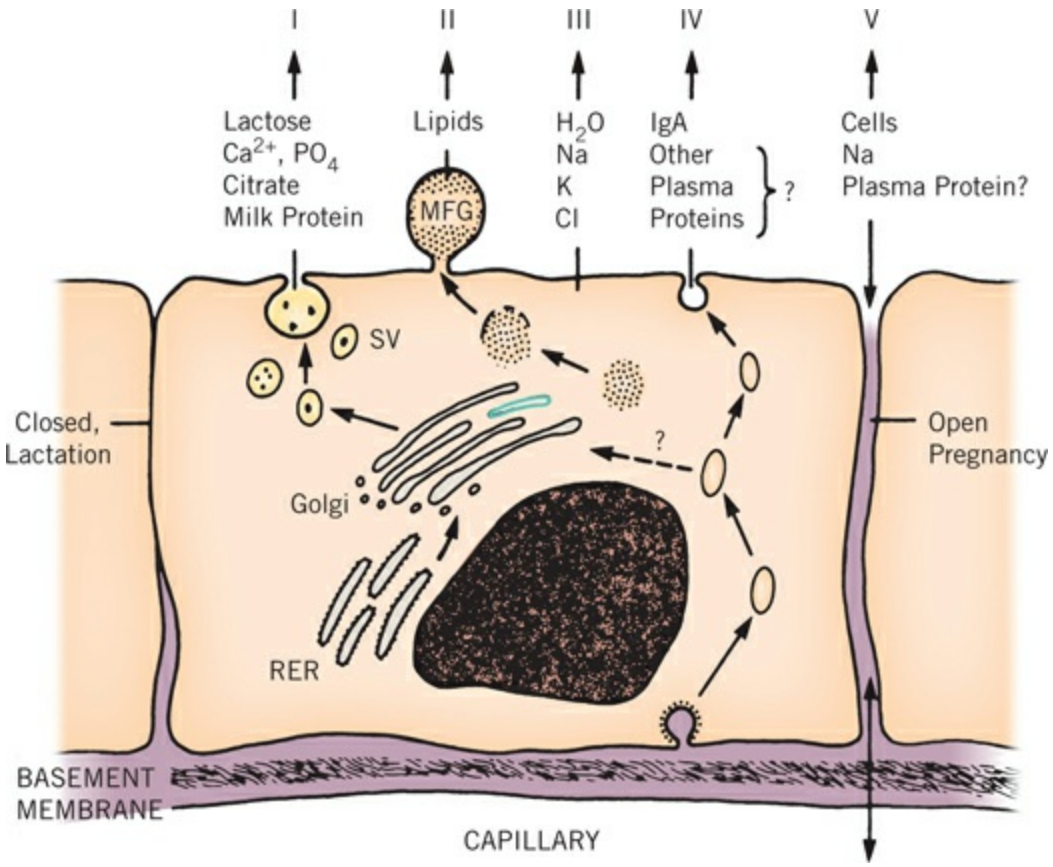


Figure 31-5 The pathways for milk synthesis and secretion in the mammary alveolus. (II) Exocytosis of milk protein and lactose in Golgi-derived secretory vesicles. (III) Secretion of ions and water across the apical membrane. (IV) Pinocytosis-exocytosis of immunoglobulins. (V) The paracellular pathway of plasma components and leukocytes. MFG, milk fat globule; RER, rough endoplasmic reticulum; SV, secretory vesicle. Adapted from Neville MC.18 Reproduced with permission from Lawrence and Lawrence.[10]

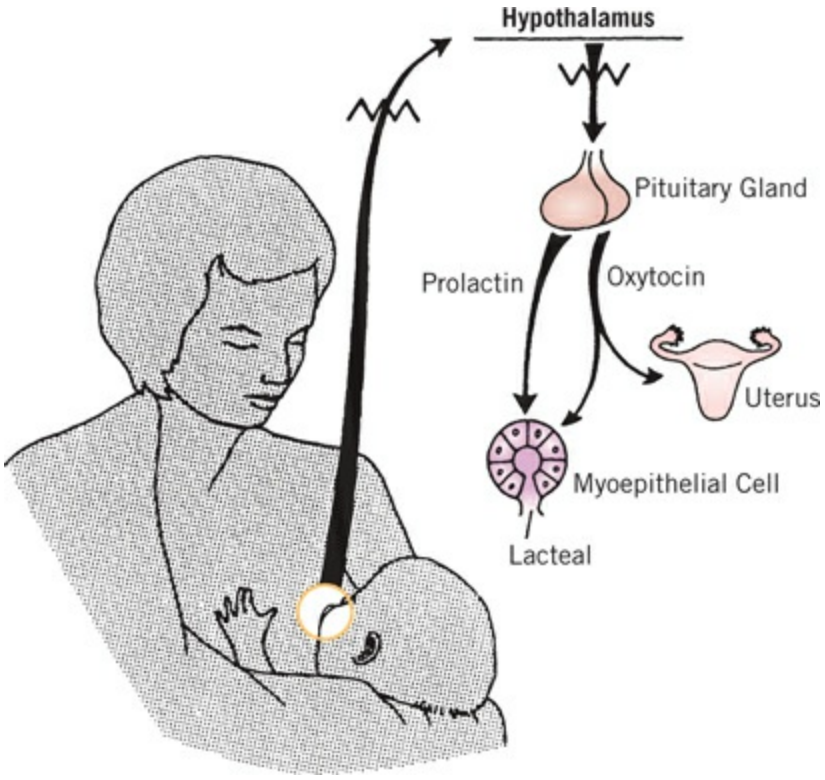


Figure 31-6 (A) Normal nipple everts with gentle pressure. (B) Inverted or tied nipple inverts with gentle pressure. Reproduced with permission from Lawrence and Lawrence.[10]

Prolactin, which is also released from the hypothalamus during sucking, stimulates the production of milk.[36] Prolactin levels during early lactation are increased 10–20 times greater than normal. The technology required to obtain prolactin levels has been available for clinical investigation, but the role of prolactin in the volume of milk produced is still not clearly defined. It is clear, however, that the surge in prolactin to about twice baseline levels is critical to the successful production of an adequate supply of milk. When evaluating prolactin during lactation, a sample of blood is drawn at baseline and then a second sample is drawn after ten minutes of breastfeeding or pumping with an electric pump.[16] The baseline should be above normal range for the laboratory and post stimulus should be increased to almost double baseline.

PRENATAL CONSIDERATIONS

Although the breast prepares for lactation independent of the mother's

decision to breastfeed, it is important to introduce the question of feeding the infant as soon as possible during pregnancy so that the mother can make an informed choice in behalf of her baby.[37] Although it has been suggested that well-educated mothers have made up their minds about how they will feed their infants long before conception occurs, there are many women who need to be informed about breastfeeding and need to receive reinforcement from their physician.[37] Many women, especially primiparas, will need considerable assistance to lactate successfully. The significant benefits of human milk to the human infant have already been reviewed in previous chapters. The psychological benefits are equally as important to both mother and child.[38]

The nutritional benefits of human milk, although legion, can in part be substituted with a modern prepared formula, but the infection protection, immunologic properties, and the psychological benefits of human milk cannot be duplicated.[21,22] In the mid-twentieth century, when bottle feeding was rampant, the single, clear, unchallenged advantage to breastfeeding that was articulated was the special interrelationship between the mother and her baby.[38] The key elements of attachment are said to include early contact, closeness, eye-to-eye contact, smell, and body warmth. Breastfeeding includes these naturally. A woman has a surge of oxytocin and prolactin during each feeding, which has been demonstrated biologically to stimulate mothering behavior.[39,40]

When a mother wishes to be free of the responsibility of breastfeeding, it is often so that she will not be tied down, will not always have to be available, and can have others feed the infant, thus depriving the infant of this special frequent closeness with the mother.

Preparation of the Breasts

Nature prepares the breasts. It is not necessary to manipulate the breasts and nipples prenatally in preparation in the normal woman. Part of the prenatal physical examination should include the breasts with respect to lactation so that any anatomic variations that may interfere with lactation can be discussed.[37] The size of the breast is not related to lactation success and is not a measure of glandular potential.[34] Women who have had benign cysts removed can still nurse successfully. Augmentation mammoplasty does not usually interfere with lactation if the nipple and duct system have been left intact, that is, the nipple has not been realigned and the implant is placed under the breast tissue on the chest wall. Unless the implant has ruptured and has caused scarring, lactation should be

successful. When breast size has been surgically diminished by reduction mammoplasty, the duct system may have been interrupted if the nipple was completely removed and replaced central to the remaining tissue. This may make lactation improbable, and this issue should be discussed with the operating surgeon. If the procedure was done leaving the nipple and areolar on a pedicle, lactation may be successful. Women who have had one breast removed surgically can successfully breastfeed, although when the mastectomy is for malignant disease, it may not be recommended because of the potential effect of continued high levels of sex steroids in the system if pregnancy occurs within five years of treatment. It should be discussed with the oncologist. Women who are in the process of treatment for breast cancer during lactation may pump and discard their milk for a few days after chemotherapy then resume feeding until the next treatment. Length of time for discarding varies with the drug employed. The time for complete clearance can be calculated as five times the half life of the drug involved.[16] Many of the cancer drugs have very short half lives so the disruption may be less than 24 hours.

Inverted nipples are the most common anatomic problem identified (Figure 31-7). Although there are stretching exercises that can be done to pull the nipple out, exercises require time, considerable dedication, and a commitment on the part of the mother to this daily manipulation. Some mothers find nipple exercises distasteful. Nipple stimulus prenatally may trigger uterine contractions and premature labor especially in the last few weeks. Another method of treatment for inverted nipples is wearing specially designed plastic shells inside the normal brassiere daily during the last six weeks of pregnancy, beginning with a few minutes a day increasing time worn to 8–10 hours after about two weeks. The continued gentle pressure on the areola, stretching the fibrous tissue, will evert the nipple through the central hole. After delivery, these shells can be worn between feedings (but not during) until the eversion is firmly established postpartum and the nipple is easily grasped by the infant. A controlled study by Alexander and colleagues found that the technique was not very effective and it often discouraged some women from even initiating breastfeeding.[41] Inverted or small nipples may best be everted by using a good hand pump or an electric pump just prior to putting the infant in position to latch on for a feeding in the first few days postpartum. Usually, the nipple will remain erect without pumping after a week of these efforts. In subsequent pregnancies, the nipples are more everted probably due to the stretching of the fibers that were tying the nipple down initially.



Figure 31-7 Breast shell in place inside a brassiere to evert the nipple. Reproduced with permission from Lawrence and Lawrence.[10]

Only gentle face soap and clear water are needed for breast care. No ointments or lotions are advised prophylactically as they irritate the skin and plug the natural pores, inhibiting the natural secretions. The sebaceous secretions of the glands of Montgomery on the areola are intended to lubricate the areola and nipple. Buffing the tissues briskly with a turkish towel or a toothbrush is neither necessary nor recommended. In a very dry climate where skin dryness is a problem, a bland ointment such as A and D ointment or purified lanolin might be prescribed in some cases.

Removing colostrum in the last few weeks of pregnancy by manual expression is not recommended as it may irritate the tissues and cause an early mastitis. Because the colostrum is discarded, it wastes a very valuable commodity, which should be left for the infant. Such manipulation of the breast may also stimulate premature contractions of the uterus. Prior to delivery, the mother should purchase and bring to the hospital a well-constructed nursing brassiere to support the breasts, especially as the milk first comes in. This will alleviate the feeling of heaviness and engorgement. Many women wear a nursing brassiere night and day, especially in the first few weeks postpartum.

A new mother may find it helpful to attend breastfeeding classes prenatally and actually see an infant at the breast before she delivers if she is totally unfamiliar with breastfeeding.[42] Many childbirth classes include breastfeeding in the curriculum. If not, the physician may wish to have the office staff provide that educational service or refer the patient to a

community breastfeeding support group, such as La Leche League.

Initiating Lactation: The First Feed

As soon after birth as possible, preferably within the first hour of life, the infant should be breastfed.[10,11] Once the infant is stable, with the airway clear and respirations established, he can be offered the breast with the mother lying on her side facing the infant, who is also lying on his side. The infant should be held close to the breast. The areola will be soft and compressible. If the mother strokes the center of the infant's lower lip with the nipple, he will quickly root, open the mouth wide, grasp the nipple and areola, and begin to suckle. The nipple and areola elongate to form a teat as they are drawn into the mouth. The infant should grasp well beyond the nipple so as to compress the areola and ductules, which lie under the areola (Figures 31-8 and 31-9). Because nipple size and areola size vary, the infant may not be able to get the entire areola into the mouth. Even at the first feeding, the infant will receive colostrum. The mother should be further instructed in the art of positioning herself comfortably and supporting her breast with her hand. Changing her position at different feedings allows the infant to grasp from different angles.[24,42,43] This will rotate the point of greatest suckling pressure and will evenly distribute the suckling pressure over the entire areola. After the first feed, a mother may lie down or sit up as she chooses. If the nipple is tender, the baby can be held on the right breast as if he were nursing on the left side, that is, facing the mother's right side with feet to her right (or the reverse on the left breast, with the infant facing the mother's left side). The key to correct positioning is having the infant face the breast. The infant can be brought close by moving the mother's arm that is holding the infant and not by pushing the infant's head toward the breast. Pushing the head toward the breast causes the infant to arch back away from the breast which is the natural arching reflex. This results when the back of the head is held. This appears to the mother as if the infant is rejecting the breast.

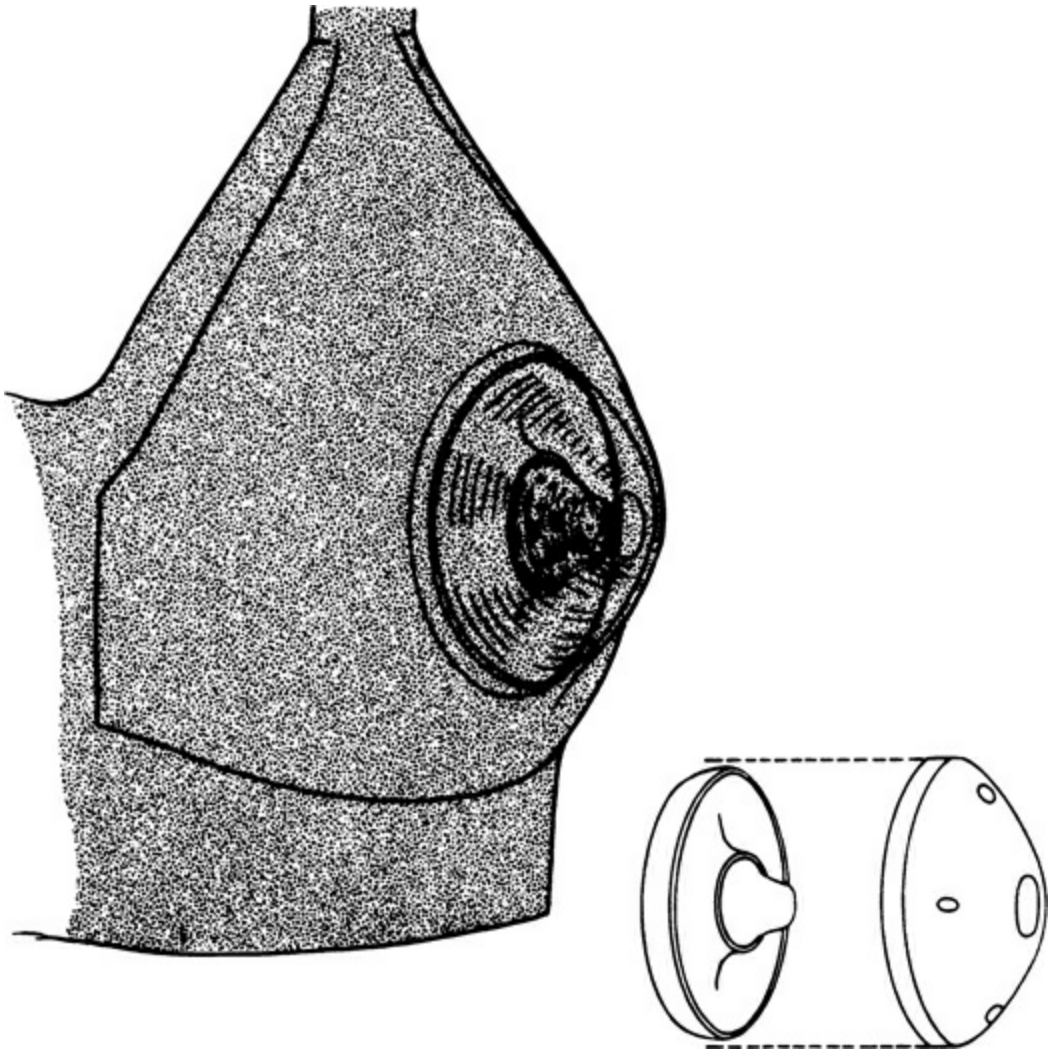


Figure 31-8 (A) As the infant grasps the breast, the tongue moves forward to draw the nipple in. (B) The nipple and the areola move toward the palate as the glottis still permits breathing. (C) The tongue moves along the nipple, pressing it against the hard palate and creating pressure, Ductules under the areola are milked and flow begins as a result of peristaltic movement of the tongue. The glottis closes. Swallow follows. Reproduced with permission from Lawrence and Lawrence.[10]

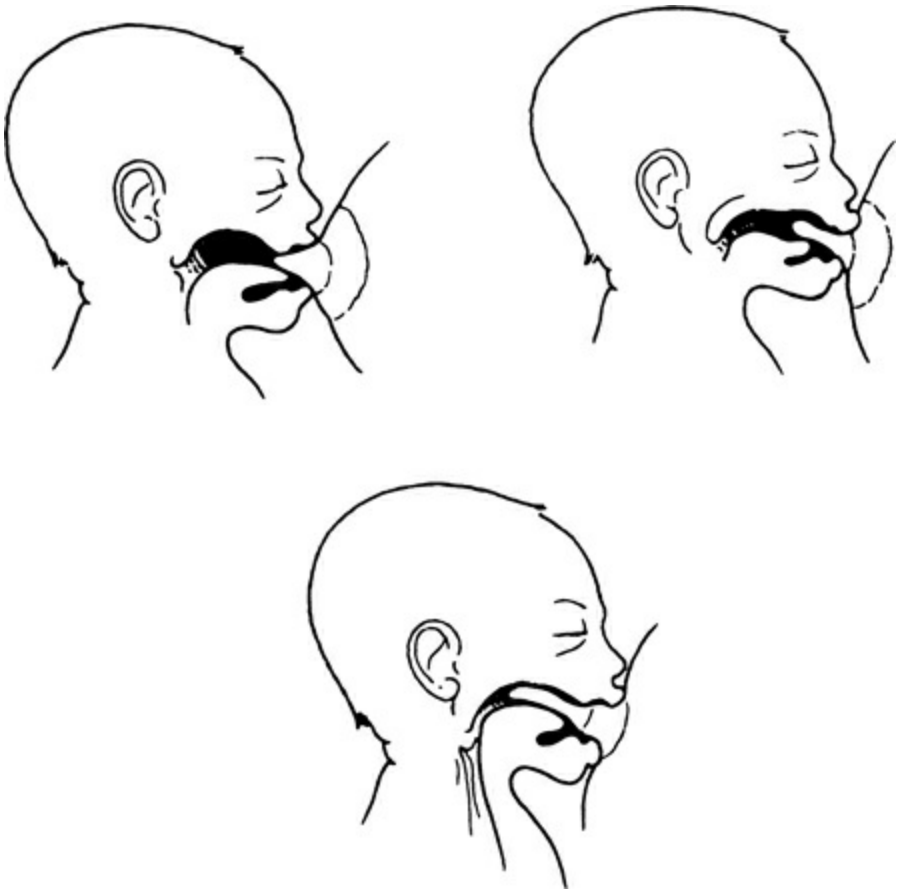


Figure 31-9 Latching on. In response to stimulating the infant's lower lip with the nipple, the mouth opens wide Reproduced with permission from Lawrence and Lawrence.[10]

The position a mother assumes while nursing should be comfortable and relaxing for her. A rocking chair is often the best for the sitting position. It is recommended that a mother may increase her comfort if she varies the hold and orientation of the baby to the breast. This includes not only lying down and sitting up, but holding the baby under the arm in a football hold or across her body so he is held by the left arm at the right breast or the reverse.[16,43,53] The infant should always be facing the breast directly regardless of the position of the rest of the body and the back of the head should not be handled.

Initially, a mother may offer both breasts at each feeding to stimulate each breast as often as possible during the first weeks. The infant, however, should nurse long enough on the first side to receive the hind milk, that is, over five minutes. In reality, he may drift off to sleep before

being switched to the second side. At the next feeding, he should be offered the other breast first. This will balance the stimulus and, thus, milk production. The infant should nurse every time he awakens and is alert and hungry, which may be as frequently as every two hours. Intervals between feedings should not be greater than four to five hours in the beginning when frequent stimulus is critical to establishing a good milk supply. If the infant sleeps six hours, he should be awakened in the first few weeks of life. Having the mother and baby cared for in close proximity as in rooming-in or by mother-baby nursing staff assignments will facilitate frequent appropriate feeding and will enhance milk production. In programs where infants are fed more, on average, 8–12 times a day than six times daily (average 10–12 times), the length of each feeding tends to be shorter. With frequent feeding, there is better milk production, less weight loss, earlier regain of birth weight, and less neonatal jaundice.[44–48] This increase in feeding frequency has not been associated with an increase in sore nipples. Sore nipples are associated with inappropriate positioning at the breast. Care should be taken not to overwhelm the mother with many suggestions for different positions, alternate hand grips, and other angles for the infant. She should find a simple way that works before leaving the hospital. If there is a problem, then different approaches can be suggested. The infant should feed when hungry with no rules for timing or intervals. Crying is a late sign of hunger. Every mother should be observed feeding her baby by a skilled observer before discharge.

Healthy mothers and their infants are being discharged in 48 hours or less in sharp contrast to the —four to five day stay of the past. Mothers with cesarean sections may leave in 36 hours. Having a helpless newborn totally dependent on a mother is an awesome, frightening, and sometimes discouraging responsibility. The mother is no longer an independent person. This responsibility may be overwhelming unless care is taken to “mother the mother” because our culture does not automatically provide maternal support.[49] In fact, our culture programs a superwoman concept in which the new mother must return to her other household chores unless the health professional intervenes. Adequate rest should be prescribed. Discussing the joint responsibilities of parenthood with both parents may facilitate a smoother transition from the sheltered hospital environment to home. Early discharge home also places a responsibility on the physician to see the breastfed infant in the first week of life at home. Provision for weight checks and assessment of jaundice should involve a home visit or an office visit within two days of discharge.[9,47] Many offices have a nurse practitioner skilled in newborn care and breastfeeding who provides this

service.

Nourishment for the lactating woman should make sense, but nurturance while providing these nutrients is equally important. Raphael has expressed it as the need for a *doula*, which is taken from the Greek language to mean "a helpful friend from across the street."[49] It means that someone must care for the mother, support her efforts to breastfeed, and make her confident in her ability to mother her infant.

Supplementation With Milk or Water

Careful study of weight loss in breastfed and formula fed infants in the first days of life indicates that the breastfed baby does not lose more weight than the formula fed baby when breastfeeding is adequately assisted. Furthermore, infants who are fed frequently at the breast in the first few days begin to gain weight at least by the fourth day. Studies correlating the method of feeding with the level of bilirubin show no significant difference between breastfeeding and formula feeding.[46–48] Studies comparing frequency of feeding in the breastfeeding group show that infants who are fed seven or more times per day have significantly lower bilirubins than those fed six or less times per day. Findings were independent of the total number of minutes per day spent nursing.[45,46] In terms of lactation physiology, the breast produces milk in response to suckling and the removal of milk by suckling or pumping. The greatest volume of milk is obtained in the first 5–10 minutes at each breast. If the infant feeds more frequently, he receives more milk, and the breast produces more in response. Animal studies by Gartner and Herschel suggest a relationship between elevated bilirubin levels and starvation.[47] Weight loss of greater than 7% requires evaluation of the breastfeeding, as does unexplained hyperbilirubinemia.[9,44] The increased use of intravenous fluids during labor or with an epidural, has been associated with increased fluid excretion by the infant and greater weight loss. If, on the other hand, the influence of giving water or milk supplements to babies who are breastfed is scrutinized, it is noted that supplementation, especially with water, is associated with increased weight loss and increased bilirubin levels in the first few days of life.[45–47,50] If the influence of water or milk supplements on babies who are breastfed is investigated from the standpoint of successful establishment of lactation, length of breastfeeding, and reasons for early weaning, it is also noted to be negative. Mothers who add supplements have more difficulty establishing a good milk supply, are more apt to wean early, and give "insufficient milk" as a reason for

weaning. Supplements interfere with successful lactation.[51]

In the first few weeks of lactation, it is important to encourage a feeding program that meets the infant's needs, that is, providing feeding when the infant is awake and hungry (the so-called demand or on-request feeding). This may be 12–16 times per day. Most babies have a period of a few hours when they want to nurse every hour and that is appropriate for several feedings. There is, however, a relationship between the fatigue and stamina of the mother that has to be balanced against the true needs of the infant. A fussy breastfed infant who has been well fed may need to be comforted by someone else. This is an important role for the father. Lactating women may not be able to comfort their own infants without offering the breast because the infant smells the milk and will root even though he is well fed. This sometimes leads to incessant non-nutritive suckling, which may be a drain on the mother's energy resources and traumatic to the nipple. Non-nutritive comforting is a significant need of most infants and can be provided by the father.

An additional side effect of supplementation is the use of a bottle and a rubber nipple, which may lead to nipple confusion on the part of the newborn.[52] The sucking mechanism utilized at the breast is the sucking reflex present at birth. The infant will have much of the areola in the mouth, compressing it against the hard palate as it elongates into a teat, maintaining the seal with the gum and lips. The tongue undulates with a peristaltic motion that also triggers the swallow and initiates peristalsis in the esophagus and the stomach. The nipple is a passive passageway for the milk to exit. When a bottle is used, the infant's jaws do little but hold the nipple in place. There is little undulating of the tongue, and milk flows easily with a little suction created by the seal. The tongue may even be thrust upward to control the flow from the unyielding rubber nipple. Because this is a different position and action, some babies are confused by switching back and forth between breast and bottle especially in the first few weeks or when the infant is slightly premature. When the tongue thrusting of bottle feeding is used with the breast, it pushes the human nipple out of the mouth.

Management at Home

Adjustments at home for a new baby are often amplified when the mother is breastfeeding, because any problem such as fussiness, colic, wakefulness, or night feedings are assumed by the mother to be due to a problem with breastfeeding. Instilling confidence in the mother's ability to care for and nourish her infant is important. Closer surveillance by the

physician in the first few days, however, is necessary to be certain that the new inexperienced mother does not interpret long sleeping periods with little feeding as adequate for proper growth. Successful breastfeeding results in fewer problems and illnesses later. Review of weight status, number of wet diapers (at least six per day), stool pattern (at least three per day in the first month), and feeding pattern is a further check on successful lactation. When a breastfed infant does not stop losing weight by five days, does not produce a stool every day, does not void adequately, or does not regain birth weight by 14 days, aggressive intervention is indicated. The physician needs to evaluate infant and the breastfeeding.[53]

Maternal Nutrition

The nursing mother should have a nutrition check to confirm her appropriate food intake. A lactating woman should have 500 extra calories over the pre pregnancy baseline, 20 extra grams of protein, and a balanced diet. Mothers who are concerned about losing weight should be counseled to consume no less than 1800 kcal/day and to consume adequate vitamins and minerals.[8,54] Maternal weight loss after the initial drop should not exceed 1–1.5 kg per month in the first six months of lactation. The most important dietary increase is calcium and phosphorus, to a total of 1200 mg/day.[55] The neonatal calcium-phosphorus requirement exceeds that of the fetus in the last trimester of pregnancy. Dairy products are the best source, but if these products are not tolerated by the mother, she needs to seek out additional sources in dark green vegetables, nuts, legumes, and certain dried fruits. Dark green leafy vegetables such as kale, cabbage, collards, and turnip greens contain readily available calcium, whereas the calcium in spinach, Swiss chard, and beet greens is bound to oxalic acid and is not absorbable.[8,54] The amount of calcium in the diet will not influence the amount in the milk, but a deficiency will lead to leaching from maternal bone and significant osteoporosis. A large placebo controlled study showed that women tolerate 4000 units of vitamin D per day and their milk levels provide sufficient vitamin D to the nursing infant. No supplements are needed for the first six months. Women need to take added vitamin D in pregnancy to provide adequate supplies to the fetus and infant at birth. The current recommendations recommend a daily intake of vitamin D of 400 IU for all infants and children beginning in the first few days of life.[57] A lactating woman does not need added iron for milk but will need to replace stores lost in pregnancy and parturition. A balanced diet should provide all other nutrients. The quality of the milk

day by day is balanced by intake and stores (Figure 31-10). The strict vegetarian is in jeopardy, however, of causing B_{12} deficiency in her offspring, unless she takes supplements, because B_{12} is not found in nature except in animal protein.



Figure 31-10 Energy use in lactation, showing availability of body stores and dietary sources. Reproduced with permission from Lawrence and Lawrence.[10]

TABLE 31-1 Herbal Teas

Ingredient	Botanical Source	Pharmacologic Principle	Use	Effects
African yohimbe bark, yohimbe	*Corynanthe yohimbe*	Yohimbine	Smoke or drink as stimulant	Mild hallucinogen
Catnip	*Nepeta cataria*	Nepetalactone	Smoke or drink as marijuana substitute	Mild hallucinogen
Gordolobo yerbal	*Senecio douglassi*	Pyrrolizidine alkaloids	Drink	veno-occlusive effects, sedation
Hops	*Humulus*	Lupuline	Smoke or drink	sedation,

	lupulus		as sedative and marijuana substitute	diuresis
Kavakava	*Piper methysticum*	Yangonin, pyrones	Smoke or drink as marijuana substitute	Mild hallucinogen
Kola nut	*Cola spp*.	Caffeine, theobromine, kolanin	Smoke, drink, or take as capsules as stimulant	Stimulant
Lobelia	*Lobelia inflate*	Lobeline	Smoke or drink as marijuana substitute	Mild euphoriant
Mandrake	*Mandragora officinarum*	Scopolamine, hyoscyamine	Drink as hallucinogen	Hallucinogen
Mate	*Ilex paraguayensis*	Caffeine	Drink as stimulant	Stimulant
Mormon tea	*Ephedra nevadensis*	Ephedrine	Drink as stimulant	Stimulant
Nutmeg	*Myristica fragrans*	Myristicin	Drink as hallucinogen	Hallucinogen
Passion flower	*Passiflora incarnate*	Harmine alkaloids	Smoke, drink, or take as capsules as marijuana	Mild stimulant
Periwinkle	*Catharanthus roseus*	Indole alkaloids	Smoke or drink as euphoriant	Hallucinogen
Snakeroot	*Rauwolfia serpentine*	Reserpine	Smoke or drink as tobacco substitute	Tranquilizer
Thorn apple	*Datura stramonium*	Atropine, scopolamine	Smoke or drink as tobacco substitute or hallucinogen	Strong hallucinogen
Valerian	*Valeriana officinalis*	Chatinine, velerine alkaloids	Drink or take as capsules as tranquilizer	Tranquilizer
Wormwood	*Artemisia absinthium*	Absinthe	Smoke or drink as relaxant	Narcotic-analgesic

Adapted from AAP Subcommittee on Hyperbilirubinemia.[44] Reproduced with permission from World Health Organization.[10]

The lactating woman does have increased needs for fluids and thus increased thirst. If a woman selects beverages that contain caffeine or other active pharmacologic principles, it could affect the infant. Beverages that either contain no caffeine or have been decaffeinated are appropriate. With the increasing interest in herbal teas, attention should be given to the

content of such teas.[58] A partial list of products is shown in Table 31-1. Many teas contain very potent glucosides which have negative pharmacologic properties; others are benign and a few even nutritious such as rose hips, which contain vitamin C.

Documenting the consumption of herbal teas by the mother or given to the infant directly should be part of the medical history. Some herbs are reputed to enhance lactation such as fenugreek. The required dose is large and soon the milk and all secretions and the infant smell like maple syrup. It helps some women but not all. There can be a cross allergy to peanuts and chickpeas that may cause colic in the infant. Comfrey has been widely used in midwifery and in lactation but is banned in many countries. The FDA has also issued a warning as its use can cause veno-occlusive disease and even be fatal especially in infants.[59]

STAGES OF BREASTFEEDING

Adaptation

Initially, there is a period of adjustment and adaptation as the mother and baby settle into a reciprocal relationship of supply-and-demand. The infant can be exclusively nourished at the breast for the first 6 months of life. During that time, there may be gradual changes in the feeding pattern as the infant matures and sleeps longer between feedings and also spends more time awake and socializing. Growth spurts are accompanied by a temporary increase in feeding frequency. This may alarm the mother if she has not been alerted to this possibility. Periods of stress or illness in the infant may be marked with temporarily increased suckling, especially non-nutritive suckling for comfort. Human milk meets all the nutrient needs of the infant for the first 6 months except for a select group of women who live in cold climates with little sunshine, have dark pigmented skin, wear occlusive clothing or use sunscreen frequently who may be vitamin D deficient.[56,60] Concern about widespread vitamin D deficiency has resulted in reconsideration of vitamin D requirements. Recommendations from the Center for Disease Control and Prevention suggest supplementing the infants with 400 units vitamin D daily by mouth with vitamin D only preparation for breastfed infants. [57] They should be supplemented as mentioned. Very low-birth-weight infants may need iron. Healthy exclusively breastfed infants do not need iron for the first 6 months of life. When weaning foods are added in the second 6 months, they should be iron containing such as iron supplemented cereal.[62]

Adding Solid Foods

The infant ideally is exclusively breastfed for the first 6 months. The single nutrient needed to add to solid foods in an exclusively breastfed infant is the need for additional dietary iron, thus introduction of iron fortified weaning foods at around 6 months is recommended, although the exact age is poorly defined.[62] At about 6 months of age it is appropriate to begin the addition of solid foods to the infant's diet for nutritional reasons.[62] Learning to take solid foods is also an important developmental milestone that involves a new use of tongue, jaw, and lips—a use that differs from suckling.[63] Beginning to take fluids from a cup is also a developmental task that should be learned around 7 months of age. The infant who is exclusively breastfed to this point needs to explore these activities and develop these skills just as a bottle fed infant would. The fluids can be water, juice, or pumped breast milk. If a mother continues to provide her milk, there is no need to introduce formula. Cow's milk when the infant is younger than 1 year of age is not recommended.

Weaning

To wean is "to transfer the young of any animal from dependence on its mother's milk to another form of nourishment" or "to estrange from former habits or associations" according to the dictionary.[63] The weaning process takes many forms, depending on the mother's schedule and beliefs and the needs of the infant. Some women plan to breastfeed for only a few months "to give the baby a good start"; other mothers wean as soon as solid foods can be started, and some continue to offer the breast for several years, even during a subsequent pregnancy and while feeding a new baby. The appropriate time for weaning should be based on nutritional and psychological needs and developmental milestones. Feeding is an important social as well as nutritional encounter, and eating solids and drinking from a cup are important social accomplishments. This does not mean the infant is taken completely off the breast. In practice, the mother is usually the instigator of weaning. The process ideally is gradual, replacing one feeding at a time with solids and the introduction of a bottle or cup, depending on the infant's age and stage of development. After the adjustment has been made to substitute one feeding, a second feeding is replaced, usually at the opposite time of day. The process is continued until there is only one nursing at night and one in the morning. These two feedings may be maintained for many months or gradually discontinued over weeks. A mother may be able to express milk from the breast for

weeks after the final feeding. An infant who has not weaned by 18–24 months usually does not spontaneously wean until 4–5 years of age.[16]

Emergency weaning because of a crisis such as illness or separation may be stormy for the dyad. The infant may reject the bottle and refuse all nourishment at first. The mother who abruptly weans may experience severe engorgement, pain, and systemic symptoms attributed to the resorption of milk and referred to as “milk fever”. Emergency weaning is facilitated by the assistance of another adult who can initiate the new feeding method and is patient and understanding with the child. Cup feeding with a small medicine cup may be a helpful alternative. In the face of recent natural disasters, the importance of keeping mothers and infants together and supporting breastfeeding as an acceptable, feasible, affordable, safe, and sustainable (AFASS) manner of continuing to provide adequate nutrition to the infant has been reemphasized.[64]

Breastfeeding and the Return to Work

Returning to work has been cited by epidemiologists as a major hurdle in the initiation and duration of breastfeeding.[65,66] Women who need to return to work or to school, often think it is best not to start breastfeeding. Before the industrial revolution, all women worked on the farm or in a cottage industry keeping their children with them. In developing countries today, women carry their infants with them to feed them whenever necessary while working. It was the industrial revolution that separated home and work and made parenting a separate role for women.

More women are employed today outside the home than ever before.[67] Women with children under 6 years of age are the fastest growing segment of the female work force in 2010 (64.2% of women with a child under 6 years old).[68] Even more startling are the number of working mothers with children younger than 3 years of age (61.1%).[68] Of the women who work during pregnancy, over 50% plan to return to work by three months postpartum. These dramatic statistics make it clear that the decision about infant feeding is an important part of this issue.[69,70] Child care also presents another consideration for the women who may well have to choose day care or some form of child care that means her infant will be in close contact with other children. In modern pediatrics, “Day Care Syndrome” is real. It is the increase in number of infections, especially diarrhea, respiratory illness, and otitis media experienced by young infants in day care.

The data are clear that breastfeeding impacts these figures. A

quantitative study has shown that extending breastfeeding from 4 months of age to 6 months decreases the risk of respiratory infection including pneumonia and otitis media even further.[19] The protective properties in human milk (Chapter 30) are even more important for the child exposed to other children early in life while mother works or attends school. A comparison of mothers absenteeism showed that those who were breastfeeding had reduced absenteeism.[69] Looking at illness rates of children whose mothers work, 75% of children who were bottle fed were ill and only 25% of those breastfed had any illness.

The feeding pattern for mothers who work: Ideally, the mother does not return to work for at least 6 weeks so she is able to establish her milk supply before having to add work to her schedule. Mother will have to decide how she will cope.[71] If her job permits her to visit her child several times a day, then she can just feed the infant at the usual times. An employer who has a day care center on the premises makes such an arrangement possible. Professional women who control their own schedules (lawyers, doctors, consultants) may be able to keep the infant on the premises under the care of a baby attendant and feed on demand. For most women, however, their jobs are more rigid and they may have to settle for an opportunity to pump their milk every three or four hours on lunch or coffee breaks and store the milk in a cooler to take home for the next day's feedings. Most women practice pumping at home several weeks ahead of time and store up a supply of milk in the freezer so they do not run out.

Employers such as hospitals, health departments, and family friendly industries like Amoco Chicago, Dow Chemical of Midland, Michigan, and the Los Angeles Department of Water and Power to name a few, have been recognized for their support of "Healthy Mothers and Healthy Babies" and their accommodations for nursing mothers. They provide a room to pump, electric pumps, refrigerators, and in some cases lactation consultants to assist with any breastfeeding issues. This support improves the incidence and duration of breastfeeding for the working woman.[71]

Pumping and Storing Milk

If the employer does not provide pumps, a mother should obtain a pump by either renting or purchasing several weeks in advance of the return to work. All pumps are not equal.[72,73] There are, however, several brands of good portable electric pumps that provide disposable attachments for those parts that contact the breast and the milk. Attachments that allow pumping

both sides simultaneously save time and for some women stimulate more milk release. Other women find double pumping overwhelming and choose to do one side at a time. Hand (manual) pumps are good for stimulating milk release and relieving engorgement but not for large volume pumping for most women. Many hospitals have lactation consultants on staff and a shop or service that rents pumps and sells other breastfeeding devices such as breast pads and storage bottles. Information about local resources should be available on the postpartum floor. If not, a mother can call La Leche League International, 1-800-LALECHE (1-800-525–3243) for a local contact person. After each pumping session, the disposable flanges, tubing and bottles used for pumping should be rinsed with cool water first and then washed in warm soapy water and thoroughly rinsed and air dried. After rinsing with cold water to remove the milk, the equipment may also be washed in an electric dish washer.

The pumped milk should be placed in a glass bottle or a firm plastic polypropylene nursing bottle that can be capped with an airtight seal without a nipple, and then used to feed the infant later. Polyethylene bags are adequate for term baby use. Storage temperatures and times have been carefully studied.[72] The container which should be labeled with name, date, and time should be placed in a refrigerator immediately or in a cooling bag or container with freezer packs if at work or school where there is no refrigerator. It is safe in a cooler bag as long as the packs remain cold (24 hours). Upon arrival home, the bottles should be placed in the refrigerator if it will be used within three days or in the freezer if stored for later use. When milk is pumped at home, milk can be placed in the refrigerator (4 degrees C) immediately and kept for five days. Actually, when there is no alternative or a bottle has been inadvertently left out, milk can be kept in a sterile container at room temperature for eight hours and then used immediately or refrigerated for a day.[16]

If milk is placed in the freezer of the refrigerator that has a separate door, it can be stored for three months if it is placed in the back to avoid thawing and freezing when the door is opened. If milk is placed in a deep freeze (−20°C), it can be kept for 6 months or if at –70°C, it is good for a year or longer.[67,72–74] The impact of freezing on the milk is minimal, destroying only the cells and their function. The effect of refrigeration is also minimal, decreasing the cells and some of their function. Nutrients are unchanged. Storing mother’s milk for her own infant does not require pasteurization. Providing donor milk to another infant does require pasteurization by regulation due to the increase in risk of infection in the present environment. Pasteurization does affect some properties destroying

cells and decreasing lipase activity and some other enzymes (see Table 31-2).[67,74]

Day Care for the Breastfeeding Infant

In choosing a day care service, care should be taken to assure that breastfeeding and breast milk are welcome. When taking an infant to day care, a mother may wish to nurse the infant just before she leaves her child or she may wish to nurse the infant at day care when she has a break from work. Further, she may wish to feed the child before she sets out for home with the infant in the afternoon. There needs to be a place to sit quietly with the infant out of the mainstream of activity. The staff should be prepared to make these accommodations and delay a feeding if mother is going to arrive shortly.

TABLE 31-2 Storage and Use of Pumped Milk for Healthy Term Infants

Place	Length of Time
Refrigerator (4°C)	5 d at home; 3 d in day care
Freezer section (separate door) refrigerator (−20°C)	3 mo
Deep freeze (manual defrost) (−20°C)	6 mo
Commercial deep freeze (−70°C)	1 y
Sterile container @room temperature (23°C) (not ideal but milk need not be discarded)	8 h
Stored in cooler bag with frozen packs (as long as packs are still cold)	Less than 24 h
Thawed previously frozen in refrigerator	24 h

The mother will probably wish to provide her stored breast milk for her infant to receive during the day. It is not necessary for the caretaker to wear gloves to handle the milk or feed the infant. It is, however, appropriate to wear gloves to change any babies' diaper. If the milk was frozen, it can be thawed in the refrigerator at day care or thawed by swirling in a container of warm water. It should not be warmed in the microwave because of possible hot spots and scalding the infant. Microwaving interferes with the anti-infective properties as well as decreasing the vitamin C content. If the infant does not empty the container, it can be refrigerated and fed later unless it has been microwaved.[16] This is not true of formula but the protective factors in

human milk will keep the bacterial count down. The day care attendants should save the containers for reuse by the mother. Thawed breast milk can be maintained in the refrigerator for 24 hours.

The milk containers should be carefully labeled with name and date of collection. The attendant should carefully confirm the name on the container before feeding. Mishaps of giving the wrong milk to the wrong infant do occur. It should be reported to both families and the day care's medical consultant with an incident report. There are no reported cases of injury following such an event.

FAILURE TO THRIVE WHILE BREASTFEEDING

Paralleling the increasing incidence of breastfeeding, there has been an increase in the number of clinical reports, including one in the *Wall Street Journal* and one in the *New York Times*, each describing a few cases of failure to thrive while breastfeeding.[50,75–78] The *New York Times* followed the dramatic story of a teenage mother prosecuted for the death of her eight–week-old breastfed son from starvation. The event followed a series of misadventures and refusal to see the child at a Medicaid clinic.[78] The majority of these cases have reflected a lack of clinical knowledge on the part of the professionals regarding the basic physiology of lactation[77] and a general failure of the health care system to provide an appropriate safety net for new and inexperienced mothers following the current early postpartum discharge practices (hospital stay ≤ two days). As cost drives the healthcare system to earlier and earlier discharge, the risk of infant problems increases since lactation will not be well established prior to discharge.[79,80] The American Academy of Pediatrics has recommended that infants be seen by the pediatrician within a week of discharge but in two days if breastfed.[9]

Failure to thrive in children has been thoroughly reviewed in Chapter 47; however, there are some critical differential factors when the infant is breastfed. Most cases of significant failure to thrive in the breastfed infant manifest themselves in the first few weeks or months of life. There is also an important distinction between failure to thrive and the slow gaining breastfed infant.[51,77] The weight curve of an adequately nourished breastfed infant from birth may well include a weight loss of 6%–8% and the regain of birth weight at 10–14 days in contrast to the formula fed infant who may lose only 3%–4% of birth weight and quickly regain birth weight by 5 to 7 days often beginning on a path to obesity.

The critical clinical distinctions between failure to thrive and slow gaining are enumerated in Table 31-3. The salient points include the slow increase in weight compared to the erratic gaining and losing pattern in failure to thrive. The slow-gaining infant is alert and active, with good skin turgor and muscle tone. He feeds frequently night and day, wets many diapers with pale dilute urine, and has a normal stool pattern. The infant looks scrawny but well.[77]

When the infant who is failing to thrive sleeps long periods between feeds; the lack of feeding may be mistakenly considered satisfied when actually he has starvation inanition. The infant often fed poorly in the first few days or for various other reasons does not stimulate good milk production. Since breast milk production depends on supply-and-demand phenomenon, when the infant sucks weakly, he receives little milk, and thus remains weak from some degree of starvation. This infant also has few wet diapers, the urine is concentrated and described as “strong” by the mother. There are few and small stools, often the green mucus of starvation. The tone and turgor are poor, the cry is weak and infrequent, and the infant looks sick. This may well be a medical emergency requiring hospitalization. The feeding pattern should be evaluated, especially focusing on the length of time spent at the first breast during a feeding to be sure it is long enough to allow the high-fat hind milk to be obtained. Sometimes the pattern of slow gaining can be reversed by limiting a feeding to a single breast to ensure high-fat, high-calorie feeds.[81] Switching back and forth between breasts several times during a feeding do not increase milk supply and can reduce the amount of high calorie fat provided to the infant.[77,81]

The diagnostic work-up of these phenomena requires the same clinical assessment that is appropriate when the infant is not breastfed and for this, the reader is referred to Chapter 47 (Failure to Thrive). Since the breastfeeding infant is part of a synchronous dyad, there are additional considerations in the differential diagnosis.[16] A suggested schema for identifying the cause of the problem is presented in Figure 31-11. The clinician should take a history oriented to the process of lactation. This history should include additional parameters that affect the success of the breastfeeding dyad, such as the mother’s perinatal history, general health, diet, habits, psychosocial state, social support system, and the attitudes of the father and family about breastfeeding. Parameters unique to the breastfed infant include any anatomic or physiologic conditions that would interfere with sucking, which is a critical link in the milk production process.

Difficulties with sucking include anatomic abnormalities that result in mechanical interference with sucking such as cleft lip, cleft palate, hypoplasia of the jaw, macroglossia, ankyloglossia (tongue "tie") and tumors or cysts of the oropharynx. These abnormalities can be identified by physical examination, which includes observation of the infant's suck. There may be neurologic interference resulting in a diminished or absent suck. Events at birth, such as maternal anesthesia or analgesia and fetal anoxia or hypoxia, may contribute to poor suckling in the immediate neonatal period and failure to provide adequate stimulus to the breast to initiate lactation. The ensuing lack of nutrition for the infant leads to hypometabolism and continued lack of vigor. Congenital cardiac anomalies may present in this manner. Other causes of neurologic deficit in sucking include trisomy 13–15, trisomy 21, and neuromuscular syndromes such as Werdnig-Hoffmann, neonatal myasthenia gravis, and congenital muscular dystrophy. Hypothyroidism, prematurity, and congenital intrauterine viral infections contribute to poor suck and lack of vigor. The greatest number of infants, however, are entirely normal but have not had sufficient assistance in establishing the proper grasp of the breast, and possibly have been further confused by being given a bottle supplement, which continues to confound their learning experience.[52] In addition to examining the infant and the maternal breast, the clinician should observe the feeding dynamics.[79,80]

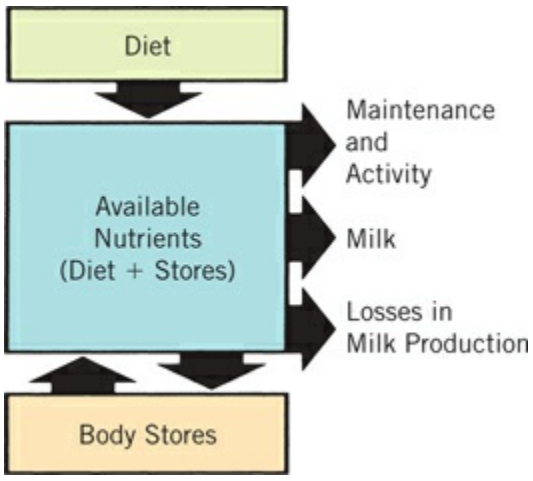


Figure 31-11 Diagnostic flow chart for failure to thrive. Reproduced with permission from Lawrence and Lawrence.[10] CNS, central nervous system; SGA, small for gestational age.

TABLE 31-3 Differential Diagnosis in Poor Weight Gain	
Slow Gainer	Failure to Thrive
Alert, healthy appearance	Apathetic or crying
Good muscle tone	Poor tone
Good skin turgor	Poor turgor
At least six wet diapers daily	Few wet diapers
Pale unconcentrated urine	“Strong” urine
Stools frequent and seedy (or, if infrequent, large and soft)	Stools infrequent and scanty
8 or more nursings daily lasting 15–20 min	Fewer than 8 feedings, often brief
Well-established let-down reflex	No signs of functioning let-down reflex
Weight gain consistent but slow	Weight erratic (loss may occur)

Reproduced with permission from World Health Organization.[10]

All physicians who counsel breastfeeding mothers should be knowledgeable about normal sucking at the breast so that observation of lactation in a diagnostic situation can be constructive. The style with which the mother approaches a feeding, her body language, may be a clue. If she is relaxed, confident, loving, and gentle with her infant, it suggests it is not maternal inexperience at fault. Her verbal interaction can be revealing. A baby suckling at the breast brings reflexive eye-to-eye contact, stroking, and verbal nuances that a seasoned lactating woman utters without consideration for the environment. The insecure, inexperienced mother will sit tensely, offering the breast gingerly, with little or no verbal communication to the infant. If the process is mechanical or punctuated by unrealistic commands to the infant, it may suggest an inability to help the infant root, grasp, and suckle properly. Rigidly timed feedings that are scheduled by the clock may result in poor milk production. The treatment rests with frequent on-request feedings that fit the infant’s biologic rhythms. Suggesting a quiet room, a rocking chair, soft music, or a relaxing beverage for the mother may all improve the situation.

The behavior of the infant when offered the breast may indicate an infant with a suckling disorder, not associated with any other neurologic symptom or long-range problem. Sucking inadequately at the breast can be altered so the infant learns the technique. The infant is identified when it is noted the infant cannot maintain the breast in the mouth unless his mother holds it there. In other words, when she takes her hand away, the breast

falls away. A normal infant sucks without help from his mother's hands if the grip is proper and the seal is adequate. When the infant does begin to suck when the breast is held in position, the suck may be a flutter or ineffective tongue actions. This may be improved by having the mother hold the breast between thumb and index finger, with fingers under the breast (palmar hold) (Figure 31-12) rather than with areolar compressed between the middle and index finger (scissor hold) (Figure 31-13). The infant's position should be adjusted so his body is turned toward the mother's body (instead of just turning his head). Thus, the breast is centered toward the infant, and this position will improve the effectiveness of the infant's efforts. The mother may have to continue to hold the breast in place for weeks until the infant perfects his technique. The mother may also have to pull his lower lip down to keep the infant from drawing the lip into the mouth and moving it along the lower surface of the breast. The lip should be held as part of the seal holding the breast in place and permitting development of some negative sucking pressure. If the mother stimulates the rooting reflex by stroking the center of the lower lip, the infant will open wide and draw the nipple and areola into the mouth to form a teat.

While the infant is learning to suckle properly, it is urgent to avoid introduction of a rubber nipple on a bottle or a pacifier.[52,79,80] This poses a problem if adequate nutrition is critical and the mother's supply needs to be stimulated to be adequate. A trial of frequent feeds, waking the infant every two hours, may suffice. Extra calories may be offered by medicine cup or Haberman feeder. When the failure to thrive has reached critical starvation, a more aggressive approach is mandatory. If hospitalization is necessary, intravenous therapy to treat dehydration may also be necessary. Hypernatremia and hypochloremia have been described, and a complete work-up including pH, electrolytes, BUN, and creatinine are essential.[29,50,53,82,83] While the infant receives intravenous therapy, the mother should be assisted to pump frequently to develop and increase her milk supply. When it is safe to begin oral feedings, the infant should be exclusively breastfed as far as sucking is concerned, and additional nourishment should be provided by intravenous line, gastric tube, or medicine cup. Thus, the infant avoids the introduction of a bottle. When the crisis has abated and full breastfeeding is appropriate, but the milk production is still inadequate, the use of a nursing supplementer (Lact-Aid) may be useful. This device permits the uninterrupted nursing at the breast while supplementary nourishment is provided via a fine capillary tube that runs along the breast into the infant's mouth (Figures 31-14 and 31-15). The tube brings the supplementary fluid from a reservoir plastic bag that

hangs around the mother's neck. The system is carefully engineered. It provides fluid only when the baby sucks; thus, it coordinates with the infant's swallowing mechanism. It is not a siphon or a pump. When used to help establish or increase milk production as with a premature infant first going to breast, the infant is usually weaned from the supplementer within one or two weeks by providing smaller and smaller volumes of supplement as maternal production increases. The supplementing device may make the critical difference when the degree of starvation is great and lactation is being preserved. It is important to point out that all too often the infant is quickly weaned to a bottle without any effort to solve the underlying lactation problem, which is unfortunate (Figures 31-14 and 31-15).

In rare cases of failure to thrive while breastfeeding, the underlying cause is actually metabolic and the infant does even less well on formula prepared from cow's milk or soy protein. In that case, it is quite possible to have the mother re-lactate. It is an art practiced in most cultures over the centuries and resorted to when the biologic mother became ill or died and another female (often the grandmother) had to assume the nourishment of the baby. In the case of premature weaning, there has already been the biologic stimulus of pregnancy and early lactation so reinstituting the milk supply occurs more easily. The lactation supplementer may be of great value in this situation, as the infant can return to the breast and be nourished with donor human milk while the mother builds up her own supply (see Figure 31-15).

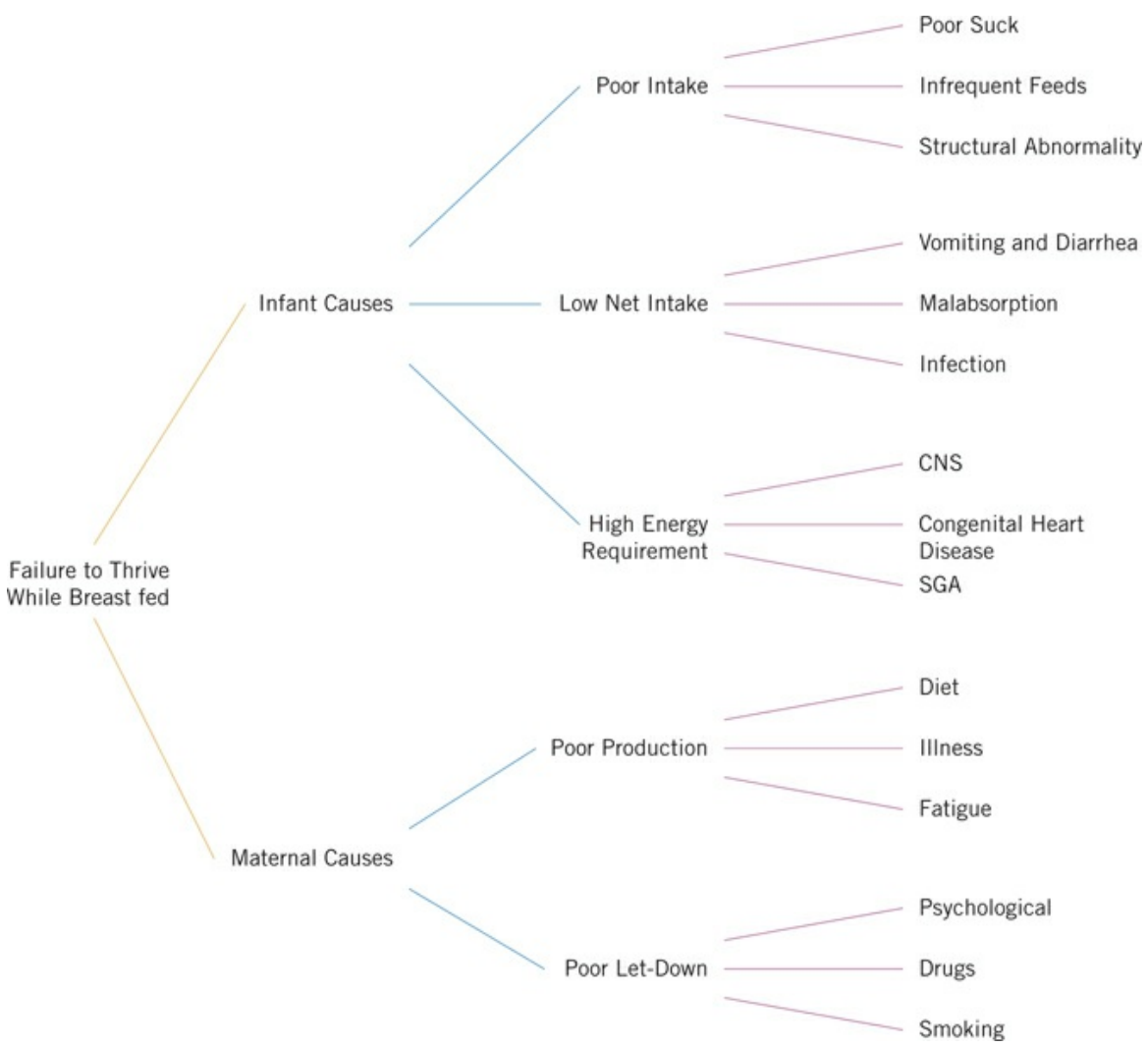


Figure 31-12 Palmar grasp (C-hold). When the palm and fingers cup the breast with support and the thumb rests lightly above the areola, the nipple projects straight ahead or slightly downward (correct). Reproduced with permission from Lawrence and Lawrence.[10]

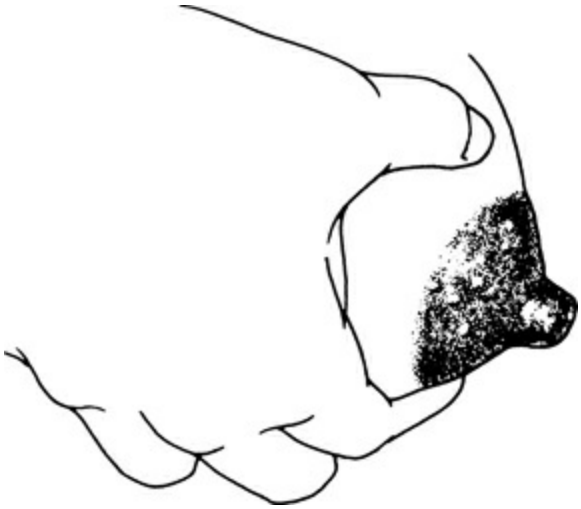


Figure 31-13 When the breast is offered to the infant, the areola is gently compressed between two fingers and the breast supported to ensure that the infant is able to grasp the areola adequately. Reproduced with permission from Lawrence and Lawrence.[10]



Figure 31-14 Lact-Aid Nursing Trainer System (Lact-Aid International, Inc.). Reproduced with permission from Lawrence and Lawrence.[10]



Figure 31-15 Lactation Supplementry by Medela, which provides additional nourishment to the infant while it suckles at the underproducing breast. Reproduced with permission from Lawrence and Lawrence.[10]

Maternal Causes of Failure to Thrive

Poor milk production may be the cause of the failure. This is usually characterized by an alert, active infant who cries hungrily and is very demanding but never satisfied. This baby demands attention and is usually

seen by the physician because of his dissatisfaction. The quiet, sleepy, starved baby gets into serious trouble before he is discovered because his sleeping is interpreted as satiation. It is rare in the United States that diet is the true cause of insufficient milk although it is appropriate to evaluate the mother's diet and make recommendations for increases or adjustments where needed (see Figure 31-11).[84] An additional 600 kcal or a minimum of 1800 kcal/d, a balance of foods with 20 gm extra protein and 400 mg extra calcium, is minimal for every mother. Many mothers feel better taking Brewer's yeast. "Mothering the mother" by caring about her diet may have a positive effect. However, the major factor in poor production is fatigue. It is the single most important element in milk production. The present day "super-mom" model that has been developed by women may be the actual destructive element. A postpartum woman needs rest to recover whether she nurses or not. When she is also nourishing an infant she needs more rest. This is often neglected when the infant needs care every three to four hours around the clock, and only the mother is involved in the feeding of the infant. When her physician suggests that the mother needs to reorder her priorities and schedule naps for herself, it may be the necessary official approval she needs to do so. A mother may need to be told it is not only okay but it is necessary for her to take care of herself in order to provide for her baby. The physician may need to prescribe rest as well as nourishment.

There is a small number of women who are unable to make sufficient milk. Some of these women have inadequate glandular tissue. Markedly asymmetric breasts, conical-shaped breasts, and extremely small ones may be in this category. Even extremely large breasts are occasionally nonfunctional. Failure of the breasts to change and enlarge during pregnancy and/or failure of the breasts to become engorged immediately postpartum are signs of inadequately functioning tissue. These signs prenatally should alert the medical team to extra vigilance as lactation is initiated.

Failure to Let Down Milk

A woman may make milk abundantly but be unable to release it. As the practitioner observes the lactation process, evidence of successful let down should be sought. If the sucking is interrupted by breaking the suction (by putting a finger in the corner of the infant's mouth), milk should continue to flow in a steady drip if not a stream. Although many women describe a tingling, and turgescence when the milk lets down; it is possible to have an effective ejection reflex without these sensations. As indicated in Figure

31-16, it is possible for pain or stress to interfere with let down. If mother has sore nipples or the infant has an improper grasp at the breast, the pain may interfere with let down.[36,16] If the adjustments and remedial actions to avoid stress and enhance confidence do not result in a change in the release of milk, it may be necessary to temporarily provide the oxytocin needed for the let-down arc.[16] Synthetic oxytocin can be prepared by the pharmacist as a nasal spray for home use utilizing the injectable oxytocin. It is packaged in a 5–10 mL nasal dropper bottles. It contains 10 USP units (IU) per milliliter of oxytocin, a polypeptide hormone of the posterior pituitary gland. A prescription is required. It is destroyed in the gastrointestinal tract; therefore, it must be used nasally on the mucous membranes where it is rapidly absorbed. Four to six drops into one nostril followed by having the infant suckle within twoto three minutes is sufficient. This is repeated using the second nares if the infant is switched to the second breast. This may also be used when using a breast pump and collecting for an infant who cannot nurse directly as in the case of a premature baby. Usually, it is only necessary to use the medication for a few days as the natural process will take over.

A rare finding in lactation failure is the lack of a prolactin surge when the breast is stimulated by the suckling or pumping. The prolactin should double over baseline upon suckling. If prolactin levels are obtained, the samples should be carefully timed so that the baseline sample is drawn from a heparin lock after the mother has recovered from the needle stick. Then she should feed the infant or pump her breasts for 10 minutes, and a second sample should be drawn. The percentage increase in prolactin over baseline should approach 50%. The baseline should be above normal for the laboratory. Replacement prolactin is not clinically available although prolactin stimulation with fenugreek or meclopromide or other galactogues may increase milk supply while the treatment is continued.[16]

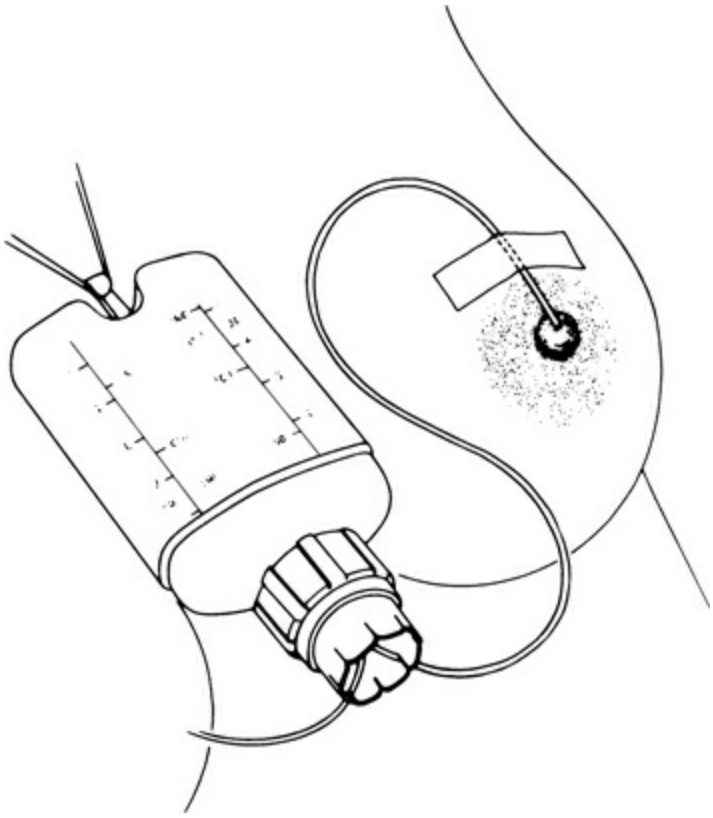


Figure 31-16 Ejection reflex are. When suckling the breast, the infant stimulates mechanoreceptors in the nipple and areola that send a stimulus along nerve pathways to the hypothalamus, which stimulates the posterior pituitary to release oxytocin. It is carried via the bloodstream to the breast and uterus. Oxytocin stimulates myoepithelial cells in the breast to contract and eject milk from the alveolus. It is secreted by the anterior pituitary gland in response to suckling. Stress such as pain and anxiety can inhibit the let-down reflex. The sight or cry of an infant can stimulate it. Reproduced with permission from Lawrence and Lawrence.[10]

Knowing When to Discontinue Breastfeeding

Although breastfeeding provides species-specific nourishment, infection protection, immunologic protection, and psychological benefits for both mother and baby, there are times when it should be discontinued. The role of the physician is a delicate one, one in which true support of breastfeeding is necessary for credibility. On the other hand, the physician must recognize when other alternatives are medically preferable. The mother will need help in accepting this. Having to wean prematurely or

before the planned date is not to be construed as maternal failure. It is still possible to nurture the infant, to be a good mother, and to have a good mother–infant relationship, even though the mother may no longer be able to breastfeed.

The indications for premature weaning are rare but include severe illness in the mother, severe galactosemia in the infant, and a few maternal drugs such as therapeutic doses of radioactive pharmaceuticals.[16,85] An important resource for all healthcare professionals related to drugs and lactation is LactMed, Drugs, and Lactation Database maintained by the National Library of Medicine.[86] Useful resources which healthcare professionals can share with mothers and families are The Nursing Mother's Companion and the New Mother's Guide to Breastfeeding.[87,88]

REFERENCES

1. US Department of Health & Human Services. *Public Service Campaign to Promote Breastfeeding Awareness Launched: Emphasizes "Babies were born to be breastfed"*. Ad Council. http://www.hhs.gov/news. Accessed June 4, 2004.
2. Italian Ministry of Health. *Communication Campaign to Promote Breastfeeding, "Mother's Milk is Never Forgotten"*. http://www.salute.gov.it/servizio/galleriaEnglish.jsp?lang=italiano&id=836&dad=s&men=campagne11&label=stilevita2011. Accessed November 1, 2012.
3. Lawrence RM, Lawrence RA. Breastfeeding: more than just good nutrition. *Pediatr Rev*. 2001;32:267–279.
4. US Department of Health & Human Services. *National Breastfeeding Week 1st–7th October 2012 "Good health begins with breastfeeding"*. http://www.healthpromotion.ie/index.php/newsView/national_breastfeeding_week_1 Accessed October 23, 2012.
5. Fourth Annual Summit on Breastfeeding. First food: the essential role of breastfeeding. *Breastfeed Med*. 2012;7:321–389.
6. Centers for Disease Control and Prevention, Division of Nutrition, Physical Activity and Obesity. *Breastfeeding Report Card–United States, 2012*. http://www.cdc.gov/breastfeeding/pdf/2012BreastfeedingReportCard.pdf. Accessed October 23, 2012.
7. U.S. Department of Health and Human Services. *Healthy People 2020, Topics and Objectives, Maternal, Infant and Child Health: MICH-21. 2012;214–215*. http://www.healthypeople.gov/2020/topicsobjectives2020/pdf/HP2020objectives.pdf. Accessed October 24, 2012.
8. Committee on Nutrition During Lactation. *Report: Nutrition During Lactation, Food and Nutrition Board, Institute of Medicine, National Academy of Sciences*. Washington, DC: National Academy Press; 1991.
9. American Academy of Pediatrics, Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129:e287–841. http://pediatrics.aappublications.org/content/early/2012/02/22/peds.2011–3552. Accessed October 24, 2012.

10. World Health Organization, Family and Reproductive Health, Division of Child Health and Development. *Evidence for the Ten Steps to Successful Breastfeeding*. Geneva, Switzerland: WHO: Publication # WHO/CHD/98.9; 1998.
11. World Health Organization. *Protecting, Promoting and Supporting Breastfeeding: The Special Role of Maternity Services. A Joint WHO/UNICEF Statement*. Geneva, Switzerland: WHO; 1989.
12. Horwood LJ, Fergusson DM. Breastfeeding and later cognitive and academic outcomes. *Pediatrics*. 1998;101:91–97.
13. Rogan WJ, Gladen BC. Breastfeeding and cognitive development. *Early Human Dev*. 1993;31:181–193.
14. Temboury MC, Otero A, Polanco I, et al. Influence of breastfeeding on infant's intellectual development. *J Pediatr Gastroenterol Nutr*. 1994;28:32–36.
15. Lawrence RA. *A Review of the Medical Benefits and Contraindications to Breastfeeding in the United States. Maternal and Child Health Technical Information Bulletin*. Arlington, VA: National Center for Education in Maternal Child Health; 1997.
16. Lawrence RA, Lawrence RM. *Breastfeeding: A Guide for the Medical Profession*. 7th ed. Maryland Heights, MI; Elsevier Mosby, Inc.; 2011.
17. Ip S, Chung M, Raman G, et al. Breastfeeding and maternal and infant health outcomes in developed countries. *Evid Rep Technol Assess*. 2007;153:1–186.
18. Ip S, Chung M, Chung G, et al. A summary of the Agency for Healthcare Research and Quality's evidence report on breastfeeding in developed countries. *Breastfeed Med*. 2009;4(suppl 1):S17–S30.
19. Chantry CJ, Howard CR, Auinger P. Full breastfeeding duration and associated decrease in respiratory tract infection in U.S. children. *Pediatrics*. 2006;117:425–432.
20. Davis MK. Breastfeeding and chronic disease in childhood and adolescence. In: Schanler RJ, ed. *Breastfeeding 2001, Part I, The Evidence for Breastfeeding. Pediatr Clin North Am*. 2001;48:125–141.
21. Hanson LA. Human milk and host defense: immediate and long term effects. *Acta Paediatr Suppl*. 1999;88:42–46.
22. Wilson A, Stewart Forsyth J, Greene S, et al. Relation of infant diet to childhood health: seven year follow up at a cohort of children in Dundee infant feeding study. *BMJ*. 1998;316:21–25.
23. Ryan AS, Wenjun Z, Acosta A. Breastfeeding continues to increase into the new millennium. *Pediatrics*. 2006;110:1103–1109.
24. Chandran L, Gelfer P. Breastfeeding: the essential principles. *Pediatri Rev*. 2006;27:409–417.
25. Neville MC. The physiological basis of milk secretion. Part I: basic physiology. *Ann NY Acad Sci*. 1990;586:1–11.
26. Vorherr H. *The Breast-Morphology, Physiology, and Lactation*. New York, NY: Academic Press, Inc.; 1974.
27. Neville MC. Physiology and endocrine changes underlying human lactogenesis II. *J Nutr*. 2001;131:3005S-3008S.
28. Neville MC, Morton J, Umemura S. Lactogenesis: transition from pregnancy to lactation. In: Schanler RJ, ed. *Breastfeeding 2001, Part I, The Evidence for Breastfeeding. Pediatr Clin North Am*. 2001;48:35–52.
29. Asnes RS, Wisotsky DH, Migel PF, et. al. The dietary chloride deficiency syndrome occurring in a breastfed infant. *J Pediatr*. 1982;100:923–924.
30. Ramsay DT, Kent JC, Hartmann RL, Hartmann PE. Anatomy of the lactating human breast redefined with ultrasound imaging. *J Anat*. 2005;206:525–535.

31. Ramsay DT, Mitoulas LR, Kent JC, et al. Milk flow rates can be used to identify and investigate milk ejection in women expressing breast milk using an electric breast pump. *Breastfeeding Med*. 2006;1:14–23.
32. Love SM, Barsky SH. Anatomy of the nipple and breast ducts revised. *Cancer*. 2004;101:1947–1957.
33. Mendelson EB. The breast. In: Rumack CM, Wilson SR, Charboreau JW, eds. *Diagnostic Ultrasound*. 2nd ed. St. Louis, MO: Mosby-Yearbook; 1998:751–789.
34. Ramsay DT, Kent JC, Owens RA, Hartmann PE. Ultrasound imaging of milk ejection in the breast of lactating women. *Pediatrics*. 2004;113:361–367.
35. Cooper AP. *Anatomy of the Breast*. London, UK; Longman, Orme, Green, Browne and Longmans; 1840.
36. Newton M, Newton NR. The let-down reflex in human lactation. *J Pediatr*. 1948;33:698–704.
37. Winikoff B, Baer EC. The obstetrician's opportunity: translating "breast is best" from theory to practice. *Am J Obstet Gynecol* 1980;138:105–117.
38. Newton NR. Psychologic differences between breast and bottle feeding. *Am J Clin Nutr* 1971;24:993–1004.
39. Uvnas-Moberg K, Eriksson M. Breastfeeding: physiological, endocrine and behavioral adaptations caused by oxytocin and local neurogenic activity in the nipple and mammary gland. *Acta Paediatr*. 1996:85:525–530.
40. Moberg, Kerstin Uvnäs. The *Oxytocin Factor: Tapping the Hormone of Calm, Love and Healing*. Cambridge, UK: Da Capo Press; 2003.
41. Alexander JM, Grant AM, Campbell MJ. Randomized controlled trial of breast shells and Hoffman's exercises for inverted and non-protractile nipples. *Br Med J*. 1992;304:1030–1032.
42. Slusser W, Powers NG. Breastfeeding update 1: immunology, nutrition, and advocacy. *Pediatr Rev*. 1997;17:111–119.
43. Powers NG, Slusser W. Breastfeeding update 2: clinical lactation management. *Pediatr Rev*. 1997;18:147–161.
44. AAP Subcommittee on Hyperbilirubinemia. Management of hyperbilirubinemia in the newborn infant 35 or more weeks of gestation. *Pediatrics*. 2004;114(1):297–316.
45. DeCarvalho M, Hall M, Harvey D. Effects of water supplementation on physiological jaundice in breastfed babies. *Arch Dis Child*. 1981;56:568–569.
46. DeCarvalho M, Klaus MH, Merkatz RB. Frequency of breastfeeding and serum bilirubin concentration. *Am J Dis Child* 1982;136:737–738.
47. Gartner LM, Herschel M. Jaundice and breastfeeding. In: Schanler RJ, ed. *Breastfeeding 2001, Part II, The Management of Breastfeeding*. *Pediatr Clin North Am*. 2001;48:389–399.
48. Hansen TW. Bilirubin production, breastfeeding and neonatal jaundice. *Acta Paediatr*. 2001;90(7):716–717.
48a. Chantry CJ, Nommsen-Rivers LA, Peerson JM, et al. Excess weight loss in first-born breastfed newborns relates to maternal intrapartum fluid balance. *Pediatrics*. 2011;127;e171.
49. Raphael D. *The Tender Gift: Breastfeeding*. New York, NY: Schocken Books; 1976.
50. Gilmore HE, Rowland TN. Critical malnutrition in breastfed infants: three case reports. *Am J Dis Child*. 1978;132:885–887.
51. Powers N. Slow weight gain and low milk supply in the breastfeeding dyad. *Perinatology*. 1999;26:399–430.
52. Neifert MR, Lawrence R, Seacat J. Nipple confusion: toward a formal definition. *J Pediatr*. 1995;126:125–129.
53. Neifert MR. Prevention of breastfeeding tragedies. In: Schanler RJ, ed.

Breastfeeding 2001, Part II, The Management of Breastfeeding. *Pediatr Clin North Am*. 2001;48:273–297.
54. Committee on Nutrition During Pregnancy and Lactation. *An Implementation Guide. Institute of Medicine, National Academy of Sciences*. Washington, DC: National Academy Press; 1992.
55. Institute of Medicine. *Dietary Reference Intakes for Calcium, Phosphorus, Magnesium, Vitamin D and Fluoride*. Washington, DC: National Academy Press; 1999.
56. Wagner C, Greer FR, the Section on Breastfeeding and the Committee on Nutrition. Prevention of rickets and vitamin D deficiency in infants, children and adolescents. *Pediatrics*. 2008;122:1142–1152.
56a. Wagner CL, Howard CR, Lawrence RA, et al. Maternal vitamin D supplementation during lactation: a viable alternative to infant supplementation. *Breastfeeding Medicine* 2006.
57. CDCP, Division of Nutrition, Physical Activity and Obesity. *Vitamin D Supplementation*. http://www.cdc.gov/breastfeeding/recommendations/vitamin_d.htm. Accessed October 25, 2012.
58. Siegel, RK. Herbal intoxication. *JAMA*. 1976;236:473–476.
59. U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition. *FDA Advises Dietary Supplement Manufacturers to Remove Comfrey Products from the Market*. www.cfsan.fda.gov/~dms/supplmnt.html. Accessed July 6, 2001.
60. Greer FA. Do breastfed infants need supplemental vitamins? In: Schanler RJ, ed. *Breastfeeding 2001, Part II, The Management of Breastfeeding*. *Pediatr Clin North Am*. 2001;48:415–423.
61. Griffin IJ, Abrams SA. Iron and breastfeeding. In: Schanler RJ, ed. *Breastfeeding 2001, Part II, The Management of Breastfeeding*. *Pediatr Clin North Am*. 2001;48:401–414.
62. World Health Organization. Global strategy for infant and young child feeding: the optimal duration of breastfeeding. 54th World Health Assembly, May 1, 2001. http://apps.who.int/gb/archive/pdf_files/WHA54/ea54id4.pdf. Accessed March 8, 2015.
63. The World Encyclopedia Americana. *Funk & Wagnalls Standard Dictionary*. Vol 2. New York, NY: Funk & Wagnalls; 1960
64. AAP, Section on Breastfeeding. *Infant Nutrition During a Disaster*. http://www2.aap.org/breastfeeding/files/pdf/InfantNutritionDisaster.pdf. Accessed October 25, 2012.
65. Fein SB, Roe B. The effect of work status on initiation and duration of breastfeeding. *Am J Public Health*. 1998;88:1042–1046.
66. United States Breastfeeding Committee. *Workplace Breastfeeding Support*. Raleigh, NC: United States Breastfeeding Committee; 2002.
67. Lawrence RA. Milk banking: the influence of storage procedures and subsequent processing on immunological components of human milk. In: Draper HH, Woodward W, eds. *Advances in Nutritional Research*. New York, NY: Kluwer Academic/Plenum Publishers; 2001:389–404.
68. US Department of Labor. *Women in the Labor Force: A Databook*. http://www.bls.gov/cps/wlf-databook-2011.pdf. Accessed October 25, 2012.
69. Cohen R, Mrtek MB, Mrtek RG. Comparison of maternal absenteeism and infant illness rates among breastfeeding and formula-feeding women in two corporations. *Am J Health Promotion*. 1995;10:148–153.
70. Meek JY. Breastfeeding in the work place. In: Schanler RJ, ed. *Breastfeeding 2001,*

Part II, The Management of Breastfeeding. *Pediatr Clin North Am*. 2001;48:461–474.
71. Cohen R, Mrtek MB. The impact of two corporate lactation programs on the incidence and duration of breastfeeding by employed mothers. *Am J Health Promotion*. 1994;6:436–441.
72. Slusser W, Frantz K. High-technology breastfeeding. In: Schanler RJ, ed. *Breastfeeding 2001, Part II, The Management of Breastfeeding*. *Pediatr Clin North Am*. 2001;48:505–516.
73. Zinaman M, Hughes V, Queenan J, et al. Acute prolactin and oxytocin responses and milk yield to infant suckling and artificial methods of expression in lactating women. *Pediatrics*. 1992;89:437–440.
74. Lawrence RA. The storage of human milk and the influence of procedures on immunological components of human milk. *Acta Paedicatr Suppl*. 1999;88(430):14–18.
75. Helliker K. Dying for milk. *Wall St J*. 1994;CCXXIV:15.
76. Roddey OF, Martin ES, Swetenburg RL. Critical weight loss and malnutrition in breastfed infants. *Am J Dis Child*. 1981;135:597–599.
77. Kent JC, Ramsey DT, Cregan MD, et al. Volume and frequency of breastfeeds and fat content of breastmilk throughout the day. *Pediatrics*. 2006;117:e387–e395.
78. Editorial. *Breastfeeding as Manslaughter*. *New York Times*, March 18, 1999 http://www.nytimes.com/1999/03/18/nyregion/metro-news-briefs-new-york-report-on-baby-s-death-prompts-delay-in-trial.html?ref=topics. Accessed March 8, 2015.
79. Howard CR, Howard FM, Lanphear B, et al. The effects of early pacifier use on breastfeeding duration. *Pediatrics*. 1999;103:e33.
80. Righard L. Are breastfeeding problems related to incorrect breastfeeding technique and use of pacifiers and bottles? *Birth*. 1998;25:40–44.
81. Woolridge MW, Ingram JC, Baum JD. Do changes in pattern of breast usage alter the baby's nutrient intake? *Lancet*. 1990;336:395–397.
82. Anand SK, Sandborg C, Robinson RG, Lieberman, E. Neonatal hypernatremia associated with elevated sodium concentration of breast milk. *J Pediatr*. 1980;96:66–68.
83. Rowland TW, Zori RT, LaFleur MD, Reiter EO. Maintenance and hypernatremic dehydration in breastfed infants. *JAMA*. 1982;247:1016–1017.
84. USDA/DHHS. *Dietary Guidelines for Americans, 2010*. http://health.gov/dietaryguidelines/2010.asp. Accessed November 4, 2012.
85. American Academy of Pediatrics Committee on Drugs. Drugs in breastmilk. *Pediatrics*. 2001;108:776
86. Toxicology Data Network of the National Library of Medicine. *LactMed: Drugs and Lactation Database*. http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen? LACTMED. Accessed March 8, 2015.

SUGGESTED READINGS

87. Huggins K. *The Nursing Mother's Companion*, 6th ed. Harvard and Boston, MA: The Harvard Common Press; 2010.
88. Meek JY, ed. *The New Mother's Guide to Breastfeeding*. AAP Section on Breastfeeding, 2nd ed. New York, NY: Bantam Books; 2011.

The Low-Birthweight Infant: Inpatient Care

32

Thibault Senterre, MD, PhD, FRCPC and Alexandre Lapillonne, MD, PhD

INTRODUCTION

The major objective when caring for low-birth-weight (LBW, <2500g) infants is to allow them to grow and develop adequately. In industrialized countries, most LBW infants who are admitted in neonatal units are small because of prematurity. In contrast, and especially in resource-poor countries, some LBW infants are born at term but have suffered from intrauterine growth restriction (IUGR). Their duration of hospitalization is usually short and their nutritional support is similar in practice from that of moderately preterm infants. Therefore, specific inpatient care of term LBW infants with IUGR falls out of the scope of this chapter that focuses on preterm infants.

Recent advances in perinatal medicine and in neonatal intensive care have dramatically increased the survival of preterm infants, especially that of very low–birth-weight (VLBW, <1500g) and extremely low-birth-weight (ELBW, <1000g) infants. Unfortunately, most of these infants develop an extrauterine growth restriction (EUGR) due to inadequate or insufficient nutritional intakes. Nowadays, this problem has emerged as a frequent co-morbidity that is associated with poorer short but also long-term outcomes.[1–3]

Nutritional requirements for preterm infants need to consider: (1) fetal growth according to gestational age (GA), focusing on lean body mass

growth and accretion of proteins, minerals, and various essential nutrients during the last trimester of gestation; (2) the risk of cumulative nutrient deficit acquired during the first weeks of life; (3) the opportunity for early catch-up growth resulting from IUGR; (4) the use of a relatively immature gastrointestinal tract to provide nutrition; and (5) the need to adapt the recommendations in relation to GA.

This chapter aims to highlight the current knowledge on the nutritional requirements of preterm infants and to emphasize on how to optimize practices by using either the enteral and parenteral route in order to reduce the incidence of EUGR and to promote optimal health of very preterm infants.

INPATIENT GROWTH

The Fetus as the Reference for Growth

Optimal growth is generally defined by the growth that is expected to be observed with optimal nutritional intakes and adequate environment. It varies in each individual according to genetic potential and health status. For the preterm infants, the objective generally admitted is to have a growth similar to that of the fetus up to the due date and similar to that of the breastfed term infant afterward.[4–6] Even if this concept is accepted by most neonatologists all around the world, it remains imperfect because many differences exist between the intrauterine fetus and the extrauterine growing preterm infant, particularly for energy expenditure, metabolic demand, and route of nutrient delivery. Fetal growth rate is extremely high during the third trimester of gestation and is much greater than during any other periods of life. If the mean fetal weight gain during the last trimester of gestation is ~15 g/kg/d, it should be emphasized that fetal weight gain decreases from ~20 g/kg/d at 24–28 weeks of gestation to ~10 g/kg/d at 39–40 weeks (Table 32-1). Furthermore, the quality of the weight gain and the protein accretion rate also changes during the last trimester of pregnancy, and therefore nutritional requirements also vary in relation to GA (Table 32-1).[7]

Measurements of Inpatient Growth

Growth of preterm infants is usually evaluated by plotting the measurement of current weight, length, and head circumference on intrauterine growth charts according to GA up to the expected date of delivery and on term infant growth charts afterward. However, using two

different charts that do not perfectly overlap between 36 and 46 weeks GA make difficult the monitoring of growth during this period. To address this issue, specific gender-related fetal-infant growth charts for preterm infants were recently designed by merging intrauterine growth curves with the World Health Organization (WHO) growth standards for term infants.[8] Since these curves are available online for unrestricted use, it is likely that they will become widely used in clinical settings (Figure 32-1). Exact z-scores and percentile have also been made available online for download (http://ucalgary.ca/fenton).

Physiological Variation of Body Weight During the First Days of Life

The initial weight loss after birth is due to the postnatal physiologic contraction of extracellular compartment implying a reduction of body water content. In adequately breastfed term infant, it usually occurs during the first 2 to 4 days of life and corresponds to 5%–10% of birth weight, which is generally regained after 5 to 10 days.[9]

Because of the larger extracellular compartment of preterm infants, physiological postnatal weight loss is more frequently around 10% of birth weight and the time to regain birth weight is also longer (7–10 days). However, because of excessive insensible water losses and inability to concentrate urine, postnatal dehydration may rapidly occur in very preterm infants. High transcutaneous evaporative water losses during the first days of life are due to the immaturity of the skin barrier of the preterm infant. They are increased by low humidity environment, high ambient temperature, phototherapy, and by the use of a radiant warmer.[9] Because of the net water loss, postnatal dehydration of preterm infants is characterized by a weight loss above 10%–15%, which is frequently associated with hypernatremia above 150 mmol/L. Such dehydration with electrolytes disturbances should be prevented since it is associated with an increased mortality, higher rate of postnatal morbidities and long-term adverse developmental outcomes. On the other hand, high postnatal water supply and fluid overload also increase the mortality rate and the risk for adverse neonatal morbidities such as patent ductus arteriosus, bronchopulmonary dysplasia and necrotizing enterocolitis.[9,10] Therefore, careful monitoring of fluid and electrolytes homeostasis is required during the first week of life of very preterm infants. This includes daily assessment of weight, recording of fluid and electrolyte intake, and quantification of urine output. Insensible water losses may be estimated by

subtracting the urine output and the weight change from the total fluid intake. Regular determination of plasma and urine electrolyte concentrations may also be helpful to adapt intakes. Prevention of cutaneous insensible water loss is also essential to maintain fluid and electrolytes homeostasis (Table 32-2).

TABLE 32-1 Weight Gain, Lean Body Mass, and Protein Gain During the Last Trimester of Pregnancy and Theoretical Energy and Protein Requirements for Enteral Nutrition are Indicated by Gestational Age Group. Before 39 Weeks GA, Requirements Are Based on Fetal Growth, Fetal Accretion Rate, and Intestinal Absorption; After 40 Weeks, Requirements Are Based on the Composition of Human Milk

	Gestational Age, wks					
Parameters (per kg/d)	**<28**	**28–31**	**32–33**	**34–36**	**37–38**	**39–41**
Fetal growth						
Weight gain (g)	20	17.5	15	13	11	10
LBM gain (g)	17.8	14.4	12.1	10.5	7.2	6.6
Protein gain (g)	2.1	2	1.9	1.6	1.3	1.2
Requirements						
Energy (kcal)	125	125	130	125	115	110
Proteins (g)	4	3.9	3.5	3.1	2.5	2
Calcium (mg)	120–140	120–140	120–140	120–140	70–120	70–120
Phosphorus (mg)	60–90	60–90	60–90	60–90	35–75	35–75

LBM, lean body mass.

The values indicated do not take into account the nutrient supply needed to compensate for any nutritional deficit.

Source: Adapted from Ziegler.[7]

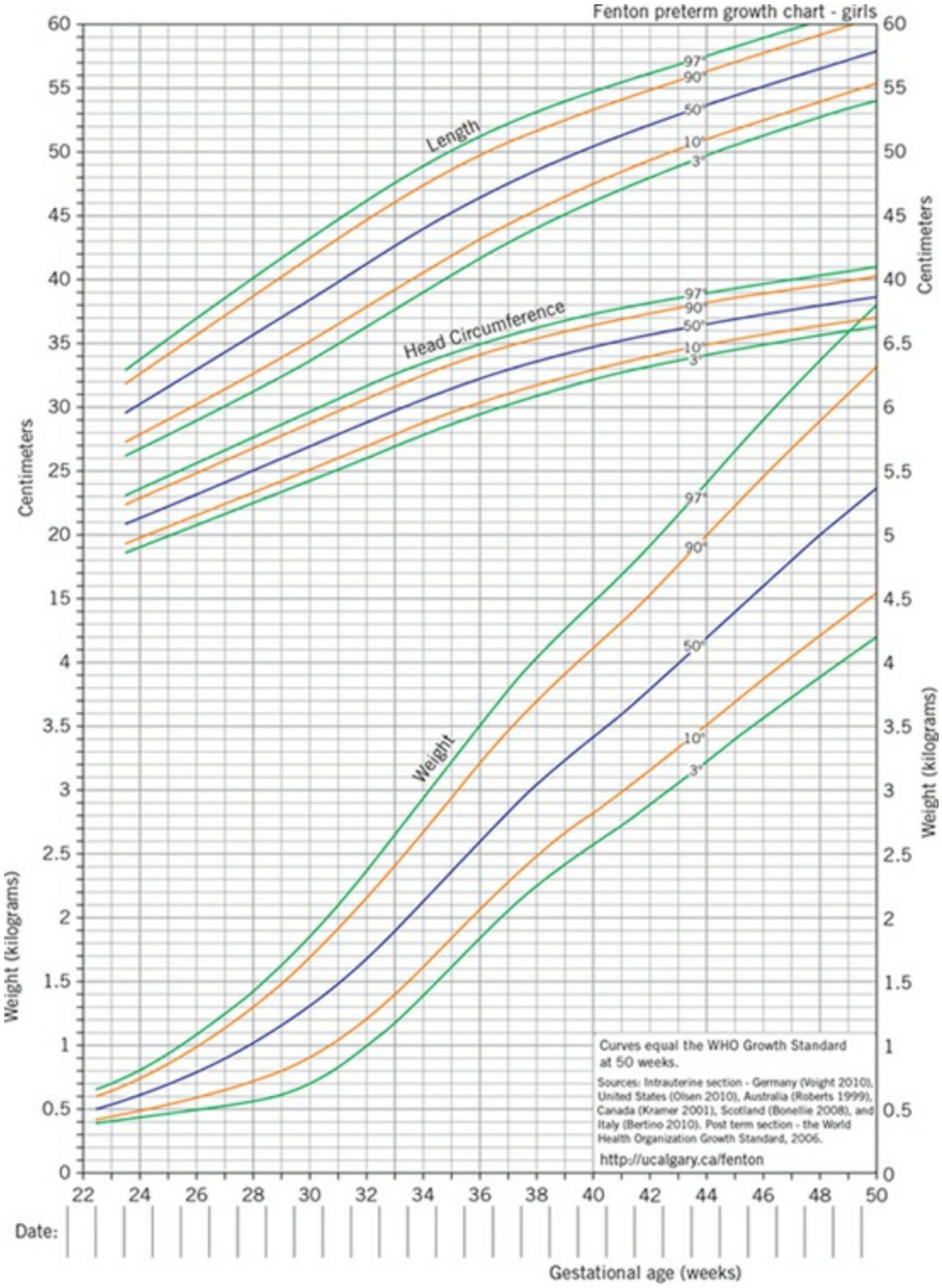
EUGR

EUGR, also called extra-uterine growth retardation, postnatal growth

restriction or postnatal growth failure, is a frequent clinical feature of preterm infants and particularly of the very, especially extremely premature ones. EUGR refers to a postnatal growth below that of the fetus of the same GA and is usually defined at hospital discharge if growth measurements are below the 10_{th} or the 3_{rd} percentile, or below -2 standard deviation (SD) on growth charts. Growth restriction can also be defined if a decrease of weight z-score >1 SD is observed during hospitalization. EUGR is inversely proportional to GA and birth weight and has been reported from all over the world at an incidence ranging from 40% to 90%.[2,11] Using weight as the unique parameter of postnatal growth may in fact underestimate the magnitude of EUGR. Indeed, linear growth is generally more severely affected than weight gain and preterm infants exhibit increased global and/or central fat mass, which can lead to lean body mass restriction.[12]

Consequences of Inadequate Perinatal Growth or Nutrition

Inadequate nutrition and/or undernutrition have long been suspected to be the main etiology of the observed EUGR of preterm infants and EUGR is observed when protein and/or energy intakes approach, but not reach, nutritional recommendations.[13] Significant relationships between poor in-hospital growth and adverse neurodevelopmental outcomes at 18 to 24 months' corrected age have been constantly demonstrated.[1,14] Furthermore, some studies suggest that this effect may last through childhood and adulthood.[15] These associations have been largely found for the smallest infants, whereas they appear insignificant in the larger babies. However, it should be noted that these associations are largely attenuated by adjustment for confounding factors (neonatal morbidity, socioeconomic factors, and level of education) and in-hospital head growth appears to better predict neurodevelopment than weight gain alone.[1,16,17]


Fenton preterm growth chart - girls
Length
Head Circumference
Weight
97°
90°
50°
10°
3°
Centimeters
Weight (kilograms)
Curves equal the WHO Growth Standard at 50 weeks.
Sources: Intrauterine section - Germany (Voigt 2010), United States (Olsen 2010), Australia (Roberts 1999), Canada (Kramer 2001), Scotland (Bonellie 2008), and Italy (Bertino 2010). Post term section - the World Health Organization Growth Standard, 2006.
http://ucalgary.ca/fenton
Date:
Gestational age (weeks)

(A)

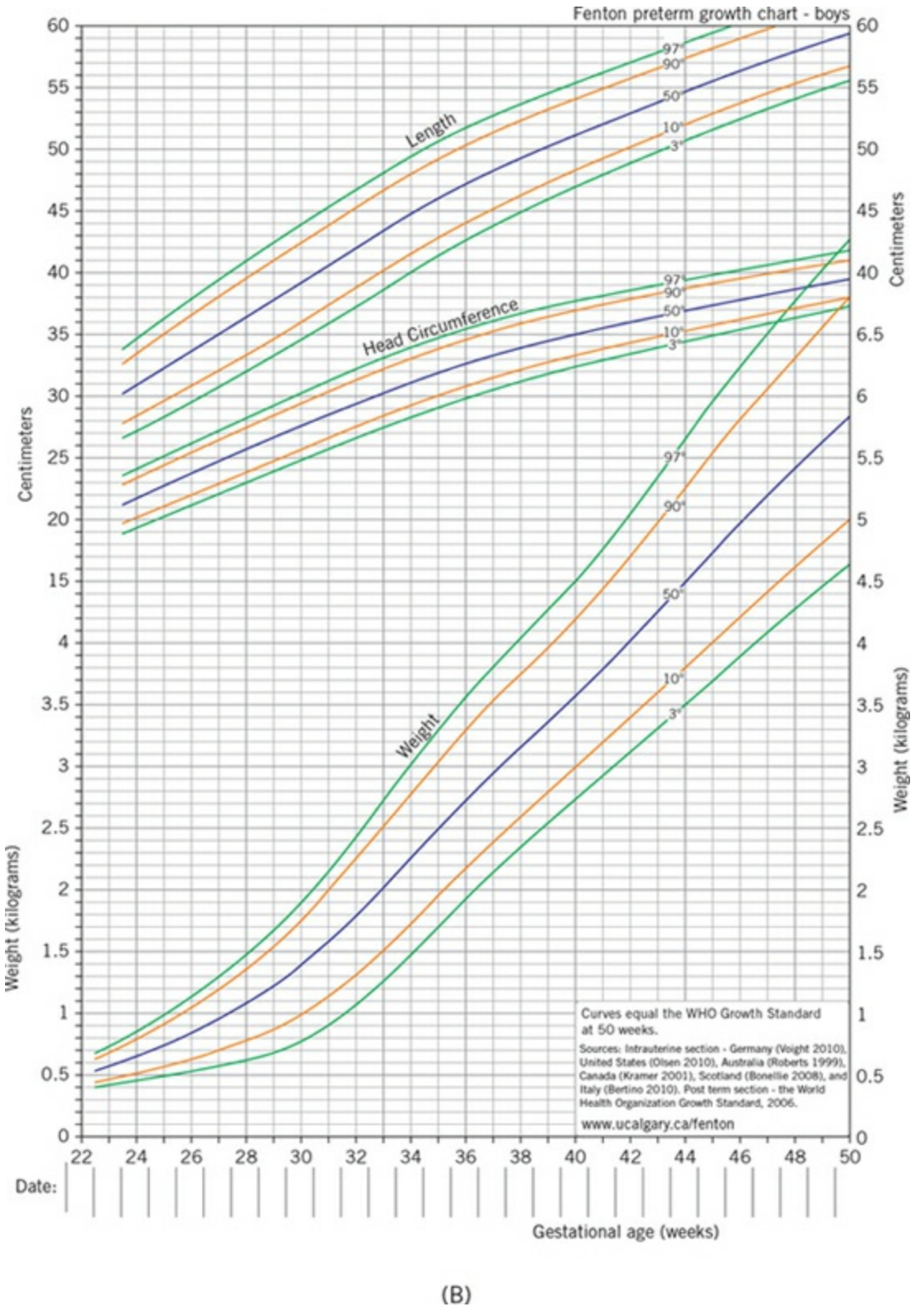


Figure 32-1 New Fenton's growth charts for preterm infants according to gender.[8]

TABLE 32-2 Factors Affecting Insensible Water Loss in Preterm Infants
Factors That Increase Insensible Water Loss
High ambient temperature or high body temperature
Use of radiant warmer
Decreased ambient humidity
Phototherapy
Mechanical ventilation, increased respiratory rate
Other: Skin injury, gastroschisis, omphalocele, surgery
Prevention of Insensible Water Loss
Use of servo-controlled incubators
Increased ambient humidity up to 80%
Humidification of inspired gases
Use of plastic cover and/or plexiglass heat shields

Protein and energy intakes are the main nutritional determinants of growth, weight gain, and body composition. Protein-energy undernutrition during critical period of development has irreversible effects on organ development and affects brain size, structure, and function.[15,18] Failure to provide sufficient protein and energy during the first week of life may have significant short- and long-term impacts on postnatal morbidities, developmental indexes throughout infancy, childhood and adolescence.[19,20]

However, other nutrients may also affect psychomotor development without any effects on growth parameters, in particular long-chain polyunsaturated fatty acids (LC-PUFAs), iron, zinc, copper, iodine, selenium, vitamin A, choline, and folate.[4–6,21] Therefore, physicians should be aware that preterm infants are at risk of nutritional deficiency that may not be reflected by altered growth rate. Additionally, breastfeeding has been shown to be beneficial for neurological development of preterm infants despite the fact that breastfed infants initially grow at a lower rate than formula fed infants.[22–24]

Epidemiological studies have described other long-term health consequences of prematurity, including an increased risk of high blood pressure, insulin resistance and other determinants of cardiovascular diseases in adulthood.[3,25,26] Growth velocity between birth and expected term seems to have no significant effect on later blood pressure and metabolic syndrome while a reduced growth during hospitalization

significantly impact later development.[1,3,14,16,17,25] To date, there is a paucity of well-designed controlled studies on the effects of nutrition during hospitalization of preterm infants on the risk of hypertension or insulin resistance. Only the duration of breastfeeding has been shown to be associated with lower blood pressure, a 10% increase in human milk consumption conferring 0.3 mmHg reduction in mean arterial blood pressure after adjustment for identifiable confounders.[27] At present, no data support the hypothesis that interventions aimed at discouraging weight gain during hospitalization and after discharge would improve adult body composition and reduce the chance of developing hypertension or type 2 diabetes. It is therefore vital that preterm infants receive sufficient nutrients to prevent EUGR and impaired neurodevelopment.

NUTRITIONAL REQUIREMENTS

The paragraphs below aim to highlight the main issues regarding nutritional specificities and requirements for enteral nutrition of the preterm infant. Recent extensive reports on guidance on appropriate nutrient intakes and recommendations on enteral nutrient supply for preterm infants have been published and are depicted in Table 32-3. Although the recommended ranges of nutrient intakes are considered reasonable, a high degree of uncertainty still remains for many nutrients.

Energy

When considering energy requirements of preterm infants, postnatal metabolic needs, energy expenditure and nutrient losses must be added to nutrient accretion to evaluate requirements. The total metabolizable energy intake (including protein) is estimate to be 90–115 kcal/kg/d since it should cover the resting energy expenditure (45–50 kcal/kg/d), the energy expenditure due to physical activity and thermogenesis (10–20 kcal/kg/d), the cost of tissue synthesis (~15 kcal/kg/d), and the energy deposited in new tissue (20–30 kcal/kg/d). Some preterm infants with chronic disease such as bronchopulmonary dysplasia may have higher energy expenditure and, thus, require additional energy intakes. Taking into account an energy absorption rate of 85% with human milk and 90% with a well-absorbed formula, the energy requirement of preterm infants receiving enteral nutrition is between 110 and 135 kcal/kg/d.[4,28]

Protein

Nitrogen balances in very preterm infants have shown that obligatory nitrogen losses due to the high protein turnover correspond to 1–1.5 g/kg/d of protein and that much higher protein supply is needed to ensure anabolism and protein accretion comparable to the fetal accretion rate (i.e., 4.0 g/kg/d at 24–25 weeks and 3.3 g/kg/d at 30–32 weeks) (Table 32-1). The protein-to-energy ratio (PER) plays a major role in the quality of growth since high PER promotes higher lean body mass accretion instead of excessive fat mass deposition that is typically occurring with high energy intakes and low PER. Since the protein and energy requirements vary according to GA, recommended PER also varies. Recently, high protein requirements (4–4.5 g/kg/d in ELBW infants and 3.5–4 g/kg/d in VLBW infants up to 1800 g) with higher PER (3.6–4.1 g/100 kcal and 3.2–3.6 g/100 kcal, respectively) have been recommended in order to take into account the frequent cumulative protein deficit that occurred during the first weeks of life in preterm infants.[4,28] It should however be noted that PER >3.6 g/100 kcal have not been tested in clinical trials.

TABLE 32-3 Recommended Enteral Nutritional Intakes for Preterm Infants and Extremely Low-Birth-Weight (ELBW, <1000 g) Expressed by 100 kcal

	Preterm Infants		ELBW Infants	
	Tsang 2005[6]	ESPGHAN 2010[4]	Tsang 2005[6]	ESPGHAN 2010[4]
Protein (g)	2.6–3.8	3.2–3.6	2.5–3.4	3.6–4.1
Carbohydrate (g)	5.4–15.5	10.5–12	6.0–15.4	10.5–12
Lipids (g)	4.1–6.5	4.4–6.0	4.1–6.5	4.4–6.0
MCT (%)	NS	<40	NS	<40
LA (mg)	462–1309	350–1400	467–1292	350–1400
ALA (mg)	NS	>50	NS	>50
LA:ALA ratio	5–15	5–15	5-15	5–15
DHA (mg)	≥16	11–27	≥16	11–27
AA (mg)	≥22	16–39	≥22	16–39
ARA:DHA ratio	NS	1–2	NS	1–2
EPA:DHA	NS	≤0.3	NS	≤0.3
Sodium (mg)	53–105	63–105	46–88	63–105

Potassium (mg)	60–106	60–120	52–90	60–120
Chloride (mg)	82-226	95–161	71–192	95–161
Calcium (mg)	77–200	110–130	67–169	110–130
Phosphorus (mg)	46–127	55–80	40–108	55–80
Magnesium (mg)	6.1–13.6	7.5–13.6	5.36–11.5	7.5–13.6
Iron (mg)	1.54–3.64	1.8–2.7	1.33–3.08	1.8–2.7
Zinc (mg)	0.77–2.73	1.0–1.8	0.67–2.31	1.0–1.8
Copper (µg)	92–136	90–120	80–115	90–120
Zinc:copper ratio	NS	<20	NS	<20
Selenium (µg)	1.0–4.1	4.5–9	0.9–3.5	4.5–9
Manganese (µg)	0.5–6.8	6.3–25	0.5–5.8	6.3–25
Fluoride (µg)	NS	1.4–55	NS	1.4–55
Iodine (µg)	7.7–54.5	10–50	6.7–46.2	10–50
Chromium (µg)	0.08–2.05	0.027–1.120	0.07–1.73	0.027–1.120
Molybdenum (µg)	0.23–0.27	0.27–4.5	0.2–0.23	0.27–4.5
Vitamin A (IU)	538–1364	1190–2440	467–1154	1190–2440
Vitamin D (IU)	115–364	727–909	100–308	727–909
Vitamin E (IU)	4.6–10.9	2–10	4.0–9.2	2–10
Vitamin K (µg)	6.2–9.1	4–25	5.3–7.7	4–25
Ascorbic acid (mg)	13.8–21.8	10–42	12.0–18.5	10–42
Pantothenic acid (mg)	0.9–1.5	0.3–1.9	0.8–1.3	0.3–1.9
Niacin (mg)	2.8–4.4	0.345–5.0	2.4–3.7	0.345–5.0
Riboflavin (µg)	192–327	180–365	167–277	180–365
Thiamin (µg)	138–218	125–275	120–185	125–275
Pyridoxine (µg)	115–191	41–273	100–162	41–273
Folic acid (µg)	19–45	32–90	17–38	32–90
Biotin (µg)	2.8–5.5	1.5–15	2.4–4.6	1.5–15
Cobalamin (µg)	0.23–0.27	0.08–0.7	0.20–0.23	0.08–0.7
Taurin (mg)	3.5–8.2	NS	3.0–6.9	NS
Carnitine (mg)	2.2–2.6	NS	1.9–2.2	NS
Choline (mg)	11.1–25.5	7–50	9.6–21.5	7–50
Inositol (mg)	25–74	4–48	21–62	4–48
Nucleotides (mg)	NS	≤5	NS	≤5

AMP (mg)	0.27–0.73	NS	0.23–0.62	NS
CMP (mg)	1.6–3.7	NS	1.4–3.2	NS
GMP (mg)	0.04–0.64	NS	0.03–0.54	NS
UMP (mg)	0.7–1.1	NS	0.6–0.9	NS

NS, not specified; MCT, medium chain triglycerides; LA, linoleic acid; ALA, alpha linoleic acid; DHA, docosahexaenoic acid; ARA, arachidonic acid; EPA, eicosapentaenoic acid; AMP, adenosine monophosphate; CMP, cytidine monophosphate; GMP, guanosine monophosphate; UMP, uridine monophosphate.

Lipids

The dietary supply of lipids provides the preterm infant with much of its energy needs. However, several factors affect fat absorption in preterm infants, which includes low enzyme secretion (gastric, pancreatic colipase-dependent triglyceride lipase, bile-salt-stimulated lipase (BSSL), pancreatic phospholipase A2), low luminal bile salt concentration, pasteurization (62.5°C for 30 minutes) of human position of fatty acids on the triglycerides, the length of carbon chain and the degree of saturation of fatty acids and pasteurization of human milk.[21] Medium chain triglycerides (MCT), which have high water solubility and are rapidly cleaved by lipases, are readily absorbed even in the presence of low intraluminal bile salts and pancreatic lipases and are therefore used in many enteral formulas for preterm infants. Further arguments for the use of MCT include carnitine-independent transport into the mitochondria and subsequent oxidation that is more rapid that for longer-chain fatty acids.[21]

Supply of LC-PUFAs are of concern because they are crucial for normal development of the central nervous system and have potential for long-lasting effects that extend beyond the period of dietary insufficiency.[21] The nervous system, and particularly the retina, is very rich in docosahexaenoic acid (DHA, C22:6n-3). Its accumulation starts in utero and proceeds during the first two years, when the growth and differentiation of the central nervous system are the most rapid. DHA supplementation of infant formula for preterm infants has consistently demonstrated better visual development compared with unsupplemented formulas. There is increasing evidence to suggest that LC-PUFAs supplementation for preterm infants is also related to improvements in more global measures of development, without any adverse effects. Given the limited and highly variable formation of DHA from alpha-linoleic acid (ALA, C18:3n-3), and because DHA is critical for normal retinal and brain development in the human, DHA is considered to be conditionally essential during early development. It is, however, important to note that

the DHA content of current preterm formulas is similar to the mean DHA content of human milk. However, recent studies have suggested that higher doses may be required to fulfill the needs and to support neurodevelopment of the preterm infants.[21,29] Finally, a concomitant intake of arachidonic acid (ARA, C20:4n-6) and a limited intake in eicosapentaenoic acid (EPA, C20:5n-3) are also recommended in order to prevent the distortion of the LC-PUFAs status and possible side effects (Table 32-4).[21]

Carbohydrates

There is a gradual increase in lactase activity with advancing gestation but overall, its activity remains low until about 36 weeks gestation. It is estimated that nearly 50%–70 % of the lactose ingested passes unabsorbed into the colon. Glucose polymers appear to be rapidly hydrolyzed and absorbed by the neonate and may be used to provide carbohydrates for preterm infants.

Minerals

Unfortified human milk (HM) provides about 6.4 mmol/L of calcium and 4.5 mmol/L of phosphorus. Assuming an intake of 160 ml/kg/d, this provides an intake of 1 mmol/kg/d of calcium and 0.72 mmol/kg/d of phosphorus, which are well below the fetal accretion rate (2.3–3.2 mmol/kg/d and 1.9–2.5 mmol/kg/d, respectively). Clinical studies have demonstrated that preterm infants fed unfortified HM develop osteopenia and metabolic bone disease of prematurity.[30] According to mineral balance studies, calcium retention is mainly dependent of calcium absorption and phosphorus availability. Calcium absorption is enhanced by vitamin D and usually varies between 50% and 65% of intakes depending on calcium salts bioavailability. Phosphorus is generally well absorbed by the gastrointestinal tract (~90%) but its availability for bone accretion is also dependent on the lean body mass accretion. Phosphate is a major constituent of the intracellular compartment and one gram of protein accretion is estimated to be associated with ~0.3 mmol of phosphorus retention. Hypophosphatemia (<1.6 mmol/L or 5 mg/dL) associated with low phosphaturia and hypercalciuria (with or without hypercalcemia) are typical biological signs of inadequate mineral intake and needs to be prevented.[30]

TABLE 32-4 Enteral Long-Chain Polyunsaturated

Fatty Acids Recommendations for Preterm Infants				
	LSRO 2002[5]	**Tsang 2005[6]**	**ESPGHAN 2010[4]**	**Lapillonne 2013[21]**
LA (C18:2n–6)				
% total energy	NS	3.2–12.8	3.2–12.8	3.2–12.8
mg/100 kcal	352–1425	352–1425	350–1400	350–1400
mg/kg/d	NS	600–11680	385–1540	385–1540
% total fatty acids	8–25	8–25	NS	NS
ALA (C18:3n–3)				
% total energy	NS	0.7–2.1	≥ 0.45	≥ 0.45
mg/100 kcal	77–228	77–228	≥ 50	≥ 50
mg/kg/d	NS	NS	≥ 55	≥ 55
% total fatty acids	1.75–4	1.75–4	≥ 0.9	≥ 0.9
LA:ALA	6–16:1	6–16:1	5–15:1	5–15:1
AA (C20:4n–6)				
mg/100 kcal	NS	NS	16–39	NS
mg/kg/d	NS	NS	18–42	18–45
% total fatty acids	≥ 0.6	0.3–0.7	NS	NS
DHA (C22:6n–3)				
mg/100 kcal	NS	NS	11–27	NS
mg/kg/d	NS	NS	12–30	12–60
% total fatty acids	≥ 0.35	0.2–0.5	NS	NS
AA:DHA	1.5-2:1	1.2–2:1	1–2:1	NS
EPA (C20:5n-3)	≤30% DHA	≤30% DHA	≤30% DHA	≤20 mg/kg/d

NS, not specified; LA, linoleic acid; ALA, linoleic acid; AA, arachidonic acid; DHA, docosahexaenoic acid; EPA, eicosapentaenoic acid.

Considering a calcium to phosphorus ratio of 1.7 in bone hydroxyapatite and a need for 1.5 to 2.2 mmol/kg/d of calcium retention to

ensure appropriate bone mineralization, an enteral intake of 3–3.5 mmol/kg/d of highly bioavailable calcium salts and 2–3 mmol/kg/d of phosphorus is usually recommended.[4,30] Additionally, a vitamin D intake of 800–1,000 IU/d is required to obtain and maintain the 25-hydroxy vitamin D plasma concentration above 80 nmol/L (30 µg/L), ideally around 180 nmol/L (70 µg/L).

Fluids and Electrolytes

Considering physiological variation of body weight during the first days of life and frequent postnatal relative oliguria, fluid requirements of most term newborns on the first day of life are 40–60 mL/kg/d whereas that of preterm infants is higher, between 60 and 80 mL/kg/d, with no or little sodium, potassium, and chloride (<1 mmol/kg/d) intakes.[6,31] During the transitional period (i.e., day 2 to day 7–10 of life), fluid and electrolyte supply need to be progressively increased to reach requirements of the stable growing infants: 120–180 mL/kg/d, 3–7 mmol/kg/d of sodium, 2–5 mmol/kg/d of potassium, and 3–7 mmol/kg/d of chloride.[6,31]

ENTERAL NUTRITION

Enteral nutrition in preterm infants is challenging for several reasons, including gastrointestinal immaturity and the risk of necrotizing enterocolitis (NEC). NEC is a severe gastrointestinal disorder secondary to immature intestinal barrier function, altered gut microbiota, and excessive gut inflammatory response. It represents a major complication of prematurity and is associated with increased mortality and later neurodevelopmental disabilities.[32] The fear of NEC frequently leads to delayed introduction of enteral feeding and slow advancement of feeds, which consequently leads to an increase need for parenteral nutrition (PN). This practice, however, is not devoid of consequences. Enteral nutrition is essential to promote gastrointestinal growth and development by providing nutrients to the mucosal epithelial cells, stimulating the secretion of local growth factors and gastrointestinal hormones, and activating gastrointestinal neural pathways. Therefore, poor feeding induces gastrointestinal mucosal atrophy, abnormal mucus secretion, increased permeability, decreased regenerative capabilities, and finally, gastrointestinal dysfunction that may secondary increase the risk of feeding intolerance, bacterial translocation and NEC.[28,33] Furthermore, prolonged need for PN has its own associated complications, which

include metabolic intolerance, sepsis, thrombosis, cholestasis and inflammatory diseases. Therefore, optimizing feeding strategy in preterm infants is essential to reduce postnatal growth retardation and neonatal morbidities but also to improve long-term outcomes.

Mother's Milk and Donor's Milk

Preterm infants' breastfeeding has numerous beneficial effects such as promoting gastrointestinal maturation and intestinal tolerance and reducing the incidence of NEC, late-onset sepsis and hospital readmissions. Association between breastfeeding and improved mental, cognitive and psychomotor development, better visual acuity, increased total and white matter brain volumes and lower incidence of retinopathy of prematurity have also been demonstrated. Finally, breastfeeding also potentiality influences the risk for later metabolic syndrome and high blood pressure through adulthood.[3,34] The biological, immunologic, and nutritional characteristics of the human milk, which are described elsewhere in this book, likely explain most of these beneficial effects.

Mothers of preterm infants frequently encounter difficulties in initiating lactation and therefore appropriate counseling is needed. Breastfeeding education during pregnancy and lactation support by well-trained perinatal caregivers is essential for the success of breastfeeding in preterm infants. Nursing staff needs to help mother developing a plan for milk expression that meet her and hers baby's needs until breastfeeding can occur. Breast stimulation must be started within the first six hours after preterm delivery. Mother may express only a few drops initially and manual milk expressions work best for this small quantity. To better establish lactation, mothers need to use a double electric pump 8 to 10 times a day during session of 10–15 minutes. Nurses and pediatricians should favor skin to skin contact and encourage mothers to achieve adequate breast milk production after one week, 600 to 800 ml/d ideally. Expressed breast milk should be refrigerated or chilled right after it has been expressed. It is widely accepted that breast milk can be stored in a refrigerator for 72 hours and in a freezer for 6 months.[28,34,35]

When mother's milk is not available for feeding preterm infants, donor milk is an alternative to provide some of the advantages of breastfeeding. Donors are screened to avoid any risk of infection (HIV, CMV, hepatitis, syphilis) or any toxic contamination (drugs, alcohol, cigarettes). Donor milk is checked microbiologically and then pasteurized to avoid any bacterial or viral contamination. Pasteurization (62.5°C for 30 minutes) provides microbiological safety but induce some oxidative losses of

unsaturated lipids and vitamins, as well as inactivation of some enzymes and immunological factors such as IgA, lactoferrin, lysozyme. Some new techniques of heat treatment of milk are under investigation to improve the bioactivity of breast milk components while maintaining the safety concerns of donor milk.[36,37] Several systematic reviews have demonstrated the advantages of donor milk versus preterm formula in very preterm infants by reducing the incidence of NEC.[38,41] Therefore, when own mother's milk is not available, pasteurized donor milk is a good alternative for feeding the preterm infants.[28]

Human Milk Fortification

Feeding the preterm infant with unfortified human milk does not meet the infant's high nutritional requirements and leads to poor weight gain, osteopenia, nutritional deficits and potentially significant adverse outcomes. Multicomponent HM fortifiers (HMF) containing protein, energy, minerals and vitamins have therefore been developed and commercialized. Their use significantly improves postnatal weight gain, linear growth and head growth.[42–44] However, several studies have documented that many preterm infants fed with standardized fortification of human milk still suffer from EUGR. In some cases, this phenomenon is due to the low protein and energy content of the human milk which is not taking into account during the standardized fortification (Table 32-5). By using milk analyzers, it is now possible to adapt the level of fortification to the protein, fat and lactose content of human milk. Such individualized fortification has been shown to significantly improve postnatal growth of preterm infants.[43,44] In the absence of milk analyzers, fortification can also be adjusted to the blood urea nitrogen (BUN) concentration (target between 3.2 and 5 mmol/L) which is an intermediate marker of the adequacy of protein supply.[42] However, this adjustment does not completely prevent some nutritional deficit since it is performed *a posteriori*.

The addition of HMF increases milk osmolarity and, for most of the commercially available HMFs, introduces bovine proteins. In the past, some concerns have been raised that it may increase the risk of NEC but systematic reviews were not able to demonstrate any significant adverse effects related to the use of HMF.[45,46] Therefore, the benefits of fortification of human milk clearly outweigh the potential risks in very preterm infants. Full fortification must be performed in order to reduce the duration of PN and to improve overall nutritional intakes. It is generally

well accepted that full fortification is safe when enteral feeding reaches 100 mL/kg/d but recent data suggest that human milk fortification can be done earlier.[28,47,48]

Preterm Infant Formula

Although fortified breastfeeding is considered as the best feeding choice for preterm infants, mother's milk is not always available and many NICUs do not have access to a human milk bank or to donor milk. Therefore, specifically adapted formula for preterm infants have been developed and commercialized according to actual guidelines that are summarized in the Table 32-3.[4–6] The advantages of the preterm formula are numerous compared to term formula since they improve nutritional intakes and reduce nutritional deficit to allow proper growth of preterm infants. However, the data available still strongly support breastfeeding as the first choice for feeding preterm infants.

Initiation and Advancement of Enteral Nutrition

The gastrointestinal tract of the preterm infants at birth is characterized by an immature motility and/or dysmotility. This frequently leads to delayed gastric emptying, bile-stained gastric residuals and difficulties in passing stools. Because of these observations and the fear of NEC, it is sometimes common practice to delay enteral feeding and to use slow feeds advancement.

TABLE 32-5 Composition of Preterm Transitional, Preterm Mature and Term Mature Mother's Milk

Nutrients (Units/dL)	Preterm Transitional (6–10 d)	Preterm Mature (22–30 d)	Term Mature (>28 d)
Total protein, g	1.9 ± 0.05	1.5 ± 0.1	1.2 ± 0.15
Energy, kcal	66 ± 6	69 ± 5	64 ± 8
Fat, g	3.4 ± 0.6	3.6 ± 0.4	3.4 ± 0.4
Carbohydrate, g	6.3 ± 0.5	6.7 ± 0.4	6.7 ± 0.5
Sodium, mmol	1.16 ± 0.60	0.88 ± 0.20	0.90 ± 0.41
Potassium, mmol	1.35 ± 0.22	1.25 ± 0.32	1.30 ± 0.20
Chloride, mmol	2.13 ± 0.22	1.48 ± 0.21	1.28 ± 0.15

Calcium, mmol	0.80 ± 0.18	0.72 ± 0.13	0.65 ± 0.15
Phosphorus, mmol	0.49 ± 0.14	0.30 ± 0.08	0.48 ± 0.08
Magnesium, mmol	0.11 ± 0.02	0.10 ± 0.03	0.13 ± 0.03
Iron, mmol	~2.3	~2.2	~2.2
Zinc, µmol	5.8 ± 1.3	3.3 ± 1.4	1.5–4.6
Copper, µmol	0.92 ± 0.21	0.80 ± 0.31	0.32–0.63
Manganese, µmol	0.11 ± 0.16	0.13 ± 0.12	0.05 ± 0.11
Iodine, µmol	-	~0.125	-
Vitamin A, IU	50–400	50-400	60–200
Vitamin D, IU	~4	~4	-
Vitamin E, mg	0.3–1.5	0.3–1.5	0.2–0.3
Vitamin K, µg	0.1–0.5	0.1–0.5	0.1–0.9
Folate, mg	~3.3	~3.3	~0.2

Value as mean ± standard deviation or range.

Source: Adapted from Tudehope.[34]

TABLE 32-6 Reasonable Strategy to Optimize Enteral Feeding Practices in Low–Birth-Weight (<2500 g) Infants According to Their Birth Weight

Birth Weight	1500–2499 g	1000–1499 g	<1000 g
Milk	HM[a]	HM[a]	HM[a]
First feeding	As soon as possible	Between 6 and 48 h of life	Between 6 and 48 h of life
Initial feeding[b]	3–4 mL/kg q2h	1 mL/kg/h or 2 mL/kg q2h	0.5 mL/kg/h or 1 mL/kg q2h
Feeding advancement	20–40 mL/kg/d	20–30 mL/kg/d	15–25 mL/kg/d
HM fortification	Before 100 mL/kg/d	Before 100 mL/kg/d	before 100 mL/kg/d
Target enteral intakes	110–135 kcal/kg/d	110–135 kcal/kg/d	110–135 kcal/kg/d
	3.0–4.0 g/kg/d	3.5–4.0 g/kg/d	4–4.5 g/kg/d

HM, human milk; MEF, minimal enteral feeding

[a]Own mother breast milk or donor human milk, but adapted preterm infant formula may also be used if there is no access to human milk.

[b]Parenteral nutrition may be consider in 1500-2499 g infants according to their clinical situation but very low birth weight (<1500 g) infants most always require parenteral nutrition to meet their

nutritional needs.

Source: Adapted from Senterre.[28]

However, there is no evidence that withholding feeding decreases NEC or improves outcomes. In fact, delaying introduction of enteral feeding significantly increases the time to establish full enteral feeding, PN needs, and PN-associated morbidities.[28,49] Therefore, enteral feeding should be initiated as early as possible (Table 32-6). To achieve this goal, minimal enteral feeding (MEF) should be considered when neonatologists are tempted to delay enteral feeding introduction or to discontinue feeding because of specific clinical conditions (i.e., significant perinatal asphyxia, significant patent ductus arteriosus, hemodynamic instability, severe intra-uterine growth retardation, ELBW). MEF is defined by the administration of a constant volume of small feeds, usually ≤24 mL/kg/d, during a prolonged period of time (i.e., three to five days) in order to promote postnatal gastrointestinal maturation. It has been shown that early MEF improves nutritional intakes and growth, and avoid PN-associated diseases without increasing the risk of NEC. Therefore, the opportunity of early MEF should be evaluated as soon as infants' cardio-respiratory function is stabilized and after a few hours in all very preterm infants.[28,50]

Compare to slow feeds advancement (i.e., <24 mL/kg/d), advancement of 25–35 mL/kg/d in VLBW infants does not increase the risk of NEC and is associated with shorter time to regain birth weight and shorter time to achieve full enteral feeding.[28,51] Thus, feeding advancement of 20–30 mL/kg/d is generally considered reasonable in 1000–1500 g preterm infants according to feeding tolerance (Table 32-6). In those weighing less than 1000 g, the safety of such feeding advancement is not well established and 15–25 mL/kg/d advancement may be preferred until further studies are performed.[28]

Feeding Administration and Feeding Tolerance

Because of immaturity, oral feeding is frequently delayed in preterm infants and a prolonged period of tube feeding is generally required. Nasogastric tubes are easier to secure than orogastric tubes but may increase the work of breathing because of partial nasal airway obstruction.[28,52] Transpyloric tube feeding is sometimes used in case of severe gastroesophageal reflux but this practice is not recommended as a routine feeding method for neonates (Table 32-6).[28,53]

Conventional tube feeding consists of delivering intermittent bolus gavage by gravity over 10–30 minutes every two or three hours. In very

preterm infants, it seems that continuous feeding reduces gastroesophageal reflux symptoms and energy expenditure, improves postnatal gastrointestinal maturation and feeding tolerance and may accelerate the achievement of full enteral feeding.[28,54] Short interval feeding methods like one hourly intermittent gavage and slow intermittent gavage over one or two hours may also be proposed.[28]

The aspiration of gastric residuals is common practice to evaluate gastrointestinal tolerance while tube feeding. Most studies have considered gastric residuals as suspect if they are >3–4 mL/kg, >20%–50% of the previous feed, or bilious-stained. However, such studies evaluating the predictive value of such gastric residuals for significant feeding intolerance or NEC have demonstrated poor results. Therefore, in absence of any significant clinical signs or symptoms, the pretext of gastric residuals <4 mL/kg or <50% of three-hours previous feed should not be considered as valid to interrupt or reduce enteral feeding.[28,55] On the other side, any sign of potential feeding intolerance requires a complete clinical examination to determine subsequent care. An abdominal radiograph may be necessary to rule out NEC but non-irradiated techniques like abdominal ultrasounds are also useful to evaluate abdominal condition of preterm infants. When feeding intolerance is suspected or confirmed, feeds need to be adapted to each infant's evolution. If enteral feeding has been interrupted but clinical evolution rules out NEC or another disease, enteral feeding should be restarted as soon as possible, even after a few hours.[28]

Probiotics and Bioactive Factors

The gut microbiota of the hospitalized preterm infants is characterized by a delayed colonization and a low diversity. The predominant aerobes species are staphylococci (mainly coagulase -negative staphylococci), enterobacteria (mainly Klebsiella spp, Enterobacter spp), and enterococci whereas clostridia are the most common anaerobes. In contrast, Bacteroides and bifidobacteria seldom colonize preterm infants. Very little is known about factors that influence the colonization of the gut of very preterm infants. Colonization by bifidobacteria appears to be not only dependent on chronological age, but also by GA, suggesting that intestinal maturity is an important factor. In contrast to term infants, breastfeeding alone is unable to promote bifidobacteria colonization in preterm infants. Therefore, there is a great deal of interest for modulating the gastrointestinal microbiota of preterm infants using dietary components and microorganisms. Several studies have demonstrated that mortality and

incidence of NEC may be significantly lower in preterm infants receiving probiotics.[56] However, uncertainty remains on safety and choice of the bacterial strain(s), and therefore, the routine use of probiotics is not yet recommended.[57]

Breast milk contains many components that may act as bioactive factors, immunonutrients and growth factors. Among them, lactoferrin is a protein that has antimicrobial, host immunomodulatory and gastrointestinal effects. Supplementation with bovine-lactoferrin suggests some benefits with potential reduction of the incidence of nosocomial infections, especially fungal sepsis but further studies are needed to assess effective benefits, safety, ideal dose, and impact on both short- and long-term outcomes before routine use can be recommended.[58]

PN

PN is required in many preterm infants until full enteral feeding is established. Despite recommendations have been published years ago, there is a large discrepancy between daily recommended intakes and actual intakes during the first few weeks of life, resulting in substantial nutritional deficits.[59] It is essential that physicians acknowledge that preterm birth is a nutritional emergency. When required, PN initiation should not be delayed and should be initiated in most neonates in the first hours of life. The Table 32-7 summarizes the most recent international recommendations for PN in preterm infants.

To safely cover resting energy expenditure, prevent negative nitrogen balance and promote anabolism, a minimum energy intake of 40–55 kcal/kg/d with at least 1.5 g/kg/d of amino acids (AA) is recommended from the first day of life in all preterm infants.[6,60] However, much higher intake, especially of AA, can be used without signs of metabolic intolerance.[61] Routine initiation of PN within two hours of life with the supply of 2.4 g/kg/d of AA, 1 g/kg/d of lipid, and 6 g/kg/d of glucose (4–5 mg/kg/d) promote adequate postnatal weight gain in very preterm infants (Figure 32-2), and significantly reduces the incidence of EUGR.[47,62]

Lipid emulsions provide essential fatty acid and non-carbohydrate energy in a low volume and with low osmolarity. Evidences have accumulated lipid emulsions can also influence numerous pathophysiologic processes including oxidative stress, immune responses and inflammation. The use of the standard 20% emulsions, which contain a lower ratio of phospholipid emulsifier/triglycerides than standard 10%

lipid emulsion is recommended for preterm infants since it allows more efficient triglyceride clearance. Triglyceride levels in serum or plasma should be monitored particularly in cases with a marked risk for hyperlipidemia (e.g., patients with high lipid dosage, sepsis, catabolism, ELBW infants). Reduction of the dosage of lipid emulsions should be considered if serum or plasma triglyceride concentrations during infusion exceed 2.85 mmol/L (250 mg/dL).[60]

TABLE 32-7 Recommendations for Parenteral Nutrition in Preterm Infants

	Tsang 2005[6]	ESPEN/ESPGHAN/ESPR, 2005[31]	Current
Amino acids			
Initiation	Day of birth	Day of birth	First hours of life
Initial dose, g/kg/d	2	≥1.5	2–3
Target dose, g/kg/d	3.5–4 (ELBW) 3.2–3.8 (VLBW)	Maximum 4	3–4 3.5–4 (VLBW)
Glucose			
Initiation	day of birth	day of birth	First hour of life
Initial dose, g/kg/d	7	5.8–11.5	6
Target dose, g/kg/d	13–17 (ELBW) 9.7–15 (VLBW)	maximum 18 g/kg/d	12–17[a]
Lipids			
Initiation	Day of birth	No later than third day	Day of birth
Initial dose, g/kg/d	1	Linoleic acid >0.25 mg/kg/d	1
Target dose, g/kg/d	3–4	3–4	3–4[b]
Energy			
Caloric target, g/kg/d	105–115 (ELBW) 90–100 (VLBW)	110–120	90–115

VLBW, very low birth weight infants; ELBW, extremely low birth weight infants; ESPEN/ESPGHAN/ESPR, European Society for Clinical Nutrition and Metabolism, European Society

of Pediatric Gastroenterology, Hepatology and Nutrition, European Society of Pediatric Research.

[a]To be reduced if hyperglycemia >8.3–10 mmol/(150–180 mg/dL).

[b]To be reduced if serum triglycerides >2.85 mmol/L > (250 mg/dL) and/or if hyperglycemia >8.3–10 mmol/L (150–180 mg/dL).

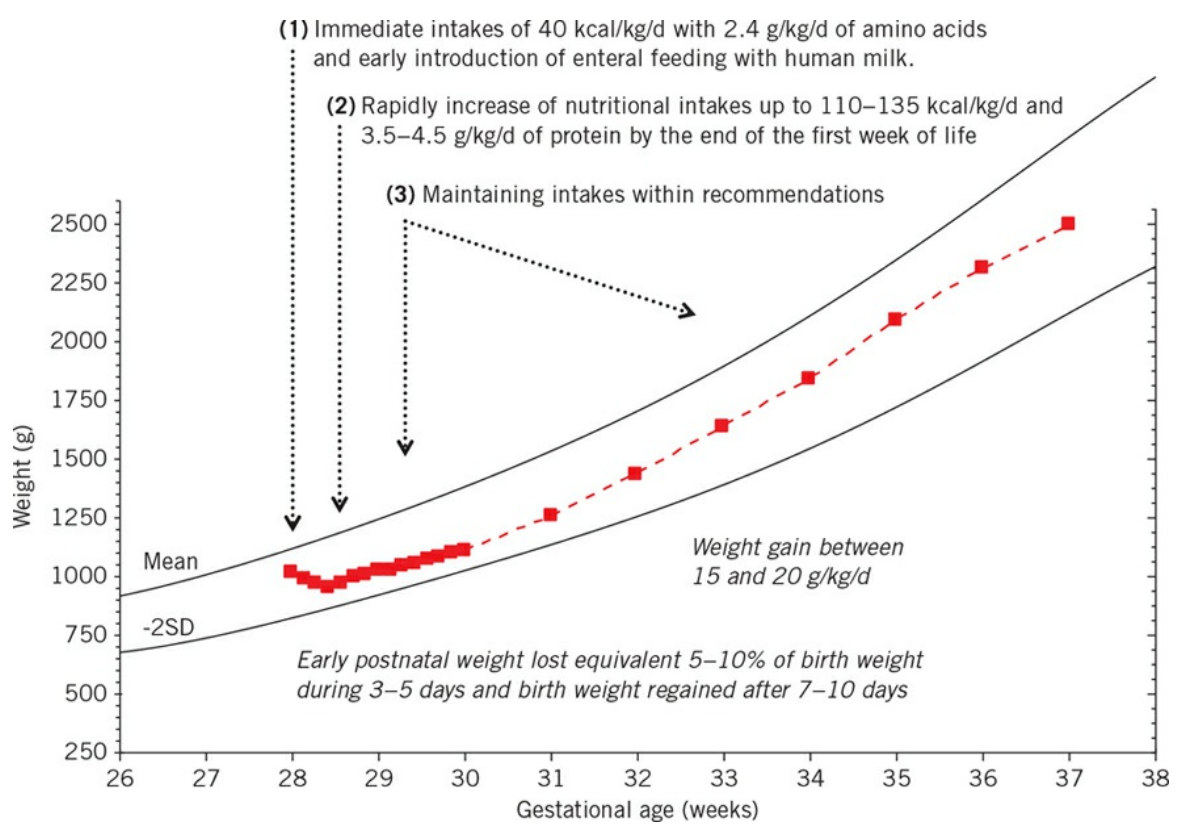


Figure 32-2 Optimizing nutritional support and postnatal growth from the first day of life onwards during postnatal hospitalization in very preterm infants. Adapted from Senterre.[47]

The adequacy of historical soybean lipid emulsions for the nutritional needs of newborn and preterm infants has been questioned, and a systematic study has demonstrated clinical benefits of lipid emulsions that are not purely soybean based (e.g., MCT-soybean, olive-soybean, or soybean-MCT-olive-fish emulsions).[63] Although these new lipid emulsions appear promising, further studies are required to determine the optimal lipid emulsions for preterm infants.[21]

Because of a combination of insulin resistance, relative insulin deficiency, high gluconeogenesis rate and the presence of risk factors (i.e., high glucose intakes, sepsis, NEC, drugs such as methylxanthines and corticosteroids, surgery, intrauterine growth retardation, and metabolic

stress) preterm infants are prone to hyperglycemia. The incidence of hyperglycemia during PN increased with decreased GA and birth weight and may reach 40%–80% in very preterm infants. Efforts are needed to avoid hyperglycemia as it has been associated with increased mortality and increased morbidity such as neurodevelopmental impairments and behavioral problems in infancy.[64] In practice, regular blood glucose monitoring is necessary. Euglycemia in neonates is usually considered for plasma glucose concentration between 2.5 and 8.3 mmol/L (45–150 mg/dL), and ideally between 2.8 and 6.7 mmol/L (50–120 mg/dL). Glucose intakes need to be adapted according to glucose tolerance up to 12–17 g/kg/d (8.5–12 mg/kg/min) but should not exceed maximal glucose oxidation capacity (18 g/kg/d, 12.5 mg/kg/min) and 60%–75% of non-protein energy intake. Any hypoglycemia in preterm infants should be treated rapidly and efficiently. In case of hyperglycemia >8.3 mmol/L, the first step is to understand the possible etiology (sepsis, stress, excessive glucose, or energy intakes) and measure potential glycosuria and the associated osmotic diuresis. Afterwards, the treatment of persistent hyperglycemia usually consists in reducing glucose infusion rate, stress factors, and lipid infusion rate. Protein intakes should not be decreased as AA infusion stimulates insulin secretion, and enhances glucose utilization. Insulin treatment can be started if hyperglycemia persists >10 mmol/L (180 mg/dL) with an initial dosage of 0.02–0.05 IU/kg/h.[60,64]

As described above, the modern nutritional approach for preterm infants is based on early initiation of enteral feeding and early PN in case of insufficient enteral intakes to rapidly meet their high nutritional requirements. The early feeding strongly influences the metabolism of potassium, phosphorus, and calcium, defining a syndrome that somehow simulates what happens in re-feeding conditions. Therefore, care should be taken to provide adequate amount of phosphorus and potassium during initiation of PN.[65]

Due to the very small diameter of blood vessels of preterm infants, the venous access is frequently uneasy and still represents an important challenge in clinical setting. Peripheral intravenous catheters do not allow the infusion of hyperosmolar PN solution, because of the risk of extravasation, dislodgement, leakage, and phlebitis. Venous umbilical catheters may be considered as a bridge procedure while awaiting placement of a long-term device. They allow administration of PN, medications and blood products during the first hours/days of life but this route should be as short as possible because of the risk of thrombotic complications. Peripherally inserted percutaneous central venous catheters

are the preferred route for prolonged PN in preterm infants. However, it must be remembered that intravascular catheters are a major source of coagulase-negative staphylococci bacteremia. This is particularly true in preterm infants for whom late-onset sepsis is one of the most common causes of morbidity and mortality.

CONCLUSION

Nutritional support in preterm and LBW infants is a major part of neonatal care and may have significant consequences on their neurodevelopment and future health. These infants have very specific nutritional requirements compared to term infants and optimizing the nutritional delivery in clinical setting is crucial. Enteral nutrition and breastfeeding needs to be initiated as soon as possible to promote gastrointestinal tract development and postnatal growth. Human milk confers many advantages to preterm infants, particularly for very preterm infants, but requires fortification to fulfill their nutritional needs. PN is an essential therapy for neonates who are not able to be completely enterally fed. Adequate protein and energy intakes are important from the first day of life and postnatal growth and metabolic homeostasis needs to be monitored closely.

REFERENCES

1. Ehrenkranz RA, Dusick AM, Vohr BR, Wright LL, Wrage LA, Poole WK. Growth in the neonatal intensive care unit influences neurodevelopmental and growth outcomes of extremely low birth weight infants. *Pediatrics.* 2006;117:1253–61.
2. Fanaroff AA, Stoll BJ, Wright LL, et al. Trends in neonatal morbidity and mortality for very low birthweight infants. *Am J Obstet Gynecol.* 2007;196:147 e1–8.
3. Lapillonne A, Griffin IJ. Feeding preterm infants today for later metabolic and cardiovascular outcomes. *J Pediatr.* 2013;162:S7–16.
4. Agostoni C, Buonocore G, Carnielli VP, et al. Enteral nutrient supply for preterm infants: commentary from the European Society of Paediatric Gastroenterology, Hepatology, and Nutrition Committee on Nutrition. *J Pediatr Gastroenterol Nutr.* 2010;50:85–91.
5. Klein CJ. Nutrient requirements for preterm infant formulas. *J Nutr.* 2002;132:1395S–577S.
6. Tsang RC, Uauy R, Koletzko B, Zlotkin SH, eds. *Nutrition of the Preterm Infant: Scientific Basis and Practical Guidelines.* Cincinnati, OH: Digital Educating Publishing, Inc; 2005;427 pages.
7. Ziegler EE. Meeting the nutritional needs of the low-birth-weight infant. *Ann Nutr Metab.* 2011;58 Suppl 1:8–18.
8. Fenton TR, Kim JH. A systematic review and meta-analysis to revise the Fenton growth chart for preterm infants. *BMC Pediatr. 2013;13:59.*

9. Fusch C, Jochum F. Water, sodium, potassium and chloride. In: Tsang RC, Uauy R, Koletzko B, Zlotkin SH, eds. *Nutrition of the Preterm Infant. Cincinnati, OH: Digital Educating Publishing, Inc; 2005:201–44.*
10. Bell EF, Acarregui MJ. Restricted versus liberal water intake for preventing morbidity and mortality in preterm infants. *Cochrane Database Syst Rev*. 2008:CD000503.
11. Clark RH, Thomas P, Peabody J. Extrauterine growth restriction remains a serious problem in prematurely born neonates. *Pediatrics*. 2003;111:986–90.
12. Cooke RJ, Griffin I. Altered body composition in preterm infants at hospital discharge. Acta *Paediatr.* 2009;98:1269–73.
13. Martin CR, Brown YF, Ehrenkranz RA, et al. Nutritional practices and growth velocity in the first month of life in extremely premature infants. *Pediatrics.* 2009;124:649–57.
14. Latal-Hajnal B, von Siebenthal K, Kovari H, Bucher HU, Largo RH. Postnatal growth in VLBW infants: significant association with neurodevelopmental outcome. *J Pediatr.* 2003;143:163–70.
15. Isaacs EB, Gadian DG, Sabatini S, et al. The effect of early human diet on caudate volumes and IQ. *Pediatr Res.*2008;63:308–14.
16. Kan E, Roberts G, Anderson PJ, Doyle LW. The association of growth impairment with neurodevelopmental outcome at eight years of age in very preterm children. *Early Hum Dev.* 2008;84:409–16.
17. Weisglas-Kuperus N, Hille ET, Duivenvoorden HJ, et al. Intelligence of very preterm or very low birthweight infants in young adulthood. *Arch Dis Child Fetal Neonatal Ed.* 2009;94:F196–200.
18. Lucas A, Morley R, Cole TJ. Randomised trial of early diet in preterm babies and later intelligence quotient. *BMJ.* 1998;317:1481–7.
19. Ehrenkranz RA, Das A, Wrage LA, et al. Early nutrition mediates the influence of severity of illness on extremely LBW infants. *Pediatr Res*. 2011;69:522–9.
20. Stephens BE, Walden RV, Gargus RA, et al. First-week protein and energy intakes are associated with 18-month developmental outcomes in extremely low birth weight infants. *Pediatrics.* 2009;123:1337–43.
21. Lapillonne A, Groh-Wargo S, Gonzalez CH, Uauy R. Lipid needs of preterm infants: updated recommendations. *J Pediatr*. 2013;162:S37–47.
22. Isaacs EB, Fischl BR, Quinn BT, Chong WK, Gadian DG, Lucas A. Impact of breast milk on intelligence quotient, brain size, and white matter development. *Pediatr Res.* 2010;67:357–62.
23. Roze JC, Darmaun D, Boquien CY, et al. The apparent breastfeeding paradox in very preterm infants: relationship between breast feeding, early weight gain and neurodevelopment based on results from two cohorts, EPIPAGE and LIFT. *BMJ Open.* 2012;2:e000834.
24. Vohr BR, Poindexter BB, Dusick AM, et al. Persistent beneficial effects of breast milk ingested in the neonatal intensive care unit on outcomes of extremely low birth weight infants at 30 months of age. *Pediatrics*. 2007;120:e953–9.
25. Hovi P, Andersson S, Eriksson JG, et al. Glucose regulation in young adults with very low birth weight. *N Engl J Med.* 2007;356:2053–63.
26. Hovi P, Andersson S, Raikkonen K, et al. Ambulatory blood pressure in young adults with very low birth weight. *J Pediatr.* 2010;156:54–9 e1.
27. Singhal A, Cole TJ, Lucas A. Early nutrition in preterm infants and later blood pressure: two cohorts after randomised trials. *Lancet.* 2001;357:413–9.
28. Senterre T. Practice of enteral nutrition in very low birth weight and extremely low birth weight infants. *World Rev Nutr Diet.* 2014;110:201–14.

29. Makrides M, Gibson RA, McPhee AJ, et al. Neurodevelopmental outcomes of preterm infants fed high-dose docosahexaenoic acid: a randomized controlled trial. *JAMA.* 2009;301:175–82.
30. Rigo J, Mohamed MW, de Curtis M. Disorders of calcium, phosphorus, and magnesium metabolism. In: Martin RJ, Fanaroff AA, Walsh MC, eds. *Fanaroff and Martin Neonatal-Perinatal Medicine*. 9th ed. St; Louis: Elsevier Mosby; 2011:1523–55.
31. Koletzko B, Goulet O, Hunt J, Krohn K, Shamir R. Guidelines on paediatric parenteral nutrition of the European Society of Paediatric Gastroenterology, Hepatology, and Nutrition (ESPGHAN) and the European Society for Clinical Nutrition and Metabolism (ESPEN), Supported by the European Society of Paediatric Research (ESPR). *J Pediatr Gastroenterol Nutr.* 2005;41 Suppl 2:S1–87.
32. Neu J, Walker WA. Necrotizing enterocolitis. *N Engl J Med.* 2011;364:255–64.
33. Neu J. Gastrointestinal development and meeting the nutritional needs of premature infants. *Am J Clin Nutr*. 2007;85:629S–34S.
34. Tudehope DI. Human milk and the nutritional needs of preterm infants. *J Pediatr.* 2013;162:S17–25.
35. Human Milk Banking Association of North America. 2011 *Best Practice for Expressing, Storing, and Handling Human Milk in Hospital, Homes, and Child Ccare Setting*. 3rd ed. Fort Worth: Human Milk Banking Association of North America (HMBANA); 2011.
36. Ewaschuk JB, Unger S, O'Connor DL, et al. Effect of pasteurization on selected immune components of donated human breast milk. *J Perinatol.* 2011;31:593–8.
37. Moro GE, Arslanoglu S. Heat treatment of human milk. *J Pediatr Gastroenterol Nutr*. 2012;54:165–6.
38. American Academy of Pediatrics. Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics. 2012;129:e827–41.*
39. Arslanoglu S, Ziegler EE, Moro GE. Donor human milk in preterm infant feeding: evidence and recommendations. *J Perinat Med. 2010;38:347–51*.
40. Quigley MA, Henderson G, Anthony MY, McGuire W. Formula milk versus donor breast milk for feeding preterm or low birth weight infants. *Cochrane Database Syst Rev.* 2008:CD002971.
41. Quigley MA, Henderson G, Anthony MY, McGuire W. Formula milk versus donor breast milk for feeding preterm or low birth weight infants. *Cochrane Database Syst Rev.* 2007:CD002971.
42. Arslanoglu S, Moro GE, Ziegler EE. Adjustable fortification of human milk fed to preterm infants: does it make a difference? *J Perinatol*. 2006;26:614–21.
43. de Halleux V, Rigo J. Variability in human milk composition: benefit of individualized fortification in very-low-birth-weight infants. *Am J Clin Nutr.* 2013;98(2):529S–35S.
44. Rochow N, Fusch G, Choi A, et al. Target fortification of breast milk with fat, protein, and carbohydrates for preterm infants. *J Pediatr*. 2013;163:1001–7.
45. Kuschel CA, Harding JE. Multicomponent fortified human milk for promoting growth in preterm infants. *Cochrane Database Syst Rev* 2004:CD000343.
46. Martin I, Jackson L. Question 1. Is there an increased risk of necrotising enterocolitis in preterm infants whose mothers' expressed breast milk is fortified with multicomponent fortifier? *Arch Dis Child.* 2011;96:1199–201.
47. Senterre T, Rigo J. Optimizing Early Nutritional Support Based on Recent Recommendations in VLBW Infants and Postnatal Growth Restriction. *J Pediatr Gastroenterol Nutr*. 2011;53:536–42.

48. Tillman S, Brandon DH, Silva SG. Evaluation of human milk fortification from the time of the first feeding: effects on infants of less than 31 weeks gestational age. *J Perinatol.* 2012;32:525–31.
49. Morgan J, Bombell S, McGuire W. Early trophic feeding versus enteral fasting for very preterm or very low birth weight infants. *Cochrane Database Syst Rev.* 2013;3:CD000504.
50. Morgan J, Young L, McGuire W. Delayed introduction of progressive enteral feeds to prevent necrotising enterocolitis in very low birth weight infants. *Cochrane Database Syst Rev*. 2011:CD001970.
51. Morgan J, Young L, McGuire W. Slow advancement of enteral feed volumes to prevent necrotising enterocolitis in very low birth weight infants. *Cochrane Database Syst Rev*. 2011:CD001241.
52. Hawes J, McEwan P, McGuire W. Nasal versus oral route for placing feeding tubes in preterm or low birth weight infants. *Cochrane Database Syst Rev.* 2004:CD003952.
53. McGuire W, McEwan P. Transpyloric versus gastric tube feeding for preterm infants. *Cochrane Database Syst Rev.* 2007:CD003487.
54. Premji SS, Chessell L. Continuous nasogastric milk feeding versus intermittent bolus milk feeding for premature infants less than 1500 grams. *Cochrane Database Syst Rev.* 2011:CD001819.
55. Mihatsch WA, von Schoenaich P, Fahnenstich H, et al. The significance of gastric residuals in the early enteral feeding advancement of extremely low birth weight infants. *Pediatrics.* 2002;109:457–9.
56. Deshpande G, Rao S, Patole S, Bulsara M. Updated meta-analysis of probiotics for preventing necrotizing enterocolitis in preterm neonates. *Pediatrics*. 2010;125:921–30.
57. Mihatsch WA, Braegger CP, Decsi T, et al. Critical systematic review of the level of evidence for routine use of probiotics for reduction of mortality and prevention of necrotizing enterocolitis and sepsis in preterm infants. *Clin Nutr.* 2012;31:6–15.
58. Embleton ND, Berrington JE, McGuire W, Stewart CJ, Cummings SP. Lactoferrin: Antimicrobial activity and therapeutic potential. Semin Fetal Neonatal Med 2013.
59. Lapillonne A, Carnielli VP, Embleton ND, Mihatsch W. Quality of newborn care: adherence to guidelines for parenteral nutrition in preterm infants in four European countries. *BMJ Open*. 2013;3:e003478.
60. Rigo J, Senterre T. Parenteral nutrition. In: Buenocore G, Bracci R, Weindling M, eds. *Neonatology A Practical Approach to Neonatal Diseases*. Milan, Italy: Springer-Verlag Italia; 2012:311–9.
61. Vlaardingerbroek H, Vermeulen MJ, Rook D, et al. Safety and efficacy of early parenteral lipid and high-dose amino acid administration to very low birth weight infants. *J Pediatr.* 2013;163:638-44 e1–5.
62. Senterre T, Rigo J. Reduction in postnatal cumulative nutritional deficit and improvement of growth in extremely preterm infants. *Acta Paediatr.* 2012;101:e64–70.
63. Vlaardingerbroek H, Veldhorst MA, Spronk S, van den Akker CH, van Goudoever JB. Parenteral lipid administration to very-low-birth-weight infants—early introduction of lipids and use of new lipid emulsions: a systematic review and meta-analysis. *Am J Clin Nutr.* 2012;96:255–68.
64. Arsenault D, Brenn M, Kim S, et al. A.S.P.E.N. Clinical Guidelines: hyperglycemia and hypoglycemia in the neonate receiving parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2012;36:81–95.

65. Bonsante F, Iacobelli S, Latorre G, et al. Initial amino acid intake influences phosphorus and calcium homeostasis in preterm infants—it is time to change the composition of the early parenteral nutrition. *PLoS One.* 2013;8:e72880.

Nutritional Management of Preterm Infants Post-Discharge

33

Mandy Brown Belfort, MD, MPH

INTRODUCTION

During the neonatal intensive care unit (NICU) hospitalization, nutritional management of the preterm infant aims to support growth and nutrient accretion equivalent to intrauterine rates. However, even when nutritional care meets or exceeds current guidelines, growth velocity during the NICU hospitalization frequently does not match that of the gestational age equivalent fetus[1] and many preterm infants are left with substantial growth and nutrient deficits at NICU discharge. Nutritional management of the post-discharge preterm infant is directed toward compensating for these deficits as well as supporting normal growth.

The first part of this chapter focuses on specific deficits that preterm infants accrue during the NICU hospitalization, typical patterns of growth that occur after discharge, and evidence regarding the impact of post-discharge growth patterns on long-term health and developmental outcomes. Next, specific post-discharge strategies to compensate for growth and other deficits at NICU discharge are reviewed.

GROWTH AND NUTRITIONAL DEFICITS AT NICU DISCHARGE

Preterm infants are typically discharged from the NICU between 35 and 40 weeks postmenstrual age (within a month of term equivalent). Nutritional

requirements after NICU discharge must address growth and nutrient deficits accrued prenatally and postnatally during the NICU hospitalization, and in addition to supporting normal post-term growth and nutrient accretion.

Deficits in Weight, Length, and Head Circumference

Prenatal deficits relate to intrauterine growth restriction, which is more common among fetuses subsequently born preterm than those who remain in-utero until term.[2] As demonstrated in several multi-center studies, postnatal weight gain, linear growth, and head growth in the NICU are substantially slower than growth of the gestational age equivalent in-utero fetus. This phenomenon is often referred to as "extrauterine growth failure." Notably, this postnatal growth failure occurs during the same developmental window as intrauterine growth failure occurs for an infant who is subsequently born growth restricted at term (Figure 33-1).

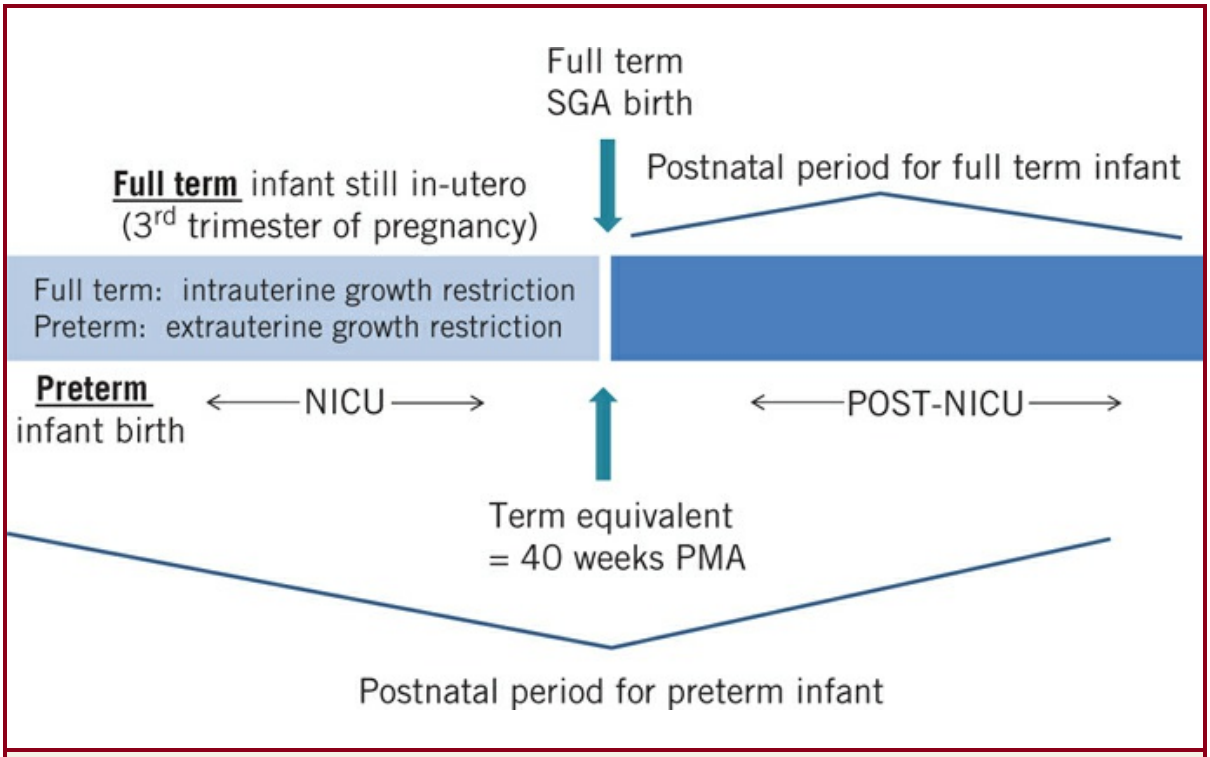


Figure 33-1 For the preterm infant, extrauterine growth restriction during the NICU hospitalization occurs with the same developmental timing as intrauterine growth restriction for the infant subsequently born small for gestational age (SGA) at term. PMA is postmenstrual age.

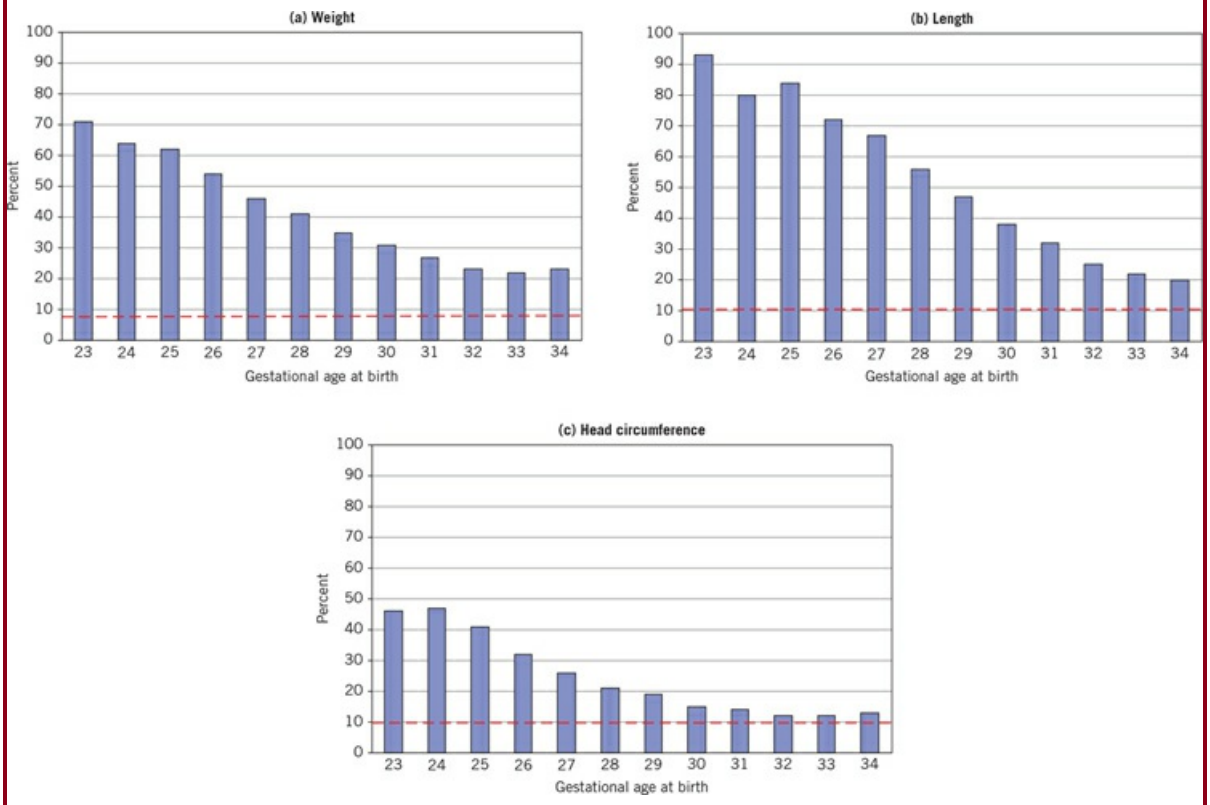


Figure 33-2 Percent of infants by gestational age with (A) weight (*n* = 23,873); (B) length (*n* = 17,203); and (C) head circumference (*n* = 20,885) <10th percentile at NICU discharge.[3] The dashed line represents the expected percent <10th percentile. The percent <10th percentile increases with decreasing gestational age at birth. From Clark.[3]

In one study[3] of infants 23–34 weeks' gestation at 124 US NICUs, 28% of infants had a discharge weight <10th percentile, 34% had length <10th percentile, and 16% had head circumference <10th percentile. The proportion with discharge size <10th percentile was higher for infants of lower gestational ages (Figure 33-2). Within gestational age categories, lower birth weight—reflective of fetal growth restriction—was associated with a higher proportion <10th percentile at discharge, underscoring the contribution of prenatally acquired deficits. Another study[4] of moderately preterm infants born at 30–35 weeks' gestation found that the mean weight *z*-score at birth was −0.16 but by discharge had fallen to −0.87; length and head circumference data were not reported. Growth outcomes varied considerably between NICUs, possibly reflecting differences in feeding practice. In sum, preterm infants are often born after a period of

intrauterine growth restriction, and postnatally in the NICU accumulate additional, substantial deficits in weight, length, and head circumference relative to the appropriately grown, full-term infant at birth.

Body Composition Differences

Deficits in body size at NICU discharge are accompanied by notable differences in body composition between preterm infants and their full-term counterparts. A recent systematic review and meta-analysis[5] summarized the data from nine studies on this topic. While total body fat percent was higher among preterm infants at term equivalent age vs. full-term infants at birth, fat-free mass was substantially lower (Table 33-1). Results of two studies[6,7] not included in the meta-analysis were largely similar. Another study[8] not included in the meta-analysis compared 67 small-for-gestational-age (SGA) preterm (≤34 weeks' gestation) infants at term equivalent age with 132 SGA full-term infants, and found that while the groups had similar weight (2403 vs. 2314 g), the preterm infants were shorter (45.3 vs. 48.1 cm) and had a substantially higher percent fat mass (14.3% vs. 5.8%) than the full-term infants. The observed fat mass difference of 8.5% in SGA infants was more pronounced than differences previously observed between appropriate-for-gestational-age (AGA) preterm and full-term infants. In sum, as compared with full-term infants at birth, preterm infants at NICU discharge have substantially greater adiposity and less fat-free mass than their full-term counterparts, and this difference may be more pronounced for SGA preterm infants. It is possible that the greater adiposity reflects an imbalance of protein: energy in the NICU diet as compared with the intrauterine environment, favoring the deposition of adipose tissue rather than lean mass.[9–11]

Energy and Protein Deficits

In addition to anthropometric differences, total energy deficits are important to quantify because postnatal energy intake is a key determinant of weight gain, in one study[12] accounting for 45% of the observed variation in weight *z*-score change from birth to 5 weeks. That study estimated that for infants born <30 weeks' gestational age, the cumulative energy deficit after five weeks in the NICU is 813 ± 542 kcal/kg. The estimated deficit was less (382 ± 263 kcal/kg) for infants born <30 weeks.

TABLE 33-1 Summary of Meta-Analyses of Studies Describing Differences in Body

Composition and Size Between Preterm Infants at Term Equivalent and Full-Term Infants at Birth				
Outcome	**Number of Studies (Participants)**	**Direction of Effect**	**Mean Effect Size (95% CI)**	**P-Value**
Total body fat %	7 (672)	Preterm > term	3% (0.25%, 5.88%)	0.03
Fat mass	5 (297)	Preterm < term	−50 g (−90 g, −10 g)	0.02
Fat free mass	3 (137)	Preterm < term	−460 g (−340 g, −270 g)	<0.0001
Weight	7 (577)	Preterm < term	−560 g (−750 g, −440 g)	<0.0001
Length	4 (383)	Preterm < term	−3.7 cm (−4.6 g, −2.8 g)	<0.0001
Head circumference	5 (450)	Preterm < term	−1 cm (−1.5 g, −0.5 g)	<0.0001

Source: From Johnson.[5]

In addition to deficits in energy, even with contemporary preterm formulas and human milk fortification, specific macro- and micronutrient deficits accrue prior to NICU discharge, which are relevant for post-NICU nutritional strategies. In one study,[12] typical protein deficits accumulated over five postnatal weeks were estimated at 23 ± 12 g/kg for infants <30 weeks, and 13 ± 15 g/kg for infants ≥30 weeks.

Mineral Deficits

Preterm infants are born before completing the third trimester of gestation, an important period for fetal accretion of minerals including calcium, phosphorus, and iron. Because postnatal enteral and parenteral nutrition rarely achieve fetal accretion rates despite routine fortification of preterm formula and human milk, at NICU discharge preterm infants may face residual deficits. Deficits in calcium and phosphorusare reflected by the lower bone mineral content seen in preterm infants at discharge, as compared with full-term newborn infants.[13] Regarding iron, in addition to missed fetal accretion, the preterm infant often experiences substantial iron losses related to phlebotomy in the NICU, exacerbating the deficit at NICU discharge.

Deficits Specific to Human Milk Fed Infants

Human milk is considered the normative food for all infants, including those born preterm[14]; however, human milk requires fortification to meet the nutritional requirements of the growing preterm infant during the NICU hospitalization. Even despite fortification, at NICU discharge, many preterm infants fed predominantly human milk face greater nutritional deficits than formula fed infants. These deficits are directly relevant to strategies for post-NICU nutrition for breast-fed infants. Specific deficits associated with the use of donor human milk are particularly important to consider given its rising use in the United States.[15]

Several contemporary single center studies suggest that at NICU discharge, human milk-fed infants have greater growth deficits than formula fed infants, despite appropriate fortification of human milk. In one,[16] as compared with infants fed <25% human milk, infants fed >75% human milk gained less weight (*z*-score change from birth to discharge −0.6 vs. −0.1 based on an external fetal growth reference,[17] Figure 33-3). In another,[18] weight, length, and head circumference were all smaller for infants who were fed predominantly breast milk in the NICU as compared with formula fed infants.

Donor human milk is used increasingly[15] during the NICU hospitalization to supplement mother's milk when it is inadequate or not available. Both observational and experimental evidence suggests that despite fortification, donor milk feeding during the NICU hospitalization is associated with poorer growth as compared with mother's milk feeding, either exclusively or with formula supplementation. In one observational study, among predominantly human-milk-fed infants, weight gain was slower for infants fed >75% donor human milk vs. >75% mother's milk (*z*-score change −0.84 vs. −0.56). In a randomized trial[19] of donor human milk vs. preterm formula to supplement mother's milk in the NICU, infants supplemented with donor milk had slower weight gain (20.1 g/kg/d vs. 17.1 g/kg/d), likely an underestimate of the true difference since 21% of infants initially assigned to the donor human milk group crossed over to the preterm formula group due to poor weight gain. Pasteurization of donor human milk inactivates several lipase enzymes normally present in breast milk,[20] which may limit digestion and absorption of fat and partially explain the slower weight gain observed in donor milk fed infants. Donor milk may also contain less DHA than mother's milk, contributing to a deficit in this important fatty acid.

Summary

The preterm infant faces growth and nutrient deficits acquired prenatally, followed by deficits acquired postnatally during the NICU hospitalization. These deficits are reflected in the preterm infant's smaller body size at term equivalent, as compared with the full term infant at birth, and are accompanied by energy deficits and deficits in accretion of protein and minerals. Preterm infants at term also have a greater proportion of adipose rather than lean mass relative to their full term counterparts. Finally, human milk fed infants, particularly those infants fed donor human milk, typically leave the NICU with more marked nutritional deficits than preterm infants fed formula. Nutritional support after NICU discharge must compensate for these deficits, in addition to supporting normal longitudinal growth.

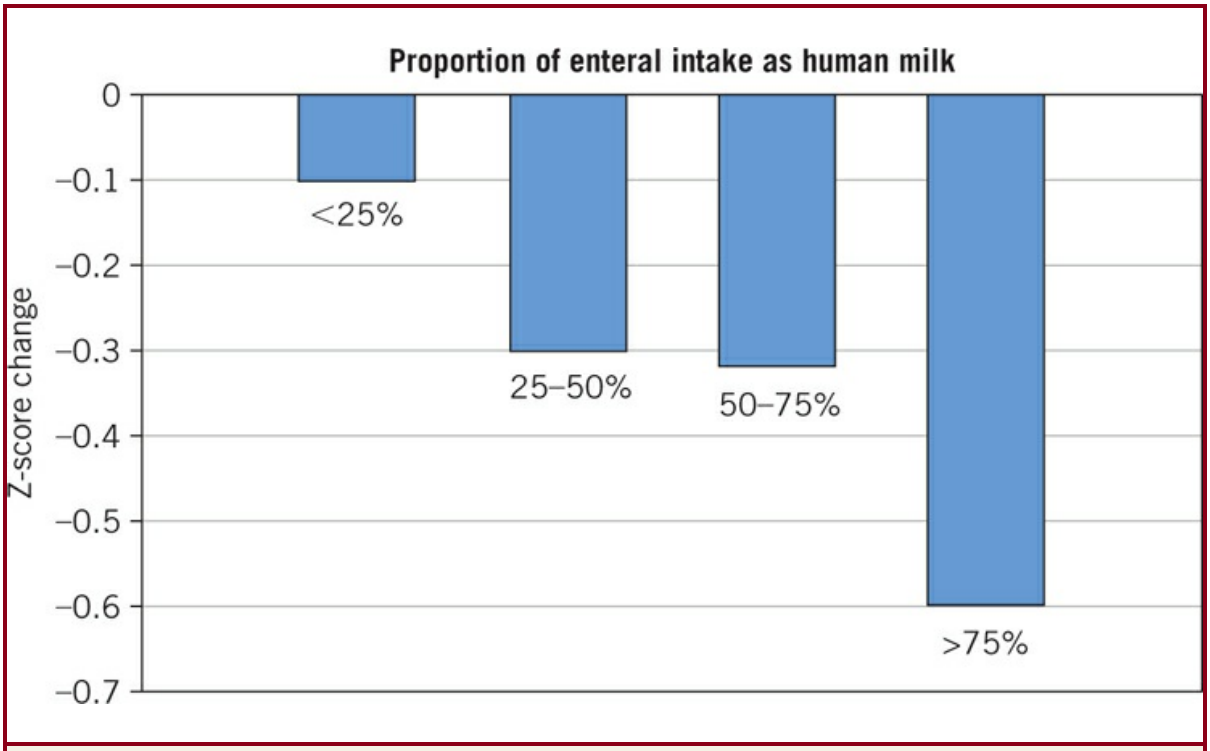


Figure 33-3 Weight z-score change from birth to NICU discharge according to proportion of enteral intake as human milk. Infants who received more human milk had a greater weight deficit at NICU discharge. From Colaizy et al.[16]

PATTERNS OF POST-DISCHARGE GROWTH

Several longitudinal studies have reported growth patterns of preterm infants post-discharge. Studies of older cohorts have the advantage of

following children to young adulthood, but reflect outdated nutritional practices. More recent studies reflect current practices, but lack long-term outcome data. Important considerations with respect to patterns post-NICU growth include timing of catch-up from deficits accrued prenatally and postnatally in the NICU; the influence of SGA status on post-NICU growth potential; sex differences; and changes in body composition over time, specifically differences in adiposity vs. lean mass.

Hack and colleagues[21] followed a US cohort of 195 VLBW infants born in 1977 to 1979 from birth to 20 years of age. In that cohort, catch up in weight after term began in early infancy, but was incomplete by 20 months (weight *z*-score increase from −1.79 to −1.16 in males; and from −1.12 to −0.82 in females). The pattern of early catch up in linear growth was similar with length *z*-scores increasing from −2.56 to −0.55 in males and −1.22 to −0.16 in females. At age 8 years, as compared with full-term controls, both males and females remained lighter (males by 4.3 kg; females, 3.6 kg) and shorter (males by 3.9 cm; females, 2.9 cm). By age 20 years, weight and height were similar for preterm vs. control females, but not males, who were 10.5 kg lighter and 3.1 cm shorter than controls. Body mass index (BMI) at age 20 years was 2.6 kg/m^2 lower for preterm vs. control males, but not different among females. Among males but not females, SGA infants were more likely to be of subnormal size at age 20 years than appropriately grown infants, for example 32% SGA vs. 6% AGA males were of subnormal weight and 21% SGA vs. 4% AGA males were of subnormal height (Figure 33-4).

Similarly, a Dutch study[22] of 1338 children born <32 weeks or <1500 g in 1983 similarly observed persistent stunting at age 10 years in SGA but not AGA infants. In a Canadian cohort of 147 extremely low birth weight (ELBW, <1000 g) born 1977 to 1982, after a period of decline in weight *z*-score from birth to 1 year, catch-up in both weight and length/height continued through infancy, childhood, and adolescence.[23] By young adulthood (age 23 years), ELBW participants remained somewhat lighter and shorter than full-term controls, but BMI was similar suggesting that gains in weight and length/height were proportional. An Australian study[24] of VLBW infants born in 1977 to 1982 also described substantial catch-up in weight and height by age 14 years relative to normal birth weight controls, but with a persistent deficit in weight (*z*-score −0.14 for infants born <1000 g) and height (*z*-score −0.43 for infants born <1000 g and −0.11 for infants born 1000–1499 g). Taken together, these data from older cohorts followed to adolescence and young adulthood suggest that catch-up in weight and height continues through adolescence, but may be

incomplete by early adulthood in some, particularly males and SGA infants.

A more contemporary cohort of 1151 US ELBW infants born in 1993 and 1994 was evaluated at 36 weeks postmenstrual age and again at 18–22 months. At 36 weeks postmenstrual age, virtually all infants had weight <10th percentile as compared with national normative standards. By 18–22 months, 40% of infants born at 600–1000 g remained <10th percentile for weight.[25] Correlates of poorer postnatal weight gain included SGA status and lower birth weight. Another US cohort of 154 ELBW infants born between 1997 and 1999 was studied through 20 months, at which time the mean weight *z*-score was −1.56, length −0.87, and head circumference −0.55, reflecting incomplete catch-up to their full-term peers.[26] A larger (*n* = 1320), population-based German VLBW cohort born in 1998 and 1999 was followed to age 6 years. As compared with a reference population, the VLBW children showed continued but incomplete catch-up in weight and length from 0.8 months through 6 years of age.[27] For example, height *z*-score increased from −0.75 to −0.62 at 6 years in that time interval. Preterm SGA infants remained lighter (weight *z*-score −1.09 vs. −0.54) and shorter (height *z*-score −0.78 vs. −0.48) than their AGA counterparts. These data suggest that even in more contemporary cohorts, VLBW and particularly ELBW infants remain substantially lighter and shorter in late infancy and at school age than their full-term counterparts.

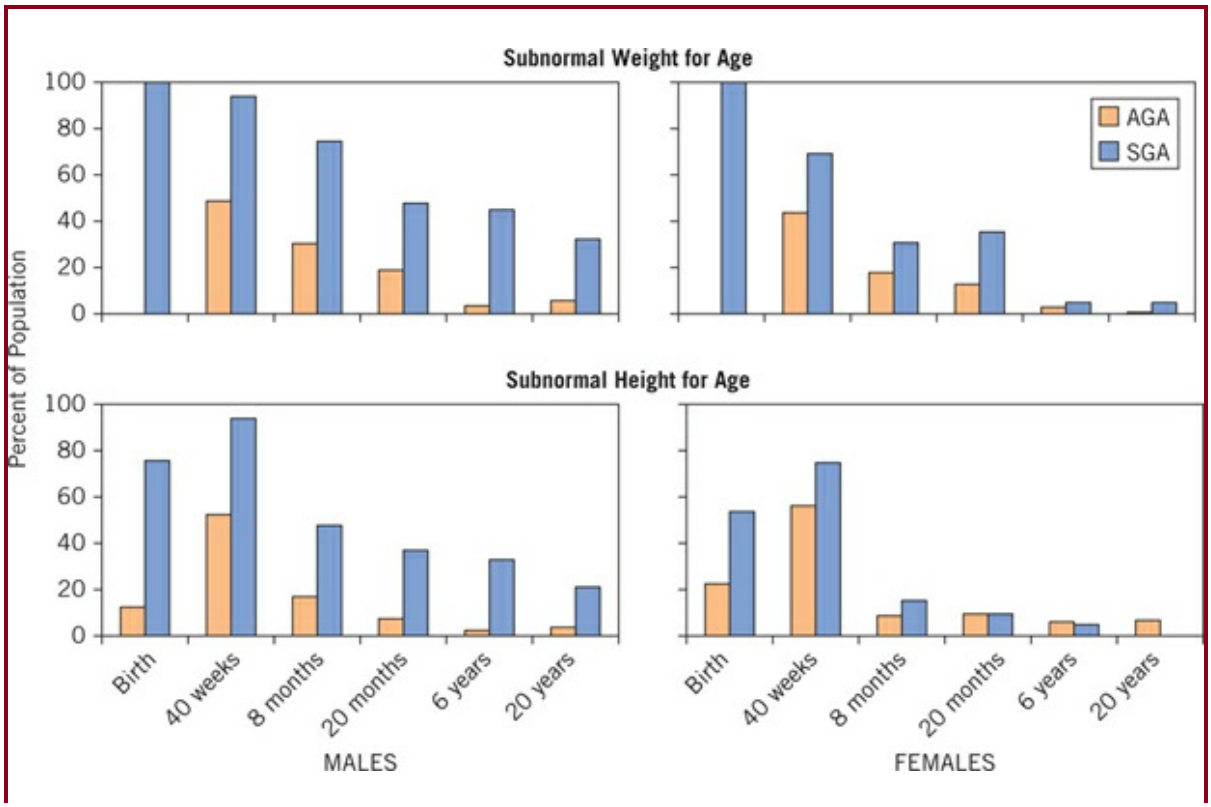


Figure 33-4 Weight *z*-score change from birth to NICU discharge according to proportion of enteral intake as human milk. Infants who received more human milk had a greater weight deficit at NICU discharge. From Hack et al.[21]

Limited data exist regarding post-NICU changes in body composition. In one study[28] of 26 preterm (<35 weeks' gestation) and 97 full-term infants—all of whom were appropriately grown at birth—assessed at term equivalent, preterm infants were lighter and shorter, and had higher body fat percent (18.7% vs. 15.1%, P <0.0001); by 4 months corrected age, weight and length were similar, and body fat percent was still higher among preterm infants, but the difference was no longer statistically significant (27.7% vs. 23.9%, P = 0.07). Further study regarding adiposity and lean mass trajectories of preterm infants after NICU discharge may be informative about optimal patterns of growth.

Summary

Many preterm infants catch up to their full term counterparts in weight and length during the first two years after NICU discharge. Most have caught up by school age, although some show continued catch-up growth through adolescence. Males and infants born SGA are at risk for deficits in weight and height that are still apparent in early adulthood. In terms of body composition, data are limited as to whether the greater fat percent seen at NICU discharge persists into the post-discharge period.

SHORT- AND LONG-TERM EFFECTS ON POST-DISCHARGE GROWTH

While longitudinal studies demonstrate that preterm infants experience poorer growth after NICU discharge than their full-term counterparts after birth, the optimal pattern of post-NICU growth is not known. Assessing the impact of early growth and nutrition on long-term health and developmental outcomes can inform recommendations for post-NICU growth targets. Specific long-term considerations include maximizing neurodevelopmental potential while also addressing emerging concerns about adverse effects of promoting rapid infant growth on later cardio metabolic health.

Post-Discharge Growth and Neurodevelopment

Weight gain during the NICU hospitalization is an important determinant of later neurodevelopmental outcome[29,30] but relatively few studies have examined the impact of infant growth after NICU discharge. In one single-center study,[31] greater weight gain and linear growth in the first two years were associated with higher developmental scores at age 2 years. In a multi-center US cohort of preterm, low-birth-weight infants born in the 1980's, higher IQ at age 8 years was associated with greater weight gain (2.1 points per SD, 95% CI 1.1, 3.1) and linear growth (2.4 points per SD, 95% CI 1.3, 3.5) from term to 4 months corrected age.[32] Another study[33] of the same cohort found that adequate postnatal weight gain was particularly important for preterm infants with fetal growth restriction. Similarly, in a contemporary (born 2001–2005) Australian cohort, greater weight gain and linear growth from term to 4 months were associated with better motor scores at 18 months.[34] Of note, post-NICU weight gain out of proportion to linear growth did not appear to benefit neurodevelopment, whereas during the NICU hospitalization, excess weight gain appeared to be beneficial.

Post-Discharge Growth and Cardio Metabolic Health

Several recent studies demonstrate that preterm infants are at heightened risk for developing cardio metabolic diseases in adulthood. Finnish, Dutch, and New Zealand groups have reported greater insulin resistance in preterm as compared with full-term adolescents and young adults,[35–37] and preterm infants appear to be 60% more likely to develop type 2diabetes.[38] A meta-analysis found that in adolescence and young adulthood, systolic blood pressure of former preterm infants is approximately 2.5 mmHg higher than full-term infants.[39] Adults born preterm are about twice as likely to be hypertensive as those born full term.[40]

It is possible that promoting rapid postnatal growth may be harmful to cardio metabolic health.[41,42] In terms of later hypertension, while little experimental evidence exists specifically regarding post-NICU growth, some[32,35] but not all[43,44] observational studies of post-NICU growth suggest a possible link with later, higher blood pressure. In terms of insulin resistance, a Dutch study of 345 infants <32 weeks and/or <1500 g reported a weak association of weight gain through 3 months of age with the log insulin concentration on fasting blood specimen at age 19 years, but no association with c-peptide or log HOMA-IR levels (homeostatic model assessment of insulin resistance).[45] Further study of a subset (*n* =

37) of the same cohort at age 21 years revealed that children in the lowest quartile of insulin sensitivity had been longer at 12 months and heavier at 24 months, as compared with children in the highest quartile of insulin sensitivity; these results reflect birth weight as well as weight gained after birth. More rapid infant weight gain in preterm infants has also been linked with obesity at age 8 years (odds ratio 2.7, 95% CI 1.9, 3.9 per 100 g additional weight gain in the first year),[46] with higher BMI (0.2 SD per SD additional weight gain from birth to 3 months), and with greater body fat and abdominal fat percentage as measured by dual-energy x-ray absorptiometry (DXA) at age 19.[47]

Summary and Recommendations

In sum, observational data suggests that promoting more rapid weight gain, and specifically linear growth, after NICU discharge may benefit long-term neurodevelopmental outcomes, but rapid weight gain, particularly out of proportion to linear growth, may also increase the risk for adverse cardiometabolic effects. We recommend that clinicians caring for preterm infants after NICU discharge monitor weight gain and linear growth over time, plotting measurements on a growth curve such as the World Health Organization (WHO) Child Growth Standards (Appendix A). The WHO Child Growth Standards have the advantage over other growth curves of providing BMI-for-age curves for boys and girls from birth to 2 years, which can help identify excess weight gain out of proportion to linear growth, even in infancy. Plotting measurements for both chronologic and corrected age is helpful in monitoring catch-up from extrauterine growth failure experienced during the NICU hospitalization (Figure 33-5). Corrected age can be calculated using online calculators such as the one found at this website http://www.vtoxford.org/research/elbw/calc/readme.aspx.

INTERVENTIONS TO SUPPORT POST-DISCHARGE GROWTH

Given the substantial deficits that preterm infants experience at NICU discharge, combined with observational evidence associating more rapid post-NICU growth with beneficial neurodevelopmental outcomes, nutritional strategies to promote more rapid growth are of interest. For formula-fed infants, various strategies of supplementing protein and minerals (particularly calcium and phosphorus) with or without additional

calories have been examined. For breast milk fed infants, strategies to supplement breast milk have been proposed, although data about the benefits and risks are sparse. Relevant outcomes to consider include both short term (growth, safety) and longer term (neurodevelopment, cardio metabolic).

Energy and Nutrient Enrichment of Infant Formula After NICU Discharge

Whereas standard term formulas are based on the composition of mature human milk, in-hospital preterm formulas are typically enriched with energy, protein, and minerals, vitamins, and trace elements in amounts estimated to support intrauterine accretion rates. Post-discharge formulas are also enriched relative to term formulas, but generally provide less energy, protein, and minerals than in-hospital preterm formulas (Table 33-2). Studies have examined both preterm and post-discharge formulas for use in preterm infants after NICU discharge.

A recent Cochrane review[48] addressed randomized trials of feeding nutrient-enriched rather than standard full-term formula after NICU discharge (Tables 33-3 and 4). Of 15 relevant trials, 10 compared a post-discharge vs. term formula, whereas 5 compared an in-hospital preterm vs. term formula. Outcomes included growth, development, and bone mineralization. As compared with infants fed term formula, at 9 months of age those fed post-discharge formula were on average 244 g heavier and 7.3 mm longer, but by 12 and 18 months, no weight or length differences were noted (Table 33-3). In contrast, at 18 months, infants fed preterm formula after NICU discharge remained 490 g heavier and 11 mm longer (Table 33-4). Feeding preterm formula also led to better head growth (by 5.4 mm at 18 months) than term formula, but post-discharge formula did not. No significant differences were noted in neurodevelopmental outcome to 12 months or in body composition. Only one study of post-discharge vs. term formula examined bone mineralization, and found increased bone mineral content by DXA at 9 months in infants fed post-discharge formula (difference 20.6 mg/cm). Cardio Metabolic outcomes such as body mass index and blood pressure were not reported. In sum, there is little evidence to support a sustained beneficial effect of post-discharge formula on growth in infancy, although there may be a short-term benefit to bone mineralization. Continued feeding of preterm formula after NICU discharge may confer short term benefits on growth. Data are lacking on long-term neurodevelopmental benefits as well as adverse cardio

metabolic effects of feeding nutrient-enriched formulas to preterm infants after NICU discharge.

Isocaloric Nutrient Enrichment of Infant Formula After NICU Discharge

Because of the concern that providing excess energy after NICU discharge could lead to excess fat but not lean mass accumulation, it is relevant to examine the effect of supplementing protein and minerals but not energy after NICU discharge. One small (n = 37) study[49] compared a formula with added protein (2.2 vs. 2 g/100 mL) and minerals (e.g., calcium, 100 vs. 80 gm; phosphorus, 60 vs. 42.5 mg) vs. a standard term formula starting in the NICU and continuing to term equivalent. The enriched and control formulas were isocaloric. At hospital discharge, infants were similar in weight, length, and head circumference. By term equivalent, infants fed the enriched formula were on average 231 g heavier, with no differences noted in length or head circumference. At hospital discharge and by term equivalent, infants fed the enriched formula had greater bone mineral content measured by DXA (difference at term equivalent, 3 g/kg body weight), but similar fat mass and lean mass percent. Whether there were longer term effects on bone mineral content or other aspects of body composition was not assessed.

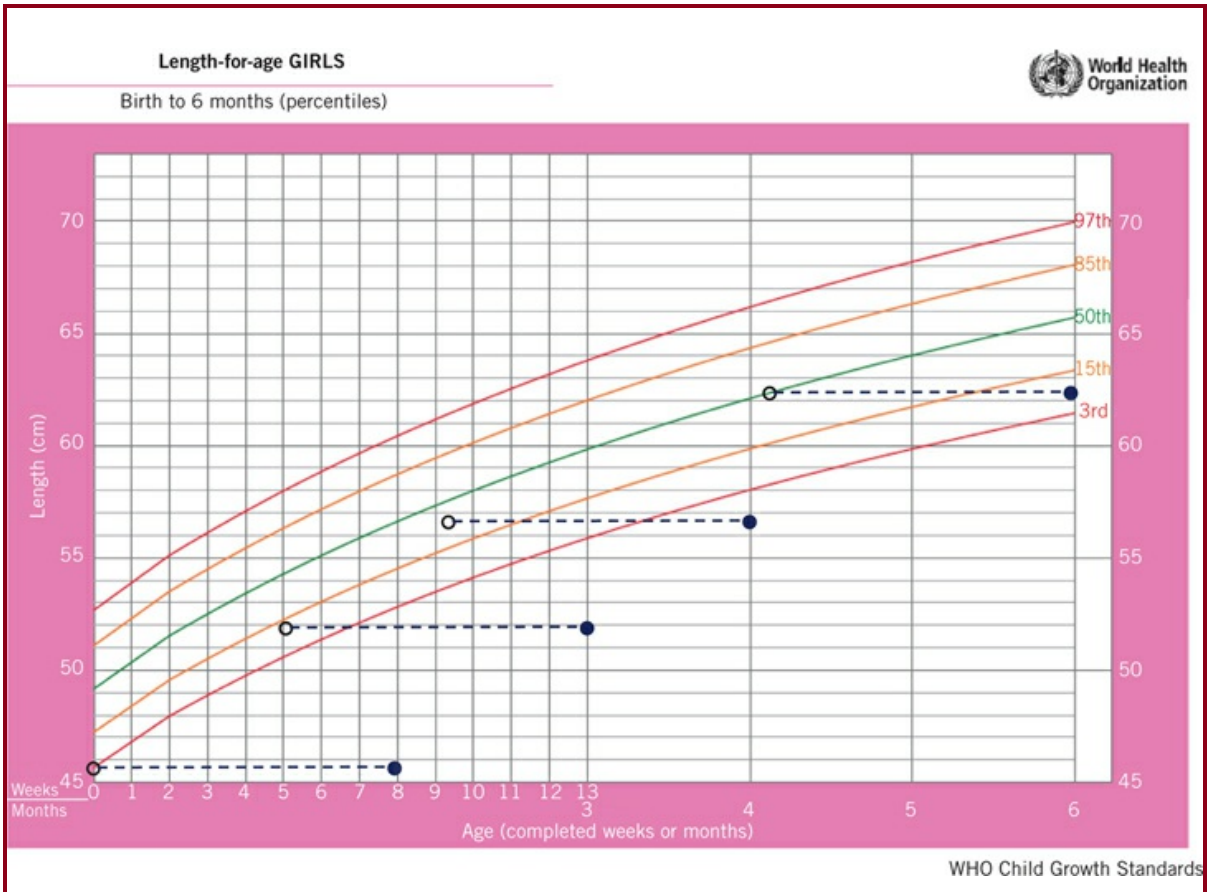


Figure 33-5 Example of post-NICU length measurements plotted for chronologic and corrected age on the World Health Organization Child Growth Standards. Filled circles indicate chronologic age and open circles indicate corrected age. In this example, the child was born at 32 weeks' gestation, or 8 weeks preterm. For 8 weeks chronologic age, length is well below the 3rd percentile, but for the corrected age of term, length is at the 3rd percentile. By 6 months chronologic age, the length-for-chronologic-age is between the 3rd and 15th percentiles, whereas for the corrected age of just over 4 months, length is at the 50th percentile.

Another study[50] compared two isocaloric formulas, one with more protein (1.70 vs. 1.47 g per 100 mL) and therefore a higher protein/energy ratio (2.75 g/100 kcal vs. 2.54 g/100 kcal). Calcium (65 vs. 50 mg), phosphorus (38 vs. 30 mg), DHA (14.0 vs. 6.7 mg), and several other nutrients were also different between the formulas. The trial was continued to 6 months of age. While weight and length at 6 months were similar between the groups, infants fed the enriched formula gained more lean

mass and less fat mass, as measured by DXA. Of note, a reference group of human milk fed infants gained less lean mass and more fat mass than either of the formula fed groups. Longer term outcomes were not reported.

Enrichment of Human Milk After NICU Discharge

While evidence is limited to support the multi-nutrient fortification of infant formula after NICU discharge, given the greater nutritional deficits accumulated by breast milk fed vs. formula fed infants during the NICU hospitalization and the relatively low protein content of mature human milk, post-discharge fortification of breast milk is considered separately. Fortifying breast milk has the potential to compensate for deficits specific to breast milk fed infants while allowing continued breastfeeding according to national and international recommendations.

TABLE 33-2 Comparison of Human Milk With Typical Term, Post-Discharge Preterm, and In-Hospital Preterm Formulas

	Mature Human Milk	Term Formula	Post-Discharge Formula	In-Hospital Preterm Formula
Energy (kcal)	650–700	680	740	810
Protein (g)	9	14	21	24
Fat (g)	35	36	39	41
Carbohydrate (g)	67	74	79	90
Calcium (mg)	200–250	530	890	1340
Phosphorus (mg)	120–140	290	490	670
Iron (mg)	0.3–0.9	12.2	13.3	14.4
Zinc (mg)	1–3	6.8	9	12.2
Sodium (mEq)	5.2–10.9	8	11.3	13.9
Vitamin A(mg)	0.3–0.6	2000	3330	10,100
Vitamin D (mcg)	0.33	410	590	2200
Vitamin E (mg)	3–8	13.5	30	51

Approximate content per liter.

Source: Adapted from Kleinman, RE, Ed. *AAP Pediatric Nutrition Handbook*, 6th edition (Elk Grove

Village, IL: American Academy of Pediatrics), 2009.

TABLE 33-3 Cochrane Meta-Analysis of Post-Discharge Versus Standard Term Formula After NICU Discharge: Effects on Growth

	Age at Follow-Up				
	3–4 Months	6 Months	9 Months	12 Months	18 Months
Weight (g)	−3.8 (−156.7, 149.2)	56.2 (−111.5, 224.0)	244.1 (17.0, 471.2)	71.5 (−344.1, 487.1)	100.0 (−246.9, 446.9)
Length (mm)	4.2 (−0.8, 9.1)	3.5 (−1.2, 8.1)	7.3 (1.8, 12.9)	−0.8 (−11.0, 9.3)	9.0 (0.3, 17.7)
Head circumference (mm)	−0.9 (−3.7, 2.0)	0.7 (−2.1, 3.6)	0.2 (−3.2, 3.5)	0.3 (−5.5, 6.0)	−3.0 (−8.2, 2.2)

Numbers are mean differences (95% confidence intervals). Positive numbers indicate a beneficial effect of the post-discharge formula.

Source: From Young.[48]

TABLE 33-4 Cochrane Meta-Analysis of In-Hospital Preterm Versus Standard Term Formula After NICU Discharge: Effects on Growth

	Age at Follow-Up				
	3–4 Months	6 Months	9 Months	12 Months	18 Months
Weight (g)	74.4 (−267.1, 415.9)	74.6 (−164.7, 313.9)	112.0 (−482.7, 706.7)	539.5 (255.0, 823.9)	490.8 (142.2, 839.4)
Length (mm)	−2.3 (−13.1, 8.6)	1.8 (−6.3, 9.9)	−3.0 (−17.0, 11.0)	5.1 (−4.2, 14.5)	11.0 (1.9, 20.1)
Head circumference (mm)	3.61 (−2.1, 9.3)	5.8 (1.3, 10.3)	8.0 (0.9, 15.2)	6.1 (1.1, 11.1)	5.4 (0.7, 10.1)

Numbers are mean differences (95% confidence intervals). Positive numbers indicate a beneficial effect of the preterm formula.

Source: From Young.[48]

In contrast to evidence that fortified formula is of little benefit to growth after discharge, limited evidence suggests that fortifying human milk in the initial months after NICU discharge has a sustained benefit to growth. In one small (n = 39) study,[51] infants <33 weeks' gestation receiving predominantly (≥80%) human milk at the time of discharge were randomized to fortify half the daily feedings with a powdered multi nutrient human milk fortifier for 12 weeks vs. no fortification. At 12 weeks, infants in the fortification group were longer and had larger head circumferences as compared with control infants. Growth benefits persisted to 1 year,[52] with infants who received fortified milk approximately 1.2 kg heavier and 3.8 cm longer than control infants. Head circumference at 1 year was 1 cm larger for infants who received fortification (not statistically significant). Results also suggested better neurodevelopment in the fortification group. At 1 year, the Bayley mental development index score was 9 points (~2/3 standard deviation) higher for intervention infants, although the study was not powered for that outcome. Early visual acuity at 4 and 6 months was also better in the fortified infants,[53] although it is unclear to what extent the subtle differences observed represent clinically important neurodevelopmental differences. Taken together, these data support fortification of human milk after NICU discharge to promote better growth and neurodevelopment; however, the extent to which benefits persist beyond infancy is unknown.

Even despite fortification, infants fed breast milk after NICU discharge appear to have slower growth, different body composition, and less mineral accretion as compared with formula fed infants. A small (n = 37) study[54] compared growth, body composition, and bone mineral content of preterm (mean birth weight 1.4 kg) infants fed standard term formula vs. fortified breast milk after discharge. Although weight, length and head circumference were similar, formula fed infants had lower fat mass compared with breast milk fed infants, which parallels findings in term infants. Formula fed infants also had substantially higher bone mineral content at 6 months (159 vs. 132 g), although not at 12 months. The ratio of bone mineral content to lean mass was similar between breast milk and formula fed infants, suggesting that greater mineral accretion paralleled greater growth of lean mass in the formula fed infants. Longer term outcomes were not reported. It remains unclear whether faster vs. slower linear growth, greater gain in fat vs. lean mass, and degree of early bone mineralization is optimal with respect to later adiposity, neurodevelopment, and bone health outcomes.

A unique consideration for post-discharge fortification of breast milk

relates to the route of feeding. During most of the NICU hospitalization, fortified breast milk is administered by gavage tube. Once an infant is able to begin feeding orally by bottle or breast (typically around 34–35 weeks postmenstrual age and near the time of hospital discharge), the benefits of bottle feeding fortified milk must be weighed against the benefits of direct breastfeeding, since milk obtained by the infant directly at the breast cannot be fortified. Specifically, nutritional benefits of human milk fortification must considered alongside maternal preference for breastfeeding vs. bottle feeding and the logistical difficulties of expressing and fortifying milk, with the potential to reduce continued breastfeeding after NICU discharge.

Summary and Recommendations

Limited evidence supports the use of nutrient enriched formula for preterm infants after NICU discharge to improve early growth, and data are lacking with respect to long-term neurodevelopmental or cardiometabolic effects of such formulas. Typical in-hospital preterm formulas appear to support growth better than “transitional” or post-discharge formulas, but are not available outside of the hospital setting. Thus, standard practice is to discharge formula-fed infants on a post-discharge formula for several months to provide extra energy, protein, and minerals to support catch up in linear growth. Evidence from small studies suggests that fortifying human milk feedings after NICU discharge may improve both growth and neurodevelopment. A combination of direct feeding at the breast and bottle feeding of expressed and fortified milk may promote adequate growth while minimizing logistical barriers to continued breastfeeding. Clinicians should monitor growth with special attention to infants with rapid weight gain out of proportion to linear growth, which should prompt a decrease in caloric supplementation.

BREASTFEEDING AFTER NICU DISCHARGE

Continued breastfeeding of preterm infants after NICU discharge depends first on successful establishment of breastfeeding during the NICU hospitalization. In the United States, as compared with women who deliver at term, women who deliver preterm (<34 weeks’ gestation) are more than twice as likely to initiate breastfeeding and continue breastfeeding to 4 weeks. However by 10 weeks, women who deliver preterm are half as likely to breastfeed,[55] suggesting that while practices to support breast milk expression and feeding in the NICU are at least partially successful,

barriers after NICU discharge undermine continued breastfeeding.

Relatively recent data suggest that more than half of VLBW infants are still receiving at least some breast milk at NICU discharge. A California study[56] of 127 NICUs found that from 2005 to 2006, 61% of VLBW infants were fed breast milk at discharge, with wide variation noted across hospitals. Heavier infants and multiple births were more likely to receive breast milk at discharge, whereas younger women and those of racial and ethnic minorities were less likely to breast feed at discharge. An older (1999–2000) study[57] of 124 US NICUs found that approximately 50% of all NICU infants were receiving some breast milk at discharge, with similar inter-NICU differences and racial/ethnic and social disparities. National dataregarding breastfeeding continuation after NICU discharge are not available.

Despite early growth, bone, and neurodevelopmental advantages of post-discharge breast milk fortification, the process of expressing and fortifying milk itself may be a barrier to continued breastfeeding after NICU discharge. In the small randomized trial[51] of post-discharge breast milk fortification, breast milk intake was similar between fortified and unfortified groups at 12 weeks, but by 6 months, the experimental fortified infants had received less breast milk (33 vs. 71 mL/kg/d) vs. infants receiving unfortified milk. Total breastfeeding duration was also shorter for experimental infants (43 vs. 51 weeks). While greater lactation support during the intervention to 12 weeks is a possible explanation for these findings, another study found that simply providing additional lactation support does not appear to improve post-discharge breastfeeding duration, at least among highly motivated and socioeconomically advantaged populations.[58] Further research is needed to identify barriers to continued breastfeeding after NICU discharge.

Summary

Mothers who successfully initiate breastfeeding during the NICU hospitalization face challenges when continuing to breastfeed after discharge, including logistical barriers related to expressing and fortifying milk at home. Early lactation support may be helpful, but additional research is needed to identify strategies to improve duration and exclusivity of breastfeeding after NICU discharge.

SUMMARY

At the time of NICU discharge, the preterm infant faces substantial deficits in body size and nutrient accretion that must be addressed by strategies for nutritional support in the post-discharge period. Infants fed human milk may face greater deficits than formula-fed infants. Following NICU discharge, catch up in weight and length/height is complete by late infancy for many preterm infants, but for some continues through school age and late adolescence. A minority of preterm infants never fully catch up in weight and height to their term-born peers. More rapid post-discharge linear growth is associated with better neurodevelopment, but excess weight gain may lead to obesity-related complications later in life. Thus, the optimal pattern of post-discharge growth is unclear. Data from well-controlled randomized trials suggests that continued supplementation of energy, protein, and minerals after NICU discharge benefits short-term weight gain, linear growth, and possibly head growth, although evidence is stronger in support of formulas typically used during the NICU hospitalization rather than typical post-discharge formulas. Infants fed human milk after NICU discharge may benefit from at least partial milk fortification in combination with direct breastfeeding. Further research is needed regarding optimal nutrient supplementation in the post-discharge period, including a focus long-term growth, neurodevelopment, and cardiometabolic outcomes.

REFERENCES

1. Martin CR, Brown YF, Ehrenkranz RA, et al. Nutritional practices and growth velocity in the first month of life in extremely premature infants. *Pediatrics*. 2009;124(2):649–657.
2. Hutcheon JA, Platt RW. The missing data problem in birth weight percentiles and thresholds for “small-for-gestational-age”. *Am J Epidemiol*. 2008;167(7):786–792.
3. Clark RH, Thomas P, Peabody J. Extrauterine growth restriction remains a serious problem in prematurely born neonates. *Pediatrics*. 2003;111 (5, pt 1):986–990.
4. Blackwell MT, Eichenwald EC, McAlmon K, et al. Interneonatal intensive care unit variation in growth rates and feeding practices in healthy moderately premature infants. *J Perinatol*. 2005;25(7):478–485.
5. Johnson MJ, Wootton SA, Leaf AA, Jackson AA. Preterm birth and body composition at term equivalent age: a systematic review and meta-analysis. *Pediatrics*. 2012;130(3):e640–e649.
6. Simon L, Borrego P, Darmaun D, et al. Effect of sex and gestational age on neonatal body composition. *Br J Nutr*. Mar 28 2013;109(6):1105–8.
7. Cooke RJ, Griffin I. Altered body composition in preterm infants at hospital discharge. *Acta Paediatr*. 2009;98(8):1269–1273.
8. Gianni ML, Roggero P, Taroni F, et al. Adiposity in small for gestational age preterm infants assessed at term equivalent age. *Arch Dis Child Fetal Neonatal Ed*. 2009;94(5):F368–F372.
9. Kashyap S, Forsyth M, Zucker C, et al. Effects of varying protein and energy

intakes on growth and metabolic response in low birth weight infants. *J Pediatr*. 1986;108(6):955–963.
10. van Goudoever JB, Sulkers EJ, Lafeber HN, Sauer PJ. Short-term growth and substrate use in very-low-birth-weight infants fed formulas with different energy contents. *Am J Clin Nutr*. 2000;71(3):816–821.
11. Costa-Orvay JA, Figueras-Aloy J, Romera G, et al. The effects of varying protein and energy intakes on the growth and body composition of very low birth weight infants. *Nutr J*. 2011;10:140.
12. Embleton NE, Pang N, Cooke RJ. Postnatal malnutrition and growth retardation: an inevitable consequence of current recommendations in preterm infants? *Pediatrics*. 2001;107(2):270–273.
13. Ahmad I, Nemet D, Eliakim A, et al. Body composition and its components in preterm and term newborns: a cross-sectional, multimodal investigation. *Am J Hum Biol*. 2010;22(1):69–75.
14. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129(3): e827–e841.
15. Cohen RS. Current issues in human milk banking. *NeoReviews*. 2007;8: e289–e295.
16. Colaizy TT, Carlson S, Saftlas AF, Morriss FH, Jr. Growth in VLBW infants fed predominantly fortified maternal and donor human milk diets: a retrospective cohort study. *BMC Pediatr*. 2012;12:124.
17. Fenton TR. A new growth chart for preterm babies: Babson and Benda's chart updated with recent data and a new format. *BMC Pediatr*. 2003;3:13.
18. O'Connor DL, Jacobs J, Hall R, et al. Growth and development of premature infants fed predominantly human milk, predominantly premature infant formula, or a combination of human milk and premature formula. *J Pediatr Gastroenterol Nutr*. 2003;37(4):437–446.
19. Schanler RJ, Lau C, Hurst NM, Smith EO. Randomized trial of donor human milk versus preterm formula as substitutes for mothers'own milk in the feeding of extremely premature infants. *Pediatrics*. 2005;116(2):400–406.
20. Henderson TR, Fay TN, Hamosh M. Effect of pasteurization on long chain polyunsaturated fatty acid levels and enzyme activities of human milk. *J Pediatr*. 1998;132(5):876–878.
21. Hack M, Schluchter M, Cartar L, et al. Growth of very low birth weight infants to age 20 years. *Pediatrics*. 2003;112(1 Pt 1):e30–e38.
22. Knops NB, Sneeuw KC, Brand R, et al. Catch-up growth up to ten years of age in children born very preterm or with very low birth weight. *BMC Pediatr*. 2005;5:26.
23. Saigal S, Stoskopf B, Streiner D, et al. Growth trajectories of extremely low birth weight infants from birth to young adulthood: a longitudinal, population-based study. *Pediatr Res*. 2006;60(6):751–758.
24. Ford GW, Doyle LW, Davis NM, Callanan C. Very low birth weight and growth into adolescence. *Arch Pediatr Adolesc Med*. 2000;154(8):778–784.
25. Dusick AM, Poindexter BB, Ehrenkranz RA, Lemons JA. Growth failure in the preterm infant: can we catch up? *Semin Perinatol*. 2003;27(4): 302–310.
26. Sices L, Wilson-Costello D, Minich N, et al. Postdischarge growth failure among extremely low birth weight infants: correlates and consequences. *Paediatr Child Health*. 2007;12(1):22–28.
27. Trebar B, Traunecker R, Selbmann HK, Ranke MB. Growth during the first two years predicts pre-school height in children born with very low birth weight (VLBW): results of a study of 1,320 children in Germany. *Pediatr Res*. 2007;62(2):209–214.

28. Ramel SE, Gray HL, Ode KL, et al. Body composition changes in preterm infants following hospital discharge: comparison with term infants. *J Pediatr Gastroenterol Nutr*. 2011;53(3):333–338.
29. Ehrenkranz RA, Dusick AM, Vohr BR, et al. Growth in the neonatal intensive care unit influences neurodevelopmental and growth outcomes of extremely low birth weight infants. *Pediatrics*. 2006;117(4):1253–1261.
30. Belfort MB, Rifas-Shiman S, Sullivan T, et al. Infant growth before and after term: effects on neurodevelopment in preterm infants. *Pediatrics*.In press.
31. Latal-Hajnal B, von Siebenthal K, Kovari H, et al. Postnatal growth in VLBW infants: significant association with neurodevelopmental outcome. *J Pediatr*. 2003;143(2):163–170.
32. Belfort MB, Martin CR, Smith VC, et al. Infant weight gain and school-age blood pressure and cognition in former preterm infants. *Pediatrics*. 2010;125(6):e1419–e1426.
33. Casey PH, Whiteside-Mansell L, Barrett K, et al. Impact of prenatal and/or postnatal growth problems in low birth weight preterm infants on school-age outcomes: an 8-year longitudinal evaluation. *Pediatrics*. 2006;118(3):1078–1086.
34. Belfort MB, Rifas-Shiman SL, Sullivan T, et al. Infant growth before and after term: effects on neurodevelopment in preterm infants. *Pediatrics*. 2011;128(4):e899–e906.
35. Rotteveel J, van Weissenbruch MM, Twisk JW, Delemarre-Van de Waal HA. Infant and childhood growth patterns, insulin sensitivity, and blood pressure in prematurely born young adults. *Pediatrics*. 2008;122(2):313–321.
36. Hovi P, Andersson S, Eriksson JG, et al. Glucose regulation in young adults with very low birth weight. *N Engl J Med*. 2007;356(20):2053–2063.
37. Hofman PL, Regan F, Jackson WE, et al. Premature birth and later insulin resistance. *N Engl J Med*. 2004;351(21):2179–2186.
38. Kajantie E, Osmond C, Barker DJ, Eriksson JG. Preterm birth--a risk factor for type 2 diabetes? The Helsinki birth cohort study. *Diabetes Care*. 2010;33(12):2623–2625.
39. de Jong F, Monuteaux MC, van Elburg RM, et al. Systematic review and meta-analysis of preterm birth and later systolic blood pressure. *Hypertension*. 2012;59(2):226–234.
40. Crump C, Winkleby MA, Sundquist K, Sundquist J. Risk of hypertension among young adults who were born preterm: a Swedish national study of 636,000 births. *Am J Epidemiol*. 2011;173(7):797–803.
41. Singhal A, Fewtrell M, Cole TJ, Lucas A. Low nutrient intake and early growth for later insulin resistance in adolescents born preterm. *Lancet*. 2003;361(9363):1089–1097.
42. Singhal A, Cole TJ, Lucas A. Early nutrition in preterm infants and later blood pressure: two cohorts after randomised trials. *Lancet*.2001;357(9254):413–419.
43. Hack M, Schluchter M, Cartar L, Rahman M. Blood pressure among very low birth weight (<1.5 kg) young adults. *Pediatr Res*. 2005;58(4):677–684.
44. Keijzer-Veen MG, Finken MJ, Nauta J, et al. Is blood pressure increased 19 years after intrauterine growth restriction and preterm birth? A prospective follow-up study in The Netherlands. *Pediatrics*. 2005;116(3):725–731.
45. Finken MJ, Keijzer-Veen MG, Dekker FW, et al. Preterm birth and later insulin resistance: effects of birth weight and postnatal growth in a population based longitudinal study from birth into adult life. *Diabetologia*. 2006;49(3):478–485.
46. Casey PH, Bradley RH, Whiteside-Mansell L, et al. Evolution of obesity in a low birth weight cohort. *J Perinatol*. 2012;32(2):91–6.

47. Euser AM, Finken MJ, Keijzer-Veen MG, et al. Associations between prenatal and infancy weight gain and BMI, fat mass, and fat distribution in young adulthood: a prospective cohort study in males and females born very preterm. *Am J Clin Nutr*. 2005;81(2):480–487.
48. Young L, Morgan J, McCormick FM, McGuire W. Nutrient-enriched formula versus standard term formula for preterm infants following hospital discharge. *Cochrane Database Syst Rev*. 2012;3:CD004696.
49. Lapillonne A, Salle BL, Glorieux FH, Claris O. Bone mineralization and growth are enhanced in preterm infants fed an isocaloric, nutrient-enriched preterm formula through term. *Am J Clin Nutr*. 2004;80(6):1595–1603.
50. Amesz EM, Schaafsma A, Cranendonk A, Lafeber HN. Optimal growth and lower fat mass in preterm infants fed a protein-enriched postdischarge formula. *J Pediatr Gastroenterol Nutr*. 2010;50(2):200–207.
51. O'Connor DL, Khan S, Weishuhn K, et al. Growth and nutrient intakes of human milk-fed preterm infants provided with extra energy and nutrients after hospital discharge. *Pediatrics*. 2008;121(4):766–776.
52. Aimone A, Rovet J, Ward W, et al. Growth and body composition of human milk-fed premature infants provided with extra energy and nutrients early after hospital discharge: 1-year follow-up. *J Pediatr Gastroenterol Nutr*. 2009;49(4):456–466.
53. O'Connor DL, Weishuhn K, Rovet J, et al. Visual development of human milk-fed preterm infants provided with extra energy and nutrients after hospital discharge. *JPEN J Parenter Enteral Nutr*. 2012;36(3):349–353.
54. Wauben IP, Atkinson SA, Shah JK, Paes B. Growth and body composition of preterm infants: influence of nutrient fortification of mother's milk in hospital and breastfeeding post-hospital discharge. *Acta Paediatr*. 1998;87(7):780–785.
55. Colaizy TT, Saftlas AF, Morriss FH, Jr. Maternal intention to breast-feed and breast-feeding outcomes in term and preterm infants: Pregnancy Risk Assessment Monitoring System (PRAMS), 2000–2003. *Public Health Nutr*. 2012;15(4):702–710.
56. Lee HC, Gould JB. Factors influencing breast milk versus formula feeding at discharge for very low birth weight infants in California. *J Pediatr*. 2009;155(5):657–662. e1–2.
57. Powers NG, Bloom B, Peabody J, Clark R. Site of care influences breastmilk feedings at NICU discharge. *J Perinatol*. 2003;23(1):10–13.
58. Pinelli J, Atkinson SA, Saigal S. Randomized trial of breastfeeding support in very low-birth-weight infants. *Arch Pediatr Adolesc Med*. 2001;155(5):548–553.

The Term Infant

34

Ekhard E. Ziegler, MD and Susan J. Carlson, MMSc, RD

THE GOALS OF INFANT FEEDING

The goals of infant feeding are to meet all nutritional needs, to aid mother-infant bonding, to aid the infant in making the transition from the predominantly liquid diet of the infant to the predominantly solid food diet of the child and adult, and to aid in establishing habits of eating in moderation. The goals can best be met by breastfeeding. However, in industrialized countries, feeding of commercially available infant formulas, although not ideal, is an acceptable substitute. Breastfeeding, besides meeting all nutritional needs, offers protection against infectious illnesses, may foster optimal neurocognitive development and may offer some protection against conditions in adulthood such as obesity and atherosclerosis. Whether feeding practices during infancy can aid in establishing the goal of eating in moderation is unknown. Nevertheless, in various animal models habits established during infancy have been difficult to extinguish later in life and it therefore seems reasonable that habits of eating in moderation should be established during infancy because they may facilitate eating in moderation later in life.

TRENDS IN INFANT FEEDING

The description of trends presented here chronicles events as they occurred in the United States. As infant feeding evolved in generally similar fashion throughout the industrialized world, the trends presented here are presumed to be representative of trends in most industrialized countries.

Historically, in situations where breastfeeding was not possible, survival of the infant was in jeopardy. The feeding of preparations based

on various mammalian milks was associated with high morbidity and mortality. Success at formula feeding gradually improved from the end of the nineteenth century through the first half of the twentieth century. The objective was to make available safe and effective feedings for situations where breast milk was not available. By the 1950s, formulas reached a point where they provided adequate nutrition and appeared to be reasonably safe. In the absence of convincing evidence of the inferiority of formula feeding, many parents and physicians adopted formula feeding, presumably because of the supposed greater convenience. The prevalence of formula feeding increased and the prevalence of breastfeeding decreased.

The decrease in the prevalence of breastfeeding was associated with earlier introduction into the infant's diet of complementary foods and cow milk. By the end of the 1950s, feeding of complementary foods was common during the first 2 months of life. Whole or reduced-fat cow milk commonly replaced formula by 4–6 months of age and often even earlier.[1]

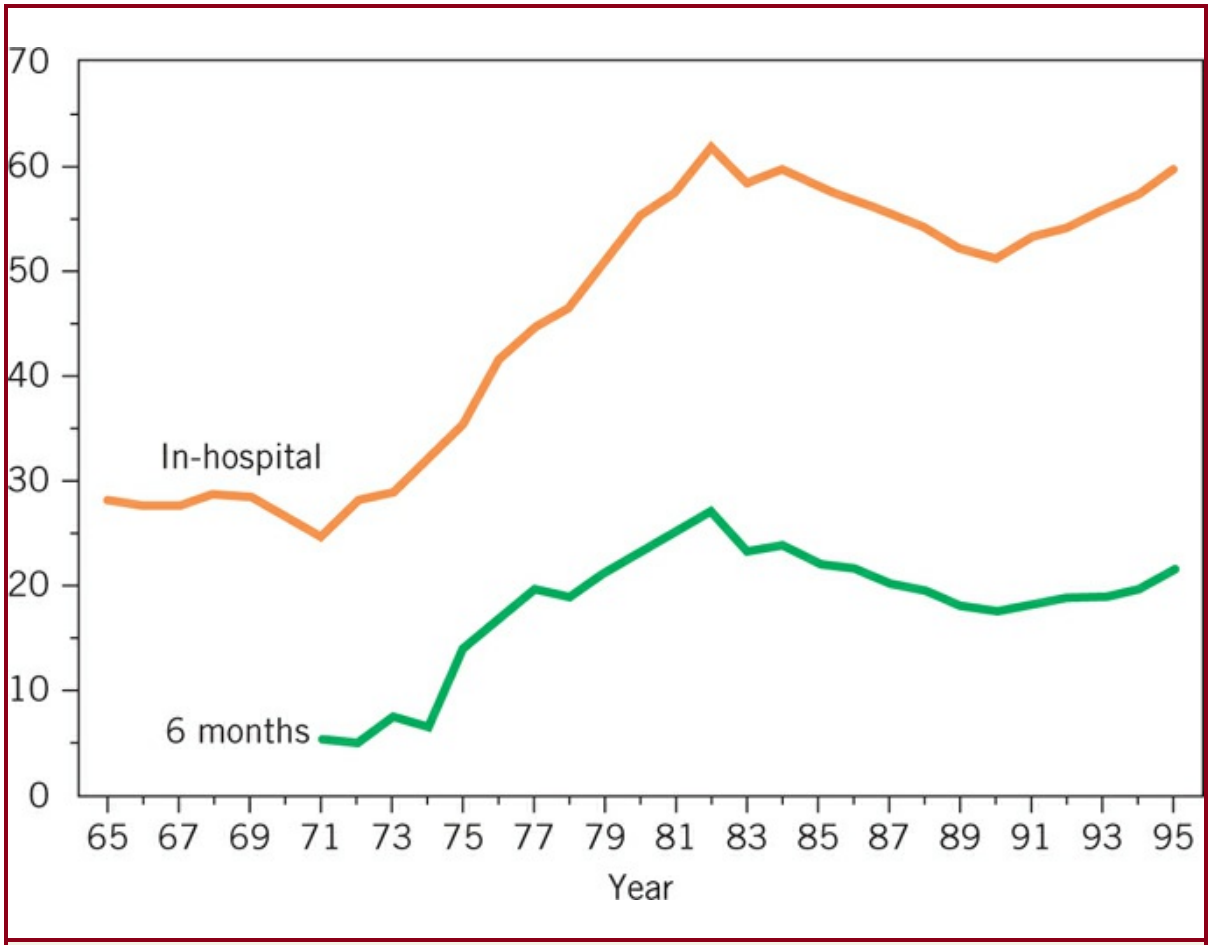


Figure 34-1 Prevalence of breastfeeding in the United States 1965-1995. Reproduced with permission from Ryan.[2]

A resurgence of breastfeeding began about 1971 (Figure 34-1). The reason(s) for the increase in breastfeeding are poorly understood. But enthusiasm for breastfeeding was sustained, at least in part, by the ever increasing number of reports on the demonstrated or assumed advantages of breastfeeding. As Figure 34-2 indicates, in 2001 about 70% of infants in the United States were breastfed for at least some time. That percentage increased to 83% by 2005–2007.[3] The relatively quick decrease in breastfeeding prevalence with age is at least partially explained by mothers returning to work. Despite the pressures of work, the number of women who breastfeed for six months and longer has continued to increase in the United States and reached 50% in 2005–2007.[3]

In association with the increase in breastfeeding, infants were increasingly fed formula after cessation of breastfeeding. Thus, formula feeding largely displaced feeding of cow milk in older infants except during the very end of the first year of life (Figure 34-3), a practice that seems to have persisted.[3] Also, complementary foods were less commonly fed during the early months of life. In 2005–2007, about 40% of mothers introduced complementary foods before 4 months.[4]

Trends in Formula Composition

The formulas fed in the United States in the late 1920s were generally prepared in the home from evaporated milk or fresh cow milk and sugar. The protein content was considerably greater than that of current formulas, the fat was butterfat, the added carbohydrate was corn syrup or sucrose and neither vitamins nor minerals were added. Prepared powdered formulas (to be diluted with water before use) were available but their use was quite limited until the 1950s. Over the years, formulas underwent a number of changes: the protein content was decreased and bovine whey proteins were included to result in a whey protein-casein ratio more like that of human milk; vegetable oils replaced butterfat; the added carbohydrate was lactose rather than corn syrup, and vitamins and minerals were included. See Appendix C for detailed composition of currently available formulas.

NUTRIENT REQUIREMENTS

The approach to estimating nutrient requirements of the infant differs considerably from that for adults. The normal adult requires regular intake of energy and specific nutrients to maintain the size and composition of the

body (i.e., requirements for “maintenance”), whereas infants and young children need, in addition, energy and nutrients for growth. The ratio of need for growth to need for maintenance is greatest for the fetus, the rapidly growing preterm infant and the term infant during the early months of life. Therefore, to obtain reasonable estimates of the requirements for energy and nutrients of the infant, it is desirable to have quantitative data on the increments in energy storage and the increments in specific nutrients needed for synthesis of new tissue. Defining the requirements of energy or nutrients for growth is much less important for children over the age of two years and for adolescents because by then the requirements for growth are quite small fractions of total requirements.

Thus, from a nutritional point of view, rapid growth is what differentiates infancy from all other ages. Nutrient requirements are exceedingly high primarily because nutrients are needed to support rapid growth. But growth is not only very rapid, it also changes dramatically in velocity as well as in composition. These dynamic changes are illustrated in Figure 34-4, which depicts rate as well as composition of growth of a “reference infant”. The rapid decline in the rate of growth during the first year of life is readily appreciated. It is also evident that during the first four months of life, infants deposit large amounts of body fat. During that period, fat accounts for as much as 40% of weight gain, and the energy necessary for deposition of this fat accounts for about 20% of energy requirements. Toward the end of the first year of life, fat deposition slows down markedly, and so does the proportion of energy requirement explained by fat deposition. Although accretion of protein as a percentage of weight gain increases with age, absolute protein accretion is highest during the first two months of life.

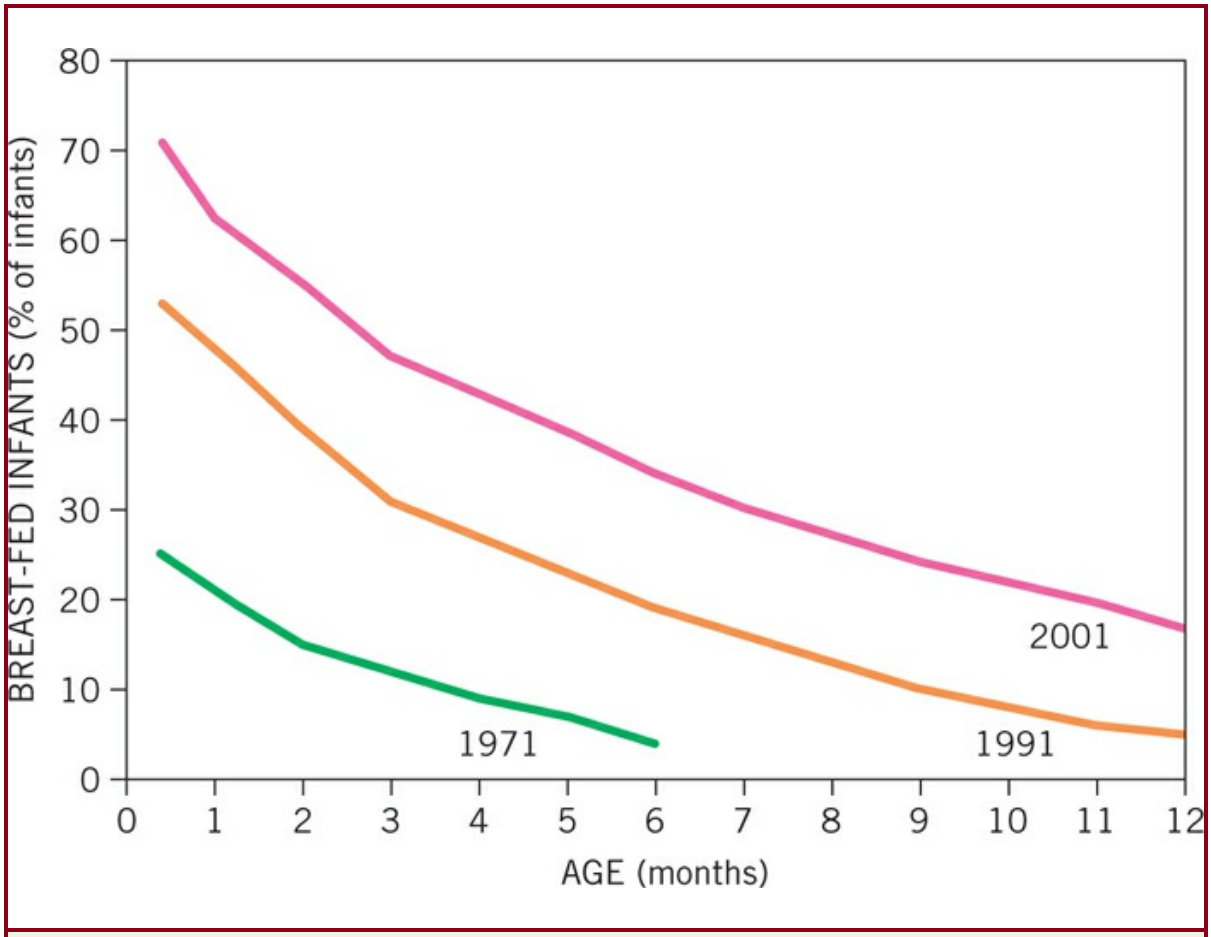


Figure 34-2 Exclusively and partially breastfed infants as a percentage of all infants in 1971, 1991, and 2001.

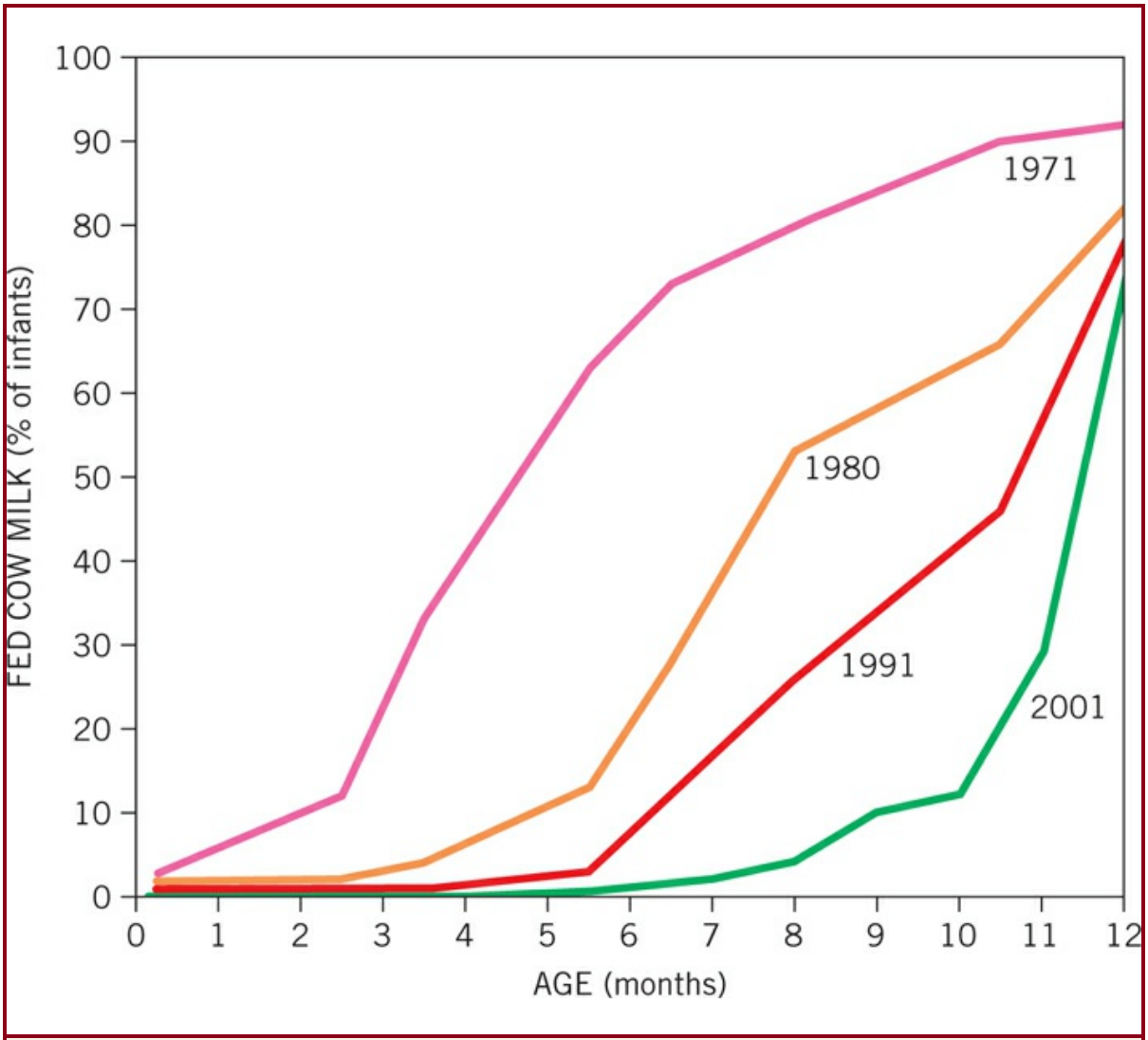


Figure 34-3 Infants fed cow milk (no breastfeeding, no formula feeding) as percent of all infants in 1971, 1980, 1991, and 2001.

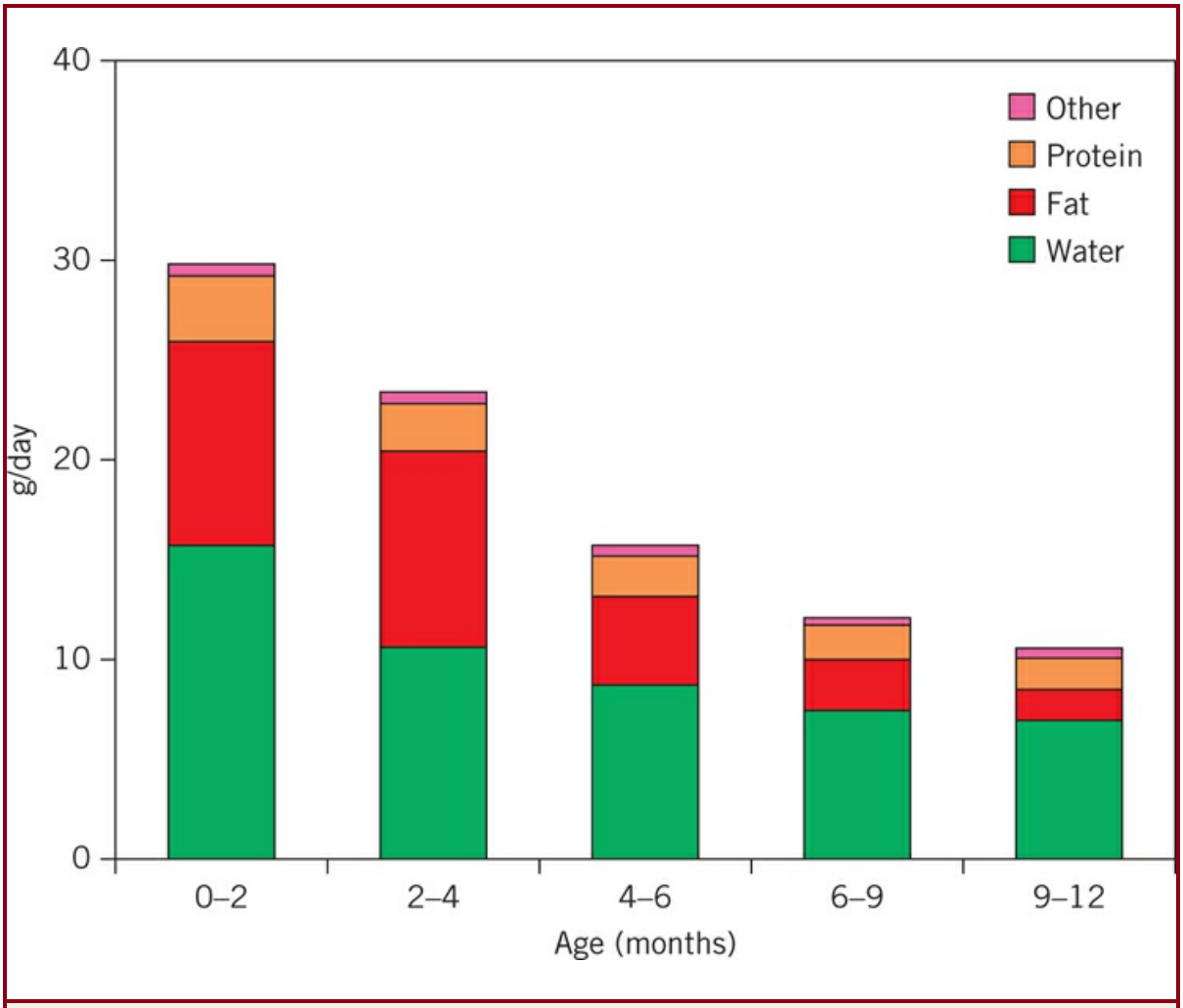


Figure 34-4 Gain in weight and in major body components of a "reference infant" (an infant with weight and length and body composition at the 50th centile for age). Based on Fomon and Nelson.[5]

Nutrient requirements of infants are obtained chiefly by two methods: The observational method and the factorial method. Each approach has its difficulties and limitations. The *observational method* relies on the model of the breastfed infant. The main difficulty with this approach is the determination of representative concentrations of nutrients in breast milk. Concentrations of many nutrients change markedly with the duration of lactation and are in many cases influenced by maternal dietary intakes. Also, the breastfed infant is not a reliable model in the case of certain nutrients, for example, iron, that are not provided in breast milk in amounts that meet the infant's needs. The *factorial method* yields good estimates for certain nutrients, such as protein, for which estimates of rates of accretion by the infant are available. Nutrient requirements, largely

because of the dynamic changes of the needs for growth, are strongly age dependent. Stating nutrient requirements in relation to energy requirements diminishes the age dependency of nutrient requirements some what, but does not eliminate it completely.

Energy Requirement

Because the infant possesses the ability to self-regulate energy intake, it may be assumed that energy intakes observed in infants receiving feedings with normal energy density represent energy requirements. Table 34-1 presents energy intakes observed in breast-fed and formula-fed infants. The data illustrate the markedly higher energy intakes during the early months of life compared to later infancy, a reflection of the deposition of large amounts of fat as energy stores. The data also show that formula-fed infants consume some what more energy than breast-fed infants, especially during the first three months of life. Therein may lie (part of) the reason for the somewhat more rapid growth of formula-fed infants. Gender-related differences in energy intake, although quite small, are nevertheless consistently present.

Protein Requirement

Table 34-2 presents estimates of requirements for protein by the factorial method as well as protein intakes observed in breast-fed infants. The close agreement between the factorial estimate and observed intake is striking. During the first month of life, growth accounts for over 50% of protein requirement. But by the end of the first year of life, the proportion needed for growth is less than 20%. It must be mentioned that other estimates by the factorial method yield some what lower estimates of protein requirements, largely because of lower assumed inevitable losses (0.56 g/kg/d).[7]

THE BREASTFED INFANT

Prevalence

Breastfeeding practices vary considerably from country to country. Not only does the rate of initiation of breastfeeding vary, but there is also large variation in the duration of exclusive breastfeeding (age of introduction of complementary feedings) as well as in the duration of any breastfeeding.[8] Besides local customs, legislation regarding protected time off work is presumed to have a major effect on the prevalence of breastfeeding.

TABLE 34-1 Energy Intakes (kcal/kg/d) of Infants

	Breast-Fed		Formula-Fed			
	Males	Females	Males		Females	
Age Interval (mo)	Mean	Mean	50th	10th–90th	50th	10th–90th
0–1	115	111	118	99–137	116	96–136
1–2	104	101	113	99–131	111	94–130
2–3	95	93	100	88–116	100	87–114
3–4	91	91	95	83–106	95	83–107
4–5	89	89	94	78–112	97	81–113
5–6	86	85	95	64–113	94	78–113
6–9	81	81				
9–12	92	92				

TABLE 34-2 Protein Requirements of Infants

Age Interval (mo)	Growth (g/kg/d)	Losses (g/kg/d)	Requirement (g/kg/d)	Intake of Breast-fed Infant (g/kg/d)	Requirement (g/100 kcal)	Gr Re (%
0–1	1.03	0.95	1.98	2.09	1.7	
1–2	0.78	0.93	1.71	1.59	1.5	
2–3	0.56	0.90	1.46	1.18	1.5	
3–4	0.38	0.89	1.27	1.06	1.4	
4–5	0.30	0.88	1.18	1.00	1.3	
5–6	0.29	0.89	1.18	0.95	1.3	
6–9	0.26	0.91	1.17		1.3	
9–12	0.20	0.94	1.14		1.2	

Source: Adapted from Fomon.[6]

Energy and Protein Intakes

The breastfed infant receives water, energy and specific nutrients in amounts that meet the needs of the infant at all times, notwithstanding the fact that the needs change with age of the infant. A prime example is protein. Through a combination of a sharp decrease in protein concentration in the first few months of lactation and an increase in milk

volume, a close match between intake of protein and protein requirement is achieved (Table 34-2). In the case of energy, whose content in breast milk does not change with the duration of lactation, the match between intake and need (Table 34-1) is achieved solely through a decrease in volume of milk consumed per kilogram of body weight. Most other nutrients fall somewhere between protein and energy, that is, a match between intake and need is brought about through a combination of concentration changes and volume increase. Formulas, on the other hand, must be designed to meet the highest possible needs, that is, those of the youngest infants in the target age group. Consequently, as the infant grows, nutrient intakes from formulas exceed needs by an increasing margin.

Although it is well established that infants generally do not obtain all the milk that is synthesized by the mammary gland, the cause of the failure to consume all the available milk is not known. One possible reason is infant fatigue since the process of obtaining milk from the breast requires considerable effort on the part of the infant. Whatever the mechanism may be, failure to consume all available milk may be one of the reasons why breast-fed infants consistently consume less energy than formula-fed infants, as shown in Table 34-1. It has been surmised that because the milk remaining in the breast is not visible to the mother, contrary to formula remaining in the bottle, she may be more prone to stop feeding at the earliest sign of satiety on the part of the infant than if the infant is fed by bottle.

Growth

Formula-fed infants grow more rapidly than breastfed infants. Differences in energy consumption may explain the difference in growth. However, inconsistent with this explanation is the fact that growth is similar during the first six weeks of life and only begins to differ significantly there after, whereas the difference in energy consumption seems to be largest during the first 2 months of life. Therefore, there may be other explanations. When the average intake is close to the average requirement for a nutrient, there is the likelihood that for some individuals intakes fall short of or exceed requirements. That is the case for protein (Table 34-2) and also for zinc, nutrients whose deficient intake can limit growth. Thus, it is possible that moderately inadequate intakes of protein or zinc, or both, occur among breast-fed infants and explain the difference in growth. But it must be stressed that the reason for the growth differential is not actually known. By the same token, it is not known whether there are any long-

term consequences, beneficial or detrimental, due to the slower growth of breast-fed infants, or due to the lower nutrient intake that is causing it. Nevertheless, it is widely assumed that the long-term health advantages of the breastfed infant are brought about by slower growth, or by the lower nutrient intakes that cause growth to be slower.

Flavor

Breast milk contains a variety of flavoring substances derived from the maternal diet, and there is clear evidence that the infant recognizes flavors and responds to them.[9] Thus, in contrast to the formula-fed infant, the breast-fed infant is exposed to a variety of tastes from the earliest age. The nutritional significance of early exposure to flavor variety, perhaps even in the long-term, is that it leads to greater acceptance and apparent enjoyment of novel flavors later on.[9]

Oligosaccharides

Breast milk contains substantial amounts (5–12 g/L) of oligosaccharides, a complex group of substances derived from lactose.[10] These oligosaccharides share similarities with epithelial cell surface carbohydrates and adhesion molecules. They exert a number of important functions, from blocking the adhesion of bacteria to intestinal cell surfaces to being responsible for the predominance of Bifidobacteria in the colonic microflora of the breastfed infant. Consonant with these important functions is the fact that oligosaccharides are only minimally digested in the upper gastrointestinal tract and undergo fermentation in the colon, thereby accounting for much of the hydrogen gas that is exhaled by breastfed infants.

Iron

Most infants are born with generous iron stores that render them independent of an exogenous supply of iron for the first 5–6 months of life. Breast milk provides a small amount of highly bioavailable iron that allows the infant to absorb about 0.15 mg each day. Beginning when iron stores are exhausted around 5–6 months of age, the infant needs to absorb approximately 0.75 mg each day in order to avoid iron deficiency. Since breast milk provides only about 0.15 mg/d of absorbed iron, the infant must absorb about 0.6 mg/d from other sources. Except for meat, which provides a highly bioavailable form of iron (heme iron), the natural iron content of most complementary foods is low. Therefore, the breast-fed

infant can obtain an adequate intake of iron, besides meat, only from iron-fortified foods, such as infant cereals, or from medicinal iron. These sources of iron have been demonstrated to be effective in preventing iron deficiency during prolonged breastfeeding.[11] However, concerns about adverse effects of medicinal iron limit its use, leaving iron-fortified cereals as the main source of iron for the breast-fed infant.

Some infants are born with diminished iron stores[11,12] and these infants may become dependent on an exogenous iron supply sooner than infants born with more generous iron stores. Because these infants may be prone to developing iron deficiency at a young age and thus be particularly vulnerable to the adverse effects of iron deficiency, it is advisable to start ensuring an adequate iron intake for all infants beginning at 4 months of age.

FORMULAS

Formulas provide macronutrients and micronutrients in amounts and proportions that permit infants to grow normally. Like breast milk, formulas provide energy in roughly equal amounts from carbohydrate and fat. Contrary to breast milk, the protein level is fixed and set at a level that meets the needs of the fastest-growing infants within the target age group. Formulas provide water in amounts that allow the infant to dispose of waste products with a satisfactory margin of safety. Owing to the higher potential renal solute load of formulas, that margin is somewhat narrower than in the breast-fed infant, a difference that is not known to be of clinical consequence.

An enumeration of the myriad substances that are provided by breast milk but are absent from formulas is beyond the scope of this chapter. A number of substances, including macro minerals, trace minerals, and vitamins, are provided in formulas in greater concentration than in breast milk. Some substances that are believed to be biologically important for the infant are added to formulas in amounts similar to breast milk. Some formulas, notably the soy protein-based formulas, provide substances that are not provided by breast milk. Of considerable nutritional importance is the presence of phytate because of its potential to inhibit the absorption of macro minerals and, especially, trace minerals. A group of substances collectively referred to as phytoestrogens is contained in soy formulas. These substances are not nutritionally important but are biologically active, exerting estrogenic as well as anti-estrogenic activities. Phytoestrogens are readily absorbed by the infant but produce no clinically

or otherwise recognizable effects. When followed into young adulthood, women who had been exposed to soy-formulas as infants had somewhat greater menstrual flow and experienced more discomfort with menstruation.[13] Although these effects are not known to be clinically important, they are biologically significant because they demonstrate that relatively brief exposure during a susceptible period can have effects that last for decades.

Nutrients

Carbohydrates

Milk-based formulas currently contain lactose as the sole carbohydrate and lack the complex mix of oligosaccharides that make up a substantial proportion of the carbohydrate of breast milk. Some formulas, however, contain longer-chain oligosaccharides, such as fructo-oligosaccharides, that exert prebiotic effects similar to those of human milk oligosaccharides. Human milk oligosaccharides are responsible for the differences in colonic flora between breast-fed and formula-fed infants. They are essentially nonabsorbable in the upper gastrointestinal tract, but the colonic micro flora is quite effective in fermenting them. The fermentation products, acetate and butyrate, are absorbed from the colon.

Fat

The fat of formulas is made up of various combinations of plant oils, selected to provide major fatty acids in approximately the proportions found in breast milk. The absorbability of the fat of formulas is similar to that of breast milk fat. Some formulas contain added long-chain polyunsaturated fatty acids (LC-PUFAs) (arachidonic acid [ARA] and docosahexaenoic acid [DHA]) in amounts similar to those present in breast milk.

Protein

Although formulas provide bulk casein and whey proteins in approximately the same proportions as breast milk, the individual whey proteins show little resemblance to those provided in breast milk. Manufacturers are able to selectively decrease and/or increase individual whey proteins and thereby alter the biologic quality and amino acid composition of the protein mix. The protein concentration of formulas is set to meet the highest needs, that is, those of the youngest infants in the

target age group. Consequently, older infants in the target group, who have lower protein needs than the younger infants, receive protein intakes that exceed their needs by some margin. In contrast, breastfed infants receive protein intakes that approximately match their needs at all times.

Iron

Formulas fortified with quite large amounts of iron (1.8 mg/100 kcal) were introduced in 1959 and were shown to be effective in preventing iron deficiency. Although non-iron-fortified formulas continued to be used, the use of formulas providing iron at 1.8 mg/100 kcal gradually increased, and in the year 2001 they accounted for about 95% of formula sales. At present, all routine formulas contain added iron at 1.8 mg/100 kcal. Iron fortification is not associated with adverse effects on gastrointestinal function.

Formula Composition

Compositional standards for routine formulas and formulas for special medical purposes are laid down in the Codex alimentarius.[14] Formulas for *routine use* are rather uniform in composition, as is evident from Table C-1 of Appendix C. Some formulas provide partially hydrolyzed whey proteins. The latter type of formula is sometimes referred to as "hypoallergenic".

A number of formulas are available that contain *no lactose*. Their composition is also shown in Table C-1, Appendix C. While those based on soy protein isolate (with methionine supplementation) have been available for many years, those based on cow milk have been introduced more recently. Although the scientific basis for the widespread use of these formulas is uncertain, it can be said that these formulas provide all essential nutrients and support normal growth. The carbohydrates that are provided in lieu of lactose include sucrose and fractions of corn syrup. The protein concentration of soy-based formulas is generally higher than that of milk-based formulas, reflecting the (presumed) somewhat lesser quality (per unit of weight) of methionine-supplemented isolated soy protein. Although all lactose-free soy-based formulas contain iron at 1.8 mg/100 kcal, this includes a substantial amount of iron provided by the soy protein isolate. Little is known about the bioavailability of this iron.

Table C-1 also lists the *extensively hydrolyzed formulas* (also referred to as predigested) and *elemental formulas*. These formulas are characterized by the absence of intact protein as well as lactose. The source of nitrogen is hydrolyzed casein or hydrolyzed whey proteins, and

in the case of elemental formulas, free amino acids. Three of the formulas provide a sizable proportion of their fat in the form of medium-chain triglycerides. Formulas designed for *specific medical indications* include formulas that are of thicker than usual consistency and are intended for infants with severe gastroesophageal reflux. Some special formulas are intended for infants recovering from diarrhea. A formula with low sodium and phosphorus content is used in infants with renal failure.

Substances Added to Formulas

Of the many bioactive substances that are present in human milk but are contained in cow milk in lower concentration or are completely absent from cow milk, some are added to formulas. Evidence that the addition ameliorates an otherwise existing deficiency is not always very convincing. However, it appears reasonable to add a substance when its presence in breast milk seems “intentional” and when its absence from the infant’s diet is associated with deficits in serum or tissue concentrations relative to the breast-fed infant. A prerequisite for the addition of any substance is, of course, the availability of the technology necessary for making the addition feasible. The list of substances added to formulas now includes taurine, carnitine, nucleotides, lutein, and long-chain polyunsaturated fatty acids.

Taurine

Taurine is an amino sulfonic acid found in high concentration in certain parts of the developing brain and in the retina. In infants taurine also plays a role in the conjugation of bile acids and it has osmoregulatory functions. Taurine is synthesized from cysteine and the term infant (but perhaps not the preterm infant) appears to possess ample capacity to carry out the conversion. Breast milk contains appreciable quantities of taurine, whereas the taurine content of cow milk and hence of cow milk-based formulas is only a fraction of that of breast milk. However, except for low plasma taurine concentrations, adverse effects due to low taurine intakes have not been demonstrated convincingly. Nevertheless, all formulas contain added taurine.

Carnitine

Carnitine plays an essential role in the entry of fatty acids into the mitochondria and hence is necessary for fatty acid oxidation. Carnitine is derived from endogenous synthesis as well as the diet. Breast milk, cow milk and hence milk-based formulas, and meat provide ample amounts of

carnitine. When carnitine is absent from the diet, plasma concentrations of infants are low and certain biochemical abnormalities are observed, whose clinical significance is uncertain. Carnitine is added to all formulas.

Nucleotides

Breast milk contains free RNA nucleosides (adenine, cytosine, guanine, and uridine) as well as their respective nucleotides (i.e., nucleosides with one sugar moiety and one or more phosphate groups attached), and some polymeric RNA. The amount of nucleosides in breast milk is considerably higher than in cow milk. It is thought that during times of high demand, that is, during rapid growth, dietary nucleosides are useful and may be conditionally essential. Animal studies have shown nucleosides to enhance T-cell function and gastrointestinal maturation. Fortification of infant formula with the four main nucleotides found in human milk led to enhanced antibody responses to *H. influenzae* type b and diphtheria.

Lutein

Lutein is a carotenoid and like other carotenoids has anti-oxidant and anti-inflammatory properties. It is highly concentrated in the macula area of the retina. It is contained in breast milk and is added to some formulas.

LC-PUFAs

These fatty acids have received considerable attention in recent years. Although the consequences, especially those in the long term, of a lack of these fatty acids in the infant's diet remain controversial, formula manufacturers are now offering many formulas with and without added LC-PUFAs. Formulas with added LC-PUFAs are specially designated. The fatty acids in question are DHA and ARA. DHA is a C22 n-3 fatty acid with six double bonds. It is derived from dietary α-linolenic acid by a series of chain elongation and desaturation steps. ARA is a C20 n-6 fatty acid with four double bonds. It is derived from dietary linoleic acid. Breast milk contains preformed DHA (about 0.2% of fatty acids) and ARA (about 0.5% of fatty acids). The plant oils used in manufacturing formulas do not contain DHA and ARA and formulas are devoid of DHA and ARA unless these fatty acids are specifically added from other sources.

Infants, including premature infants, have the ability to synthesize DHA and ARA from their respective precursors, but the synthetic capacity appears to be insufficient to keep up with the high demands for growth. DHA is needed for incorporation into cortical gray matter and the retina. In infants who are fed formulas devoid of DHA, concentrations in gray

matter and retina are substantially lower than in breast-fed infants.[15] The same is true for concentrations in plasma and erythrocyte membrane lipids. Infants fed DHA-free formulas show evidence of impaired retinal function during the early months of life. However, this effect seems to be transient and is no longer demonstrable in older infants. The presence of DHA and ARA in formulas is considered desirable because these fatty acids are present in breast milk and because their absence from the infant's diet produces abnormal concentrations in brain, retina, erythrocytes, and plasma.

WEANING AND COMPLEMENTARY FOODS

Weaning is the withdrawal or displacement of breastfeeding. The weaning period begins with the first introduction of a food other than breast milk (water and vitamin-mineral supplements are not considered to be food) and extends until the complete discontinuation of breastfeeding. We do not apply the term weaning to the withdrawal and displacement of formula. Solid and semi-solid foods that are fed in addition to breastfeeding are referred to as complementary foods or weaning foods.

Age of Introduction of Complementary Foods

Evidence that infants can tolerate complementary foods before the currently recommended age of its introduction was provided by the experience in the 1950s and 1960s, when various complementary foods were fed to many infants even during the early weeks of life without any apparent ill effects.[1] One reason for deferring the introduction of complementary foods to 4–6 months of age is that the infant acquires around 4 months the ability to sit with support and have adequate neuromuscular control of trunk and neck. But another reason is that complementary foods are thought to interfere with the immune-protective effects of breastfeeding. This is the main reason why the WHO in 2003[16] recommended that exclusive breastfeeding should continue to six months and that complementary foods should therefore be introduced after six months. Some professional organizations, such as the American Academy of Pediatrics,[17] have endorsed the WHO recommendation, citing evidence that exclusive breastfeeding even in developed countries provides better protection against diarrhea and lower respiratory disease[18,19] than mixed feeding (breastfeeding plus complementary foods). There is also the possibility of long-term benefits such as protection against obesity later in

life.[20] Other organizations[21,22] and individuals[23] have argued that the evidence favoring exclusive breastfeeding for six months is weak and that the absence of complementary foods places the breastfed infant at risk of micronutrient deficiency, for which there is some evidence in the case of iron.[24,25] From the point of view of allergy prevention, it seems that introduction of complementary foods between four and six months is preferable over earlier or later introduction.[21] Weighing infectious morbidity risk against the risk of iron deficiency, the authors favor introduction of complementary foods between four and six months.

Choices of Complementary Foods

There is no evidence that the sequence of introduction of various complementary food items is a matter of great importance. It is convenient to begin with cereal because a small amount of the dry powder can be mixed with breast milk or formula at quite low expense. Initially, only a small amount, less than a teaspoonful, should be given at one time. Whether pureed fruits or vegetables are then introduced seems of little consequence. However, single foods rather than combinations should be offered. It is generally recommended that one food be fed for several days before introducing another. It is desirable to work toward acceptance of variations in tastes and textures.

The exclusively breastfed infant consumes an adequate intake of protein but, once complementary food begins to comprise a substantial portion of the total energy intake, it is important to include some high protein foods. Deficiency of iron and zinc in breastfed infants is possible. Meats, especially red meats, are a good source of protein, zinc, and a highly bioavailable form of iron. We recommend for the breastfed infant that soft-cooked meats be introduced within the first month or two of complementary feeding (by 5 or 6 months of age). We consider the risk of iron and zinc deficiency to be greater than the risk of sensitization to meat proteins.

With the exception of providing sources of iron and zinc of high bioavailability for the breastfed infant, the goal of feeding complementary foods is to begin the transition from the infant's primarily liquid diet to the predominantly solid food diet of the child. Therefore, as infancy progresses, the variety of flavors and textures offered to the infant should gradually be increased.

Fruit juices should be fed sparingly and only by cup. Fruit juices are associated with adverse reactions and do not provide nutrients that are

lacking in the diet of the breastfed or formula-fed infant.

COW MILK

Feeding of cow milk during the first year of life is undesirable because such feeding is associated with the development of iron deficiency. It is unclear which of the several mechanisms is mainly accountable for the iron deficiency. It has long been recognized that ingestion of large quantities of cow milk may result in low intakes of other foods that contain more generous amounts of iron. In addition, there is considerable evidence that ingestion of cow milk causes occult gastrointestinal blood loss.[26] Finally, cow milk provides high intakes of casein and calcium, potent inhibitors of iron absorption that may contribute to the development of iron deficiency.

The potential renal solute load provided by cow milk is undesirably high.[27] Feeding of whole cow milk does not interfere with maintenance of water balance in normal infants under most circumstances. But in a hot environment or during febrile illness, especially if associated with decreased fluid intake, the infant is at greater risk of dehydration if fed cow milk than if breastfed or fed formula. The margin of safety is greater with breastfeeding or feeding of infant formulas than with feeding of cow milk.[27]

REFERENCES

1. Fomon SJ. Infant feeding in the 20th century: Formula and beikost. *J Nutr*. 2001;131:409S–420S.
2. Ryan AS. The resurgence of breastfeeding in the United States. *Pediatrics*. 1997;99:E4–E12.
3. Grummer-Strawn LM, Scanlon KS, Fein SB. Infant feeding and feeding transitions during the first year of life. *Pediatrics*. 2008;122:S36–S42.
4. Clayton HB, Li R, Perrine CG, Scanlon KS. Prevalence and reasons for introducing infants early to solid foods: variations by milk feeding type. *Pediatrics*. 2013;131:e1108–e1124.
5. Fomon SJ, Nelson SE. Body composition of the male and female reference infants. *Annu Rev Nutr*. 2002;22:1–17.
6. Fomon SJ. Requirements and recommended dietary intakes of protein during infancy. *Pediatr Res*. 1991;30:391–395.
7. Dewey KG, Beaton G, Fjeld C, et al. Protein requirements of infants and children. *Eur J Clin Nutr*. 1996;50(suppl 1):S119–S150.
8. World Health Organization. *The WHO Global Data Bank on infant and young child feeding*. http://www.who.int/nutrition/databases/infantfeeding/en/index.html
9. Menella JA, Jagnow CP, Beauchamp GK. Prenatal and postnatal flavor learning by human infants. *Pediatrics*. 2001;107:e88–e93.

10. Bode L. Human milk oligosaccharides: prebiotics and beyond. *Nutr Rev*. 2009;67:S183–S191.
11. Ziegler EE, Nelson SE, Jeter JM. Iron status of breastfed infants is improved equally by medicinal iron and iron-fortified cereal. *Am J Clin Nutr*. 2009;90:76–87.
12. Tamura T, Goldenberg RL, Hou J, et al. Cord serum ferritin concentration and mental psychomotor development of children at five years of age. *J Pediatr*. 2002;140:165–170.
13. Strom BL, Schinnar R, Ziegler EE, et al. Exposure to soy-based formula in infancy and endocrinological and reproductive outcomes in young adulthood. *JAMA*. 2001;286:807–814.
14. Codex alimentarius. http://www.codexalimentarius.org
15. Makrides M, Neumann MA, Byard RW, et al. Fatty acid composition of brain, retina, and erythrocytes in breast- and formula-fed infants. *Am J Clin Nutr*. 1994;60:189–194.
16. World Health Organization. *Global Strategy for Infant and Young Child Feeding*. Geneva, Switzerland: WHO; 2003.
17. Eidelman AI, Schanler RJ, American Academy of Pediatrics Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129:e827–e841.
18. Chantry CJ, Howard CR, Auinger P. Full breastfeeding duration and associated decrease in respiratory tract infection in US Children. *Pediatrics*. 2006;117:425–432.
19. Quigley MA, Kelly YJ, Sacker A. Breastfeeding and hospitalization for diarrheal disease and respiratory infection in the United Kingdom Millennium Cohort Study. *Pediatrics*. 2007;119:e837–e842.
20. Schack-Nielsen L, Sorensen TIA, Mortensen EL, Michaelsen KF. Late introduction of complementary feeding, rather than duration of breastfeeding, may protect against adult overweight. *Am J Clin Nutr*. 2010;91:619–627.
21. Agostoni C, Decsi T, Fewtrell M, et al, for ESPGHAN Committee on Nutrition. Complementary feeding: a commentary by the ESPGHAN Committee on Nutrition. *J Ped Gast Nut*. 2008;46:99–110.
22. EFSA Panel on Dietetic Products, Nutrition and allergies (NDA). Scientific opinion on the appropriate age for introduction of complementary feeding of infants. *EFSA J*. 2009;7(12):1423.
23. Fewtrell M, Wilson DC, Booth I, Lucas A. Six months of exclusive breast feeding: how good is the evidence? *Br Med J*. 2011;342:c5955.
24. Dewey KG, Cohen RJ, Rivera LL, Brown KH. Effect of age of introduction of complementary foods on iron status of breast-fed infants in Honduras. *Am J Clin Nutr*. 1998;67:878–884.
25. Dube K, Schwartz J, Mueller MJ, et al. Complementary food with low (8%) or high (12%) meat content as source of dietary iron: a double- blinded randomized controlled trial. *Eur J Nutr*. 2010;49:11–18.
26. Ziegler EE, Fomon SJ, Nelson SE, et al. Cow milk feeding in infancy: further observations on blood loss from the gastrointestinal tract. *J Pediatr*. 1990;116:11–18.
27. Fomon SJ, Ziegler EE. Renal solute load and potential renal solute load in infancy. *J Pediatr*. 1999;134:11–14.

35

Complementary Feeding and Health Outcomes

Mary S. Fewtrell, MA, MD, FRCPCH

INTRODUCTION

Complementary foods are defined by the World Health organization (WHO) as any food or liquid other than breast milk.[1] They are required during the second part of the first year of life for both nutritional and developmental reasons, and to enable the transition from milk feeding to family foods. The ability of breast milk to meet macro and micronutrient requirements becomes limited at this point, whereas from a developmental perspective, infants develop the ability to chew and start to show an interest in foods other than milk.

Current recommendations on the age at which complementary foods should be introduced are based on consideration of the optimal duration of exclusive breast-feeding (EBF) in terms of nutritional adequacy and effects on health outcomes. Following a systematic review[2] and expert consultation in 2001, WHO recommended that all infants should be exclusively breast-fed for six months followed by the introduction of

complementary foods alongside continued breast-feeding.[3] However, because human milk substitutes (HMS; infant formulas and follow-on formulas) are defined by WHO as complementary foods, this definition can be confusing: in high-income countries where many infants receive HMS from the first weeks of life it is not clear whether recommendations based on the duration of EBF should apply to these infants with respect to the introduction of other nonmilk foods. In recent years, debate on the optimal age for introducing complementary foods in different situations has largely overshadowed discussion on the optimal content of the diet during this period, and the age at which specific foods should be introduced.

This chapter will discuss evidence for health effects of complementary feeding practices, considering timing, content, and feeding behavior, and will focus on infants in higher income countries. Nutritional considerations and issues for infants living in lower income settings are considered in Part V, Chapter 43.

TIMING OF COMPLEMENTARY FEEDING

Complementary feeding practices are strongly influenced by social and cultural factors, and vary markedly between countries. There is general agreement that complementary foods should not be given before 17 weeks of age, as early introduction may be associated with an increase in fatness, wheeze, and eczema later in childhood.[4] Recommendations for the age at which complementary foods should be introduced are generally based on infant developmental readiness, nutritional requirements, and the health effects of different practices. In terms of developmental readiness, gastrointestinal and renal functions are likely to be sufficiently mature by around 4 months of age to enable infants to process some complementary foods. With regard to neurodevelopmental readiness, it is probable that there is a range at which infants attain the necessary motor skills to cope safely with complementary foods, but these skills typically appear during the 4- to 6-month period. It is however important to recognize that the skills required will depend on the type of complementary foods that are to be introduced, and that the neurodevelopmental readiness required for an infant to safely accept and swallow pureed complementary foods from a spoon differs from that required to handle semisolids or to self-feed, as currently advocated in the “baby-led weaning” approach currently popular in some higher income countries (see below).

Scientific Evidence to Support Recommendations on the Duration of EBF and the Introduction of Solid Foods for Infants in Developed/High-Income Countries

It is important not to confuse evidence for the optimal duration of EBF with that for breast-feeding *per se*.[5] Current advice on the length of EBF is based on a global recommendation introduced by the WHO in 2001 following a systematic review and expert consultation on the health effects of EBF for six months compared to EBF for 3–4 months with continued partial breast-feeding alongside the introduction of complementary foods. This review did not address the introduction of solid foods in infants who are receiving infant formula (a significant proportion in many high-income countries for 5–6 months), since the latter is defined by the WHO as a complementary food. The review[2] initially included 16 eligible studies, seven of which were from developing countries. With the exception of two randomized trials in Honduras, all of the studies were observational and therefore subject to the many well-described methodological problems associated with this type of study,[6,7] notably confounding factors associated with the decision to introduce complementary food. An update of the systematic review was published in 2012, including a literature review to June 2011.[8] A total of 23 eligible studies were identified—11 (2 randomized) from developing countries and 12 (all observational) from developed countries. No new randomized trials had been conducted since the original review, and the conclusions were largely unaltered, with no new findings of relevance specifically to infants in developed countries. The review's main conclusions were that EBF for six months, compared with EBF for 3–4 months with partial breast-feeding thereafter, is associated with:

- a significantly reduced risk of gastroenteritis (studies from Belarus, Iran and Nigeria; adjusted odds ratio 0.61; 95% confidence interval 0.41–0.93 for infants in Belarus)
- no apparent growth deficits
- no apparent relation with the development of allergy
- poorer iron status for infants in developing country settings where newborn iron stores may be suboptimal: in the Honduras randomized trial, infants exclusively breast-fed for six (vs. four) months had lower mean hemoglobin (difference–5.00 g/L; 95% confidence interval–8.46 to –1.54), and ferritin concentrations
- delayed return of menses (studies from Honduras, Bangladesh, and Senegal) and more rapid postpartum weight loss in mothers

(Honduras)
- no effect on height, weight, body mass index (BMI), dental caries, cognitive ability, or behavior at 6.5 years (Belarus)

Thus, for infants in higher income countries, the most relevant health benefit demonstrated was a reduced risk of gastrointestinal infection in infants EBF for six months. This finding came from an observational analysis of data from the PROBIT study, a large cluster-randomized trial of a breast-feeding promotion intervention.[9] The protective benefit of EBF against gastrointestinal infection in this study was confined to the 3–6-month age period during which feeding practices differed between the two groups.

In a second systematic review, also published in 2001 but examining the health effects of the timing of introduction of solids in both breast-fed and formula-fed infants, Lanigan et al[10] identified 33 eligible studies, and concluded there was no compelling evidence to support changing from the existing recommendation to introduce solids at 4–6 months; in their opinion, 13 studies supported a change, 13 did not, and the remaining 7 did not provide evidence for either position.

In 2009, a review was commissioned by the European Union (EU) and conducted by an Expert panel for the Nutrition, Dietetics, and Allergy Committee of the European Food Safety Authority (EFSA), to address the issue of when complementary foods can be safely introduced into the diets of infant in the EU.[11] Their main conclusions were:

- As long as complementary feeding is introduced after four months of life it does not constitute a problem for the digestive system or the renal function of the infant.
- The age of introduction of complementary feeding seems not to have a strong impact on growth velocity.
- It is justified to assume that EBF by well-nourished mothers for six months can meet a healthy infant's need for energy, protein, and for most vitamins and minerals with the exception of iron and zinc, particularly in situations where the maternal diet is poor.
- Breast-feeding protects against infectious morbidity. This effect is proportional to the length of breast-feeding. The available evidence suggests that early introduction (<3 months) of complementary feeding may increase the risk of infectious morbidity. There seems to be no effect of introduction of complementary feeding after four months on the risk of infectious morbidity.

- There is no convincing scientific evidence that avoidance or delayed introduction of potentially allergenic foods reduces allergies in infants considered at increased risk for the development of allergy or in those not considered to be at increased risk. The available data do not permit a conclusion on the appropriate age for introduction of complementary feeding with respect to allergy prevention or reducing the risk of allergy.
- The available data suggest that gluten containing complementary feeding could be introduced after four months in small amounts preferably while the infant is still breast-fed.
- The age of introduction of complementary feeding seems not to have a clear impact on the risk of obesity.

Overall, the Panel concluded that the introduction of complementary food into the diet of healthy term infants in the EU between the age of 4 and 6 months is safe and does not pose a risk for adverse health effects. Following the publication of this opinion, the U.K. Department of Health requested an expert review and comment on the data relevant to the age at introduction of gluten. The conclusion,[12] based on a consideration of the same data examined by the EFSA panel, was that the evidence was not sufficient to support EFSA's conclusion on the introduction of gluten into the infant diet not later than 6 completed months of age with the aim of reducing the risk of subsequent development of coeliac disease and type 1 diabetes. This example of expert groups arriving at different conclusions based on the same available scientific evidence illustrates well the problem of developing recommendations when the data are subject to so many limitations.

A synthesis of available data on the health effects of six months EBF with a particular focus on infants in higher income countries is presented below, including studies that were not eligible for the Kramer systematic review. The discussion also includes some data published since the last update of the systematic review, notably results from the only randomized controlled trial to compare four versus six months EBF in infants living in a high-income country.

Infection

Five observational studies in developed countries have now provided evidence on the duration of EBF and risk of infection. Questionnaire based data from the National Health and Nutrition Examination Survey III cohort[13] showed that U.S. infants who were EBF for more than six months

had lower risk of pneumonia and recurrent otitis media than those breast-fed for 4–6 months. A Spanish study found the risk of hospital admission for all infant infections was decreased with longer EBF; although, this advantage was seen principally before three months, with little apparent benefit thereafter.[14] A German study[15] found infants exclusively breast-fed for six months, compared with less than four months, had less gastroenteritis. Duijts et al[16] reported that, compared with never-breast-fed infants, those who were breast-fed exclusively until the age of 4 months and partially thereafter had lower risks of upper and lower respiratory tract and gastrointestinal infections until the age of 6 months and of lower respiratory tract infections between the ages of 7 and 12 months. Similar tendencies were observed for infants who were exclusively breast-fed for six months or longer, whereas partial breast-feeding, even for six months, did not result in significantly lower risks of these infections. Finally, the large U.K. Millennium cohort study suggested that 53% of admissions for gastroenteritis and 27% of those for chest infection could be prevented each month by EBF, with a smaller protective effect from partial breast-feeding.[17] Importantly for practice, in subsequent analyses, it was shown that it was the introduction of infant formula, not solid foods, that predicted an increased likelihood of hospital admission.[18]

Nutritional Adequacy

An important issue is whether EBF for six months feeding reliably supports adequate growth—traditionally used as a measure of overall nutritional status. The Kramer systematic review[2,8] suggested no apparent growth deficits associated with six months EBF for infants in either developing or developed country settings, although with the caveat that data were insufficient to exclude small effects. However, because infants who are EBF for six months represent a minority in all settings, it is important to consider whether generalizing findings from this small and potentially biased subgroup to the whole infant population is appropriate. Reilly and Wells[19] conducted a systematic review of infant energy requirements, breast milk output, and energy content, and calculated that many mothers who exclusively breast-fed would not support their infant's energy requirements to six months. Subsequently, Neilsen et al[20] conducted an observational study measuring breast milk production and energy content noninvasively using stable isotopes in 47 British infants who were EBF for six months. Milk intakes were higher than published values and increased significantly between time points (923 (SD 122) g/d

at 15 weeks, 999 (SD 146) g/d at 25 weeks of age, mean increase 61 g/d (95% confidence interval: 23–99). Infant growth was normal using WHO Child Growth Standards, and energy intakes were adequate compared with reference requirements. Behavioral data also indicated no evidence of strain on breast-feeding practices. These results show that in mother–infant pairs who successfully EBF for six months, physiological adaptations occur in milk production to support normal infant growth. However, it is still not certain whether these findings can be generalized to the whole population since the mothers in this study were a small and highly selected —notably, well educated—subgroup.

Most recently, in the first randomized trial of the duration of EBF to be conducted in a high-income country, Wells et al[21] reported significantly higher breast milk intake at age 6 months in a cohort of 100 Icelandic infants randomized to six months versus four months EBF; breast milk intake was 83 g/d (95% CI: 19, 148 g/d) greater (mean ± SD: 901 ± 158 g/d vs. 818 ± 166 g/d), equivalent to 56 kcal/d. Infants who received complementary foods obtained 63 ± 52 kcal/d from these foods, suggesting complementary foods displaced breast milk in the early introduction group. Estimated total energy intakes were similar and anthropometric outcomes and body composition did not differ significantly between groups.

Another issue of uncertainty is whether six months EBF reliably meets infant iron requirements for infants in higher income settings. This is of concern given the association of iron deficiency anemia with adverse mental, motor, and psychosocial outcomes.[22,23] In the Icelandic randomized trial comparing four versus six months EBF, infants in the six month group had significantly lower serum ferritin at age 6 months[24] although the proportion of infants with iron deficiency, anemia, or iron deficiency anemia did not differ in this small study. Although this finding suggests a positive effect of earlier introduction of solids on iron stores, even in a population considered at fairly low risk of iron deficiency, the biological significance of the findings is uncertain. Two observational studies from the United States[25] and Germany[26] have also reported poorer iron status in infants EBF for six months versus those EBF for shorter periods or formula-fed. In countries where there is no routine screening for iron deficiency it would currently be impossible to detect such adverse effects.

It is important to emphasize that there are a number of potential methods for avoiding poor infant iron status other than introducing iron-rich solid foods, including delayed clamping of the umbilical cord—which

has proven efficacy and safety even in Swedish infants considered at low risk for iron deficiency[27]—and iron supplementation of infants. However, these methods are unlikely to be systematically implemented without recognition of the issue and a specific action plan.

Allergy and Coeliac Disease

The Kramer systematic review did not find a link between EBF duration and allergic disease. However, this is an area of ongoing research and discussion. Paradoxically, many developed countries have rising rates of food allergy, despite increasing advice to restrict and delay exposure to potentially allergenic foods, including cows' milk, egg, fish, gluten, peanut, and seeds. Moreover, countries where peanuts are commonly used as weaning foods, such as Israel[28] have low incidences of peanut allergy. These observations have prompted further research on immune tolerance to foods. It has been proposed that the development of immune tolerance to an antigen may require repeated exposure, perhaps during a critical early window, and perhaps modulated by other dietary factors including breast-feeding. A 2008 review[29] reported an increased risk of allergy if solids were introduced before 3–4 months. After four months, the evidence was weak, but suggested an increased risk with delayed introduction of certain allergens. Recently, Nwaru et al[30] reported data from a large Finnish cohort (n = 3781) investigating associations between the duration of breast-feeding and timing of introduction of complementary foods and the development of asthma and allergies by the age of 5 years. They concluded that a longer total duration of breast-feeding rather than its exclusivity was protective against nonatopic asthma, whereas earlier introduction of certain allergenic foods (wheat, rye, oats, and barley cereals at 5.5 months or less; fish at 9 months or less; and egg at 11 months or less) was associated with a decreased risk of asthma, allergic rhinitis, and atopic sensitization in childhood; these findings were unchanged by adjustment for confounders and for potential reverse causality.

Two randomized trials in the United Kingdom are currently investigating the hypothesis that the earlier introduction of allergenic foods may reduce the subsequent risk of allergy: the Learning Early About Peanut Allergy study which addresses the timing of exposure to peanuts in high risk infants, and the Enquiring About Tolerance study which is investigating early (from three to four months) versus delayed (beyond six months) introduction of six potentially allergenic foods in a general

population of infants, not selected for risk of atopy. Data from these studies will add significantly to the evidence base.

The specific issue of the relationship between the introduction of gluten-containing cereals and subsequent development of celiac disease has been much studied, particularly in Sweden where the incidence of early onset coeliac disease increased following advice to delay introduction of gluten until age 6 months, and fell to previous levels after the recommendation reverted to four months. Subsequent analyses suggest that gluten should ideally be introduced in small quantities alongside continued breast-feeding and these data are generally more robust than data on the *timing* of introduction.[31] Based on this, and other observational studies, many expert groups have suggested it is prudent to advise mothers to introduce gluten at a time when they are still predominantly breast-feeding (supported by the ESPGHAN CoN[32] and the AAP[33]). However, the recent publication of two randomized trials reporting that neither the timing of introduction of gluten or breastfeeding influenced the proportion of infants who went on to develop celiac disease[34,35] may lead to reappraisal of this advice; and also illustrates the limitations of observational data as a basis for practice.

Longer Term Health Outcomes

There are limited data on later health outcomes in relation to the duration of EBF or the age at introduction of solid foods. In Scottish infants, the introduction of solids before 3–4 months was associated with increased fatness, wheeze, and eczema at age 7 years.[1] A 6.5-year follow-up of the Belarus PROBIT cohort[36] showed no effect of EBF for six months versus three months on blood pressure, cognition, atopy, and dental caries. However, in a Danish birth cohort[37] earlier introduction of solids was associated with late emergence of a higher risk of overweight at 42 years. A recent systematic review examining the association between the timing of complementary feeding and risk of obesity in infancy and childhood[38] identified 24 studies that met the inclusion criteria, providing data from over 34,000 participants for interpretative analysis. No clear association between the age of introduction of solid foods and obesity was found.

Balance of Risks and Benefits

Data on the health effects of the timing of introduction of complementary feeding for infants in high-income countries suggest there may be a balance of risks and benefits. Clearly for infants living in situations where

there is limited access to clean water or safe, nutritious complementary foods, with significant risk of morbidity and mortality from infectious diseases, the protective effect of more prolonged EBF would override any other considerations. Available data suggest that more prolonged EBF also protects against gastrointestinal and respiratory infection, and hospitalization for such infections, in infants living in higher income settings. However, more prolonged EBF may not be nutritionally optimal for all infants, particularly for iron, and delayed introduction of certain allergenic foods, especially at the point where breast-feeding has decreased or ceased, may be associated with increased risk of allergy. Hence, the risks and benefits should be considered in each setting. Perhaps the most urgent requirement in this field is for new data, preferably, as suggested by Kramer and Kakuma,[2] from randomized trials, although these are difficult to conduct in practice especially given strong opinions on the topic which make it difficult to identify suitable subjects and investigators.

CONTENT OF THE COMPLEMENTARY FEEDING DIET

Most current guidelines for the gradual introduction of different foods during the complementary feeding period are based on cultural factors and food availability rather than scientific evidence. Although in developing countries the focus is still predominantly on providing adequate nutrients to support growth and development (see Part V, Chapter 43), in more affluent environments achieving a better balance of nutrients and avoiding excess may be more important. Nutritional recommendations for the complementary feeding period are based on the concept that breast milk will not meet full requirements for energy, protein, and micronutrients beyond about 6 months of age and assume that complementary foods are combined with breast milk; but this is not the case for a significant proportion of infants in many high-income countries. Deriving appropriate population-based recommendations for intakes of specific nutrients or foods is problematic in the situation where the nutrient intake from milk varies significantly between infants depending on the type of milk they are consuming. For example, the iron intake from breast milk, infant formula, and cow milk varies markedly with the consequence that it is almost impossible to define an “average” requirement that is required from complementary foods. For this reason, it is important to consider infant diets on an individual basis, taking into account the main source of milk. Theoretically, the requirement for good quality complementary foods—

particularly those rich in iron and zinc—may be more important for breast-fed infants, since an infant fed entirely on HMS will have higher intakes of these nutrients from milk at this point. In practice, however, most authorities consider it undesirable and impractical to have different recommendations for breast-fed and formula-fed infants.[32]

Specific Nutrients or Foods

Energy

Energy requirements remain high throughout the whole of the first year of life. The fat content of the diet is an important determinant of its energy density; it should not be less than 25% of energy intake and a higher proportion may be required if the infant has a poor appetite, recurrent infections, or is fed infrequently. It is generally advised that reduced fat cow's milk and other low fat products should not be introduced too early as this will reduce the energy density of the diet. However, consideration should be given to the rest of the infant's diet, and to his or her growth. In countries with high rates of child obesity, it may be advantageous to accustom children to low-fat products from a fairly early age. The Finnish STRIP study, a large ($n > 1000$) trial in which the intervention group were given dietary advice to reduce the intake of saturated fats from infancy throughout childhood has reported beneficial effects on plasma lipid profile and endothelial function, particularly in boys, with no adverse effects on growth to age 14 years or on cognitive development at age 5 years. The intervention resulted primarily in changes in dietary fat quality rather than quantity and also resulted in the consumption of generally healthier diets later in childhood.[35]

Iron and Zinc

More than 90% of iron requirements during the complementary feeding period in a breast-fed infant must be provided by complementary foods. Potential strategies for achieving this include the use of fortified weaning foods, iron-fortified infant formulas and follow-on formulas, foods naturally rich in bioavailable iron such as meat, or the use of supplements. The most suitable strategy or combination of strategies will vary in different circumstances and, as discussed above, the iron needed from complementary foods will depend on the infant's main source of milk since infant formulas and follow-on formulas are supplemented.

Cow milk is a very poor source of iron, and it is generally recommended that it should not be used as the main drink before 12

months of age.[32] In a prospective observational cohort of 928 British term infants, those consuming cow milk as the main drink had an increased incidence of anemia at 12 months and low serum ferritin at 8 and 12 months. More than 25% of infants having breast milk and cow milk as their main drink and 41% of infants having more than six breast feeds per day had iron intakes below the lower reference nutrient intake, whereas infants consuming more than 600 mL/d of cow milk or formula or taking more than six breast feeds per day had lower intakes of solids.[39] In a study of 138 high birth weight Icelandic infants, iron status indices at 12 months were negatively associated with cow milk consumption above 500 mL/d at 9–12 months, but were positively associated with consumption of iron-fortified breakfast cereals, fish, and meat.[40]

Meat has been highlighted as a good source of bioavailable iron and zinc, as well as protein. In a randomized trial in the United States, meat was shown to be acceptable to mothers and infants as an early CF and associated with a trend toward better behavioral outcomes at 12 month using the Bayley Scales of Infant Development,[41] whereas Danish infants randomized to receive a higher meat intake had a smaller decrease in hemoglobin in late infancy.[42] An observational cohort study in the United Kingdom also reported that meat intake from 4 to 12 months was positively and significantly associated with weight gain up to 12 months and with psychomotor development at 22 months.[43]

Protein

Protein intake in relation to energy intake is low while an infant is exclusively breast-fed (protein:energy ratio (PE) 5%) but increases markedly in infants in high-income countries during the complementary feeding period, particularly if complementary foods are consumed along with cow milk (20% PE) or infant formulas (7%–8% PE).[44] Protein intakes during this period are often two to three times higher than the theoretical requirements. In one study, figures for PE% in 9–12-month-old infants from four European countries ranged from 15.0 to 15.7, whereas infants in Italy had a very high mean intake of 19.5 PE%, a large proportion of which (13.0 PE%) came from animal foods.[45]

It has been suggested that there should be upper limits for protein intake during the first years of life, mainly based on concerns that a high protein intake may increase the risk of later obesity. However, although there is increasing evidence linking more rapid early growth to later cardiovascular risk, and randomized trials have shown adverse effects of

higher protein infant formulas on later fatness[46,47] and blood pressure,[48] these trials focused on protein intake from milk, and no randomized study has specifically addressed the issue of the protein content of complementary foods. It is also probable that different protein sources have different effects; and that dairy proteins may specifically influence IGF-1 concentrations and hence growth patterns.[49,50]

Salt and Sugar

Two randomized trials have shown that higher intakes of salt in early infancy are associated with higher blood pressure at the time,[51,52] and follow-up of one study suggested the effect persisted during adolescence.[52] Furthermore, infants may become accustomed to a salty taste which could affect their subsequent food preferences and intake. Requirements for sodium can be met by breast-feeding and subsequently from complementary foods without the need for added salt. Hence, it is generally agreed that salt should not be added to food during the complementary feeding period. Similarly, sugar is associated with the development of dental caries and with development of a preference for sweet foods. Its use should be restricted, and it is important to ensure good dental hygiene practices as early as possible.

Vegetarian Diets

If infants receive a vegetarian diet it is important that the diet includes a sufficient amount of milk (about 500 mL/d) and dairy products. It is recommended that vegan diets should be discouraged in infancy because of the risk of vitamin B_{12} deficiency that can affect neurodevelopment.[32]

TASTE AND FOOD ACCEPTANCE

An important but poorly researched area is the potential effect of the complementary feeding diet on food acceptance and subsequent food preferences. There may be “windows” in infancy during which certain tastes are more readily accepted. Infants are predisposed to like high-energy foods, to prefer sweet and salty tastes and to reject new foods. However, these predispositions may be modified by early dietary experience, and parents play an important role in establishing good dietary habits. Mennella and colleagues[53,54] have demonstrated that exposure to specific tastes *in utero* or during early infancy enhances preference for foods with those tastes into early childhood. They argue that taste

preferences are innate, but can be magnified through childhood. Preferences for healthy foods can be developed; for example, repeated early exposure to the taste of some vegetables enhances liking for those vegetables, suggesting the importance of optimizing dietary variety and including healthy foods during complementary feeding.

In the large UK ALSPAC cohort, children who were given home-cooked fruit or vegetables more often at 6 months of age were more likely to be eating a higher proportion of fruit and vegetables at 7 years than those who were given home-cooked fruit and vegetables less often. Interestingly, this effect was not seen in relation to the consumption of ready-prepared fruits or vegetables at 6 months.[55] However, this was an observational study and it is not clear whether this finding reflects differences in the flavor of home-prepared versus commercial foods, or perhaps the fact that infants given prepared fruit and vegetables came from families where subsequent opportunities to consume these foods may have been more limited.

In another analysis using data from the ALSPAC cohort, the authors developed an index reflecting the degree of adherence to complementary feeding guidelines, and explored associations between index scores and sociodemographic factors and dietary pattern scores at the age of 3 years.[56] The Complementary Feeding Utility Index consisted of 14 components, including breast-feeding duration, feeding to appetite, timing of introduction to solids, exposure to iron-rich cereals, fruit and vegetable intake, exposure to high-fat/-salt/-sugar foods including sugary drinks, food texture, and meal/snack frequency. Associations were found among index score, maternal age, education, social class, maternal smoking history, and prepregnancy BMI. After adjustment for sociodemographic factors, the index score was associated negatively associated with "processed" and positively associated with "healthy" dietary pattern scores at age 3 years, suggesting that an index of complementary feeding practices reflecting adherence to recommendations may be predictive of later dietary patterns.

METHOD OF FEEDING

In addition to the timing and content of the complementary feeding diet, it is important to consider whether the manner in which foods are given to the infant might be important. Traditionally, infants have generally been spoon-fed with first complementary foods in the form of purees, with subsequent introduction of semisolid and finger foods. However, alongside

recommendations to delay the introduction of solid foods until 6 months, there has been an increasing tendency to avoid the initial "puree" stage altogether and progress straight to finger foods. In the "Baby Led Weaning" method, which is currently very popular among parents in countries such as the United Kingdom and New Zealand, the infant feeds himself hand-held foods instead of being spoon-fed by an adult, sharing family foods and mealtimes. It has been suggested that this approach may encourage improved eating patterns and reduce the risk of overweight and obesity by providing the infant with greater control over his/her intake and encouraging more responsive parenting.[57] However, given the self-selected nature of parents and infants who currently follow this practice, it is not yet clear whether these associations are causal. Furthermore, data are lacking on whether infants who are weaned using this approach obtain sufficient nutrients, including energy and iron, or eat a more diverse range of foods. These important issues ideally need to be tested in a randomized controlled trial.

Emmett and colleagues[58] investigated the impact of the age at introduction of lumpy solid foods in a follow-up of 7821 children from the prospective U.K. ALSPAC cohort. Dietary intake and feeding difficulties were assessed at 7 years of age. Children were divided into three groups based on the age at which they were first introduced to "lumpy" solids: 12.1% before 6 months of age, 69.8% between 6 and 9 months of age, and 18.1% after 9 months. Children introduced to lumpy solids after the age of 9 months ate less of many of the food groups at 7 years, including all 10 categories of fruit and vegetables, than those introduced to lumpy foods between 6 and 9 months. In addition, they were reported as having significantly more feeding difficulties.

PARENTAL FACTORS

Parents clearly play a major role during the complementary feeding process, both in terms of decisions on the timing and content of the diet, and also the way in which the infant is fed, setting rules and expectations, and providing a role model. It is increasingly recognized that parenting style, defined as the way parents interact with a child in terms of attitudes and behaviors across different domains of parenting, including feeding can influence the infant's feeding behavior.[59] In a recent review of literature examining relationships between parenting styles and feeding behaviors and the fruit and vegetable consumption of preschool children, Blissett[60] concluded that an authoritative feeding style, which is typified by

emotional warmth and responsiveness but high expectations for children's dietary adequacy and behavior, accompanied by practices such as modeling consumption of fruit and vegetables, making these foods available within the home, moderately restricting unhealthy alternative snack foods, and encouraging children to try fruit and vegetables, is associated with better consumption in the childhood years. However, most published studies are observational and intervention studies are ideally needed to determine if changing parental feeding style and practices can favorably influence offspring food choice and feeding behavior.

CONCLUSION

The timing, content, and method used to deliver complementary feeding have potentially important effects on health outcomes, nutritional status and subsequent food choices, and feeding behaviors. Research in this field has tended to be dominated by the timing of introduction of complementary foods in the context of the optimal duration of EBF, which has the effect of excluding a large proportion of infants in many high-income countries who receive breast milk substitutes from early infancy. Greater focus on the content of the diet during this period would be beneficial in achieving adequate intakes of nutrients such as iron, while avoiding excess, and promoting the development of healthy dietary habits. The majority of studies in this field have been observational, and intervention trials are much needed to establish causal relationships which can be used as a basis for recommendations.

REFERENCES

1. World Health Organization. *Complementary Feeding of Young Children in Developing Countries: A Review of Current Scientific Knowledge*. Geneva, Switzerland: World Health Organization; 1998. (WHO/NUT/98.1.)
2. Kramer MS, Kakuma R. Optimal duration of exclusive breastfeeding. *Cochrane Database Syst Rev*. 2002;1:CD003517. doi: 10.1002/14651858.CD003517.
3. 55th World Health Assembly. *Infant and Young Child Nutrition*. Geneva, Switzerland: World Health Organization; 2002 (WHA55.25). Internet: http://www.who.int/gb/ebwha/pdf_files/WHA55/ewha5525.pdf
4. Wilson AC, Forsyth JS, Greene SA, et al. Relation of infant diet to childhood health: seven year follow up of cohort of children in Dundee infant feeding study. *BMJ*. 1998;316(7124):21–25.
5. Fewtrell MS. The evidence for public health recommendations on infant feeding. *Early Hum Dev*. 2011;87:715–721.
6. Bauchner H, Leventhal JM, Shapiro ED. Studies of breastfeeding and infections. How good is the evidence? *JAMA*. 1986;256:887–892.

7. Kramer MS. Does breast feeding help protect against atopic disease? Biology, methodology, and a golden jubilee of controversy. *J Pediatr*. 1988;112:181–190.
8. Kramer MS, Kakuma R. Optimal duration of exclusive breastfeeding. *Cochrane Database Syst Rev*. 2012;8:CD003517. doi: 10.1002/14651858.CD003517.pub2.
9. Kramer MS, Guo T, Platt RW, et al. Infant growth and health outcomes associated with 3 compared with 6 mo of exclusive breastfeeding. *Am J Clin Nutr*. 2003;78:291–295.
10. Lanigan JA, Bishop J, Kimber AC, Morgan J. Systematic review concerning the age of introduction of complementary foods to the healthy full-term infant. *Eur J Clin Nutr*. 2001;55:309–320.
11. EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA), Scientific Opinion on the appropriate age for introduction of complementary feeding of infants. *EFSA J*. 2009;7(12):1423 [38 pp.]. doi:10.2903/j.efsa.2009.1423. www.efsa.europa.eu © European Food Safety Authority, 2009.
12. *SACN/COT Statement on the Timing of Introduction of Gluten* http://www.sacn.gov.uk/pdfs/sacn_cot_ _into_.pdf Updated 2011.
13. Chantry CJ, Howard CR, Auinger P. Full breastfeeding duration and associated decrease in respiratory tract infection in US children. *Pediatrics*. 2006;117:425–432.
14. Paricio Talayero JM, Lizan-Garcia M, Otero Puime A, et al. Full breastfeeding and hospitalization as a result of infections in the first year of life. *Pediatrics*. 2006;118:e92–e99.
15. Rebhan B, Kohlhuber M, Fromme H, et al. Breastfeeding duration and exclusivity associated with infants' health and growth: data from a prospective cohort study in Bavaria, Germany. *Acta Paediatrica*. 2009;98:974.
16. Duijts L, Jaddoe VW, Hofman A, Moll HA. Prolonged and exclusive breastfeeding reduces the risk of infectious diseases in infancy. *Pediatrics*. 2010;126:e18–e25.
17. Quigley MA, Kelly YJ, Sacker A. Breastfeeding and hospitalization for diarrheal and respiratory infection in the United Kingdom Millenium Cohort Study. *Pediatr*. 2007;119:e837–e842.
18. Quigley MA, Kelly YJ, Sacker A. Infant feeding, solid foods and hospitalisation in the first 8 months after birth. *Arch Dis Child*. 2009;94:148–150.
19. Reilly JJ, Wells JCK. Duration of exclusive breastfeeding: introduction of complementary feeding may be necessary before 6 months of age. *Br J Nutr*. 2005;94:869–872.
20. Nielsen SB, Wells JC, Slater C, et al. Administering labelled water to exclusively breast-fed infants in studies involving stable isotope dilution techniques. *Isotopes Environ Health Stud*. 2011;47:18–25.
21. Wells JC, Jonsdottir OH, Hibber PL, et al. Randomized controlled trial of 4 compared with 6 mo of exclusive breastfeeding in Iceland: differences in breast-milk intake by stable-isotope probe. *Am J Clin Nutr*. 2012;96:73–79.
22. Lozoff B, Jimenez E, Hagen J, et al. Poorer behavioural and developmental outcome more than 10 years after treatment for iron deficiency. *Pediatrics*. 2000;105:E51.
23. Halterman JS, Kaczorowski JM, Aligne CA, et al. Iron deficiency and cognitive achievement among school-aged children and adolescents in the United States. *Pediatrics*. 2001;107:1381–1386.
24. Jonsdottir OH, Thorsdottir I, Hibberd PL, et al. Timing of the introduction of complementary foods in infancy: a randomized controlled trial. *Pediatrics*. 2012;130:1038–1045.

25. Chantry CJ, Howard CR, Auinger P. Full breastfeeding duration and risk for iron deficiency in US infants. *Breastfeed Med.* 2007;2:63–73.
26. Dube K, Schwartz J, Mueller MJ, et al. Iron intake and iron status in breastfed infants during the first year of life. *Clin Nutr*. 2010;29:773–778.
27. Andersson O, Hellström-Westas L, Andersson D, Domellöf M. Effect of delayed versus early umbilical cord clamping on neonatal outcomes and iron status at 4 months: a randomised controlled trial. *BMJ*. 2011;343:d7157. doi: 10.1136/bmj.d7157.
28. Du Toit G, Katz Y, Sasieni P, et al. Early consumption of peanuts in infancy is associated with a low prevalence of peanut allergy. *J Allergy Clin Immunol*. 2008;122:984–991.
29. Prescott SL, Smith P, Tang M, et al. The importance of early complementary feeding in the development of oral tolerance: concerns and controversies. *Pediatr Allerg Immunol*. 2008;19:375.
30. Nwaru BI, Erkkola M, Ahonen S, et al. Age at the introduction of solid foods during the first year and allergic sensitization at age 5 years. *Pediatrics*. 2010;125:50–59.
31. Olsson C, Hernell O, Hörnell A, et al. Difference in celiac disease risk between Swedish birth cohorts suggests an opportunity for primary prevention. *Pediatrics*. 2008;122:528–534.
32. Agostoni C, Decsi T, Fewtrell MS, et al. Complementary feeding: a commentary by the ESPGHAN Committee on Nutrition. ESPGHAN Committee on Nutrition. *J Paediatr Gastroenterol Nutr*. 2008;46:99–110.
33. American Academy of Pediatrics Section on Breastfeeding. Breastfeeding and the use of human milk. *Pediatrics*. 2012;129:e827–e841.
34. Vriezinga SL, Auricchio R, Bravi E, et al. Randomized feeding intervention in infants at high risk for celiac disease. *NEJM*. 2014;371:1304–15.
35. Lionetti E, Castellaneta S, Francavilla R, et al. Introduction of gluten, HLA status, and the risk of celiac disease in children. *NEJM*. 2014;371:1295–303.
36. Kramer MS, Matush L, Bogdanovitch, et al. Health and development outcomes in 6.5-y-old children breastfed exclsuively for 3 or 6 mo. *AJCN*. 2009;90:1070.
37. Schack-Nielsen L, Sorensen TIA, Mortensen EL, Michaelsen KF. Late introduction of complementary feeding, rather than duration of breastfeeding, may protect against adult overweight. *AJCN*. 2010;91:1–9.
38. Moorcroft KE, Marshall JL, McCormick FM. Association between timing of introducing solid foods and obesity in infancy and childhood: a systematic review. *Mat Child Nutr*. 2011;7:3–26.
39. Magnussen CG, Niinikoski H, Juonala M, et al. When and how to start prevention of atherosclerosis? Lessons from the Cardiovascular Risk in the Young Finns Study and the Special Turku Coronary Risk Factor Intervention Project. *Pediatr Nephrol*. 2012;27:1441–1452.
40. Hopkins D, Emmett P, Steer C, et al. Infant feeding in the second 6 months of life related to iron status: an observational study. *Arch Dis Child*. 2007;92:850–854.
41. Thorsdottir I, Gunnarsson BS, Atladottir H, et al. Iron status at 12 months of age—effects of body size, growth and diet in a population with high birth weight. *Eur J Clin Nutr*. 2003;57:505–513.
42. Krebs NF. Meat as first complementary food for breastfed infants: feasibility and impact on zinc intake and status. *J Pediatr Gastroenterol Nutr*. 2006;42:207–214.
43. Engelmann MDM, Sandstrom B, Michaelsen KF. Meat intake and iron status in late infancy: an intervention study. *J Pediatr Gastroenterol Nutr*. 1998;26(1):26–33. doi: 10.1097/00005176-199801000-00005.

44. Morgan J, Taylor A, Fewtrell T. Meat consumption is positively associated with psychomotor outcome in children up to 24 months of age. *J Pediatr Gastroenterol Nutr*. 2004;39(5):493–498.
45. Michaelsen KF, Larnkjær A, Mølgaard C. Amount and quality of dietary proteins during the first two years of life in relation to NCD risk in adulthood. *Nutr Metab Cardiovasc Dis*. 2012;22:781–786.
46. Koletzko B, von Kries R, Closa R, et al; European Childhood Obesity Trial Study Group. Lower protein in infant formula is associated with lower weight up to age 2 y: a randomized clinical trial. *Am J Clin Nutr*. 2009;89:1836–1845.
47. Singhal A, Kennedy K, Lanigan J, et al. Nutrition in infancy and long-term risk of obesity; evidence from two randomised controlled trials. *Am J Clin Nutr*. 2010;92:1133–1144.
48. Singhal A, Cole TJ, Fewtrell MS, et al. Promotion of faster weight gain in infants born small for gestational age. Is there an adverse effect on later blood pressure? *Circulation*. 2007;115:213–220.
49. Socha P, Grote V, Gruszfeld D, et al. Milk protein intake, the metabolic-endocrine response, and growth in infancy: data from a randomized clinical trial. *Am J Clin Nutr*. 2011;94(suppl 6):1776S–1784S.
50. Larnkjaer A, Hoppe C, Mølgaard C, Michaelsen KF. The effects of whole milk and infant formula on growth and IGF-I in late infancy. *Eur J Clin Nutr*. 2009;63:956–963.
51. Hofman A, Hazelbrock A, Valkenburg HA. A randomized trial of sodium intake and blood pressure in newborn infants. *J Am Med Assoc*. 1983;250:370–373.
52. Geleijnse JM, Hofman A, Witteman JCM, et al. Long-term effects of neonatal sodium restriction on blood pressure. *Hypertension*. 1997;29:913–917.
53. Mennella JA, Trabulsi JC. Complementary foods and flavor experiences: setting the foundation. *Ann Nutr Metab*. 2012;60(suppl 2):40–50.
54. Mennella JA, Lukasewycz LD, Castor SM, Beauchamp GK. The timing and duration of a sensitive period in human flavor learning: a randomized trial. *Am J Clin Nutr*. 2011;93:1019–1024.
55. Coulthard H, Harris G, Emmett P. Long-term consequences of early fruit and vegetable feeding practices in the United Kingdom. *Public Health Nutrition*. 2010;13(12):2044–2051.
56. Golley RK, Smithers LG, Mittinty MN, et al. An index measuring adherence to complementary feeding guidelines has convergent validity as a measure of infant diet quality. *J Nutr*. 2012;142:901–908.
57. Cameron SL, Heath A-LM, Taylor RW. How feasible is baby-led weaning as an approach to infant feeding? A review of the evidence. *Nutrients*. 2012;4:1575–1609.
58. Coulthard H, Harris G, Emmett P. Delayed introduction of lumpy foods to children during the complementary feeding period affects child's food acceptance and feeding at 7 years of age. *Matern Child Nutr*. 2009;5:75–85.
59. Ventura A, Birch L. Does parenting affect children's eating and weight status? *Int J Behav Nutr Phys Act*. 2008;5:15.
60. Blissett J. Relationships between parenting style, feeding style and feeding practices and fruit and vegetable consumption in early childhood. *Appetite*. 2011;57:826–831.

Energy and Substrate Regulation in Obesity

36

Daniel J. Hoffman, PhD Susan B. Roberts, PhD

Overweight and obesity are widely considered to be the major public health problems in affluent countries[1,2] and are becoming increasingly common in some economically disadvantaged societies as well.[3,4] Previously considered a problem mainly occurring in adults, overweight and obesity are now prevalent in children, with an estimated 18% of children aged 2–19 years thought to be affected.[5]

DEFINITION OF OBESITY

The definition of obesity is deceptively simple; it is a state of excessive accumulation of body fat. However, the translation of this simple definition into working standards for excess body fat and body weight has been complicated for several reasons. First, because body fatness is a continuum, the cutoff point for the definition of obesity is necessarily arbitrary.[1] In addition, the harmful effects of obesity result primarily from the excessive accumulation of fat, but there is no simple method for the direct measurement of this body compartment in individuals of any age. These concerns notwithstanding, the Centers for Disease Control and Prevention (CDC) have published body mass index (BMI; kg/m^2) percentiles for children aged up to 19 years (see Appendix), together with useable definitions for "risk of overweight" and "overweight."[6] These

definitions recognize that BMI is a better indicator of relative weight than weight for height, even if it is not precisely aligned with body fatness in all population groups.[7]

The CDC charts provide an improved set of reference growth data that have been updated and smoothed using statistical techniques. However, as reviewed elsewhere,[8] it should be noted that the latest national survey data for children ages 6 years and over were not included in the data set used to calculate the BMI percentiles because of the marked increase in weight of children ages 6 years and older in the latest survey (Third National Health and Nutrition Examination Survey [NHANES III]) compared with previous surveys. The inclusion of the latest survey data would have shifted the percentiles up and resulted in a relative underclassification of the prevalence of overweight. Because the 85th percentile of BMI is used to classify risk of overweight and the 95th percentile is used to classify overweight,[7] the data used effectively define overweight as a fixed percentage relative to population surveys before NHANES III. Thus, the BMI charts can be considered more as recommendations for the healthy range of BMI rather than as current population values because they are designed primarily to provide a reference for the diagnosis of overweight. They can also potentially be used to diagnose underweight (e.g., at the 10th or 5th percentiles), but accepted definitions for underweight are needed. One concern with using BMI to identify overweight children is the potential for misclassification.[8] The relationship between weight and height and body fat content can be weak,[9] and especially in children younger than five years when the high BMI percentiles are very close together, factors such as enhanced muscular development, large head size, and a high torso-to-leg ratio could all falsely elevate the BMI of a child with a healthy amount of body fat into an overweight range. For example, differences in head circumference between a 3-year-old child on the 10th head circumference percentile and one on the 90th percentile translate into a body weight difference of approximately 0.75 kg (assuming that the head weighs 1.7 g/cm^3 and is a sphere). A 3-year-old child's BMI would therefore vary by approximately 0.85 units depending on whether head circumference was on the 10th or 90th percentile. This could mean the difference between the 75th and 85th to 90th percentile for BMI—in other words, the difference between no weight concern and a diagnosis of risk of overweight.

The effect of the relative length of torso and legs can also be predicted using theoretical calculations. Assuming that the proportion of standing

height attributed to torso varies from 0.35 to 0.38 and that for a given contribution to height, torso weight is twice that of legs or neck,[10] BMI would vary by approximately 0.4 units. These calculations highlight the potential for misclassification of individuals, especially those between the 85th and 95th percentiles. Until improved methods for determining body fatness are routinely available, some individual judgment is needed in applying standard BMI definitions for overweight in individual children.

At the same time that the 2000 CDC percentiles were published, Cole et al[11] reported internationally derived, age-specific BMI values for children equivalent to BMI definitions of overweight (BMI = 25 or more) and obesity (BMI = 30 or more) in adults. The adult cutoff values of 25 and 30 were originally developed based on increased health risks.[12] The theoretical advantage of creating equivalent values for children is that they would provide absolute standards for fatness rather than standards relative to population fatness at any given period of survey.[13,14] It should be noted that it is not currently known whether equivalent percentiles in childhood confer similar risk to those found in adults, but because BMI tends to track from childhood through adult life, the approach seems a reasonable one.

As reviewed elsewhere,[8] Cole et al[11] obtained national survey data from six countries (Brazil, the United Kingdom, Hong Kong, the Netherlands, Singapore, and the United States). For each country, the percentiles equivalent to BMI values of 25 and 30 at age 18 years were determined, and then BMI values at different childhood ages were obtained for the same percentiles. There was substantial agreement in childhood BMI values among the six countries; thus, mean values for different ages were computed to give age-specific BMI cutoffs equivalent to adult BMIs of 25 and 30 kg/m^2. Figure 36-1 shows a comparison of the CDC 85th and 95th percentiles with the overweight and obesity cutoff points for BMI provided by Cole et al.[11] For both boys and girls, the 2000 CDC definition of overweight (95th percentile) is markedly higher than the Cole et al[11] definition of overweight, except for the period between age 4 and 5 years. In other words, by the current U.S. definition, many fewer children in the United States and worldwide are overweight. In fact, the 2000 CDC definition of risk of overweight (85th percentile) most closely approximates the Cole et al[11] definition of overweight, with the Cole et al[11] values tending to be slightly higher for ages 2–10 years and slightly lower for 17 years and older. In addition, the CDC BMI definition of overweight is not similar to the Cole et al[11] definition of either overweight or obesity, but the CDC 97th BMI percentile is similar to the Cole et al[11]

definition of obesity for the period between 7 and 14 years.

These comparisons raise the important issue that greater agreement is needed among national and international agencies with regard to definitions of what age-specific BMI values appropriately define overweight and obesity. In the meantime, the 2000 CDC percentiles for BMI might be most appropriate for assessing different degrees of overweight in U.S. children, and the BMI cutoffs of Cole et al[11] could be used for international comparisons of obesity prevalence.

RISKS OF OBESITY

The short-term risks of overweight in children and adolescents extend to reduced scholastic performance, psychological problems, and a number of significant health risks, including hyperlipidemia.[15] In addition, obese children are at increased risk of becoming overweight or obese adults compared with children of normal weight[16] and therefore are at long-term risk of the substantially increased morbidity and mortality, psychological problems, reduced economic achievement, and discrimination that are seen in obese adults.[17] In a summary of 17 studies investigating the long-term risks of childhood overweight for the development of adult obesity, Serdula et al[18] observed that 26%–41% of overweight preschool children go on to become obese adults, with the risk of adult obesity increased to 2.0–2.6 times that of nonoverweight children.

The risk of adult obesity in overweight children increases progressively with age. Data from Braddon et al[19] on changes in the risk of adult obesity according to the age of childhood manifestation are shown in Figure 36-2. In that study, which was conducted in a cohort of English children, the percentage of obese adults who were overweight at age 7 years was only 2.6% for men and 7.5% for women, whereas values increased to 13.9% for men and 25.5% for women who were overweight at age 14 years. In addition, these figures might substantially underestimate the true relationship between early fatness and later obesity because 28.6% of obese men and 36.5% of obese women were already obese or overweight by age 7, a substantially increased percentage relative to the group of children who were categorized as overweight, and again there was an increase in the number of obese adults who were overweight in childhood as age increased (Figure 36-2).

A further issue relevant to the risks of childhood overweight is that there is evidence to suggest that severe overweight developing early in life

is particularly likely to lead to adult obesity. Whitaker et al[16] showed that childhood overweight was increasingly likely to predict adult obesity from age 3 years and older. Similarly, Abraham and Nordsieck[20] reported that 72.2% of severely overweight children (weighing more than 120% of the average for the population) became obese adults; a similar result was found by others.[21] Although these data illustrate the substantial risk of childhood overweight leading to adult obesity, it is also important to note that many overweight children do not become obese adults. Data from Poskitt[22] indicate that only one in nine overweight infants remain obese as adults in England, whereas other studies similarly indicate that a minority of overweight children remain overweight.[19]

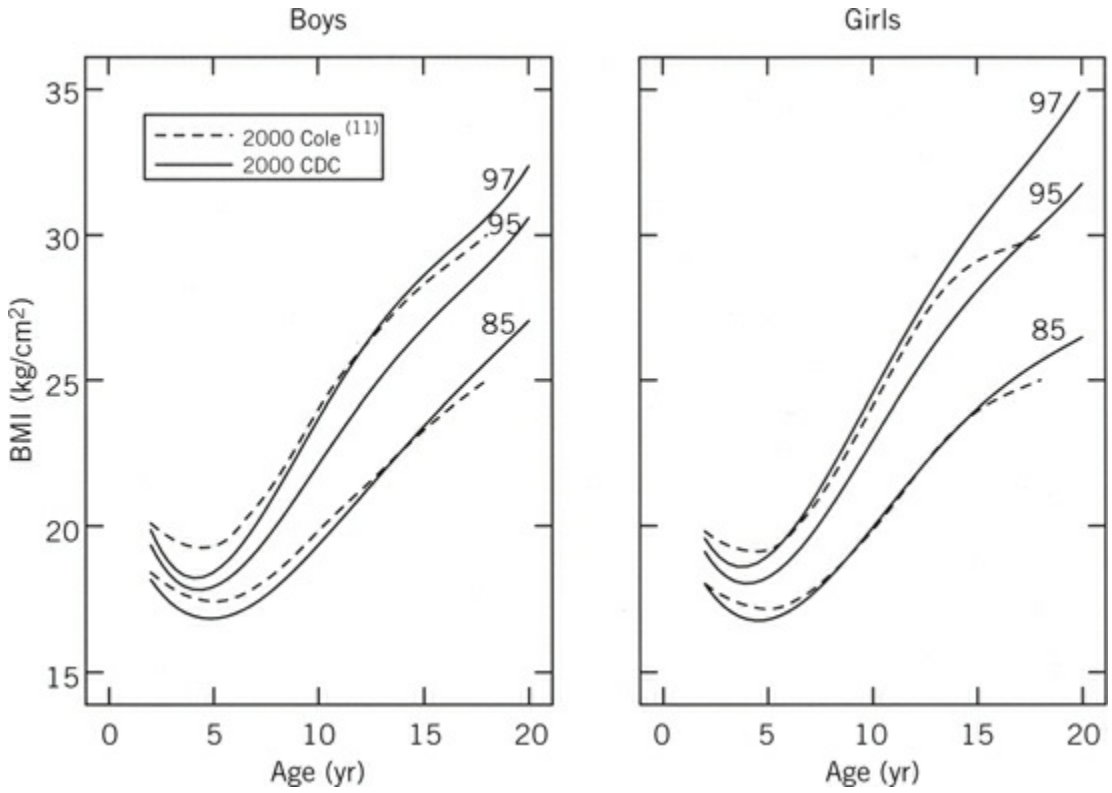


Figure 36-1 Comparison of Cole et al's[11] BMI definitions of overweight and obesity during childhood (broken line) with 2000 CDC 85th and 95th percentiles (defined as risk of overweight and overweight, respectively) and the CDC 97th percentile (*solid line*). (Adapted from Roberts and Dallal.[8])

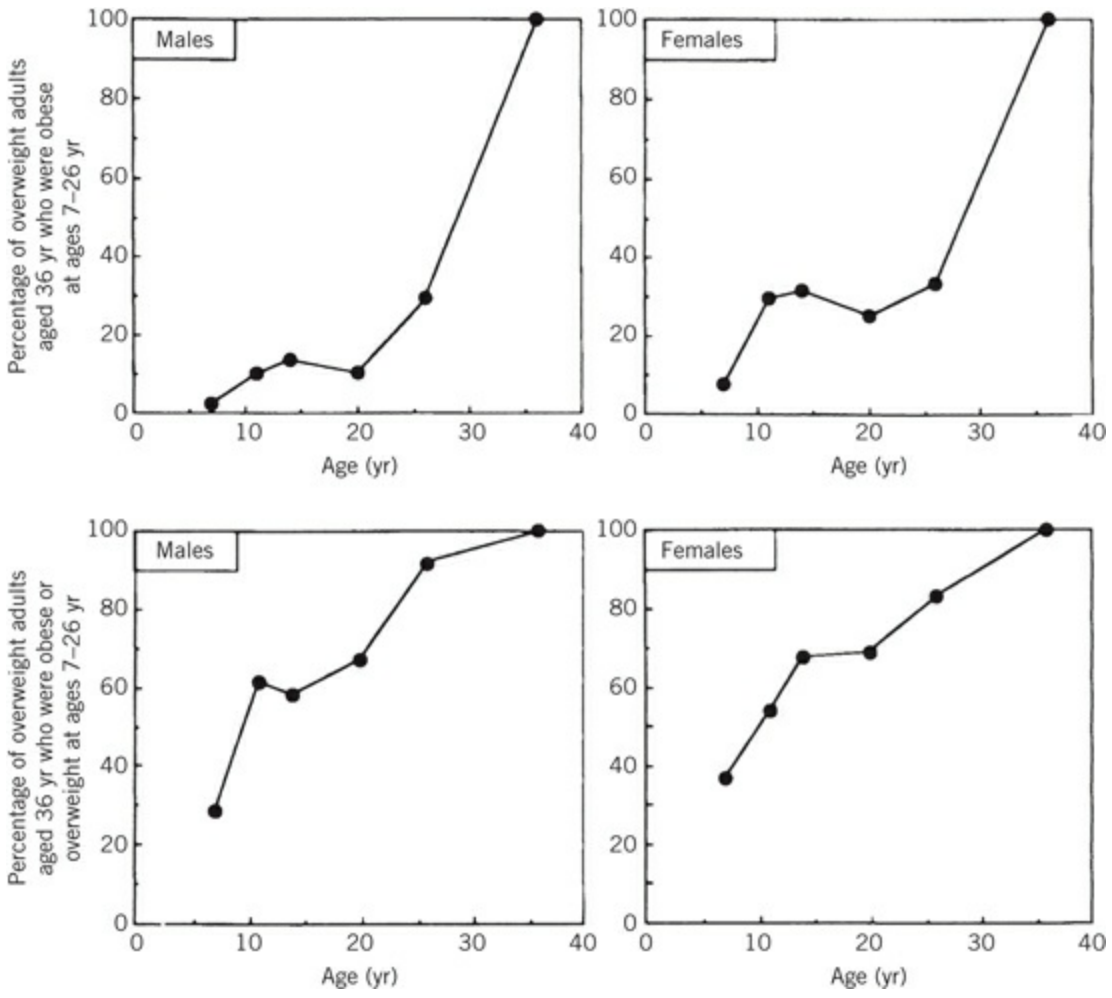


Figure 36-2 Percentage of obese men and women aged 36 years who were already obese (top) or either obese or overweight (bottom) by ages 7–26 years. (Adapted from Braddon et al.[19])

Recent studies have also raised the issue of whether, in addition to increasing the risk of adult obesity, obesity in childhood could also exert an independent influence on adult morbidity and mortality. In an analysis of longitudinal anthropometric data from the Third Harvard Growth Study (1922–1935), Must et al[23] observed that over a 55-year period, men who were overweight in adolescence had significantly increased all-cause mortality (relative risk 1.8) compared with men who were lean during adolescence. This effect of adolescent weight, confirming a similar result found previously by Mossberg,[24] persisted even when weight at age 53 years was controlled. The implication of this finding is that the effects of adolescent weight on morbidity and mortality resulted from the adolescent

obesity directly rather than from the effects of adolescent obesity on adult weight.[24] There was no association between adolescent weight and adult mortality in females in the study of Must et al,[23] and gender was not distinguished in the study by Mossberg.[24]

The mechanisms by which adolescent weight might exert an influence on adult morbidity independent of adult fatness in men and possibly in women are not known. However, reports of increased plaque deposition in the arterial walls of overweight children suggest that there could be long-term effects of early body weight on morphology or metabolism that are not easily reversible. Consistent with this suggestion is the fact that overweight girls have been shown to have a reduced age of menarche, a risk factor for developing breast cancer.[25–27] This effect appears to be primarily related to increased body size rather than to differences in dietary intake in the obese.[28–30] A study by Meyer et al[31] suggested that the lowered age of menarche in overweight girls might be associated with increased weight rather than with increased fat mass.

It is important to note that not all studies of the relationship between childhood weight and adult morbidity have reported positive associations. The study of Barker et al[32] reported an opposite trend, with a negative association between weight at 1 year of age and vascular disease in adult life.[32] Further studies are needed to clarify this issue and in particular to determine whether the long-term effects of early obesity could be important at some ages during childhood but not at other ages.

GENETIC VERSUS ENVIRONMENTAL FACTORS IN THE DEVELOPMENT OF OBESITY

Studies in Animals

There is substantial evidence that obesity in animals can be transmitted genetically.[33] Several genetically obese animal strains exist, such as the Zucker rat and the ob/ob mouse, which have substantially more adipose tissue than their lean counterparts. The fat gain in genetically obese animals is caused by a variety of mechanisms, including hyperphagia and an increased efficiency of energy use. The increased energy efficiency can be attributed primarily to a reduced thermogenesis for body temperature regulation. It is likely that some degree of imbalance in the autonomic nervous system contributes to the defective thermogenesis, as indicated by reduced sympathetic nervous drive in response to mildly cold ambient

temperatures and in response to overeating.[34] In addition, parabiotic studies, in which the circulatory systems of lean and genetically obese animals are joined, have indicated that unidentified circulating factors (or the ability to respond to them correctly) are also important in the development of obesity.[35]

Animal studies also demonstrate that obesity can result from environmental factors, most particularly the availability of high-fat or high-variety diets. Some—but not all—strains of laboratory rodents gain substantial weight and fat when offered cafeteria diets containing a variety of snack foods such as cookies, cheese, cakes, processed meats, nuts, and so forth. Similar results have also been produced by offering synthetic high-fat diets, as well as by providing sucrose solutions in addition to standard rat chow. It could be noteworthy that physical activity does not have as much of an influence on body fat content as does diet in these rodent species. The fact that only some strains of animals, and only some animals within a strain, are susceptible to this diet-induced obesity[36,37] strongly indicates that "environmentally induced" obesity occurs only in genetically susceptible individuals and should therefore really be viewed as a genotype–environment effect. In relation to this observation, it is important to note that the willingness to overconsume food is not entirely a result of absolute standards of taste in animals. Instead, in studies that have separated the process of eating from the process of digestion by the use of esophageal cannulae, it has been shown that unidentified postingestion factors play a dominant role in determining food preferences.[36]

Studies in Humans

Studies on the importance of genetic inheritance versus environmental factors in the development of overweight and obesity have been harder to conduct in humans. It is widely accepted that body fat mass and fat distribution are subject to significant familial transmission[38–43]; in other words, the body compositions of children to a large extent reflect those of their parents. There remains a great deal of uncertainty, however, over the relative importance of genetic inheritance and environmental transmission (transmission between generations through the sharing of common environmental circumstances) in determining body fat content. Reported values for genetic heritability range from negligible to 0.8, implying that genetic inheritance accounts for between 0% and 80% of the variability in body fat mass in the community, and rates of environmental transmission range almost as much.[39,41,43–55]

This uncertainty over the relative importance of genetic and environmental transmission probably relates in large part to the methods that have been used to measure body composition and to the populations that have been studied. Most previous studies have used simple anthropometric indicators of body fat mass, such as weight corrected for height or one or more skinfold thicknesses.[39,43–48,50–55] However, the relationship between anthropometric parameters and fat mass can be very weak.[9] In addition, muscle mass, bone size, and height appear to be strongly inherited.[43] This could help explain the genetic inheritance of indicators of apparent fatness based on weight and could indicate that weight measurements will tend to result in an overestimation of the genetic inheritance of fat mass.

Bouchard et al.[49,56] used hydrostatic weighing (a gold standard method) for assessment of body composition. Data from these investigations indicate that there are significant genetic and environmental components to the transmission of body density, with genetic inheritance accounting for 25% and environmental transmission for 30%. Elder et al[57] recently used several different methods and reported that the heritability of body fatness was 60%, and unique environmental factors accounted for 20%. Similar results were obtained by Dubois et al.[58] Thus, it appears that the family environment is less important than genetic inheritance in determining body fat mass.

Other lines of evidence from studies that effectively separate genetic inheritance from environmental factors confirm the existence of significant environmental factors in human overweight and obesity. For example, the body fat content of unrelated married couples becomes more similar over time.[40] In addition, the prevalence of obesity varies by the season of the year (it is highest in the fall and winter) and location within a country.[59] As further evidence, surveys show that the prevalence of childhood obesity has increased in the United States during the past two decades despite a relatively stable population pool.

METABOLIC PROGRAMMING IN THE DEVELOPMENT OF OBESITY

There is increasing awareness of the possibility that there might be critical periods during the life span when diet and other environmental factors exert a long-term influence on body fat content and the risk of adult obesity in humans,[60–64] a potential event that has been termed “metabolic

programming." As emphasized by Lucas,[60,61] the fact that this tremendously important issue has not yet been resolved reflects the relative lack of randomized dietary intervention trials with long-term follow-up in infants and children. More studies of this type are needed despite the complex ethical issues involved. Nonetheless, a body of evidence has developed to the point that several potential mechanisms to explain "metabolic programming" now exist.[64] Perhaps more important is the fact that several studies now suggest that maternal influences on such programming may not only have epigenetic mechanisms, but such epigenetic changes may persist beyond one generation.[65]

Evidence consistent with the view that metabolic programming of adult body fatness might exist at different ages comes from several sources, in particular indicative of the possibility that either long-term energy insufficiency or long-term energy excess in childhood can lead to excess body fat content in adult life. In 1976, Ravelli et al[66] published the first study suggesting that nutrition of the mother during the intrauterine period might have a long-term influence on the risk of obesity. Specifically, the investigators followed the incidence of obesity among men whose mothers lived in Holland during the Dutch "Hunger Winter," a time when the food supply was reduced in specific areas of the country at the end of the World War II. A significant *increase* in the incidence of obesity at age 19 years was noted among the male offspring of mothers whose energy intake was reduced to famine levels (estimated at approximately 600 kcal/d) specifically during the first trimester of pregnancy, whereas a significant *decrease* in the risk of obesity was observed in women who had undergone energy deprivation during the third trimester. Although "metabolic programming" of intrauterine malnutrition can explain this result, it is also possible that other factors were important. For example, the mothers who were deprived of food might have been particularly concerned about feeding their infants substantial amounts of food after giving birth. In addition, the food provided by the Allied Forces after the famine was extremely high in fat, and the possibility cannot yet be ruled out that this might have had an important long-term effect. More recent studies in humans are consistent with the Ravelli data, suggesting, for example, that low birth weight is associated with significantly greater abdominal adiposity later in life.[67]

Recent studies in animal models have also reported that malnutrition during the early part of pregnancy can be associated with increased body fat in adult life,[68,69] with leptin resistance suggested as one possible

mechanism[70] supporting the belief that long-term metabolic changes can be induced through diet. Although the mechanism for this effect is not known, Anguita et al[69] suggest that morphologic changes in the hypothalamus might be important.

Similar to in utero energy insufficiency, childhood stunting (which is widely considered an indicator of chronic energy insufficiency during early childhood and typically occurs before age 5) has also been correlated with high BMI in several transitional countries, such as Brazil, China, and South Africa.[4,71–74] The risk for obesity and overweight for adults who were stunted as children and adolescents varies among countries but is consistently positive.

The relative risk for obesity for an adult who was stunted in childhood is 1.7 in Brazil, 2.6 in South Africa, and 7.8 in Russia.[71] At the same time, one study from South Africa reported no association between stunting at age 2 years and indicators of excess fat mass (BMI and waist circumference) at age 8 years.[75] Thus, controversies regarding these associations remain, mostly due to the fact that mechanisms to explain these associations remain unclear; however, recent studies have offered possible metabolic as well as behavioral evidence that might explain part of this association.

In perhaps the first piece of evidence that early nutrition has long-term effects on metabolism that predispose a child to subsequent obesity, Hoffman et al[76] reported that nutritionally stunted children have a higher respiratory quotient and lower fat oxidation compared with healthy children from the same environment (Figure 36-3). When these children were studied four years later, it was found that the stunted children had gained more body fat as a proportion of body weight compared to the control children (Martins, PA 2005). This would suggest that not only are they metabolically predisposed to obesity, but the effects appear well before adulthood. In the same series of experiments, stunted children also appeared to have impaired regulation of food intake compared with control children from the same environment,[77] resulting in greater energy consumption per kilogram of body weight when the stunted children were exposed to an abundant, ad libitum diet (Table 36-1). The results of this study need to be replicated but are consistent with the hypothesis that exposure to food insecurity early in life can disrupt normal perceptions of hunger and satiety. Additional support for suggesting that metabolic adaptations occur following chronic undernutrition comes from a study of Buryat tribes in Siberia where it was reported that adults who were stunted

compared to their peers had a lower rate of fat oxidation,[78] consistent with the study from Brazil. Taken together, these studies provide multiple lines of evidence consistent with the suggestion that undernutrition during early life can have long-term metabolic effects that can promote obesity later in life.

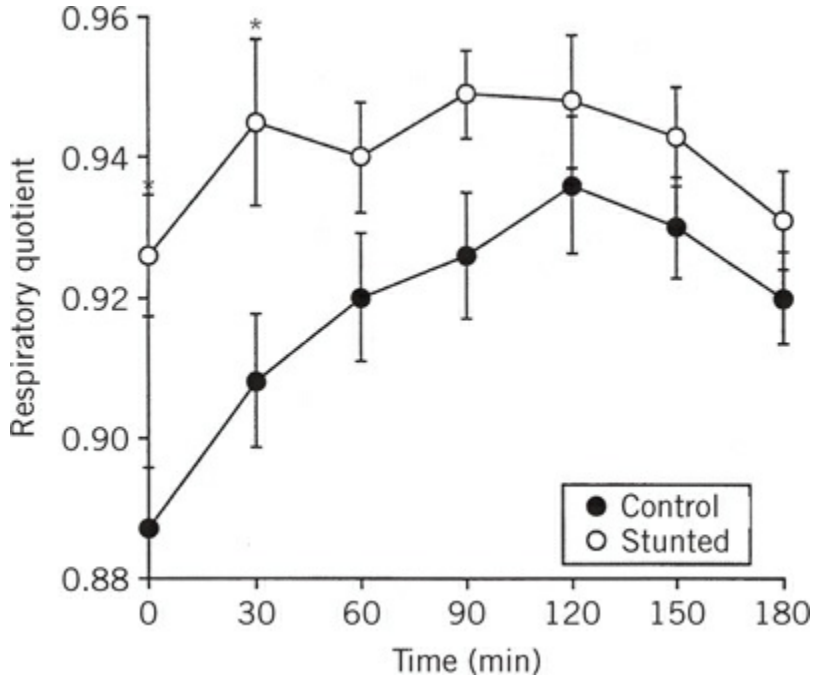


Figure 36-3 Fasting and postprandial respiratory quotient (RQ) in stunted and control children.[60] Values are means ±SEM. The stunted children has a significantly higher fasting RQ compared with control children (t = 0) and postprandially at t = 30 min ($P < 0.05$). The average postprandial RQ was not significantly different between the two groups ($P = 0.10$). (Reproduced with permission from Hoffman et al.[69])

TABLE 36-1 Energy Intake and Ratio of Energy Intake to Resting Energy Expenditure

	Girls		Boys	
Variable	**Control (n = 15)**	**Stunted (n = 13)**	**Control (n = 14)**	**Stunted (n = 14)**
Energy kJ/kg body weight*	60 ± 15	74 ± 14	67 ± 18	81 ± 9
Energy intake (kcal/d)/resting energy expenditure*	1.67 ± 0.4	1.89 ± 0.3	1.69 ± 0.4	1.93 ± 0.2

Adapted from Hoffman et al.[70]
± SD.
*$P < 0.05$ between groups.

The first months after birth have received considerable attention as another potential time when metabolic programming, through the choice of diet (breast milk or formula) and the time of weaning, could have a long-term influence on body fatness. In particular, although several studies have suggested no effect of formula feeding on body fatness, the more recent studies have consistently indicated that breast-fed infants tend to become leaner adults.[79–87] Additionally, the duration of breast-feeding may have a u-shaped relationship with adult body fatness, indicated by recent finding that although 5–7 months breast-feeding is associated with lower rates of adult overweight than 0–2 months of overfeeding, breast-feeding of greater than 8 months was also associated with elevated rates of overweight at 60 years of age.[88] The finding of a tendency for formula feeding to promote later body fatness in full-term babies is consistent with preliminary findings from randomized studies in preterm infants[89] and also with the well-known effect of early overfeeding in rodent models (resulting in a permanent increase in body weight and fat content).[90] Additionally, similar results have been demonstrated in nonhuman primates.[91]

Further studies are needed to determine whether metabolic or behavioral influences of formula versus breast-feeding are responsible for the differences observed. However, of potential relevance to this issue is the fact that Singhal et al[89] conducted a prospective study of preterm infants who were fed either a standard infant formula, banked breast milk, or an enriched preterm formula (5 g protein/L and 9 g fat/L greater than the standard formula) to examine long-term effects on circulating leptin.

Between the ages of 13 and 16 years, the children were measured for body fatness and leptin concentrations. It was found that the children who had received the preterm formula had a 30% higher ratio of leptin to fat mass compared with both of the other groups,[89] suggesting that the excess energy intake or altered nutrient composition of the preterm formula might have programmed higher circulating leptin during adolescence.

ENERGY AND SUBSTRATE REGULATION IN OBESITY

Energy Regulation

Average nonoverweight children consume more than two million kilojoules a year, but their body fat stores increase only modestly if growth is normal and energy expenditure approximates energy intake. A change of only 10%, either in intake or output, can lead to a 15-lb change in body fat in a single year, in addition to that deposited as a normal consequence of growth. The majority of children, who are of normal weight, do not exhibit such undesirable changes in body weight and fat, implying that normal energy regulation must be remarkably sensitive.

Until recently, our understanding of the underlying cause of this sensitive energy regulation was dominated by the guiding principle of energy balance:

$$\text{Energy stored} = \text{Energy intake} - \text{Energy expenditure}$$

Thus, it has been recognized that high levels of body fat (i.e., energy) cannot have been accumulated without energy intake being unusually high or energy expenditure being unusually low, or a combination of these two options. Similar calculations to those given above can illustrate that the dysregulation of energy in obesity needs to be only very small for excess adipose tissue to accumulate. For example, a child who becomes 100 lb overweight by the age of 15 years needs to have eaten less than an extra approximately 100 MJ/d between the ages of 5 and 15 years. There has been considerable interest in defining whether energy intake or energy expenditure is the primary factor determining this energy dysregulation in obese individuals, as well as seeking to identify metabolic mechanisms that determine the separate regulation of these parameters. As described below, most of the investigations conducted in children have been cross-sectional in nature, that is, comparing measurements of energy intake and expenditure between lean and obese individuals or examining the relationship between these parameters in groups of individuals differing widely in body composition. It is also important to recognize that there may have been changes in the relative importance of excess energy intake and low-energy expenditure over time, reflecting shifting food behavior patterns and physical activity norms.

Energy Intake

The energy intake of an individual is usually understood to be that energy available to the body after obligatory losses of energy in urine and stools. Although complex metabolic balance studies are needed to precisely determine this “metabolizable energy,” it can also be predicted from the

dietary intakes of protein, fat, carbohydrate, and alcohol using the standard conversion factors of 4.0, 9.0, 4.0, and 7.0 kcal/g, respectively, which take into account the obligatory losses.

The regulation of food intake clearly plays an important part in the sensitive energy regulation in children of normal weight. More than 70 years ago, Davis[92] demonstrated that very young children of normal body weight are capable of choosing a balanced diet and maintaining normal growth without dietary intervention from adult caregivers. This remained true over periods of several days or longer, although food choices at individual meals were frequently extremely erratic. More recently, Birch et al[93] and Shea et al[94] provided additional strong evidence in support of an important role for the regulation of food intake in the energy regulation of young children of normal body weight. These investigators showed that although there might be substantial meal-to-meal fluctuations in energy and nutrient intakes, prompt and appropriate compensation occurs spontaneously so that the 24-hour energy intake remains relatively stable. A further report by Birch et al[95] also demonstrated that young children are able to compensate for the inclusion of low-calorie fat substitutes in a meal to maintain a constant energy intake.

These studies clearly demonstrate that in children of normal body weight, energy regulation through the regulation of food intake occurs and plays an important role in the maintenance of a stable pattern of body composition change throughout childhood. However, a caveat to these studies is that the observed capacity for normal energy regulation may occur only when relatively healthy foods are offered, and the ability of young children to accurately regulate energy intake when offered many highly energy dense foods of low nutritional value has not been systematically explored.

Individual studies of differences in reported energy intake between overweight and nonoverweight children have been conflicting, but taking all of the data together, reported energy intakes of overweight children are not significantly greater than those of nonoverweight children of the same age and are significantly less when expressed in relation to body weight.[29,94–113] However, it is known that overweight individuals, including adolescents,[114] underreport their energy intake to a greater degree than do individuals of normal weight, which indicates that the finding of normal food intake in overweight children might be owing to methodological error.

In contrast to the studies of reported energy intake, studies from psychological investigations of eating behavior in overweight children

suggest that already overweight children consume more total energy than do nonoverweight subjects. Studies by several groups have suggested that overweight children and adolescents are faster eaters, chew less, prefer high-fat foods, and do not exhibit the normal pattern of decelerating eating toward the end of a meal, as seen in children of normal weight.[115–119] They may also consume more sugar sweetened beverages. Furthermore, obese parents have been reported to give more encouragement to their children to eat than do parents of nonobese children and serve them larger portions of food.[120] These findings are also consistent with results from studies in adults.[121] In addition, overweight children may have an abnormal pattern of energy consumption within the day, with a reduced percentage of calories being consumed at breakfast, which is a meal widely recognized to be associated with prevention of weight gain in adults.[122] As described below, studies of total energy expenditure (TEE) might be more reliable for estimating the energy intake of overweight children than studies of reported energy intake and confirm that already overweight children consume more total energy than children of normal weight. A recent summary of TEE data in children concluded that high energy intake rather than low TEE could explain secular increases in body weight during the period 1985–1995, a suggestion that is consistent with increases in per capita food supply during this time.[123]

Energy Expenditure

The TEE of an individual, which is equal to the metabolizable energy intake when body energy stores remain constant, can be divided into compartments that relate to the different types of energy expenditure measurements that are made. Typically, three major compartments are defined. The resting energy expenditure is the energy expended at rest after an overnight fast and is the major compartment, comprising at least half of the TEE in a typical individual. The thermic effect of feeding is the increase in energy expenditure above baseline resulting from the consumption and processing of food and comprises 7%–15% of TEE on average. This thermic effect of feeding has been further subdivided into facultative and obligatory components, where obligatory energy expenditure is the expenditure for necessary aspects of digestion and assimilation, whereas the facultative component has been proposed as a form of energy wasting that can perhaps serve to burn off surplus dietary energy. The final component of TEE is the energy expended for physical activity and arousal, which is generally the second largest component of

TEE and the most variable because to a large extent it is under voluntary control.

The role of energy expenditure in the regulation of energy balance has been controversial in the past but is perhaps less so currently. Studies in both adults[124–128] and adolescents[129] have suggested that humans have a relatively limited capacity for dissipating surplus dietary energy by thermogenesis. In these studies, subjects have been overfed, and changes in energy expenditure and energy storage have been measured to assess the fate of the excess dietary energy. In all carefully controlled studies, the ability to dissipate excess energy, seen as an increase in TEE or components of energy expenditure, has represented a relatively minor component of the excess dietary intake.[124–128]

Thus, the majority of the surplus energy was stored, primarily in the form of additional body fat. Studies of this kind highlight the importance of the regulation of food intake, rather than thermogenesis, in the short-term regulation of energy balance.

It is possible, however, that energy expenditure plays other critical roles in energy regulation that these studies do not address. In particular, it has been speculated that low-energy expenditure in one or more of the components of expenditure could have an association with increased body fatness over the long-term. This could be a result of the low-energy expenditure promoting increased fatness or of an underlying mechanism promoting fat gain that precipitates low-energy expenditure to provide the surplus energy needed for the weight gain. Studies of differences in components of energy expenditure between obese and nonobese children are discussed below. These cross-sectional studies do not define the causal role of energy expenditure in obesity but do serve to define the energy expenditure of children who are already obese in relation to normal values. Prospective studies that have investigated the role of energy expenditure in the weight gain in obesity are discussed in the section “Roles of Energy Intake and Expenditure in the Development of Obesity.”

Concerning resting energy expenditure, a summary of studies in otherwise healthy overweight and nonoverweight children reveals that the overweight individuals do not have reduced resting energy expenditure. In relation to age, the resting energy expenditure of overweight children appears to be within the normal range at 8–10 years and increases subsequently compared with values of normal children (Figure 36-4). Values for energy expenditure per unit of fat-free mass (the metabolically active component of the body because fat mass is relatively metabolically inactive) are somewhat lower in overweight compared with nonoverweight

children. However, this is an inappropriate method of expressing resting energy expenditure in individuals of different sizes because there is a wide variability in the energy expenditure of different tissues that comprise the lean mass. Tissues such as brain and heart have a very high-energy expenditure and tissues such as muscle have a low-energy expenditure. It appears that overweight and nonoverweight individuals have more similar amounts of tissues with high-energy expenditure than they have tissues with low-energy expenditure. Thus, it is more appropriate to examine the relationship between fat-free mass and resting energy expenditure than to compare resting energy expenditure per unit of fat-free mass between different groups.

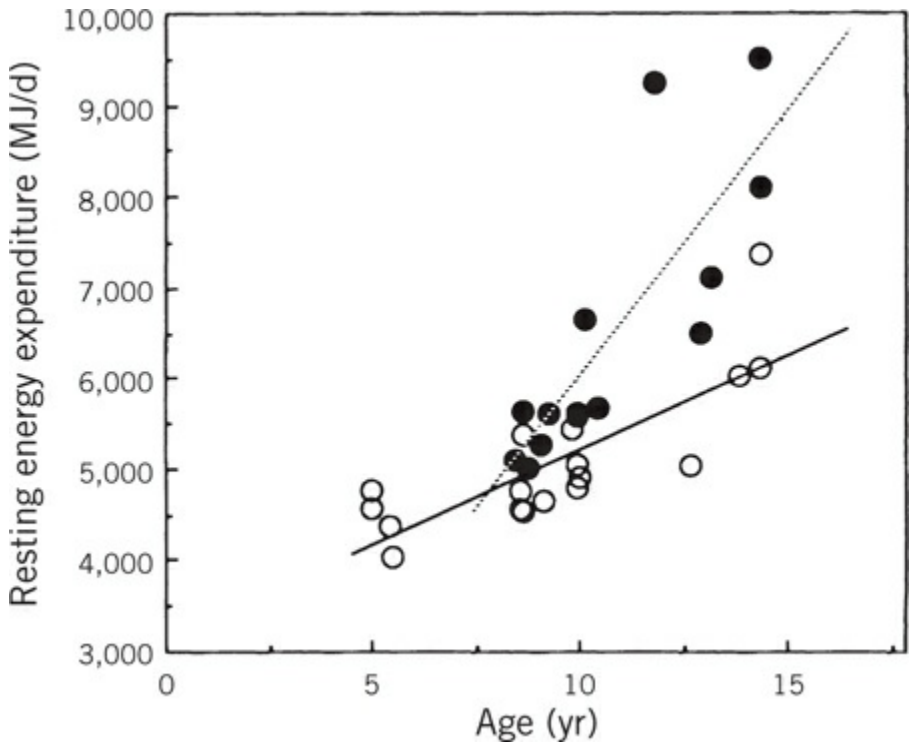


Figure 36-4 Relationship between resting energy expenditure (in kJ/d, where 1 kJ = 0.239 kcal) and age in nonoverweight (*solid line*) and overweight (*broken line*) boys and girls aged 5–15 years.[171] Data points are group means reported between 1989 and 1994.[92,103,122,132,137,141,144,172–178] There was a significant effect of obesity status on the relationship between resting energy expenditure and age ($P < 0.001$), and the relationship between resting energy expenditure and age was significant in both nonoverweight ($r^2 = 0.630$, $P < 0.000$) and overweight children ($r^2 = 0.682$, $P < 0.000$).

As seen in Figure 36-5, neither the intercept nor the slope of the relationship differs between obese and nonobese individuals, demonstrating that there is no difference in the resting energy expenditure of overweight and nonoverweight children when the data are appropriately normalized for metabolically active tissue. This result, obtained by combining data from studies in which data from at least two groups of subjects were available, is compatible with results from regression analyses in a group of children with a wide range of body compositions,[130] as well as with results of numerous studies in adults.[131]

Several studies on the thermic effect of feeding demonstrate a normal energy expenditure in overweight compared with nonoverweight children and adolescents,[129,132,133] a result that contrasts with the numerous studies that indicate a reduced thermic effect of feeding in obese adults.[134,135] There is one report of a reduced thermic effect of feeding in obese children,[136] and the duration of obesity has been implicated as a possible explanation for differences in results among different studies.[132] This explanation could also help explain the fact that the thermic effect of feeding is reduced in obese adults.

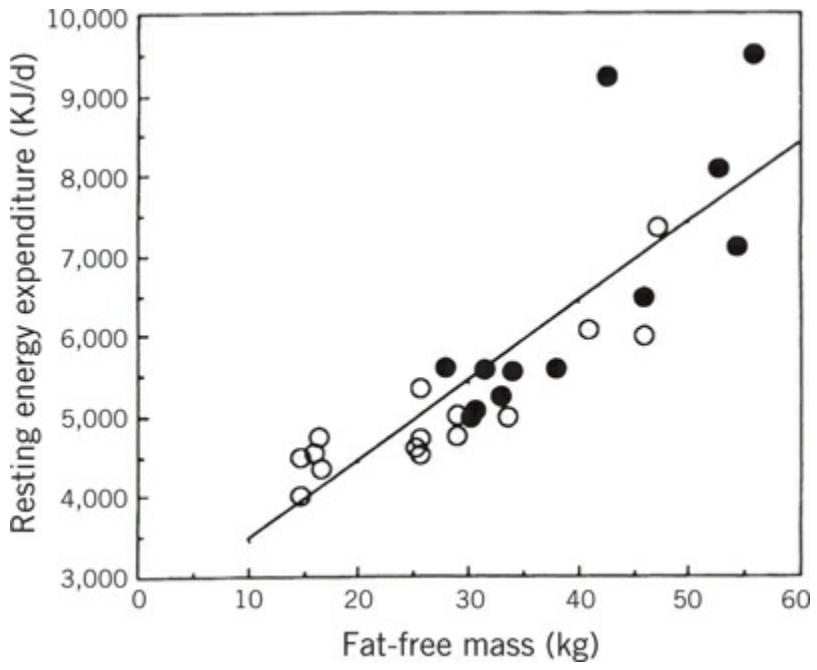


Figure 36-5 Relationship between resting energy expenditure and fat-free mass in nonoverweight and overweight boys and girls aged 5–15 years.[171] Data points are group means (in kJ/d, where 1 kJ = 0.239 kcal)

reported between 1989 and 1993.[103,122,132,141,144,172–178] There was no significant effect of overweight status on resting energy expenditure in multiple regression analyses, including fat-free mass as an independent variable. The relationship between resting energy expenditure and fat-free mass is significant (r^2 = 0.712, P < 0.000).

Concerning physical activity, several studies have reported no difference in physical activity between lean and overweight children,[71,87,137,138] whereas other studies have reported low[114,123,139–142]or even high[143] levels of physical activity in overweight children relative to nonoverweight controls.

Television viewing has also been investigated separately as a marker for low physical activity and for its relationship to body composition in children.[144] Although the mechanism by which television viewing might influence body fat mass is not known, a reduction in physical activity is strongly implicated as part of the mechanism because television viewing is a sedentary activity that might replace more strenuous ones, together with the possibility that dietary intake could be increased as well.[145,146] Studies in this area have not produced consistent results, with some studies reporting a negative association between the time spent watching television and body fatness[145,147] and other studies failing to substantiate this result.[138,148]

This controversy might reflect the fact that inactivity during television viewing can potentially be compensated for by strenuous activity at other times; more research is needed to elucidate the relationship between body fat mass and physical activity in children as well as in adults. A further unresolved aspect of the literature on television viewing is that there are reports of both a decrease[137] and a normal[149] resting energy expenditure during television viewing in obese children compared with nonobese controls.

On the basis of the studies described above, it cannot be concluded that the obese child is necessarily inactive, or that inactivity is an important cause of obesity. However, this conclusion does not exclude an important role for physical activity in obese children, and further studies are needed to clarify this important issue.

Studies on TEE as determined using the doubly labeled water technique[150,151] have been conducted as a way to examine the combined effects of different components of energy expenditure in normal-weight and overweight children and also as a more accurate way to assess energy

expenditure for physical activity (when combined with measurements of resting energy expenditure). Using this method, several studies in children and adults have indicated an inverse association between energy expenditure for physical activity and body fat mass when values for physical activity are normalized for body size by the use of the ratio of total to resting energy expenditure as a physical activity index.[106,152,153] and one recent study reported no difference between lean and overweight children despite differences in sedentary behavior and sports participation.[154] The mechanism of this association is not known but is likely to include the fact that inactivity promotes insulin resistance and could enhance fat oxidation.[155] Although this ratio of total to resting energy expenditure is reduced with increasing body fat content, the TEE for physical activity is typically high. This is because resting energy expenditure and the energy costs of most activities increase with body size. These consistent findings contrast with the substantial controversy over the effects of obesity on physical activity when studies using reported estimates of physical activity are considered.

Doubly labeled water measurements of TEE have also been used to examine differences in energy intake and energy requirements between normal-weight and overweight children. The recent Dietary Reference Intakes for Macronutrients[156] summarized all available data on TEE in childhood to report that there is no substantial difference between normal-weight and overweight children in the relationship between TEE and body weight, height, and age, with equations suitable for all children based on summaries of all available doubly labeled water data:

$$\text{TEE (kcal/d)} = 89 \times \text{weight (kg)} - 100 \text{ (boys and girls 0–2 years)}$$

$$\text{TEE} = 88.5 - 61.9 \times \text{age (years)} + \text{PA} \times 26.7 \times \text{weight (kg)} + 903 \times \text{height (mm)} \text{ (boys 3–18 years)}$$

$$\text{TEE} = 135.3 - 30.8 \times \text{age (years)} + \text{PA} \times 10.0 \times \text{weight (kg)} + 934 \times \text{height (mm)} \text{ (girls 3–18 years)}$$

where PA is a physical activity coefficient used for children 3 years and older:

PA = 1.00 if physical activity level is sedentary.

PA = 1.13 if physical activity level is low.

PA = 1.27 if physical activity level is active.

PA = 1.45 if physical activity level is very active.

TEE does not equal energy intake in children because there is additional consumed energy used for growth that is deposited in the form of lean tissue and fat. However, except during the early months of life and during the peak of pubertal development, energy used for energy deposition is only 20–25 kcal/d[156]; thus, TEE closely reflects energy intake. The equations given above indicate that children who are overweight do indeed need more energy to maintain current body weight than do normal-weight children. For example, a child more than 3 years of age who is already overweight by 10 kg compared with normal-weight children will have increased TEE and hence energy intake of 100–267 kcal/d. Table 36-2 summarizes estimated energy requirements for children of reference weights, based on the above equations with an increment added for the energy deposition in normal growth.

A final issue to consider when comparing differences in energy expenditure between lean and overweight individuals is the energy expenditure response to overfeeding. As detailed above, an ability to burn off surplus dietary energy has been suggested as potentially important in the regulation of energy balance. However, in a study of overweight and nonoverweight adolescents, Bandini et al[157] found no difference between groups in the ability to dissipate excess energy intake. This finding is consistent with the view, outlined above, that diet-induced thermogenesis plays only a small role in the regulation of energy balance.

Substrate Balance

As an extension of the energy balance principles detailed above, it has been suggested that body carbohydrate balance could play a critical role in overall energy regulation.[157] The basic principle behind this theory of energy regulation, which is in keeping with the earlier glucostatic theory of energy regulation, is that balance of the primary energy substrates, fat and carbohydrate, have to be considered separately.[158] Thus:

TABLE 36-2 Estimated Energy Requirements[a]

Age	Reference Weight (kg)	Activity Level (kcal/d)			
		Sedentary	Low Active	Active	Very Active
Boys					
1 mo	4.4	472	(No activity level specified before 3 y)		
6 mo	7.9	645			
12 mo	10.3	844			
18 mo	11.7	961			
24 mo	12.7	1,050			
3 y	14.3	1,162	1,324	1,485	1,683
6 y	20.7	1,328	1,535	1,742	1,997
9 y	28.6	1,530	1,787	2,043	2,359
12 y	40.5	1,798	2,113	2,428	2,817
15 y	56.3	2,223	2,618	3,013	3,499
18 y	67.2	2,383	2,823	3,263	3,804
Girls					
1 mo	4.2	438	(No activity level specified before 3 y)		
6 mo	7.2	593			
12 mo	9.5	768			
18 mo	11.0	899			
24 mo	12.1	997			
3 y	13.9	1,080	1,243	1,395	1,649
6 y	20.2	1,247	1,451	1,642	1,961
9 y	29.0	1,415	1,660	1,890	2,273
12 y	41.6	1,617	1,909	2,183	2,640
15 y	52.0	1,731	2,057	2,363	2,870
18 y	56.2	1,690	2,024	2,336	2,858

[a] Values are estimated average requirements from Dietary Reference Intakes.[143]

$$\text{Fat stored} = \text{Fat intake} - \text{Fat oxidation}$$
$$\text{Carbohydrate stored} = \text{Carbohydrate intake} - \text{Carbohydrate oxidation}$$

Because the energy in carbohydrate stores is very small in relation to that in fat stores, the carbohydrate stores have a high turnover rate and are liable to be depleted quickly and frequently. The essential requirement of key tissues for glucose suggests that signals should exist to efficiently monitor and correct body carbohydrate balance. An extension of this theory of nutrient regulation is that dietary macronutrient composition plays an important role in determining body fat content.[158] This is because the higher the dietary fat content, the more total food (and energy) needs to be consumed to maintain carbohydrate intake and therefore carbohydrate stores, and the fatter the individual will be because of increased energy intake.

Flatt[158] has also suggested that the increase in body fat induced by a high-fat diet could be self-limiting because reduced insulin sensitivity and increased circulating free fatty acids tend to occur with increased body fat

content, promoting increased fat oxidation and preservation of carbohydrate stores, resulting in a stabilization of hunger and satiety signals from body carbohydrate stores and stability of food intake and body fat stores. This model provides a potential explanation for body fat regulation that does not rely on the concept of a novel "signal" from fat stores that detects and corrects a body set point.

Support for the importance of nutrient balance in the overall regulation of energy balance can be derived from several sources, although there is currently little information available on infants and children. Flatt[158] has demonstrated in mice the critically important idea that energy balance is indeed adjusted on a day-to-day basis to maintain stable body carbohydrate balance, through adaptive fluctuations in food intake. Studies in humans have indicated that carbohydrate balance is more closely regulated than fat balance.[159,160] Concerning the effect of dietary composition on energy intake and body composition, animal studies have repeatedly demonstrated that hyperphagia and obesity can be induced in many strains of normal-weight rodents by feeding a high-fat diet as described above.[36]

In humans, results from some,[161–164] although not all,[164–167] short-term intervention studies in adults indicate that energy intake increases with consumption of a high-fat diet. The fact that not all human studies have found an increase in energy intake with consumption of a high-fat diet does not invalidate the nutrient balance theory of energy regulation because a high-fat diet could induce positive energy balance by decreasing energy expenditure, rather than increasing energy intake as suggested by both animal and adult human studies.[168,169] Also consistent with the nutrient balance model are the data from cross-sectional studies that indicate that consumption of a high-fat diet is frequently associated with high body fat mass in adults.[170–174] Thus, as predicted by the nutrient balance model above, obese and overweight adults tend to consume a high-fat diet than do nonobese subjects and, at least in the short-term, might be hyperphagic.

Some,[142,143] but not all,[87] studies in children and adolescents support the view that high dietary fat intake is associated with obesity. It is possible that this controversy could be owing to the fact that the fiber content of the diet and its energy density might have significant effects on food intake. Although high-fat diets tend to be low in fiber and have a high energy density, this is not always so, and differences in these aspects of the diet (unreported in the studies described above) might have been important. However, an impairment in total fat oxidation is unlikely to

explain an association between dietary fat intake and the development of obesity in review of the several recent reports in children[175–177] that rates of total fat oxidation are equivalently responsive to change in dietary intake in both lean and overweight children. There is also some suggestion that endogenous fuels are less readily oxidized in overweight to lean individuals[178] which requires further study. An additional important consideration in relation to dietary intake is that on theoretical grounds, saturated fat can be expected to have more adverse effects on body fat mass than other fats do because of its influence on insulin sensitivity.[155] and indeed several recent studies have reported lower oxidation of saturated fats than mono-or polyunsaturated fats.[179,180]

ROLES OF ENERGY INTAKE AND EXPENDITURE IN THE DEVELOPMENT OF OBESITY

As described above, the majority of published investigations into the causes of early overweight have been cross-sectional in nature, that is, they have related measurements of body fat mass to energy expenditure or other parameters measured at the same time. This approach provides important information on energy intake, energy expenditure, and substrate oxidation in individuals who are already obese but does not address the question of what happens during the development of the obese state. A finding, for example, of high energy intake in overweight children does not preclude the possibility that energy intake was normal or even low during the period of excess weight gain. Recent prospective studies have addressed the issue of causality in overweight by investigating whether energy expenditure is low in individuals with increased susceptibility to overweight (because of parental obesity). Although, the results have been conflicting, with reports of both low-energy expenditure[181–185] and normal energy expenditure[186–188] in the pre-overweight state the current weight of evidence indicates that low-energy expenditure is a contributing factor in some circumstances.[189]

SUMMARY AND IMPLICATIONS FOR THE TREATMENT OF OBESITY

The excess fat mass that characterizes obesity is attributable to both

genetic susceptibility and environmental factors and results from energy intake exceeding energy expenditure plus the energy requirement for normal growth over a prolonged period of time. However, this increment in energy intake over energy expenditure can be as little as 400 MJ/d or less if it persists over a period of years. The studies summarized in this chapter do not provide definitive evidence as to whether this differential between energy intake and energy expenditure is caused primarily by decreased energy expenditure or by increased energy intake. However, the weight of evidence currently indicates that both overeating and under-expending can be primary causes of overweight in childhood, and that once a child is overweight, energy intake is higher than that of normal-weight children. In addition, dietary fat, in particular saturated fat, can promote fat deposition through its influence on energy intake.

Further studies are needed in this area, in particular to delineate the usual roles of high energy intake and low-energy expenditure as causal factors in excessive weight gain leading to overweight in childhood. Current prescriptions for the treatment of childhood overweight, taking into account the degree of caution that is appropriate for a disorder in which the treatment can itself have detrimental consequences, should be based on individualizing for each child a decrease in energy intake relative to energy expenditure by a combination of moderate dietary therapy and promotion of energy expenditure from physical activity (see Chapter 37, "Evaluation and Management of Obesity").

REFERENCES

1. Committee on Diet and Health, Food and Nutrition Board, Commission on Life, National Research Council. *Diet And health: Implications for Reducing Chronic Disease*. Washington, DC: National Academy Press; 1989.
2. Livingstone MBE. Childhood obesity in Europe: a growing concern. *Public Health Nutr*. 2001;4:109–116.
3. Popkin BM. The nutrition transition in low-income countries: an emerging crisis. *Nutr Rev*. 1994;52:285–298.
4. Sawaya AL, Dallal G, Solymos G, et al. Obesity and malnutrition in a shantytown population in the city of Sao Paulo, Brazil. *Obes Res*. 1995;3:107S–115S.
5. Department of Health and Human Services, Centers for Disease Control and Prevention. http://www.cdc.gov/index.htm.
6. Kuczmarski RJ, Ogden CL, Grummer-Strawn LM, et al. *Advance Data From Vital and Health Statistics*. No. 314, Vol. 314. Hyattsville, MD: National Center for Health Statistics; 2000.
7. Himes JH, Dietz WH. Guidelines for overweight in adolescent preventive services: recommendations from an expert committee. *Am J Clin Nutr*. 1994;59:307–316.
8. Roberts SB, Dallal GE. The new childhood growth charts. *Nutr Rev*. 2001;59:31–

36.
9. Garn SM, Leonard WR, Hawthorne VM. Three limitations of the body-mass index. *Am J Clin Nutr*. 1986;44:996.
10. Trowbridge FL, Marks JS, de Romana GL, et al. Body composition of Peruvian children with short stature and high weight-for-height. II. Implications for the interpretation for weight-for-height as an indicator of nutritional status. *Am J Clin Nutr*. 1987;46:411–418.
11. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition for child overweight and obesity worldwide: international survey. *BMJ*. 2000;320:1240–1243.
12. World Health Organization. *Obesity: Preventing and Managing The Global Epidemic. Report of a WHO Consultation on Obesity; 1997 June 3–5; Geneva*. Geneva, Switzerland: World Health Organization; 1998.
13. Dietz WH, Robinson TN. Use of the body mass index (BMI) as a measure of overweight in children and adolescents. *J Pediatr*. 1988;132:191–193.
14. Bellizzi MC, Dietz WH. Workshop on childhood obesity: summary of the discussion. *Am J Clin Nutr*. 1999;70:173S–175S.
15. Dietz WH. Prevention of childhood obesity. *Pediatr Clin North Am.* 1986;33:823–833.
16. Whitaker RC, Wright JA, Pepe MS, et al. Predicting obesity in young adulthood from childhood and parental obesity. *N Engl J Med*. 1997;337:869–873.
17. Wadden TA, Stunkard AJ. Psychosocial consequences of obesity and dieting. In: Stunkard AJ, Wadden TA, eds. *Obesity: Theory and Therapy*. New York: Raven Press; 1993:163–177.
18. Serdula MK, Ivery D, Coates RJ, et al. Do obese children become obese adults? A review of the literature. *Prev Med*. 1993;22:167–177.
19. Braddon FEM, Rodgers B, Wadsworth MEJ, Davies JMC. Onset of obesity in a 36 year birth cohort study. *BMJ*. 1986;293:299–303.
20. Abraham S, Nordsieck M. Relationship of excess weight in children and adults. *Public Health Rep*. 1960;75:263–273.
21. Rimm IJ, Rimm AA. Association between juvenile onset obesity and severe adult obesity in 73,532 women. *Am J Public Health*. 1976;66:479–481.
22. Poskitt EME. Do fat babies stay fat? *BMJ*. 1977;6052:7–9.
23. Must A, Jaques PF, Dallal GE, et al. Long-term morbidity and mortality of overweight adolescents. A follow-up of the Harvard Growth Study of 1922 to 1935. *N Engl J Med*. 1992;327:1350–1355.
24. Mossberg H-O. 40-year follow-up of overweight children. *Lancet*. 1989;ii:491–493.
25. Staszewski J. Age at menarche and breast cancer. *J Natl Cancer Inst.* 1971;47:935–940.
26. Kelsey JL. A review of the epidemiology of human breast cancer. *Epidemiol Rev*. 1979;1:74–109.
27. MacMahon B, Trichopoulos D, Brown J. Age at menarche, urine estrogens, and breast cancer risk. *Int J Cancer*. 1982;30:427–431.
28. Moisan J, Meyer F, Gingras S. A nested case-control study of the correlates of early menarche. *Am J Epidemiol*. 1990;132:953–961.
29. Maclure M, Travis LB, Willet W, MacMahon B. A prospective cohort study of nutrient intake and age of menarche. *Am J Clin Nutr.* 1991;54:649–656.
30. Post GB, Kemper HCG. Nutrient intake and biological maturation during adolescence. The Amsterdam growth and health longitudinal study. *Eur J Clin Nutr*. 1993;47:400–408.

31. Meyer F, Moisan J, Marcoux D, Bouchard C. Dietary and physical determinants of menarche. *Epidemiology*. 1990;1:377–381.
32. Barker DJP, Osmond C, Winter PD, et al. Weight in infancy and death from ischaemic heart disease. *Lancet*. 1989;2(8663):577–580.
33. Bray GA, York DA. Hypothalamic and genetic obesity in experimental animals: an autonomic and endocrine hypothesis. *Physiol Rev*. 1979;59:719–809.
34. York DA. Genetic models of animal obesity. In: Bjorntorp P, Brodoff BN, eds. *Obesity*. Philadelphia, PA: JB Lippincott; 1992:233–240.
35. Harris RB, Hervey E, Hervey GR. Body composition of lean and obese Zucker rats in parabiosis. *Int J Obes*. 1987;11:274–280.
36. Sclafani A. Dietary-induced overeating. *Ann N Y Acad Sci*. 1989;575:281–291.
37. West DB, Boozer CN, Moody DL, Atkinson RL. Dietary obesity in nine inbred mouse strains. *Am J Physiol*. 1992;262(6 Pt 2):R1025–R1032.
38. Biron P, Mongeau JG, Bertrand D. Familial resemblance of body weight and height in 374 homes with adopted children. *Pediatrics.* 1977;91:555–558.
39. Garn SM, Bailey SM, Cole PE. Similarities between parents and their adopted children. *Am J Phys Anthropol*. 1979;45:539–544.
40. Garn SM, Cole PE, Bailey SM. Living together as a factor in family-line resemblances. *Hum Biol*. 1979;51:565–587.
41. Savard R, Bouchard C, Leblanc C, Tremblay A. Familial resemblance in fatness indicators. *Ann Hum Biol*. 1983;10:111–118.
42. Heller R, Garrison RJ, Havlik RJ, et al. Family resemblances in height and relative weight in the Framingham Heart Study. *Int J Obes*. 1984;8:399–405.
43. Devor EJ, McGue M, Crawford MH, Lin PM. Transmissible and nontransmissible components of anthropometric variation in the Alexanderwohl Mennonites: resolution by path analysis. *Am J Phys Anthropol*. 1986;69:83–92.
44. Borjeson M. The aetiology of obesity in children. *Acta Paediatr Scand.* 1976;65:279–287.
45. Hartz A, Giefer E, Rimm AA. Relative importance of the effect of family environment and heredity on obesity. *Ann Hum Genet (Lond)*. 1977;41:185–193.
46. Bray GA. The inheritance of corpulence. In: Cioffi LA, ed. *The Body Weight Regulatory System: Normal and Disturbed Mechanisms*. New York: Raven Press; 1981:185–195.
47. Annest JL, Sing CF, Biron P, Mongeau JG. Familial aggregation of blood pressure and weight in adoptive families. *Am J Epidemiol*. 1983;117:492–506.
48. Byard PJ, Sharma K, Russell JM, Rao DC. A family study of anthropometric traits in a Punjabi community: an investigation of familial transmission. *Am J Phys Anthropol*. 1984;64:97–104.
49. Bouchard C, Savard R, Despres JP, et al. Body composition in adopted and biological siblings. *Hum Biol*. 1985;57:61–75.
50. Stunkard AJ, Sorensen TIA, Hanis C, et al. An adoption study of human obesity. *N Engl J Med*. 1986;314:193–198.
51. Price RA, Cadoret RJ, Stunkard AJ, Troughton E. Genetic contributions to human fatness: an adoption study. *Am J Psychiatry*. 1987;144:1003–1008.
52. Newman B, Selby JV, Quesenberry CP, et al. Nongenetic influences of obesity on other cardiovascular disease risk factors: an analysis of identical twins. *Am J Public Health*. 1990;80:675–678.
53. Stunkard AJ, Harris JR, Pedersen NL, McClearn GE. The body mass index of twins who have been reared apart. *N Engl J Med*. 1990;322:1483–1487.
54. Fabsitz RR, Carmelli D, Hewitt JK. Evidence for independent genetic influences on obesity in middle age. *Int J Obes*. 1992;16:657–666.

55. Sorensen T, Holst C, Stunkard A. Correlations of body mass index of adult adoptees and their biological and adoptive relatives. *Int J Obes*. 1992;16:227–236.
56. Bouchard C, Perusse L. Heredity and body fat. *Annu Rev Nutr*. 1988;8:259–277.
57. Elder SJ, Roberts SB, McCrory MA, et al. Effect of body composition methodology on heritability estimation of body fatness. *Open Nutr J*. 2012;6:48–58.
58. Dubois L, Ohm Kyvik K, Girard M, et al. Genetic and environmental contributions to weight, height, and BMI from birth to 19 years of age: an international study of over 12,000 twin pairs. *PLoS One*. 2012;7(2):e30153.
59. Dietz WHJ, Gortmaker SL. Factors within the physical environment associated with childhood obesity. *Am J Clin Nutr*. 1984;39:619–624.
60. Lucas A. Does early diet program future outcome? *Acta Paediatr Scand.* 1990;365:58–67.
61. Lucas A. Programming by early nutrition in man. In: Bock GR, Whelan J, eds. *The Childhood Environment and Adult Disease*. London, UK: John Wiley & Sons; 1991:38–55.
62. Dietz WH. Critical periods in childhood for the development of obesity. *Am J Clin Nutr*. 1994;59:955–959.
63. Martorell R, Stein AD, Schroeder DG. Early nutrition and later adiposity. *J Nutr*. 2001;131:874S–880S.
64. Rabadan-Diehl C, Nathanielsz P. From mice to men: research models of developmental programming. *J Dev Orig Health Dis*. 2013;4(1):3–9.
65. Galliano D, Bellver J. Female obesity: short-and long-term consequences on the offspring. *Gynecol Endocrinol*. 2013;20:20.
66. Ravelli GP, Stein ZA, Susser MW. Obesity in young men after famine exposure in utero and early infancy. *N Engl J Med*. 1976;295:349–353
67. Novotny R, Vijayadeva V, Grove J, et al. Birth size and later central obesity among adolescent girls of Asian, White, and Mixed ethnicities. *Hawaii J Med Public Health*. 2013;72(2):50–55.
68. Jones AP, Simson EL, Friedman MI. Gestational undernutrition and the development of obesity in rats. *J Nutr*. 1984;114:1482–1484.
69. Anguita RM, Sigulem DM, Sawaya AL. Intrauterine food restriction is associated with obesity in young rats. *J Nutr*. 1993;123:1421–1428.
70. Shin BC, Dai Y, Thamotharan M, et al. Pre-and postnatal calorie restriction perturbs early hypothalamic neuropeptide and energy balance. *J Neurosci Res*. 2012;90(6):1169–1182.
71. Popkin BM, Richards MK, Monteiro CA. Stunting is associated with overweight in children of four nations that are undergoing the nutrition transition. *J Nutr*. 1996;126:3009–3016.
72. Sawaya AL, Grillo LP, Verreschi I, et al. Mild stunting is associated with higher susceptibility to the effects of high-fat diets: studies in a shantytown population in Sao Paulo, Brazil. *J Nutr*. 1998;128:415S–420S.
73. Schroeder DG, Martorell R, Flores R. Infant and child growth and fatness and fat distribution in Guatemalan adults. *Am J Epidemiol*. 1999;149:177–185.
74. Florencio T, Ferreira HS, de França AP, et al. Obesity and undernutrition in a very-low income population in the city of Maceio, northeastern Brazil. *Br J Nutr*. 2001;86:277–284.
75. Cameron N, Wright MM, Griffiths PL, et al. Stunting at 2 years in relation to body composition at 9 years in African urban children. *Obes Res*. 2005;13(1):131–136.
76. Hoffman DJ, Sawaya AL, Verreschi I, et al. Why are nutritionally stunted children at increased risk of obesity? Studies of metabolic rate and fat oxidation in

shantytown children from Sao Paulo, Brazil. *Am J Clin Nutr*. 2000;72:702–707.
77. Hoffman DJ, Roberts SB, Verreschi I, et al. Regulation of energy intake may be impaired in nutritionally stunted children from the shantytowns of Sao Paulo, Brazil. *J Nutr*. 2000;130:2265–2270.
78. Leonard WR, Sorensen MV, Mosher MJ, et al. Reduced fat oxidation and obesity risks among the Buryat of Southern Siberia. *Am J Hum Biol*. 2009;21(5):664–670.
79. Wilkinson PW, Pearlson J, Parkin JM, et al. Obesity in childhood: a community study in Newcastle upon Tyne. *Lancet*. 1977;1(8007):350–352.
80. Dine MS, Gartside PS, Glueck CJ, et al. Where do the heaviest children come from? A prospective study of white children from birth to 5 years of age. *Pediatrics*. 1979;63:1–7.
81. Vobecky JS, Vobecky J, Shapcott D, Demers P-P. Nutrient intake patterns and nutritional status with regard to relative weight in early infancy. *Am J Clin Nutr*. 1983;38:730–738.
82. Kramer MS, Barr RG, Leduc DG, et al. Infant determinants of childhood weight and adiposity. *J Pediatr*. 1985;107:104–107.
83. Warrington S, Storey DM. Comparative studies on Asian and Caucasian children. 2. Nutrition, feeding, practices and health. *Eur J Clin Nutr*. 1988;42:68–80.
84. Agras WS, Kraemer HC, Berkowitz RI, Hammer LD. Influence of early feeding style on adiposity at 6 years of age. *J Pediatr*. 1990;116:805–809.
85. Strubak V, Skultetyova M, Hromadova M, et al. Late effects of breast-feeding and early weaning: seven-year prospective study in children. *Endocrine Regul*. 1991;25:53–57.
86. Elliot KG, Kjohede CL, Gournis E, Rasmussen KM. Duration of breastfeeding associated with obesity during adolescence. *Obes Res*. 1997;5:538–541.
87. von Kries R, Koletzko B, Sauerwald T, et al. Breast feeding and obesity: cross sectional study. *BMJ*. 1999;319:147–150.
88. O'Tierney PF, Barker DJ, Osmond C, et al. Duration of breast-feeding and adiposity in adult life. *J Nutr*. 2009;139(2):422S–425S.
89. Singhal A, Farooqi IS, O'Rahilly S, et al. Early nutrition and leptin concentrations in later life. *Am J Clin Nutr*. 2002;75:993–999.
90. Hahn P. Effect of litter size on plasma cholesterol and insulin and some liver and adipose tissue enzymes in adult rodents. *J Nutr*. 1984;114:1231–1234.
91. Lewis DS, Bertrand HA, McMahan CA, et al. Influence of preweaning food intake on body composition of young adult baboons. *Am J Physiol*. 1989;275:R1128–R1135.
92. Davis CM. Self selection of diet by newly weaned infants: an experimental study. *Am J Dis Child*. 1928;36:651–79.
93. Birch LL, Johson SL, Andresen G, et al. The variability of young children's energy intake. *N Engl J Med*. 1991;324:232–235.
94. Shea S, Stein AD, Basch CE, et al. Variability and self-regulation of energy intake in young children in their everyday environment. *Pediatrics*. 1992;90:542–546.
95. Birch LL, Johnson SL, Jones MB, Peters JC. Effects of a nonenergy fat substitute on children's energy and macronutrient intake. *Am J Clin Nutr*. 1993;58:326–333.
96. Stefanik PA, Heald FP, Mayer J. Caloric intake in relation to energy output of obese and non-obese adolescent boys. *Am J Clin Nutr*. 1959;7:55–62.
97. Durnin JVGA, Lonergan ME, Good J, Ewan A. A cross-sectional nutritional and anthropometric study, with an interval of 7 years, on 611 young adolescent schoolchildren. *BMJ*. 1974;32:169–178.
98. Morgan KJ, Zabik ME, Leveille GA. The role of breakfast in nutrient intake of 12-year-old children. *Am J Clin Nutr*. 1981;34:1418–1427.

99. Persson LA, Carlgren G. Measuring children's diets: evaluation of dietary assessment techniques in infancy and childhood. *Int J Epidemiol*. 1984;13:506–517.
100. Griffiths M, Rivers JPW, Payne PR. Energy intake in children at high and low risk of obesity. *Hum Nutr Clin Nutr*. 1987;41C:425–430.
101. Epstein LH, Wing RR, Cluss P, et al. Resting metabolic rate in lean and obese children: relationship to child and parent weight and percent-overweight change. *Am J Clin Nutr*. 1989;49:331–336.
102. Bandini LG, Schoeller DA, Cyr HN, Dietz WH. Validity of reported energy intake in obese and nonobese adolescents. *Am J Clin Nutr*. 1990;54:421–425.
103. Butte NF, Wong WW, Garza C, et al. Energy requirements of breast-fed infants. *J Am Coll Nutr*. 1991;10:190–195.
104. Drake MA. Anthropometry, biochemical iron indexes, and energy and nutrient intake of preschool children: comparison of intake at day care center and at home. *J Am Diet Assoc*. 1991;91:1587–1588.
105. Miller WC. Diet composition, energy intake, and nutritional status in relation to obesity in men and women. *Med Sci Sports Exerc*. 1991;23:280–284.
106. Nicklas TA, Webber LS, Berenson GS. Studies of consistency of dietary intake during the first four years of life in a prospective analysis: Bogalusa Heart Study. *J Am Coll Nutr*. 1991;10:234–241.
107. Pelto GH, Urgello J, Allen LH, et al. Household size, food intake and anthropometric status of school-age children in a highland Mexican area. *Soc Sci Med*. 1991;33:1135–1140.
108. Stein AD, Shea S, Basch CE, et al. Variability and tracking of nutrient intakes of preschool children based on multiple administrations of the 24-hour dietary recall. *Am J Epidemiol*. 1991;134:1427–1437.
109. Albertson AM, Tobalmann RC, Engstrom A, Asp EH. Nutrient intake of 2-to 10-year-old American children: 10-year trends. *J Am Diet Assoc*. 1992;92:1492–1495.
110. Livingstone MBE, Prentice AM, Coward WA, et al. Validation of estimates of energy intake by weighed dietary record and diet history in children and adolescents. *Am J Clin Nutr*. 1992;56:29–35.
111. Rasanen L, Ylonen K. Food consumption and nutrient intake of one-to two-year-old Finnish children. *Acta Paediatr Scand*. 1992;81:7–11.
112. Maffeis C, Schutz Y, Zoccante L, et al. Meal-induced thermogenesis in lean and obese prepubertal children. *Am J Clin Nutr*. 1993;57:481–485.
113. Nicklas TA, Bao W, Webber LS, Berenson GS. Breakfast consumption affects adequacy of total daily intake in children. *J Am Diet Assoc*. 1993;93:886–891.
114. Nicklas TA, Webber LS, Srinivasan R, Berenson GS. Secular trends in dietary intakes and cardiovascular risk factors of 10-y-old children: the Bogalusa Heart Study (1973–1988). *Am J Clin Nutr*. 1993;57:930–937.
115. Schoeller DA, Fjeld CF. What have we learned from the doubly labeled water method? *Annu Rev Nutr*. 1991;11:355–375.
116. Marston AR, London P, Cooper LM. A note on the eating behaviour of children varying in weight. *J Child Psychol Psychiatry*. 1976;17:221–224.
117. Drabman RS, Cordua GD, Hammer D, et al. Development trends in eating rates of normal and overweight preschool children. *J Child Dev*. 1979;50:211–216.
118. Barkeling B, Ekman S, Rossner S. Eating behaviour in obese and normal weight 11-year-old children. *Int J Obes*. 1992;16:355–360.
119. Kosova EC, Auinger P, Bremer AA. The relationships between sugar-sweetened beverage intake and cardiometabolic markers in young children. *J Acad Nutr Diet*.

2013;113(2):219–227.
120. Fisher JO, Birch LL. Fat preferences and fat consumption of 3-to 5-year-old children are related to parental adiposity. *J Am Diet Assoc*. 1995;95:759–764.
121. Ray JW, Klesges RC. Influences on the eating behavior of children. In: Williams CL, Kimm SYS, eds. *Childhood Obesity*. Vol 699. New York: New York Academy of Sciences; 1993:57–69.
122. Spitzer L, Rodin J. Human eating behavior: a critical review of studies in normal weight and overweight individuals. *Appetite*. 1981;2:293–329.
123. Bellisle F, Rolland-Cachera M-F, Deheeger M, Guilloud-Bataille M. Obesity and food intake in children: evidence for a role of metabolic and/or behavioral daily rhythms. *Appetite*. 1998;11:111–118.
124. Swinburn BA, Jolley D, Kremer PJ, et al. Estimating the effects of Energy imbalance on changes in body weight in children. *Am J Clin Nutr*. 2006;83:859–863.
125. Dauncy MJ. Metabolic effects of altering the 24 h energy intake in man, using direct and indirect calorimetry. *Br J Nutr*. 1980;43:257–269.
126. Ravussin E, Schutz Y, Acheson KJ, et al. Short-term, mixed-diet overfeeding in man: no evidence for "luxuskonsumption." *Am J Physiol Endocrinol Metab*. 1985;249:E470–E477.
127. Forbes GF, Brown MR, Welle SL, Lipinski BA. Deliberate overfeeding in women and men: energy cost and composition of the weight gain. *Br J Nutr*. 1986;56:1–9.
128. Roberts SB, Young VR, Fuss P, et al. Energy expenditure and subsequent nutrient intakes in overfed young men. *Am J Physiol*. 1990;259:R461–R469.
129. Bandini LG, Schoeller DA, Edwards J, et al. Energy expenditure during carbohydrate overfeeding in obese and nonobese adolescents. *Am J Physiol*. 1989;256:E357–E367.
130. Goran MI, Kaskoun M, Johnson R. Determinants of resting energy expenditure in young children. *J Pediatr*. 1995;126:679,680.
131. Ravussin E, Bogardus C. Relationship of genetics, age, and physical fitness to daily energy expenditure and fuel utilization. *Am J Clin Nutr*. 1989;49:968–975.
132. Molnar D, Varga P, Rubecz I, et al. Food-induced thermogenesis in obese children. *Eur J Pediatr*. 1985;144:27–31.
133. Maffeis C, Schutz Y, Pinelli L. Postprandial thermogenesis in obese children before and after weight reduction. *Eur J Clin Nutr*. 1992;46:577–583.
134. Segal KR, Lacayanga I, Dunaif A, et al. Impact of body fat mass and percent fat on metabolic rate and thermogenesis in men. *Am J Physiol*. 1989;256:E573–E579.
135. Segal KR, Edano A, Tomas MB. Thermic effect of a meal over 3 and 6 hours in lean and obese men. *Metabolism*. 1990;39:985–992.
136. Katch VL, Moorehead CP, Becque MD, Rocchini AP. Reduced short-term thermic effect of a meal in obese adolescent girls. *Eur J Appl Physiol Occup Physiol*. 1992;65:535–540.
137. Klesges RC, Shelton ML, Klesges LM. Effects of television on metabolic rate: potential implications for childhood obesity. *Pediatrics*. 1993;91:281–286.
138. Wolf AM, Gortmaker SL, Cheung L, et al. Activity, inactivity, and obesity: racial, ethnic, and age differences among schoolgirls. *Am J Public Health*. 1993;83:1625–1627.
139. Waxman M, Stunkard AJ. Caloric intake and expenditure of obese boys. *J Pediatr*. 1980;96:187–193.
140. Berkowitz RI, Agras WS, Korner AF, et al. Physical activity and adiposity: a longitudinal study from birth to childhood. *J Pediatr*. 1985;106:734–738.
141. Sunnegardh J, Bratteby L-E, Hagman U, Samuelson G. Physical activity in

relation to energy intake and body fat in 8-and 13-year old children in Sweden. *Acta Paediatr Scand*. 1986;75:955–963.
142. Eck LH, Klesges RC, Hanson CL, Slawson D. Children at familial risk for obesity: an examination of dietary intake, physical activity and weight status. *Int J Obes*. 1992;16:71–78.
143. Gazzaniga JM, Burns TL. Relationship between diet composition and body fatness, with adjustment for resting energy expenditure and physical activity, in preadolescent children. *Am J Clin Nutr*. 1993;58:21–28.
144. Robinson TN. Television viewing and childhood obesity. *Pediatr Clin North Am.* 2001;48:1–8.
145. Dietz WH Jr, Gortmaker SL. Do we fatten our children at the television set? Obesity and television viewing in children and adolescents. *Pediatrics*. 1985;75:807–812.
146. Coon KA, Goldberg J, Rogers BL, Tucker KL. Relationships between use of television during meals and children's food consumption. *Pediatrics*. 2001;107:E7.
147. Vasques C, Mota M, Correia T, Lopes V. Prevalence of overweight/obesity and its association with sedentary behavior in children. *Rev Port Cardiol*. 2012;31(12):783–788.
148. Robinson TN, Hammer LD, Killen JD, et al. Does television viewing increase obesity and reduce physical activity? Cross-sectional and longitudinal analyses among adolescent girls. *Pediatrics*. 1993;91:273–280.
149. Dietz WH, Bandini LG, Morelli JA, et al. Effect of sedentary activities on resting metabolic rate. *Am J Clin Nutr*. 1994;59:556–559.
150. Schoeller DA. Measurement of energy expenditure in free-living humans by using doubly labeled water. *J Nutr*. 1988;118:1278–1289.
151. Roberts SB. Use of the doubly labeled water method for measurement of energy expenditure, total body water, water intake, and metabolizable energy intake in humans and small animals. *Can J Physiol Pharmacol*. 1989;67:1190–1198.
152. Roberts SB, Heyman MB, Evans WJ, et al. Dietary energy requirements of young adult men, determined by using the doubly labeled water method. *Am J Clin Nutr*. 1991;54:499–505.
153. Goran MI, Carpenter WH, Poehlman ET. Total energy expenditure in 4-to 6-yr-old children. *Am J Physiol*. 1993;264:E706–E711.
154. Salbe AD, Weyer C, Lindsay RS, et al. Assessing Risk factors for obesity between childhood and adolescence: I. Birth weight, Childhood adiposity, parental obesity, insulin, and leptin. *Pediatrics*. 2002;110 (2 Pt 1):299–306.
155. Parker DR, Weiss ST, Troisi R, et al. Relationship of dietary saturated fatty acids and body habitus to serum insulin concentrations: the Normative Aging Study. *Am J Clin Nutr*. 1993;58:129–136.
156. National Academy of Sciences. *Dietary Reference Intakes for Macronutrients*. Washington, DC: National Academy of Sciences; 2002.
157. Bandini LG, Schoeller DA, Dietz WH. Energy expenditure in obese and nonobese adolescents. *Pediatr Res*. 1990;27:198–203.
158. Flatt JP. Importance of nutrient balance in body weight regulation. *Diabetes Metab Res Rev*. 1988;4:571–581.
159. Flatt JP, Ravussin E, Acheson KJ, Jequier E. Effects of dietary fat on postprandial substrate oxidation and on carbohydrate and fat balances. *J Clin Invest*. 1985;76:1019–1024.
160. Abbott WGH, Howard BV, Christin L, et al. Short-term energy balance: relationship with protein, carbohydrate, and fat balances. *Am J Physiol*.

1988;255:E332–E337.
161. Lissner L, Levitsky DA, Strupp BJ, et al. Dietary fat and the regulation of energy intake in human subjects. *Am J Clin Nutr*. 1987;46:886–892.
162. Tremblay A, Plourde G, Despres JP, Bouchard C. Impact of dietary fat content and fat oxidation on energy intake in humans. *Am J Clin Nutr*. 1989;49:799–805.
163. Kendall A, Levitsky DA, Strupp BJ, Lissner L. Weight loss on a low-fat diet: consequences of the imprecision of the control of food intake in humans. *Am J Clin Nutr*. 1991;53:1124–1129.
164. Sheppard L, Kristal AR, Kushi LH. Weight loss in women participating in a randomized trial of low-fat diets. *Am J Clin Nutr*. 1991;54:821–828.
165. Foltin RW, Fischman MW, Moran TH, et al. Caloric compensation for lunches varying in fat and carbohydrate content by humans in a residential laboratory. *Am J Clin Nutr*. 1990;52:969–980.
166. DeGraaf C, Hulshof T, Weststrate J, Jas P. Short-term effects of different amounts of protein, fat and carbohydrate on satiety. *Am J Clin Nutr*. 1992;55:33–38.
167. Foltin RW, Rolls BJ, Moran TH, et al. Caloric, but not macronutrient, compensation by humans for required-eating occasions with meals and snacks varying in fat and carbohydrate. *Am J Clin Nutr*. 1992;55:331–342.
168. Storlien LH, James DE, Burleigh KM, et al. Fat feeding causes widespread in vivo insulin resistance, decreased energy expenditure, and obesity in rats. *Am J Physiol*. 1986;251:E576–E583.
169. Prewitt TE, Schmeisser D, Bowen PE, et al. Changes in body weight, body composition, and energy intake in women fed high and low fat diets. *Am J Clin Nutr*. 1991;54:304–310.
170. Castro DE. Macronutrient relationships with meal patterns and mood in the spontaneous feeding behavior of humans. *Physiol Behav*. 1987;39:561–569.
171. Dreon ME, Frey-Hewitt B, Ellsworth N, et al. Dietary fat: carbohydrate ratio and obesity in middle aged men. *Am J Clin Nutr*. 1988;47:995–1000.
172. Romieu I, Willet WC, Stampfer MJ. Energy intake and other determinants of relative weight. *Am J Clin Nutr*. 1988;47:406–412.
173. Miller WC, Lindeman AK, Wallace J, Niederpruem M. Diet composition, energy intake and exercise in relation to body fat in men and women. *Am J Clin Nutr*. 1990;52:426–430.
174. Tucker LA, Kano MJ. Dietary fat and body fat: a multivariate study of 205 adult females. *Am J Clin Nutr*. 1992;56:616–622.
175. Sunehag AL, Toffolo G, Treuth MS, et al. Effects of dietary macronutrient content on glucose metabolism in children. *J Clin Endocrinol Metab*. 2002;87:5168–5178.
176. Treuth MS, Sunehag AL, Trautwein LM, et al. Metabolic adaptation to high-fat and high-carbohydrate diets in children and adolescents. *Am J Clin Nutr*. 2003;77:479–489.
177. Maffeis C, Schutz Y, Grezzani A, et al. Meal-induced thermogenesis and obesity: is a fat meal a risk factor for fat gain in children? *J Clin Endocrinol Metab*. 2001;86:214–219.
178. Sun M, Schutz Y, Maffeis C. Substrate metabolism, nutrient balance and obesity development in children and adolescents: a target for intervention? *Obes Rev*. 2004;5:183–188.
179. Piers LS, Walker KZ, Stoney RM, et al. The influence of the type of dietary fat on postprandial fat oxidation rates: monosaturated (olive oil) vs saturated fat (cream). *Int J Obes Relat Metab Disord*. 2002;26:814–821.
180. DeLany JP, Windhauser MM, Champagne CM, Bray GA. Differential oxidation of individual dietary fatty acids in humans. *Am J Clin Nutr*. 2000;72:905–911.

181. DeLany JP, Bray GA, Harsha DW, Volaufova J. Energy expenditure in African American and white boys and girls in a 2-y follow-up of the Baton Rouge Children's Study. *Am J Clin Nutr*. 2004;79:268–273.
182. Treuth MS, Butte NF, Sorkin JD. Predictors of body fat gain in nonobese girls with a familial predisposition to obesity. *Am J Clin Nutr*. 2003;78:1212–1218.
183. Griffiths M, Payne PR. Energy expenditure in small children of obese and non-obese parents. *Nature*. 1976;260:698–700.
184. Roberts SB, Savage J, Coward WA, et al. Energy expenditure and energy intake in infants born to lean and overweight mothers. *N Engl J Med*. 1988;318:461–466.
185. Goran MI, Carpenter WH, McGloin A, et al. Energy expenditure in children of lean and obese parents. *Am J Physiol*. 1995;268:E917–E924.
186. Goran MI, Kaskoun M, Johnson R. Determinants of resting energy expenditure in young children. J *Pediatr*. 1994;125:362–367.
187. Davies PS, Wells JC, Fieldhouse C, et al. Parental body composition and infant energy expenditure. *Am J Clin Nutr*. 1995;61:1026–1029.
188. Treuth MS, Butte NF, Wong W. Effects of familial predisposition to obesity on energy expenditure in multiethnic prepubertal girls. *Am J Clin Nutr*. 2000;71:893–900.
189. DeLany JP. Role of energy expenditure in the development of pediatric obesity. *Am J Clin Nutr*. 1998;68:950S–955S.

Evaluation and Management of Childhood Obesity

37

Alison G. Hoppin, MD and Janey S. Pratt, MD

INTRODUCTION

At the beginning of the twenty-first century, poverty and under-nutrition have worsened in some countries, while over-nutrition and obesity have reached epidemic levels in many others.[1] Currently, obesity is the most important nutritional problem globally, and the associated medical problems account for substantial morbidity, mortality, and healthcare costs in developed countries. In developing countries undergoing rapid changes in economic and technological development, the population is exposed to rapid changes in diet and energy balance; this "nutrition transition" presents unique health challenges.[2] Understanding the mechanisms causing obesity and associated medical problems will be essential to reverse the global trend toward obesity and its threats to public health.

EPIDEMIOLOGY

Relevance and Significance

In 2012, about 15% of children and adolescents in the United States were overweight, and 18% were obese.[3] Hispanic, Black, and American-Indian children are disproportionately affected. The prevalence of obesity in US children and adolescents tripled between the 1960s and 2000, but subsequently plateaued. Meanwhile, obesity rates continue to rise in some subpopulations (e.g., Black and American-Indian girls in California), while

declining in others (e.g., children in New York City). Similar but more gradual increases in obesity were seen in most resource-rich countries worldwide during the last part of the twentieth century,[1] and in a few of these populations obesity rates are beginning to plateau or decrease slightly.[4] The highest rates of childhood obesity are in North and South America and much of Europe, with somewhat lower rates in Nordic countries, Russia and Eastern Europe.

Determining the specific causes of this rapid increase in rates of obesity is essential, yet remarkably complex. Both genetic and environmental factors have been shown to contribute significantly to this problem. In general, genetic factors explain a large part of the variation of body weight within a given population in a common environment, while environmental factors tend to explain changes in obesity over time in that population. The study of Pima Indians provides an important example of the interaction between environmental and genetic factors.[5] The Pima Indians who live in the southwestern United States are strongly predisposed to obesity and diabetes, and these traits assort in patterns indicating genetic inheritance. The genetically similar Pima living as subsistence farmers in Mexico have substantially lower rates of obesity and diabetes. Genetic factors clearly explain a large part of the obesity among Pima Indians in this country, while environmental factors likely explain the dramatic difference in rates of obesity between the two Pima populations.

Epidemiologists have used cohort studies and case-control designs to determine which environmental factors may contribute to obesity. Such studies have pointed to dietary trends, sedentary lifestyle, decreases in structured physical activity, psychosocial stressors, and cultural trends as likely contributors to the obesity epidemic.[1,2] A number of dietary factors have been proposed to play important roles. These include marked increases in the proportion of foods consumed from restaurants and fast food outlets, increasing portion size served at restaurants, increased consumption of sugar-sweetened beverages, consumption of energy-dense foods (i.e., lower water content, usually correlated with higher fat and carbohydrate content), and lower intake of fruits and vegetables.[6,7] Other factors include decreases in structured physical activity, particularly for children, increasing sedentary activities (particularly television viewing and computer use), and shorter and fragmented sleep. However, it is important to note that no single factor among these has been shown to play a pivotal role in the increasing prevalence of obesity.

Tracking

"Tracking" describes the risk for a disease state persisting over time. In the case of obesity, there is a moderate risk of childhood obesity persisting into adulthood, and that risk increases if the child remains overweight as he or she grows older (Figure 37-1). As an example, in one population with a high frequency of obesity, there was substantial tracking within weight categories from late childhood (age 9–11) to young adulthood (19–35 years).[8] Tracking varied by gender and ethnicity: 73% of European-American women tracked within their weight category, while 49% of European-American men tracked. Among African-American participants, 60% of both men and women remained in the same weight category between childhood and adulthood.

A child's risk for having obesity in adulthood also depends on the weight status of his parents. An obese preschool-aged child has a 33% risk of becoming an obese adult if neither of his or her parents are obese, but an 83% risk if either parent is obese.[9] Before three years of age, the primary predictor of obesity in adulthood was the parents' obesity status, and the child's obesity status was not an important indicator of the risk of adult obesity. By contrast, after seven years of age, the child's own obesity status became the more important predictor of his or her risk for obesity in adulthood.

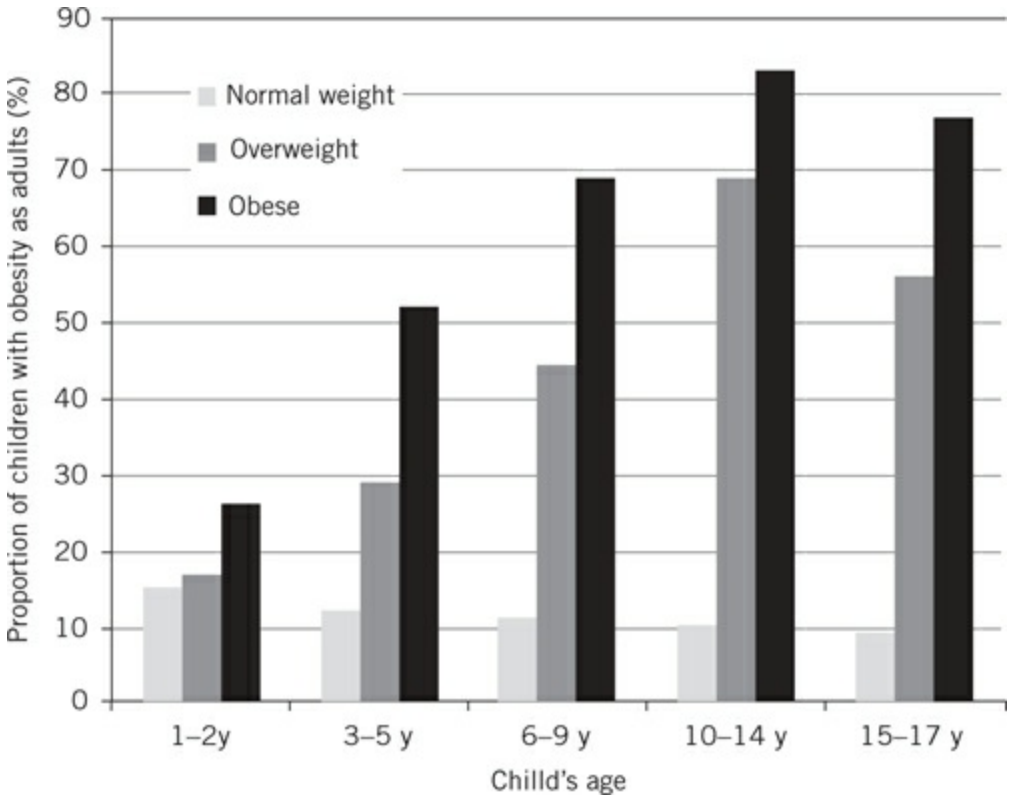


Figure 37-1 Tracking of obesity from childhood to adulthood. Proportion of children who will become obese as adults, as predicted by childhood weight status.[9]

Obesity in childhood thus confers a higher risk of obesity in adulthood. Moreover, adults who were obese in childhood have a greater risk of morbidity and mortality, independent of their BMI in adulthood, family history of cardiovascular diseases or cancer, and smoking.[10,11] In longitudinal studies, overweight adolescents were shown to have an increased risk for developing obesity-related medical problems in adulthood, including cardiovascular disease, diabetes, and breast cancer, as compared to adults with more recent onset of obesity.[12,13]

PATHOPHYSIOLOGY OF OBESITY

Heritability

Studies of twins and adoptees provide useful estimates of the role of heritable factors in determining an individual's body weight.[14] Adoption studies tend to generate the lowest heritability estimates (30%), whereas

twin studies provide highest heritability estimates (70%). The variability in estimates of heritability depends in part on definitions of obesity used in each study: more severe obesity tends to have a greater heritability factor than lesser variations in BMI.

Observations of a genetic contribution to obesity gave rise to the "thrifty genotype hypothesis,"[15] which posits that genes predisposing to energy conservation were preserved as a survival characteristic in times of famine, but become a liability in environments with plentiful food and low required physical activity. Possible mechanisms through which genetic polymorphisms can translate to differences in body weight regulation are discussed below.

Metabolic Programming

Several decades ago, David Barker and colleagues showed that poor fetal growth, resulting in *low* birthweight, is associated with an increased risk for type 2 diabetes and other elements of the metabolic syndrome. They proposed that poor nutrition during gestation imprints permanent changes in glucose and insulin metabolism.[16] Interestingly, many more recent studies have also shown clear associations between *high* birthweight and several components of the metabolic syndrome,[17] suggesting that newborns at both ends of the weight spectrum are at risk for obesity-associated disease. This concept of "metabolic programming" has now been supported by numerous studies in other human populations and in animals.

The metabolic programming hypothesis has been extended to include the possibility that postgestational influences can participate to create a lifelong metabolic phenotype.[18] For example, a study of adults in Finland showed that the development of type 2 diabetes mellitus was associated with the combination of low birthweight followed by accelerated gain in height and weight during childhood and with high maternal BMI.[19] Other studies have demonstrated an association between childhood weight gain and cardiovascular disease markers in adulthood.[20] Indeed, populations in transition from conditions of low-to high-nutrition may be at the greatest risk for such obesity-related complications because of the combination of fetal undernutrition and childhood overnutrition.[2]

A few studies strongly support the idea that the intrauterine environment affects obesity risk independently from genetic factors, thus distinguishing the concept of a "thrifty phenotype" from that of the "thrifty genotype." In a study of the effects of wartime famine in the Netherlands,

infants exposed to famine in utero had higher rates of obesity and diabetes in adulthood as compared to a genetically similar cohort not exposed to famine, and this effect was largely independent of birthweight.[21] Furthermore, the timing of the famine exposure during gestation appeared to have important effects, as fetuses exposed to famine during the first trimester of gestation were more severely affected than those exposed later in gestation. These observations are supported by animal studies, which also demonstrate lasting effects of intrauterine and postpartum nutrition. Other studies have shown that children of mothers who had undergone gastric bypass surgery prior to pregnancy have reduced risk of obesity as compared to the offspring of mothers who had not undergone the surgery.[22]

The mechanism underlying the observed associations, including the contributions of maternal hyperglycemia, insulinemia, and postnatal growth to the development of later complications, is an important subject for future studies. See Chapter 26, “Human Growth and Disease in Later Life,” for further discussion of the influence of the prenatal environment on obesity and other nutritional diseases.

Genetics

Mature animals and adult humans have a strong tendency to maintain a stable body weight over time, due to a close but sometimes imperfect matching of energy intake with energy expenditure. Animal studies in which energy intake is manipulated reveal powerful homeostatic mechanisms defending body weight. Similarly, the poor long-term results of weight reduction therapies in humans (about 95% of adults regain all weight after dieting), suggest there are mechanisms that defend a highly individualized “set point” for body weight. When an individual has a heritable or acquired susceptibility to positive energy balance, superimposed on these native homeostatic mechanisms, he or she has a tendency to become obese.

Currently, identifying specific genetic causes of obesity is rarely possible or useful during the course of clinical care, with the exception of the small minority of patients with syndromic obesity in whom understanding of the syndrome may inform clinical management. For example, identifying individuals with leptin deficiency allows for a specific and effective therapy, but this defect is extraordinarily rare (fewer than 10 individuals identified to date), so general screening for this disorder is not necessary or appropriate. These and other genetic causes of obesity are detailed below.

Nonetheless, a basic understanding of the genetic contributors to obesity is helpful in guiding the clinical evaluation of a patient. Furthermore, ongoing research in this field is rapidly expanding knowledge about the mechanisms underlying weight regulation and obesity. It is likely that some of these mechanisms will lead to better-targeted interventions, so that in the future, pharmacologic and/or behavioral interventions may be guided by an individual patient's genetic profile.

Common Obesity

The predisposition to obesity is a heritable trait, as outlined above. However, the specific genetic cause or causes of obesity cannot be determined for the vast majority of individuals with obesity. Genome-wide association studies have identified a variety of loci that contribute to obesity risk, including the fat mass and obesity-associated gene *(FTO)*, which is most strongly associated with obesity, conferring about 1.7-fold increased risk. Other loci are near genes that encode neuronal regulators of appetite or energy balance, including melanocortin 4 receptor *(MC4R)*, brain-derived neurotrophic factor *(BDNF)*, pro-opiomelanocortin *(POMC)*, and transmembrane protein 18 *(TMEM18)*.[23] Each of these loci contributes only a small increased risk of obesity. Thus, the genetic predisposition to common obesity is likely to be determined by a combination of multiple risk alleles acting in concert, including some that have yet to be discovered.

Monogenic Causes of Obesity

Genetic studies on individuals with very severe or early-onset obesity have identified specific mutations in genes known to be involved in the regulation of body weight. Mutations in MC4R are the most common of these; heterozygous mutations are present in 4% to 6% of individuals with early-onset obesity.[14] Individuals with MC4R mutations tend to have increased linear growth, hyperinsulinemia and normal cognition; hypertension is rare. A few individuals have been identified with homozygous or compound heterozygous MC4R mutations, and they have a more severe phenotype. Other genes with mutations that are strongly associated with obesity include *POMC*, leptin *(LEP)*, leptin receptor *(LEPR)* and proprotein convertase 1 *(PCSK1*, also known as prohormone convertase 1). These mutations are extremely rare, each identified in fewer than 20 individuals worldwide, most from consanguineous families (Table

37-1).

TABLE 37-1 Selected Monogenic Causes of Obesity

Gene	Characteristics
Melanocortin 4 receptor (MC4R)	4%–6% of individuals with early-onset obesity are heterozygous for an MC4R mutation. Hyperinsulinemia, hyperphagia, increased linear growth. Low prevalence of hypertension.
Leptin (LEP) or leptin receptor (LEPR)	Fewer than 20 cases of each reported, usually associated with consanguinity. Severe early-onset obesity, hyperphagia, hypogonadism.
Pro-opiomelanocortin (POMC)	Fewer than 10 cases reported. ACTH deficiency with adrenal crisis, early-onset obesity, red hair.
Proprotein convertase 1 (PCSK1)	Fewer than 10 cases reported. Low plasma insulin but high proinsulin; hypogonadotropic hypogonadism, diarrhea.

Obesity Syndromes

Approximately 30 human genetic syndromes whose phenotype includes obesity have been identified and cataloged in the Online Mendelian Inheritance in Man (OMIM) database, including Albright, Alström, Bardet-Biedl, Beckwith-Wiedemann, Carpenter, Cohen and Prader-Willi syndromes. Many of these syndromes have been mapped to one or more chromosomal locations.

The most common syndrome with severe obesity is Prader-Willi syndrome characterized by infantile hypotonia, hyperphagia, hypogonadotropic hypogonadism, and cognitive deficits. This disorder is caused by loss of paternally expressed genes in chromosome 15q11-13, a region containing several candidate genes for which mechanistic explanations for the Prader-Willi phenotype(s) are being sought.[24] The syndrome occurs in approximately 1 in 25,000 live births. Because the defect almost always arises sporadically, recurrence within a family is extremely rare. Interestingly, individuals with Prader-Willi syndrome also have elevated levels of ghrelin, providing a potential mechanism for their marked hyperphagia.

The Bardet-Biedl syndrome requires a mutation at one of 15 loci[14] and includes polydactyly and retinopathy. It is rare, occurring in less than 1 in 100,000 live births. Molecular studies suggest that the syndrome may be caused by loss of function of ciliated hypothalamic neurons. Alström syndrome (retinopathy, deafness, often with cardiomyopathy and renal

failure), and Cohen syndrome (hypotonia, retinopathy, and cognitive deficits) have been mapped to one location each, and candidate genes have been identified but the mechanism through which mutations in these genes causes obesity is still unknown. Each of these syndromes has characteristic findings as outlined in Table 37-2 and in the OMIM database (www.omim.org). These syndromes usually can be distinguished from common obesity by a thorough medical history and physical examination.

EVALUATION

Definitions of Obesity

Any definition of obesity is useful only if it predicts medical disability or complications. Since most medical complications of obesity are associated with excess body fat and not muscle mass, measures of obesity represent an attempt to estimate the adipose compartment. At present, there is no precise clinically practical method to measure body fat, so most metrics use measurements of body weight as a surrogate for adiposity. Such approaches are imperfect, as they may misclassify a patient with an unusual proportion of fat to lean body mass, but they are inexpensive and practical for use in the clinical setting and in epidemiological studies.

Current definitions of overweight and obesity rely on calculations of the body mass index (BMI), defined as the weight in kilograms divided by the height in meters squared (kg/m^2). Although BMI does not directly measure body fat, it has been established as a useful predictor of adiposity in adults, adolescents, and children. Like other estimates of adiposity, BMI is a moderately good predictor of risks for present or future medical complications of obesity.[25] BMI in children is not only correlated with other predictors of body fat but also with blood pressure, lipid levels, fatty liver disease, and markers of insulin resistance.

TABLE 37-2 Selected Genetic Syndromes With an Obesity Phenotype

Syndrome	Characteristics
Albright hereditary osteodystrophy (pseudohypoparathyroidism type 1a) (MIM #103580)	Short stature, skeletal defects, subcutaneous ossifications, impaired sense of smell, variable intellectual disability.
Alström (MIM #203800)	Retinopathy (causing early nystagmus and progressive blindness), neurosensory deafness, truncal obesity with insulin resistance. Often

	complicated by cardiomyopathy, renal or hepatic failure and pulmonary fibrosis.
Bardet-Biedl (MIM #209900)	Retinopathy, polydactyly or syndactyly, mild intellectual disability, hypogonadism, structural or functional renal disorders.
Beckwith-Wiedemann (MIM #130650)	Features are variable, but often include macrosomia +/-hemihypertrophy, macroglossia, ear creases, omphalocele or umbilical hernia, neonatal hyperinsulinemia and hypoglycemia; risk of embryonal tumors.
Cohen (MIM #216550)	Prominent incisors, microcephaly, moderate–to-severe intellectual disability.
Prader-Willi (MIM #176270)	Infantile hypotonia and feeding problems; intellectual disability, short stature, hypogonadism. Obesity usually develops in early childhood.

In adults, standard ranges of BMI can be used to define risk categories for obesity. However, during childhood and adolescence, BMI varies with age and gender. Therefore, BMI percentiles are used to define risk categories for children and adolescents, and BMI growth charts or standard deviation scores are used to track an individual against established standards for clinical purposes.

In 2000, the Center for Disease Control and National Center for Health Statistics (CDC/NCHS) established new growth charts using data from five large surveys representing the U.S. population.[26] These growth charts do not include data from the previous decade because of the sharp rise in BMI during that period. They are similar but not identical to those developed by the International Obesity Task Force (IOTF).[27] For the IOTF charts, data were drawn from six different countries, and the curves were smoothed to meet the adult cutoff points for overweight and obesity at 18 years of age. Both reference standards are strong predictors of obesity and coronary heart disease risk factors in adulthood.[28] In April 2006, the World Health Organization (WHO) released new growth curves for exclusively breastfed infants and young children. The WHO BMI curves cover a wider age range (from infancy through age 5 years), and provide different norms for children than the CDC/NCHS standards. On the WHO curves, the normal range is generally defined as between two standard deviations (SDs) below and above the mean, which corresponds approximately to the 2nd and 98th percentiles. The CDC now recommends use of the WHO curves for infants and children up to two years of age, and the CDC/NCHS curves for children and adolescents 2 years and older.[29] See Chapter 2, “Clinical Assessment of Nutritional Status,” for further

discussion of the derivation and applicability of these WHO standards.

BMI also varies with pubertal stage, reflecting changes in fat-free as compared to fat mass. In addition, there is some evidence that BMI varies with ethnicity. Therefore, research studies that aim for the most precise estimates of obesity risk using BMI should consider not only age and gender, but also pubertal stage and ethnicity. However, for clinical purposes, the WHO/CDC/NCHS or IOTF growth references are adequate for determining obesity-related risk groups.

Body Mass Index

The initial step for the assessment of the overweight child is to calculate the BMI from measured weight and height, then plot the result on standard BMI growth curves to determine the child's weight category.

By general consensus, weight categories in children between 2 and 18 years of age are defined by the following BMI ranges, for age and sex[27,30]:

- Underweight: BMI < 5th percentile
- Normal weight: BMI between the 5th and 85th percentiles
- Overweight: BMI between the 85th and 95th, or BMI ≥ 25 (whichever is lower)
- Obese: BMI ≥ 95th percentile, or BMI ≥ 30 (whichever is lower)
- Severe obesity: BMI ≥ 99th percentile.[12,30] An alternate definition of severe obesity is a BMI ≥ 120% of the 95th percentile values, or a BMI ≥ 35 (whichever is lower).[31]

Additional important information is derived from plotting serial values of BMI, to assess the child's growth trend. A child whose BMI has risen across two major percentile curves is at risk for becoming overweight or obese even if the current BMI does not yet fall into one of these categories. Similarly, a child whose BMI is in the obesity range but with a flat or decreasing trend is displaying an improvement in weight status relative to his or her peers.

Finally, it is valuable to determine the weight status of the child's biologic parents by documenting their measured or reported height and weight, and calculating their BMIs. This is because the child's long-term risk of obesity is strongly affected by the weight status of his or her parents. Assessment of the parents' weight status by visual impression is frequently inaccurate, even by experienced observers.

Medical Assessment

The two main goals of the medical evaluation of the overweight child is to (1) assess for specific causes of obesity, including medication-related weight gain and potential obesity-associated syndromes or endocrinopathies and (2) to diagnose possible comorbidities associated with obesity. The family history should assess obesity and related medical problems in first-and second-degree relatives, to help establish the genetic factors underlying the weight disorder, and the potential future medical risks.

Evaluating for Causes of Obesity

Syndromic obesity should be considered, but in most cases, these syndromes can be distinguished on the basis of their unique features (listed in Table 37-2 and described in more detail in the genetics section above).[14] Specific laboratory testing is valuable for confirmation of the diagnosis in children with suspicious clinical features of a syndrome, but is not appropriate for general screening of unselected patients. For Prader-Willi syndrome (PWS), a cognitive deficit and history of hypotonia during infancy are particularly strong predictors of the syndrome. The presence of these characteristics in a child with severe early-onset obesity is an appropriate indication for performing genetic testing for PWS, which is highly sensitive and specific.[24]

Endocrine causes of obesity are rare, and usually can be excluded by the clinical history and physical examination. Hypothyroidism tends to cause a delay in linear growth, and its contribution to weight gain is typically modest. Therefore, hypothyroidism is unlikely in a child with good linear growth and gradual onset of obesity. Similarly, Cushing's disease causes a marked delay in linear growth. It can cause substantial weight gain, usually abrupt in onset, as well as central fat distribution and a suprascapular fat pad. If Cushing's disease is suspected because of these clinical features, the most appropriate screening test is a 24-hour urine collection for free cortisol.

The medical history should include assessment of concomitant medical diseases for their potential to contribute to weight gain, such as a history of a brain tumor or significant head trauma that might cause hypothalamic dysfunction. A variety of drugs used for the treatment of psychiatric disease, epilepsy, diabetes, and migraines are associated with weight gain. The atypical antipsychotic drugs and mood stabilizers are particularly likely to promote weight gain. However, the weight-promoting effects of other commonly used classes of drugs, including tricyclic antidepressants, selective serotonin reuptake inhibitors, anticonvulsants, and antidiabetic

agents are less widely recognized. Identifying these as a likely trigger of weight gain may prompt consideration of alternate drugs (Table 37-3). There is limited empiric information about the weight-related effects of many psychoactive drugs, and their long-term effects on weight are particularly unclear. Most of these drugs should be considered potential contributors to weight gain even if they are said to be "relatively" weight neutral.

TABLE 37-3 Drugs that Promote Weight Gain

Atypical antipsychotics

Clozapine ++++
Olanzapine ++++
Risperidone ++

Mood stabilizers
Divalproex sodium +++
Lithium +++

Tricyclic antidepressants
Amitriptyline ++
Imipramine ++

Selective serotonin reuptake inhibitors (SSRIs)
Paroxetine ++
Sertraline +

Anticonvulsants
Valproate ++
Carbamazepine +
Gabapentin +

Antidiabetic agents
Insulin ++
Thiazolidinediones (rosiglitazone) +

Screening for Comorbidities

The pathophysiology of medical conditions related to obesity (Table 37-4) is discussed in Chapter 38, "The Metabolic Syndrome in Children and Adolescents," but the laboratory assessment of these conditions is discussed below.[30] Most guidelines recommend screening for dyslipidemia, steatohepatitis, and evidence of glucose intolerance, particularly as specific treatments for some of these disorders are increasingly considered.

Blood testing should be done in a fasting state if practical. Total cholesterol, very low-density lipoprotein, low-density lipoprotein, high-density lipoprotein, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) have been recommended to screen for possible

hypothyroidism, dyslipidemia, and steatohepatitis, respectively (Table 37-5).

The American Diabetes Association recommends screening for type 2 diabetes in children 10 years or older who are overweight or obese and have two additional risk factors (including clinical evidence of insulin resistance, a family history of diabetes, or member of a diabetes-prone ethnic minority).[32] Fasting or random glucose levels, or hemoglobin A1C (A1C) are used to screen for diabetes mellitus or increased diabetes risk (sometimes called "prediabetes") (Table 37-6).[30,32] A1C is particularly useful for screening because it does not require fasting. However, the accuracy of A1C as a screening test is not well established in adolescents. All of these screening tests lack both sensitivity and specificity for diagnosing type 2 diabetes or impaired glucose tolerance in children, so oral glucose tolerance testing should be used when possible in high-risk individuals. Fasting insulin levels are measured by some clinicians to provide information on carbohydrate metabolism and insulin resistance, but this test is not reliable for predicting risk for diabetes.

TABLE 37-4 Comorbidities of Obesity in Children

Endocrine
Type 2 diabetes
Polycystic ovary syndrome (PCOS)

Cardiovascular
Hypertension
Dyslipidemia
Ventricular hypertrophy, early atherosclerotic disease

Respiratory
Asthma
Obstructive sleep apnea

Gastrointestinal
Gastroesophageal reflux disease (GERD)
Fatty liver disease
Constipation and encopresis

Orthopedic
Slipped capital femoral epiphysis (SCFE)
Blount's disease

Neurologic
Pseudotumor cerebri

Psychologic
Depression, anxiety, social exclusion

Further laboratory testing may be useful in selected cases but can be

expensive and some tests are not readily available. Sleep studies can and should be performed if there are clinical symptoms of sleep apnea, including consistent snoring and/or nocturnal enuresis, especially in a child with severe obesity. Radiographic evaluation is necessary when slipped capital femoral epiphysis (SCFE) or Blount disease is suspected. SCFE usually presents with hip and/or knee pain, and Blount's disease with visible bowing of the knees and/or pain. Indirect calorimetry can be used to predict the energy deficit necessary for weight loss. This is not necessary for routine dietary counseling, but might be useful when poor compliance or abnormally low energy requirements are suspected and at times may be useful to provide concrete caloric goals to support dietary changes. Consultations with specialists in sleep disorders, neurology, otorhinolaryngology, pulmonology/allergy, endocrinology, genetics, ophthalmology, and surgery may be necessary to manage specific complications.

Nutritional Assessment

Nutritional evaluation should include a history of the onset of obesity, as well as of weight loss attempts. Assessing dietary intake by recalled food frequency does not provide a good estimate of energy intake, as underreporting of food intake by obese subjects is well described. However, it helps to evaluate diet composition, which may help identify targets for dietary change. In addition, the diet recall may be useful to assess the diet for deficiencies in micronutrients, particularly calcium and vitamin D, as well as vitamins A, E, β-carotene, folic acid, and other B vitamins; excessive energy intake does not always mean adequate intake of micronutrients.

TABLE 37-5 Laboratory Evaluation for Comorbidities of Obesity in Children and Adolescents

Laboratory Test	Indications
Lipid profile	All children with overweight or obesity (>10-year-old, repeat every two years)
Fasting glucose and/or Hemoglobin A1C	All children with overweight or obesity and two risk factors* (>10-year-old, repeat every two years)
Alanine aminotransferase (ALT) Aspartate aminotransferase (AST)	Generally recommended in children with obesity
Blood pressure (BP)	All children with overweight or obesity

Use an adequate sized cuff Interpret using age-based blood pressure norms	Prehypertension: BP 90–95th percentile or ≥120/80 mmHg Hypertension: BP ≥ 95th percentile or ≥140/90 mmHg
Sleep study (polysomnogram)	Moderate-severe obesity and any symptoms suggestive of sleep apnea (habitual snoring, mouth breathing, nocturnal enuresis)
Testosterone and Sex hormone binding globulin (SHBG) OR free testosterone	Girls with abnormal menses or hirsutism
Glucose tolerance test	If fasting glucose > 100, or Hemoglobin A1C > 5.7, or symptoms of diabetes
Depression screen	All children with obesity

* The American Diabetes Association recommends diabetes screening in children overweight children with two additional risk factors (including clinical evidence of insulin resistance, a family history of diabetes, or member of a diabetes-prone ethnic minority).

TABLE 37-6 Laboratory Assessment for Diabetes Mellitus

Test	Increased Risk for Diabetes (Prediabetes)	Diabetes Mellitus
Fasting plasma glucose	100–124 mg/dL (5.6–6.9 mmol/L)	≥ 125 mg/dL (7.0 mmol/L)
Random plasma glucose		≥ 200 mg/dL (11.1 mmol/L) with symptoms
Hemoglobin A1c	5.7%–6.4%	≥ 6.5%
Oral glucose tolerance test: Plasma glucose 2 hours after 75 g glucose load	140–199 mg/dL (7.8–11 mmol/L)	≥ 200 mg/dL (11.1 mmol/L)

To identify possible areas for intervention, the amount and type of dairy products, fruits, vegetables, and legumes should be assessed, with particular attention to the consumption of sugar-sweetened beverages including juices and soda. Excessive consumption of sugar-sweetened beverages or juice has been associated with excess weight gain and increased rate of obesity, and is an important early target for dietary change.[7,30,33] Similarly, the frequency of use of fast-food restaurants and of other restaurant meals should be examined, because these meals often contain excessive caloric content and set inappropriate standards for

portion size. The diet history should also explore usual meal patterns, with attention to whether there is regular skipping of meals (especially breakfast), binge pattern of eating, and the social context of meals, particularly if there are routine family dinners. Each of these factors may become a target for intervention to establish regular meal-based eating patterns.

Finally, the potential role of exercise and sedentary behaviors should be evaluated, with particular attention to determining the frequency of television and computer use. Several studies have suggested causal effects of television viewing on obesity, and the American Academy of Pediatrics recommends limiting television viewing to one to two hours per day.[30] Assessment and encouragement of moderate to vigorous physical activity (e.g., participation in sports programs) is essential. It is also worthwhile to inquiring about and encourage lifestyle exercise, in the form of outdoor play or regular walking (e.g., walking to school); this is often the main type of physical activity for young children or older children who are deconditioned.

Behavioral Assessment

The psychosocial complications of obesity are often subjective and difficult to measure in a standard fashion, but undoubtedly represent one of the greatest burdens of obesity in children and adolescents. In many cases, psychosocial issues are best understood as consequences of the disease, brought on by feelings of discouragement and criticism by family, peers, or self. In other instances, the psychological issues precede or exacerbate the obesity.[30] Regardless of the causality, it is important to acknowledge and assess these behavioral and psychological issues in each individual to best target treatment.

Depressive symptoms, anxiety, binge-eating disorder, decreased self-esteem, and problems with social interactions have generally been found more frequently in obese children and adolescents than in their lean peers. Negative psychosocial associations also persist into early adulthood, when obesity is associated with lower educational attainment and household income and lower rates of marriage, independent of baseline education and aptitude. Many of these problems can be attributed to the widespread and culturally entrenched bias against obese individuals in our society.[7,30]

Optimal assessment of the psychosocial issues contributing to and stemming from obesity in a particular patient has not been established, and varies greatly between centers and providers. Whether or not standardized

instruments are used, some effort should be made to assess mood, motivation, school and social performance, self-image, and eating attitudes and behaviors. It is also important to assess these issues in parents or close caretakers of the patient, because factors such as motivation, mood disorders, and eating disorders in a parent will have substantial impact on the child's attitudes, behaviors, and ability to respond to treatment. The family's financial resources and level of cognitive stimulation in the household also predict the development of obesity, and may also affect the family's ability to respond to treatment. Assessing the readiness of the patient and family for making lifestyle changes is an important foundation for patient-centered motivational counseling.

TREATMENT

Nutritional Therapy

Scientific study of interventions to prevent and treat obesity is challenging because of the many factors that influence weight gain, which often cannot be controlled or measured, and difficulty in standardizing interventions. Moreover, short-term interventions are of questionable relevance for this chronic disease, yet prolonged interventions are impractical and costly.

Nonetheless, the preponderance of evidence suggests that lifestyle interventions can have modest effects on weight status.[34] In general, the more effective interventions were multimodal (combining diet, physical activity, and/or behavioral modification), included the family (especially for younger children), and had a moderate or highly intensive schedule (at least 25 contact-hours). Moreover, younger children seem to be more likely to respond as compared with older children. The majority of studies do not provide long-term outcomes, and many issues such as optimal macronutrient composition of diets and strategies to change food preference have not been adequately studied.

As rates of obesity rise, obese children represent an increasing proportion of hospital inpatients. Whether or not the child is hospitalized for an obesity-related condition such as gallstones, diabetes, sleep apnea, or orthopedic problems, his or her nutritional needs must be considered. It is particularly important to recognize that the obesity is a chronic problem, and will not resolve by attempting weight loss acutely during the inpatient stay. Moreover, acute severe caloric restriction is inappropriate and can lead to metabolic problems including refeeding syndrome, even in children with adequate or excessive energy stores. Guidelines for nutritional care of

the obese adult inpatients have been developed,[35] but this issue has not been well examined in children. It may be appropriate to prescribe a modest reduction in caloric intake for obese inpatients, guided by indirect calorimetry or by calculations based on adjusted body weights, and using a stress factor that is appropriate to the child's condition. The inpatient stay may also present a valuable opportunity for nutritional education, and for engaging the patient and family in a therapeutic plan to address the obesity beyond the hospital stay.

Weight Goals

The goal of an intervention is to reach and maintain a healthy weight without affecting linear growth. An appropriate goal for children younger than 5 years of age who are overweight or obese is weight maintenance, which will lead to gradual reduction in BMI percentiles over time.[30] In this age category, only severely obese children (e.g., BMI ≥ 99th percentile) should target actual weight loss (up to 1 lb per month). For children 6 to 11 years of age who are obese, it is appropriate to target gradual weight loss (up to 1 lb per month for BMI 95–99th percentile, and up to 2 lb per week for BMI ≥ 99th percentile). For adolescents 12 to 18 years who are overweight, it is reasonable to target weight maintenance or gradual weight loss; for those who are obese or severely obese, weight loss of up to 2 lb per week is reasonable.

These weight targets are based on expert opinion, and may not be realistic for many patients. Some patients may respond better to more modest and achievable weight goals, which can be readjusted as the patient progresses.

Lifestyle Goals

Dietary recommendations for adults and children with obesity typically target a reduction in dietary fat and energy intake as a balanced, hypocaloric diet (characterized by less energy than required to maintain weight but not less than 1200 kcal [5040 kJ]/d).[33] Counseling for children with obesity generally focuses on diet quality and quantity, rather than specific caloric goals. Nonetheless, a general estimate of energy requirements and caloric goals may be helpful as an initial step in counseling. Estimated energy requirements (EER) for normal growth in healthy children without obesity and low activity are approximately 1000 kcal/d for children 1 to 3 years and increase to about 1500 kcals by 8 years of age (adding up to 300 kcal/d if the child has a high activity level). EER

for adolescents with low activity levels are approximately 2000 kcal/d for girls, and 2500–2800 kcal/d for boys (adding up to 600 kcal/d if the adolescent has a high activity level). Children with obesity will probably have energy requirements somewhat higher than these estimates, because their basal metabolic rate increases with body weight. However, use of the age-based EER is sufficient to provide a general estimate of the child's caloric needs.

For weight loss in obese adolescents, it is reasonable to target a caloric goal approximately 500 kcals less than the EER, but not less than 1200 kcal/d. For younger children with obesity, it is reasonable to target the EER itself. However, any caloric goal is approximate, and dietary counseling should be guided by monitoring the change in weight and BMI over time. Energy requirements in children with obesity are discussed in detail in Chapter 36.

Specific dietary goals include limiting beverages and foods with high caloric density and low nutritional value, including sugar-sweetened beverages, baked goods, and fried foods, while encouraging whole grains, fruits and vegetables.[30,33] Simple behavioral measures such as meal planning and label reading during grocery shopping are also generally encouraged to support the implementation of these nutritional guidelines. Strong evidence indicates that limiting television viewing and other recreational screen time helps to prevent or treat obesity. The mechanism has not been established, but may include reductions in caloric intake. Eating breakfast and having meals as a family are generally recommended, based on moderate-quality evidence that these behaviors are inversely associated with obesity. Moderate quality evidence indicates that physical activity helps to prevent or treat obesity if sufficient intensity and frequency is achieved; guidelines suggest promoting a variety of options for moderate-to-vigorous physical activity, including team/group, individual, lifetime and recreational forms. Ensuring adequate sleep time for children also is recommended because of associations between short or fragmented sleep and obesity, although a causal effect has not been established. Breastfeeding is encouraged because it may have a protective effect on the subsequent development of obesity, although the effect is now thought to be weak. Despite the uncertainty about the protective effects of breastfeeding and adequate sleep, these behaviors are recommended because of their other established benefits and low risk of harm.[7]

These general goals have been captured in patient-focused messaging, which typically includes the following elements[36]:

5—Five servings of fruits and vegetables daily
2—Less than two hours of recreational screen time daily
1—At least one hour of moderate-to-vigorous physical activity daily
0—Zero servings of sugar-sweetened beverages daily

Breastfeeding, ensuring adequate sleep, and monitoring BMI are sometimes added to the messaging scheme.

Specific Diets

The "traffic-light diet" is an example of a structured, balanced, hypocaloric diet in a simple format, and has been used effectively for preadolescent and preschool children, in combination with a variety of behavioral interventions.[33] It uses a simple color-coding scheme to categorize foods into categories for free consumption (low-density foods; "green"), moderate consumption (moderate-density and protein-containing foods; "yellow"), and very limited consumption (foods with high caloric density, and/or with high sugar or fat content; "red"). The prescribed caloric content of the diet is generally between 900 and 1300 kcal/d (3780–5040 kJ/d).

The Protein-Sparing Modified Fast diet (PSMF) provides high-quality lean protein while strictly limiting total calories. It has been used to treat severe obesity in a variety of settings including hospitalized patients and school-based interventions. This diet has been effectively used in settings where short-term weight loss is medically necessary, but there are no data to suggest that it reliably improves obesity in the long term.[33] The diet typically consists of 600 to 800 kcal/d, including 2 g/kg/d protein to a maximum of 100 g/d, with supplements to ensure recommended intake of vitamins and minerals for about 12 weeks. This is followed by a maintenance diet (balanced macronutrients, 1,200 kcal/d). A PSMF diet typically yields substantially more short-term weight loss than that achieved by less restrictive measures, but by 15 months follow-up, the weight loss achieved is similar to patients on a steady balanced hypocaloric diet. Potential complications of the PSMF include protein losses, hypokalemia, inadequate calcium intake, cholelithiasis, and intravascular volume depletion with orthostatic hypotension.

While the caloric content of a diet has been shown to relate to treatment success at one year, to date, there is little evidence to suggest that alterations in *specific macronutrients* yield long-term weight reduction.[7,33] A variety of popular diets have arisen around alterations of specific macronutrients, with or without limitations on total caloric intake.

Many of these show good short-term weight loss, but there is little evidence that they achieve long-term improvements in weight status in adult or pediatric populations, although individual patients may benefit. Several dietary approaches have focused on limiting carbohydrates, and in general, this approach has modest efficacy, which is similar to that achieved with low-fat diets at one year.[33,37] The most rigorously studied such diet emphasizes foods with a low glycemic index to reduce insulin release and excursions of blood sugar. A few studies have shown short-term decreases in weight, appetite, and some cardiovascular disease risk factors during a hypocaloric low-glycemic-index diet as compared with a hypocaloric low-fat diet in young adults.[38] However, other studies have not shown beneficial effects of targeting low-glycemic-index foods, and none have examined long-term outcomes of this type of diet.

The results of studies that focus on adult populations should be interpreted with caution, since there is compelling evidence that at least some “lifestyle” approaches to obesity are more effective in children than they are in adults.[39] Some dietary approaches might prove to have long-term results in children even if none can be demonstrated for adults.

Behavior Therapy

As discussed above, some interventions in childhood have demonstrated long-term improvement of obesity with 5 or 10 years follow-up, thus children appear to have a more consistent and durable response to therapy than adults in the same family, or adult populations in general.[34] The importance of including behavior therapy in the treatment of obesity in children has been demonstrated, at least in a family-based setting.[33] Although many interventions use combinations of dietary, exercise, and behavioral interventions, it is notable that the few studies showing long-term benefits have had a rigorous and structured behavioral component.

Commonly used behavioral techniques include self-monitoring of food intake and weight, modeling, positive reinforcement (praise or rewards), contingency management (certain behaviors are paired with predictable, reinforcing responses), and stimulus control (learning to avoid situations that are cues to overeat).[33,40] Some studies have tested specific elements of these behavioral techniques. These have shown superiority of family-focused over patient-focused treatment, of positive reinforcement over restrictive or critical approaches, and of frequent (daily) over less frequent (weekly) positive reinforcement. The value of problem-solving techniques, such as preplanning and relapse prevention, has not consistently been

shown. Thus, there is ample evidence to support the use of behavioral modification techniques in the treatment and possibly the prevention of obesity in children. By contrast, there is little data to support the use of behavior "micromanaging" techniques such as rate of eating and bite size.

Behavior therapy should be thought of as a tool to achieve long-term changes in diet and exercise. For example, the behavioral techniques typically are applied to specific lifestyle goals, such as reducing intake of sugar-sweetened beverages and increasing physical activity, as described in the previous section. Expert consensus suggests that reinforcement (praise or rewards) be applied to achievement of goals for healthy *behaviors*, rather than to weight goals.[30]

Success in behavior change is highly dependent on the motivational state of the patient and/or family. The technique of motivational interviewing has been applied with some success to encourage lifestyle changes for weight management.[7,40] This premise of this technique is that counseling will be most successful if it is tailored to the "stage of change" of the patient. If the patient or family has low interest in making lifestyle change, initial efforts should be focused on helping them examine their priorities and how their current behaviors support or undermine their priorities, rather than focusing on action.

Weight Loss Drugs

Many of the drugs used for the treatment of obesity in adults in the past were characterized by unproven claims, highly variable efficacy, or dangerous side effects. Nonetheless, the incomplete but growing understanding of the mechanisms underlying the homeostatic control of body weight and the increased rigor applied to clinical drug trials hold promise for the development and use of pharmacologic agents to treat this chronic disease. In the short term, drugs can be important for a patient whose medical condition requires acute weight loss. Drugs may also have a role in treating the chronic component of obesity, because the use of drugs to promote weight loss may keep an individual patient engaged in the "lifestyle" component of treatment. Thus, pharmacotherapy for obesity is not at odds with lifestyle-changing approaches. As increasingly specific drugs are developed, efficacy and safety improve, and pharmacotherapy may ultimately be an essential tool to treat an otherwise refractory and devastating disease. Whether pharmacologic treatment is cost-effective depends in large part on whether it prevents the medical complications of obesity and associated costs of medical care.

Current options for the pharmacologic treatment of obesity are quite

limited, and any pharmacologic intervention should be paired with a rigorous and simultaneous lifestyle intervention.

Orlistat (Xenical®) is the only drug approved by the US Food and Drug Administration (FDA) for treating obesity in adolescents (ages 12 to 16 years). It acts by inhibiting gastrointestinal lipases, reducing fat digestion and absorption by about 30%. Its utility is limited by very modest efficacy (BMI after treatment 0.9 kg/m^2 less than placebo), frequent dosing (three times daily), and gastrointestinal side effects (oily stools or abdominal pain in up to 50% of patients).[41] Weight regain typically occurs upon discontinuation of the medication. Concurrent efforts to reduce energy intake and increase exercise are important.

Metformin improves insulin sensitivity and also promotes modest weight loss in adults and adolescents.[41] By contrast to other diabetic agents, metformin does not increase insulin secretion, but decreases hepatic glucose production and improves insulin sensitivity in both diabetic and nondiabetic individuals. It has very modest weight-reducing effects, yielding up to 3 kg/m^2 more weight loss than placebo in adherent patients. There is some evidence that metformin may be useful in ameliorating psychotropic drug-induced weight gain; in a meta-analysis of trials in adolescents with antipsychotic drug-induced weight gain, metformin yielded a 4% reduction in body weight.[42] Metformin is approved by the FDA for treatment of type 2 diabetes mellitus and is a first-line agent for adolescents with this disease. Use of metformin for treatment of prediabetes or obesity is off-label; given its very modest weight-reducing effects, its use for these conditions is questionable.

Methylphenidate and similar stimulants used for attention-deficit disorder are having increased dopaminergic tone, which tends to blunt appetite. However, the effects of these drugs on appetite tend to be short-term, the effects on weight are inconsistent and poorly studied, and may vary by patient population.[41] For example, one study showed no effect of methylphenidate on appetite among obese adult males. Moreover, these drugs have multiple adverse side effects including agitation, insomnia, tachycardia, and hypertension. Therefore, they are not recommended or FDA-approved for weight loss, despite their utility for treating attention deficit disorder in children and adults.

Phentermine, diethylpropion, phendimetrazine, and benzphetamine are noradrenergic agents with appetite-suppressant effects that have been studied and approved only for adult patients and for short-term use (12 weeks or less).[41] These drugs are considered to have some potential for

abuse and are listed on schedule III or IV of the Drug Enforcement Agency. They are not recommended or FDA approved for weight loss in children and adolescents, because of their substantial side effects, including potential for abuse.

Topiramate is an anticonvulsant drug that enhances signaling of gamma-aminobutyric acid (GABA), an important inhibitory neurotransmitter of the central nervous system. In a randomized dose-ranging trial in adults with obesity, the average weight loss among patients treated with topiramate was 2.2 to 5.8 kg or 0.9 to 1.9 kg/m^2 (among study completers).[43] The proportion of subjects losing 10% or more of their body weight was 16% to 29% among those treated with topiramate, versus 7% among those treated with placebo. Thus, topiramate appears to have modest but clinically significant weight-reducing effects. However, its use is limited by side effects, including difficulty with concentration, memory and cognition, psychomotor slowing, mood changes with nervousness, anxiety, and depression. It is not recommended or FDA-approved for weight loss in children and adolescents.

In 2012, a combination of phentermine and topiramate (Qsymia®) was approved by the FDA for weight loss in obese adults. In a one-year clinical trial, the combination drug yielded 10.2 kg weight loss on the higher dose of drug, versus 1.4 kg with placebo.[44] In addition to the cognitive and psychological side effects of topiramate noted above, concerns were raised about teratogenicity and increases in resting heart rate, leading to a "risk evaluation and management strategy" (REMS) that was mandated with the FDA approval.[45] No trials of phentermine-topiramate have been performed in pediatric populations. Thus, it is not recommended or FDA-approved for weight loss in children and adolescents.

Weight Loss Surgery

During the past 25 years, surgically induced weight loss has emerged as an important option for adults with severe obesity. In contrast to poor long-term success rates for nonsurgical treatments of obesity, surgical approaches generally produce durable and substantial weight loss. In large series of adults undergoing weight loss surgery, the mean reduction in excess body weight was 67%, corresponding to a 34% reduction in BMI.[46] The weight loss typically was maximal around one year postoperatively, followed by modest weight regain, then stable weight for up to 16 years of follow up. Diabetes, dyslipidemia, hypertension, sleep apnea, and other weight-related comorbidities resolved or improved substantially, and

mortality risk was reduced by 89% at five years postoperatively. Weight loss and comorbidity outcomes in adolescents appear to be similar to those in adults, although long-term follow up data are sparse in the adolescent population.[47]

The three most common operations performed today are the Roux-en-Y gastric bypass (RYGB), laparoscopic adjustable gastric banding (LABG), and laparoscopic sleeve gastrectomy (LSG) (Figure 37-2). Each of these procedures reduces gastric capacity, but RYGB and LSG also reduce appetite through entero-endocrine mediators. Weight loss outcomes are generally better for RYGB and LSG as compared with LAGB, and LAGB also requires periodic readjustment of pressure to produce an optimal effect. LAGB and LSG have somewhat fewer complications in the immediate postoperative period as compared with RYGB. However, LAGB also has a high rate of re-operation for complications including band slippage (as high as 25% in some series). About 10% of patients undergoing RYGB have important complications, which include dumping syndrome, anastomotic strictures or ulcers, internal hernias, kidney stones, episodic hypoglycemia, and gallstone formation requiring cholecystectomy.[46] Anastomotic leaks, staple-line disruptions, and pulmonary embolism can occur but have been reduced to 1% to 2% each by modifications in surgical technique and postoperative care.

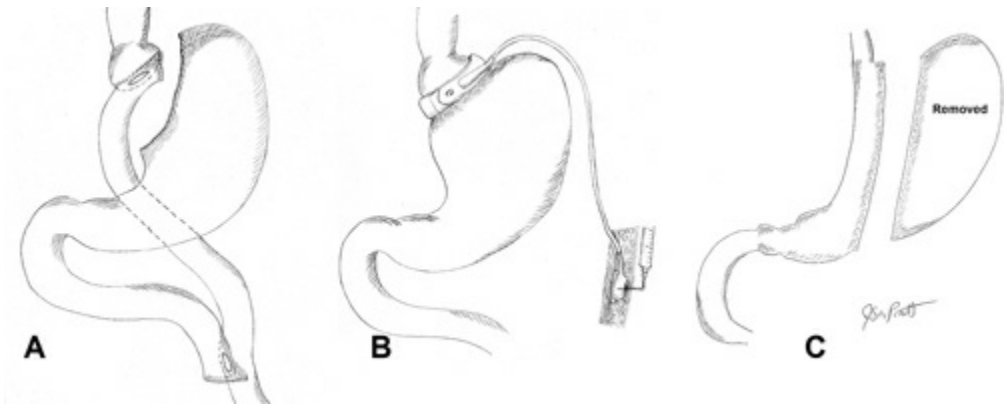


Figure 37-2 Types of weight loss surgeries. *PANEL A:* Roux-en-Y gastric bypass (RYGB). *PANEL B:* Laparoscopic adjustable gastric banding (LAGB). *PANEL C:* Laparoscopic vertical sleeve gastrectomy (LSG).

Patient Selection

The weight loss surgical procedures outlined above clearly have benefits that outweigh the modest risks for appropriately selected patients. Nonetheless, there is still some uncertainty regarding optimal patient selection. To date, no psychological or physiological factors have been defined that will determine which patients are most likely to suffer weight regain, medical complications, or psychological complications after weight loss surgery.

Currently, most guidelines suggest the following weight thresholds for selecting either adolescents or adults for weight loss surgery[48]:

- BMI ≥ 35 with major medical comorbidities (such as severe sleep apnea, type 2 diabetes, severe and progressive fatty liver disease, or pseudotumor cerebri).
- BMI ≥ 40 with more minor medical comorbidities (such as mild sleep apnea, dyslipidemia, hypertension, or impaired glucose tolerance). This threshold effectively selects for severe obesity among adolescents, because it is well above the 99th BMI percentile curve for all ages. Moreover, it is a more stringent selection criteria in younger as compared with older adolescents.

The guidelines also suggest the following additional considerations for selecting adolescents for weight loss surgery:

- Physical maturity—Demonstrated by completion of 95% of skeletal growth, or sexual maturity (Tanner) stage IV. Despite this guideline, it should be recognized that for some younger patients with serious complications of obesity, the benefits of surgery may still outweigh the small risk that the surgery might attenuate linear growth.
- Psychological and emotional maturity—Demonstrated by clear understanding of the risks and benefits of surgery, mature motivations for undergoing the procedure, and good compliance with preoperative therapy.
- Stable mental health—Psychological disorders should be fully evaluated and stabilized before consideration of weight loss surgery. Patients with substance use disorders can be considered on a case-by-case basis after they have been in remission for one year.

These represent the minimum considerations when evaluating an adolescent for weight loss surgery. Many other factors may enter the decision, including family and other support systems, adequacy of efforts

to improve weight status through non-surgical means, and the timing of the elective surgery relative to life events.

Nutritional Considerations

None of these surgical procedures causes significant malabsorption of dietary *macronutrients* or energy. However, it is very important to monitor patients postoperatively to ensure good dietary balance and to reduce the risk of vitamin deficiencies and protein malnutrition. Ample intake of fluids (e.g., 90 ounces daily) is encouraged to avoid dehydration, particularly in the early postoperative period. Most patients will also need to avoid sugar-sweetened beverages and other concentrated carbohydrates, to optimize weight loss and avoid dumping syndrome.[49]

Malabsorption of certain *micronutrients*, particularly iron, vitamin D and vitamin B_{12}, is common and requires postoperative monitoring and treatment. Case reports have described clinical vitamin deficiencies after weight loss surgery, of thiamine (vitamin B_1) causing beriberi, vitamin C causing scurvy, and of pyridoxine (vitamin B_6) causing pellagra. Deficiencies of zinc, selenium and copper are less well documented. Guidelines generally recommend supplements consisting of a multivitamin with iron, with additional vitamin D and calcium, and many programs also routinely supplement with vitamin B_{12}.[49] In addition, it is important to routinely monitor hemoglobin, iron, vitamin B_{12}, thiamine, and 25-hydroxyvitamin D after weight loss surgery, and provide additional supplementation as needed. Some guidelines also suggest monitoring pyridoxine, folic acid, vitamins A, C and E, zinc and magnesium. These nutritional concerns are most important for patients who have had RYGB, because the procedure bypasses some of the absorptive portion of the small intestine. Nonetheless, the same supplementation and monitoring usually is recommended for patients undergoing LAGB and LSG.

In female patients with regular menses, the risk of anemia is high, so it is common to recommend suppression of menses with hormonal contraceptives. Pregnancy is discouraged for the first 12 to 18 months after weight loss surgery, representing the phase of active weight loss. Outcomes of pregnancy after this period are typically good: pregnancy after weight loss surgery carries fewer risks for both the mother and the fetus than pregnancy in women with severe obesity (BMI $\geq$ 40). Female patients who become pregnant can continue their regular vitamin supplements, but should be monitored carefully for development of

anemia.

Future Directions

With the exception of surgery, current treatments for established obesity have disappointing long-term outcomes. However, research is likely to lead to advances in several important arenas. First, public recognition of the obesity problem should lead to public education and public programs designed to prevent obesity during childhood. Such population-wide approaches are more likely to be effective than treatment approaches targeting individual patients with therapies that are weakly effective and resource-intensive. The identification of which preventative strategies will be most effective is important before they can be implemented widely. Second, improved understanding of the genetic determinants and neural pathways underling the homeostatic control of body weight is likely to lead to pharmacologic interventions that are more specific, and therefore safer and more effective. Third, rigorously scientific and detailed analysis of specific environmental factors contributing to obesity, including issues of diet composition and meal patterns, may lead to more focused and effective dietary and behavioral interventions to treat obesity. Finally, education of the public and healthcare community to recognize and reverse the commonly held bias against obese individuals should help to minimize the stigma and psychological burden associated with the disease.

USEFUL WEB SITES

Nutrition and Exercise

- Body and Mind—An informational and motivational website designed by the Centers for Disease Control (CDC) for children 9 to 13 years old: http://www.bam.gov/
- ChooseMyPlate—An interactive website designed by the United States Department of Agriculture (USDA), that includes a calculator for a customized daily food plan using food groups and portions: www.choosemyplate.gov
- 5210 Let's Go—Patient messaging and toolkits for providers, to support brief focused counseling by providers of primary care. Designed by the Maine Youth Overweight Collaborative: www.letsgo.org.

Nutritional Supplements and Alternative Therapies

National Center for Complementary and Alternative Medicine (NCCAM)—An agency for scientific research and education about complementary and alternative medicine; sponsored by the National Institutes of Health: http://nccam.nih.gov/health/weightloss

Quackwatch—A peer-reviewed journal that analyzes the claims of alternative medicine: http://www.quackwatch.com

Genetics

- GeneReviews—A database of reviews covering many genetic diseases and syndromes, as well as information about when and how to perform genetic testing. Sponsored by the National Center for Biotechnology Information (NCBI): www.genetests.org
- Online Mendelian Inheritance in Man (OMIM)—An online catalog of human genes and genetic disorders: http://www.omim.org/

REFERENCES

1. Janssen I, Katzmarzyk PT, Boyce WF, et al. Comparison of overweight and obesity prevalence in school-aged youth from 34 countries and their relationships with physical activity and dietary patterns. *Obes Rev*. 2005;6:123.
2. Mattei J, Malik V, Wedick NM, et al. A symposium and workshop report from the Global Nutrition and Epidemiologic Transition Initiative: nutrition transition and the global burden of type 2 diabetes. *Br J Nutr*. 2012;108:1325.
3. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of childhood and adult obesity in the United States, 2011-2012. *JAMA*. 2014;311:806.
4. Olds T, Maher C, Zumin S, et al. Evidence that the prevalence of childhood overweight is plateauing: data from nine countries. *Int J Pediatr Obes*. 2011;6:342.
5. Ravussin E, Valencia ME, Esparza J, et al. Effects of a traditional lifestyle on obesity in Pima Indians. *Diabetes Care*. 1994;17:1067.
6. Krebs NF, Himes JH, Jacobson D, et al. Assessment of child and adolescent overweight and obesity. *Pediatrics*. 2007;120 (suppl 4):S193.
7. Huang JS, Barlow SE, Quiros-Tejeira RE, et al. Consensus statement: childhood obesity for pediatric gastroenterologists. *J Pediatr Gastroenterol Nutr*. 2013;56:99–109.
8. Deshmukh-Taskar P, Nicklas TA, Morales M, et al. Tracking of overweight status from childhood to young adulthood: the Bogalusa Heart Study. *Eur J Clin Nutr*. 2006;60:48.
9. Whitaker RC, Wright JA, Pepe MS, et al. Predicting obesity in young adulthood from childhood and parental obesity. *N Engl J Med*. 1997;337:869.
10. Must A, Phillips SM, Naumova EN. Occurrence and timing of childhood overweight and mortality: findings from the Third Harvard Growth Study. *J Pediatr*. 2012;160:743.
11. Bjørge T, Engeland A, Tverdal A, Smith GD. Body mass index in adolescence in relation to cause-specific mortality: a follow-up of 230,000 Norwegian

adolescents. *Am J Epidemiol*. 2008;168:30.
12. Freedman DS, Mei Z, Srinivasan SR, et al. Cardiovascular risk factors and excess adiposity among overweight children and adolescents: the Bogalusa Heart Study. *J Pediatr*. 2007;150:12.
13. Juonala M, Magnussen CG, Berenson GS, et al. Childhood adiposity, adult adiposity, and cardiovascular risk factors. *N Engl J Med*. 2011;365:1876.
14. O'Rahilly S, Farooqi IS. Genetics of obesity. *Philos Trans R Soc Lond B Biol Sci*. 2006;361:1095.
15. Neel JV. Diabetes mellitus: a "thrifty" genotype rendered detrimental by "progress"? *Am J Hum Genet*. 1962;14:353.
16. Hales CN, Barker DJ. Type 2 (non-insulin-dependent) diabetes mellitus: the thrifty phenotype hypothesis. *Diabetologia*. 1992;35:595.
17. Renom Espineira A, Fernandes-Rosa FL, Bueno AC, et al. Postnatal growth and cardiometabolic profile in young adults born large for gestational age. *Clin Endocrinol (Oxf)*. 2011;75:335.
18. Gardner DS, Hosking J, Metcalf BS, et al. Contribution of early weight gain to childhood overweight and metabolic health: a longitudinal study (EarlyBird 36). *Pediatrics*. 2009;123:e67.
19. Eriksson JG, Osmond C, Kajantie E, et al. Patterns of growth among children who later develop type 2 diabetes or its risk factors. *Diabetologia*. 2006;49:2853.
20. Leunissen RW, Kerkhof GF, Stijnen T, Hokken-Koelega AC. Effect of birth size and catch-up growth on adult blood pressure and carotid intima-media thickness. *Horm Res Paediatr*. 2012;77:394.
21. Roseboom T, de Rooij S, Painter R. The Dutch famine and its long-term consequences for adult health. *Early Hum Dev*. 2006;82:485.
22. Smith J, Cianflone K, Biron S, et al. Effects of maternal surgical weight loss in mothers on intergenerational transmission of obesity. *J Clin Endocrinol Metab*. 2009;94:4275.
23. Speliotes EK, Willer CJ, Berndt SI, et al. Association analyses of 249,796 individuals reveal 18 new loci associated with body mass index. *Nat Genet*. 2010;42:937.
24. Cassidy SB, Schwartz S, Miller JL, Driscoll DJ. Prader-Willi syndrome. *Genet Med*. 2012;14:10.
25. Friedemann C, Heneghan C, Mahtani K, et al. Cardiovascular disease risk in healthy children and its association with body mass index: systematic review and meta-analysis. *BMJ*. 2012;345:e4759.
26. Kuczmarski RJ, Ogden CL, Guo SS, et al. 2000 CDC Growth Charts for the United States: methods and development. *Vital Health Stat 11*. 2002;(246):1–190.
27. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition for child overweight and obesity worldwide: international survey. *BMJ*. 2000;320:1240.
28. Janssen I, Katzmarzyk PT, Srinivasan SR, et al. Utility of childhood BMI in the prediction of adulthood disease: comparison of national and international references. *Obes Res*. 2005;13:1106.
29. Grummer-Strawn LM, Reinold C, Krebs NF, Centers for Disease Control and Prevention (CDC). Use of World Health Organization and CDC growth charts for children aged 0-59 months in the United States. *MMWR Recomm Rep*. 2010;59:1.
30. Barlow SE, Expert Committee. Expert committee recommendations regarding the prevention, assessment, and treatment of child and adolescent overweight and obesity: summary report. *Pediatrics*. 2007;120 (suppl 4):S164.
31. Flegal KM, Wei R, Ogden CL, et al. Characterizing extreme values of body mass

index-for-age by using the 2000 Centers for Disease Control and Prevention growth charts. *Am J Clin Nutr*. 2009;90:1314.
32. American Diabetes Association. Standards of medical care in diabetes–2014. *Diabetes Care*. 2014; 37 Suppl 1:S14.
33. Spear BA, Barlow SE, Ervin C, et al. Recommendations for treatment of child and adolescent overweight and obesity. *Pediatrics*. 2007;120 (suppl 4): S254.
34. Whitlock EP, O'Connor EA, Williams SB, et al. Effectiveness of weight management interventions in children: a targeted systematic review for the USPSTF. *Pediatrics*. 2010;125:e396.
35. Choban PS, Flancbaum L. Nourishing the obese patient. *Clin Nutr*. 2000;19:305.
36. *National Initiative for Children's Healthcare Quality (NICHQ)* http://www.nichq.org/areas_of_focus/childhood_obesity_topic.html. Accessed February 23, 2013.
37. Hession M, Rolland C, Kulkarni U, et al. Systematic review of randomized controlled trials of low-carbohydrate vs. low-fat/low-calorie diets in the management of obesity and its comorbidities. *Obes Rev*. 2009;10:36.
38. Thomas DE, Elliott EJ, Baur L. Low glycaemic index or low glycaemic load diets for overweight and obesity. *Cochrane Database Syst Rev*. 2007;(3):CD005105.
39. Epstein LH, Valoski AM, Kalarchian MA, McCurley J. Do children lose and maintain weight easier than adults: a comparison of child and parent weight changes from six months to ten years. *Obes Res*. 1995;3:411.
40. Ross MM, Kolbash S, Cohen GM, Skelton JA. Multidisciplinary treatment of pediatric obesity: nutrition evaluation and management. *Nutr Clin Pract*. 2010;25:327.
41. Sherafat-Kazemzadeh R, Yanovski SZ, Yanovski JA. Pharmacotherapy for childhood obesity: present and future prospects. *Int J Obes (Lond)*. 2013;37:1.
42. Björkhem-Bergman L, Asplund AB, Lindh JD. Metformin for weight reduction in non-diabetic patients on antipsychotic drugs: a systematic review and meta-analysis. *J Psychopharmacol*. 2011;25:299.
43. Bray GA, Hollander P, Klein S, et al. A 6-month randomized, placebo-controlled, dose-ranging trial of topiramate for weight loss in obesity. *Obes Res*. 2003;11:722.
44. Gadde KM, Allison DB, Ryan DH, et al. Effects of low-dose, controlled-release, phentermine plus topiramate combination on weight and associated comorbidities in overweight and obese adults (CONQUER): a randomised, placebo-controlled, phase 3 trial. *Lancet*. 2011;377:1341.
45. Colman E, Golden J, Roberts M, et al. The FDA's assessment of two drugs for chronic weight management. *N Engl J Med*. 2012;367:1577.
46. Christou NV, Sampalis JS, Liberman M, et al. Surgery decreases long-term mortality, morbidity, and health care use in morbidly obese patients. *Ann Surg*. 2004;240:416.
47. Stefater M, Jenkins T, Inge T. Bariatric surgery for adolescents. *Pediatr Diabetes*. 2013;14:1.
48. Pratt JS, Lenders CM, Dionne EA, et al. Best practice updates for pediatric/adolescent weight loss surgery. *Obesity (Silver Spring)*. 2009;17:901.
49. Fullmer MA, Abrams SH, Hrovat K, et al. Nutritional strategy for adolescents undergoing bariatric surgery: report of a working group of the Nutrition Committee of NASPGHAN/NACHRI. *J Pediatr Gastroenterol Nutr*. 2012;54:125.

The Metabolic Syndrome in Children and Adolescents

38

Martin Wabitsch, MD, PhD

INTRODUCTION

The coincidence of the risk factors *visceral obesity, hypertension, dyslipidemia, insulin resistance,* and possibly *disturbed glucose regulation* in an individual is associated with a significantly increased risk of the occurrence of cardiovascular disease and type 2 diabetes mellitus as well as with an increased mortality. Longitudinal studies have shown that the occurrence of these risk factors in childhood and adolescence leads to an early onset arteriosclerosis. The clinical manifestation of arteriosclerosis, however, can only be recognized in advanced state. Clinical symptoms occur usually only in marked stenosis (>70%) of the coronary arteries or of the cerebral or leg arteries. Before the manifestation of the arteriosclerosis can be shown clinically, there are usually subclinical modifications of the vessels for many years, which are manifested as endothelial dysfunction, fatty streaks, swelling of the intima and media as well as plaques.

For many years, it has been recognized that several risk factors occur more than coincidentally together and can cause diseases like myocardial infarction and stroke. Nevertheless, the term metabolic syndrome (MetS) has found its way into clinical medicine only in recent years and was only recently established in the pediatrics and adolescent medicine.[1,2]

In the 1950s, Jean Vague recognized that the co-occurrence of obesity with abdominal body fat distribution, hypertension, diabetes, and gout

leads to arteriosclerosis. The American diabetologist Gerald M. Reaven described in 1988 the so-called syndrome X, a finding complex consisting of insulin resistance, hyperinsulinemia, hypertriglyceridemia, reduced high-density lipoprotein (HDL)-cholesterol, hypertension, and disturbed glucose tolerance. Finally, Kaplan spoke memorably about a "deadly quartet" consisting of abdominal obesity, hypertension, impaired glucose tolerance, and hypertriglyceridemia. The MetS was characterized in 1991 in pediatrics and adolescent medicine and was examined and discussed afterward by physicians of national and international pediatric societies.[3]

DEFINITIONS

The symptom complex consisting of obesity with abdominal body fat distribution, dyslipidemia (hypertriglyeridemia, reduced HDL-cholesterol), insulin resistance, and impaired glucose tolerance as well as arterial hypertension is known in adults as the MetS.

The corresponding symptoms occur in individuals more than coincidental in combinations ("clustering"). A similar constellation of risk factors can already be found in obese children.

Additional typical findings associated with MetS, which can occur in adolescence, are nonalcoholic fatty liver disease, hyperuricemia, and polycystic ovary syndrome, as well as microalbuminuria.

Interestingly, a clustering of cardiovascular risk factors is also often observed in other family members. In the parental and grandparental generations, secondary complications or endpoints (early onset myocardial infarction, stroke) may be present. This information about the family history is important for the assessment of the health risk of the affected child or adolescent.

The term "tracking," which is often used in relation with MetS, means the persistence of the individual risk factors from childhood to adulthood as well as an increasing expression of the risk factors with age. These correlations can be interpreted as follows: if evidence for MetS is found in one patient, these symptoms will persist quite likely in later life and the individual's clinical symptoms will develop with age. An exception is insulin resistance, which is more common in adolescence than in young adulthood.

So far, there is no generally accepted definition for MetS in childhood and adolescence with an indication for limit values for the individual components. The goal of developing a definition for the MetS in childhood and adolescence, which will have high clinical relevance, is barely

accessible at the moment. The reason is that a health risk for MetS has yet to be described, and there are insufficient long-term data to help us define individual reference values and their combination to match a prognostic risk. Beyond that, in childhood and adolescence it is not possible to indicate reference values for the individual components of MetS based on the development of these components that depend on age and puberty.

In 2007, the International Diabetes Federation (IDF) has published a proposal for a definition of the MetS in childhood and adolescence (Table 38-1). This rather conservative definition of MetS for childhood and adolescence is easily applied in practice and takes into account the developmental aspects (tracking). Until better definitions of MetS are available, the IDF definition of MetS for childhood and adolescence is proposed for use in practice.

TABLE 38-1 Consensus Definition of the Metabolic Syndrome in Childhood and Adolescence of the International Diabetes Federation (IDF)

Age 6 to <10 years

- Obesity > 90th percentile as assessed by waist circumference
- Metabolic syndrome cannot be diagnosed, but further measurements should be made if family history of metabolic syndrome, type 2 diabetes mellitus, dyslipidemia, cardiovascular disease, hypertension, or obesity
- IDF suggests that a strong message for weight reduction should be delivered for those with abdominal obesity

Age 10 to <16 years

- Obesity > 90th percentile (or adult cut-off if lower) as assessed by waist circumference
- Triglycerides > 1.7 mmol/L
- HDL-cholesterol < 1.03 mmol/L
- Blood pressure > 130 mm Hg systolic or >85 mm Hg diastolic
- Glucose > 5.6 mmol/L (oral glucose tolerance test recommended) or known type 2 diabetes mellitus

Age > 16 years

Use existing IDF criteria for adults.
According to the new IDF definition, for a person to be diagnosed with MetS, he or she must have:
Central obesity (defined as waist circumference with ethnicity-specific values) plus any two of the following four factors:

- Raised TG level: >150 mg/dL (1.7 mmol/L), or specific treatment for this lipid abnormality
- Reduced HDL cholesterol: <40 mg/dL (1.03 mmol/L) in males and <50 mg/dL (1.29 mmol/L) in females, or specific treatment for this lipid abnormality
- Raised blood pressure: systolic BP > 130 or diastolic BP >85 mm Hg, or treatment of previously diagnosed hypertension
- Raised fasting plasma glucose (FPG): >100 mg/dL (5.6 mmol/L), or previously diagnosed type 2 diabetes (if above 5.6 mmol/L or 100 mg/dL, OGTT is strongly recommended but is not necessary to define presence of the syndrome).

If BMI is >30 kg/m², central obesity can be assumed and waist circumference does not need to be measured.

TABLE 38-1B Upper Limits for Waist Circumference With Ethnicity-Specific Values

Country/Ethnic Group		Waist Circumference
Europids[a] In the United States, the ATP III values (102 cm male; 88 cm female) are likely to continue to be used for clinical purposes	Male Female	>94 cm >80 cm
South Asians Based on a Chinese, Malay, and Asian-Indian population	Male Female	>90 cm >80 cm
Chinese	Male Female	>90 cm >80 cm
Japanese[b]	Male Female	>90 cm >80 cm
Ethnic South and Central Americans		Use South Asian recommendations until more specific data are available.
Sub-Saharan Africans		Use European data until more specific data are available.
Eastern Mediterranean and Middle East (Arab) populations		Use European data until more specific data are available.

Children younger than 6 years were excluded from the definition because of insufficient data for this age-group.

[a]In future epidemiological studies of populations of Europid origin, prevalence should be given using both European and North American cut-off points to allow better comparisons.

[b]Subsequent data analyses suggest that Asian values (male 90 cm; female 80 cm) should be used for Japanese populations until more data are available.

The assessment of the cardiovascular risk in children and adolescents with reference for the presence of MetS and possibly the indication of a therapy should be stratified—independent of an exact specification for the definition of MetS.

REFERENCE VALUES FOR THE INDIVIDUAL COMPONENTS OF MetS

The German working group of obesity in children and adolescents recommends in their guidelines[4] the use of the following reference values for the diagnostic assessment of cardiovascular risk factors, which are conjunct in the MetS.

Visceral Obesity

In accordance with several national and international guidelines, obesity is defined by the reference values for body mass index (BMI). An assessment of abdominal fat distribution can be made by measuring the waist circumference. For this, the patient should be lightly dressed (in underwear), upright, standing straight view. Arms hanging sideways to the body and the body weight is evenly distributed on both legs. The patient should inhale and exhale quietly. For the measurement (measuring accuracy ± 1 mm), a nonelastic measuring tape is applied in the area of the strongest medial indentation of the body side structure between the lower costal arch and the iliac crest. If an indentation of the waist region is not recognizable, the waist circumference should be measured between iliac crest and costal arch. The measuring tape is applied horizontally to the ground and under tension (not denting the abdominal wall), the measured value is read after expiration of breath by the patient. To minimize errors, the measurement should be repeated, and the mean value should be calculated from the two measurements.

Waist circumference is also subject to age—and gender-specific alterations. In 2008, Kromeyer-Hauschild et al[5] have published age-and gender-specific percentiles for waist circumference of 6-to 18-year-old German children and adolescents (Figures 38-1A and B). In adults, an abdominal obesity with a waist circumference ≥80 (female) and ≥94 cm (male) is defined.[6] In contrast, there is no uniform value limit determination in the literature for children and adolescents. The waist circumference and the BMI are highly correlated.[5] To identify increased abdominal fat accumulation in childhood and adolescence, a waist

circumference above the 90th percentile is recommended based on the definition of obesity in children and adolescents (BMI > 90th percentile[7]).

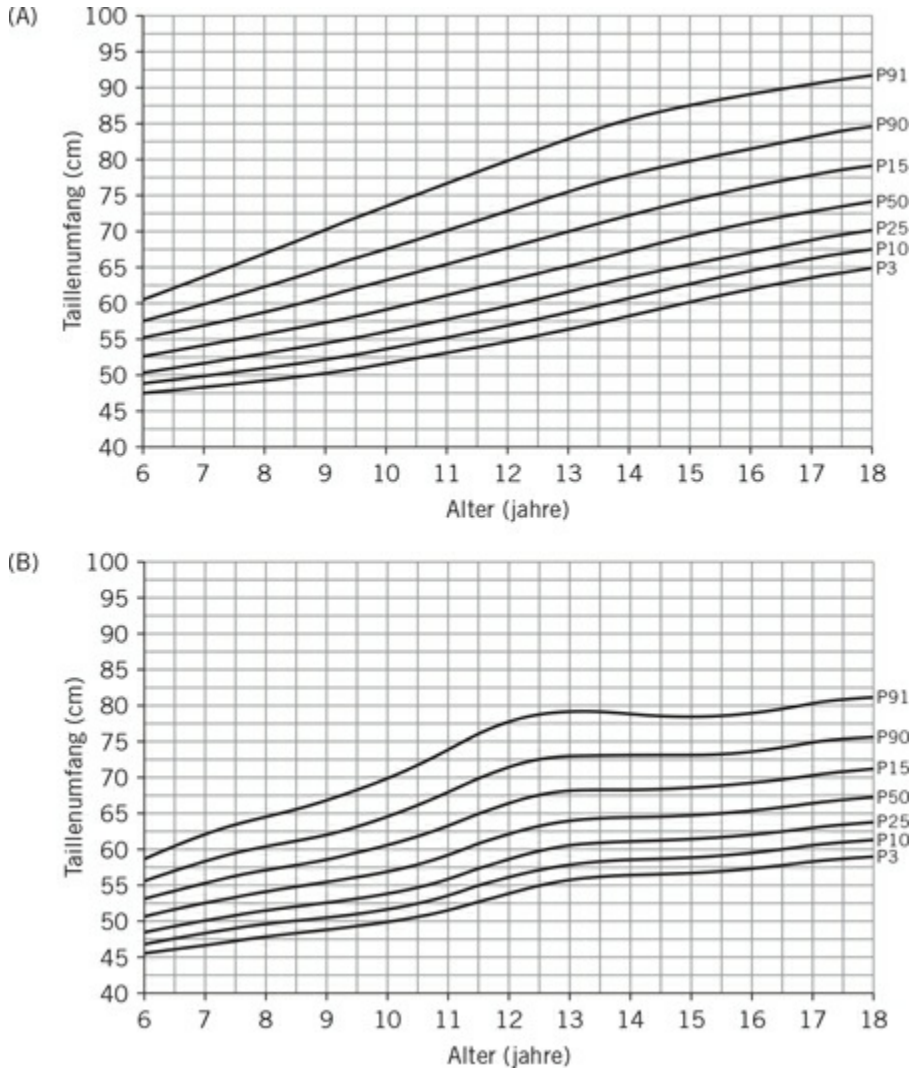


Figure 38-1 A, Percentile graph for waist circumference (boys 6–18 years) by Kromeyer-Hauschild et al.[5] B, Percentile graph for waist circumference (girls 6–18 years) by Kromeyer-Hauschild et al.[5]

Dyslipidemia

The determination of a fasting lipid levels (total cholesterol; low-density lipoprotein [LDL], HDL, triglycerides) is part of the basic diagnostic for all obese children and adolescents as well as overweight children and adolescents with special anamnestic risk factors, for example, a positive family history for early cardiovascular disease. Reference values are

available for the diagnostic evaluation of the lipid levels (Table 38-2).

Values outside of the desirable range should be observed in their further development of the patient. The assessment of the altered lipid values can only occur by taking into account of all cardiovascular risk factors. Any necessary dietary or pharmacological interventions should be made in accordance with the guidelines of the American Academy of Pediatrics.

Arterial Hypertension

The blood pressure measurement should be made in an upright sitting position, on the right arm in a quiet environment after a 5-minute period of rest. It is important to use the correct cuff width (one that covers at least 80% of the upper arm length). The measured values must be interpreted on the basis of sex-, age-, and height-specific reference values (examples of reference values are given in Tables 38-3A and B). The classification of hypertension in children and adolescents with measurement frequency and therapy recommendations is shown in Table 38-4.

With unclear or marginal results and in confirmation of the diagnostic of an arterial hypertension a 24-hour blood pressure measurement may be performed. The assessment of the 24-hour blood pressure measurement is also based on appropriate reference values.

Insulin Resistance and Glucose Regulation Disorder

An often clearly recognizable clinical indication for hyperinsulinemia is acanthosis nigricans, a dirty-brownish hyperpigmentation of the skin along with a coarsening of skin appearance, which typically occurs in the neck and armpits (Figure 38-2).

The determination of fasting serum insulin concentration allows calculation of insulin resistance indices (e.g., homeostasis model assessment of insulin resistance or quantitative insulin sensitivity check index); for calculation, see the guidelines of the German Working Group on Childhood Obesity (AGA).[4] Values outside of the desirable range should be followed during the further development of the child. It must also be considered that the method for insulin determination has not yet been standardized. The gold standard for the measurement of insulin resistance is the euglyemic-hyperinsulinemia clamp, a time-consuming, invasive, and expensive method, which is only suitable for scientific research.

The recommendations of the American Diabetes Association should be

applied to children and adolescents from age 10 or at onset of puberty (whichever comes first) to 19 years to screen for type 2 diabetes (see www.diabetes.org).

ETIOLOGY AND PATHOGENESIS

A strong genetic predisposition exists for the manifestation of individual components of MetS and for MetS itself. Table 38-5 shows estimated figures for heritability of the individual components. This genetic predisposition is polygenic and is because of polymorphisms in different genes. A single modification may thereby have a very small effect, and only the combination with further polymorphism leads to phenotypic changes. A systematic literature search with following meta-analysis identified eight polymorphisms, which are associated with MetS. These varieties were found in the following genes: *FTO*, *TCF7L2*, *IL-6*, *APOA5*, *APOC3*, and *CETP*.[8] All these polymorphisms were also associated with individual components, for example, insulin resistance. Twin studies have shown that approximately half of the variance in insulin sensitivity and secretion may be attributed to genetic factors.

TABLE 38-2 Acceptable, Borderline-High, and High Plasma Lipid, Lipoprotein, and Apolipoprotein Concentrations for Children and Adolescents According to the American Academy of Pediatrics (see www.aap.org)

Category	Low mg/dL[a]	Acceptable mg/dL	Borderline-High mg/dL	High mg/dL[a]
TC	—	<170	170–199	≥200
LDL cholesterol	—	<110	110–129	≥130
Non-HDL cholesterol	—	<120	120–144	≥145
Apolipoprotein B	—	<90	90–109	≥110
Triglycerides				
0–9 y	—	<75	75–99	≥100
10–19 y	—	<90	90–129	≥130

HDL cholesterol	<40	>45	40–45	—
Apolipoprotein A-1	<115	>120	115–120	—

[a]Low cut-off points for HDL cholesterol and apolipoprotein A-1 represent approximately the 10th percentile. The high and borderline-high cut-off points represent approximately the 95th and 75th percentiles, respectively.

TABLE 38-3A Standard Value for Blood Pressure Measurement in Boys According to the American Academy of Pediatrics (see www.aap.org)

Age (y)	BP (Percentile[a])	SBP (mm Hg) Percentile of Height							DBP (mm Hg) Percentile of Height						
		5th	10th	25th	50th	75th	90th	95th	5th	10th	25th	50th	75th	90th	95th
6	50th	91	92	94	96	98	99	100	53	53	54	55	56	57	57
	90th	105	106	108	110	111	113	113	68	68	69	70	71	72	72
	95th	109	110	112	114	115	117	117	72	72	73	74	75	76	76
	99th	116	117	119	121	123	124	125	80	80	81	82	83	84	84
7	50th	92	94	95	97	99	100	101	55	55	56	57	58	59	59
	90th	106	107	109	111	113	114	115	70	70	71	72	73	74	74
	95th	110	111	113	115	117	118	119	74	74	75	76	77	78	78
	99th	117	118	120	122	124	125	126	82	82	83	84	85	86	86
8	50th	94	95	97	99	100	102	102	56	57	58	59	60	60	61
	90th	107	109	110	112	114	115	116	71	72	72	73	74	75	76
	95th	111	112	114	116	118	119	120	75	76	77	78	79	79	80
	99th	119	120	122	123	125	127	127	83	84	85	86	87	87	88
9	50th	95	96	98	100	102	103	104	57	58	59	60	61	61	62
	90th	109	110	112	114	115	117	118	72	73	74	75	76	76	77
	95th	113	114	116	118	119	121	121	76	77	78	79	80	81	81
	99th	120	121	123	125	127	128	129	84	85	86	87	88	88	89
10	50th	97	98	100	102	103	105	106	58	59	60	61	61	62	63
	90th	111	112	114	115	117	119	119	73	73	74	75	76	77	78
	95th	115	116	117	119	121	122	123	77	78	79	80	81	81	82
	99th	122	123	125	127	128	130	130	85	86	86	88	88	89	90
11	50th	99	100	102	104	105	107	107	59	59	60	61	62	63	63
	90th	113	114	115	117	119	120	121	74	74	75	76	77	78	78
	95th	117	118	119	121	123	124	125	78	78	79	80	81	82	82
	99th	124	125	127	129	130	132	132	86	86	87	88	89	90	90
12	50th	101	102	104	106	108	109	110	59	60	61	62	63	63	64
	90th	115	116	118	120	121	123	123	74	75	75	76	77	78	79
	95th	119	120	122	123	125	127	127	78	79	80	81	82	82	83
	99th	126	127	129	131	133	134	135	86	87	88	89	90	90	91
13	50th	104	105	106	108	110	111	112	60	60	61	62	63	64	64
	90th	117	118	120	122	124	125	126	75	75	76	77	78	79	79
	95th	121	122	124	126	128	129	130	79	79	80	81	82	83	83
	99th	128	130	131	133	135	136	137	87	87	88	89	90	91	91
14	50th	106	107	109	111	113	114	115	60	61	62	63	64	65	65
	90th	120	121	123	125	126	128	128	75	76	77	78	79	79	80
	95th	124	125	127	128	130	132	132	80	80	81	82	83	84	84
	99th	131	132	134	136	138	139	140	87	88	89	90	91	92	92
15	50th	109	110	112	113	115	117	117	61	62	63	64	65	66	66
	90th	122	124	125	127	129	130	131	76	77	78	79	80	80	81
	95th	126	127	129	131	133	134	135	81	81	82	83	84	85	85
	99th	134	135	136	138	140	142	142	88	89	90	91	92	93	93
16	50th	111	112	114	116	118	119	120	63	63	64	65	66	67	67
	90th	125	126	128	130	131	133	134	78	78	79	80	81	82	82
	95th	129	130	132	134	135	137	137	82	83	83	84	85	86	87
	99th	136	137	139	141	143	144	145	90	90	91	92	93	94	94
17	50th	114	115	116	118	120	121	122	65	66	66	67	68	69	70
	90th	127	128	130	132	134	135	136	80	80	81	82	83	84	84
	95th	131	132	134	136	138	139	140	84	85	86	87	87	88	89
	99th	139	140	141	143	145	146	147	92	93	93	94	95	96	97

[a] The 90th percentile is 1.28 SD, the 95th percentile is 1.645 SD, and the 99th percentile is 2.326 SD over the mean.

TABLE 38-3B Standard Value for Blood Pressure Measurement in Girls According to the American Academy of Pediatrics (see www.aap.org)

Age (y)	BP (Percentile[a])	SBP (mm Hg) Percentile of Height							DBP (mm Hg) Percentile of Height						
		5th	10th	25th	50th	75th	90th	95th	5th	10th	25th	50th	75th	90th	95th
6	50th	91	92	93	94	96	97	98	54	54	55	56	56	57	58
	90th	104	105	106	108	109	110	111	68	68	69	70	70	71	72
	95th	108	109	110	111	113	114	115	72	72	73	74	74	75	76
	99th	115	116	117	119	120	121	122	80	80	80	81	82	83	83
7	50th	93	93	95	96	97	99	99	55	56	56	57	58	58	59
	90th	106	107	108	109	111	112	113	69	70	70	71	72	72	73
	95th	110	111	112	113	115	116	116	73	74	74	75	76	76	77
	99th	117	118	119	120	122	123	124	81	81	82	82	83	84	84
8	50th	95	95	96	98	99	100	101	57	57	57	58	59	60	60
	90th	108	109	110	111	113	114	114	71	71	71	72	73	74	74
	95th	112	112	114	115	116	118	118	75	75	75	76	77	78	78
	99th	119	120	121	122	123	125	125	82	82	83	83	84	85	86
9	50th	96	97	98	100	101	102	103	58	58	58	59	60	61	61
	90th	110	110	112	113	114	116	116	72	72	72	73	74	75	75
	95th	114	114	115	117	118	119	120	76	76	76	77	78	79	79
	99th	121	121	123	124	125	127	127	83	83	84	84	85	86	87
10	50th	98	99	100	102	103	104	105	59	59	59	60	61	62	62
	90th	112	112	114	115	116	118	118	73	73	73	74	75	76	76
	95th	116	116	117	119	120	121	122	77	77	77	78	79	80	80
	99th	123	123	125	126	127	129	129	84	84	85	86	86	87	88
11	50th	100	101	102	103	105	106	107	60	60	60	61	62	63	63
	90th	114	114	116	117	118	119	120	74	74	74	75	76	77	77
	95th	118	118	119	121	122	123	124	78	78	78	79	80	81	81
	99th	125	125	126	128	129	130	131	85	85	86	87	87	88	89
12	50th	102	103	104	105	107	108	109	61	61	61	62	63	64	64
	90th	116	116	117	119	120	121	122	75	75	75	76	77	78	78
	95th	119	120	121	123	124	125	126	79	79	79	80	81	82	82
	99th	127	127	128	130	131	132	133	86	86	87	88	88	89	90
13	50th	104	105	106	107	109	110	110	62	62	62	63	64	65	65
	90th	117	118	119	121	122	123	124	76	76	76	77	78	79	79
	95th	121	122	123	124	126	127	128	80	80	80	81	82	83	83
	99th	128	129	130	132	133	134	135	87	87	88	89	89	90	91
14	50th	106	106	107	109	110	111	112	63	63	63	64	65	66	66
	90th	119	120	121	122	124	125	125	77	77	77	78	79	80	80
	95th	123	123	125	126	127	129	129	81	81	81	82	83	84	84
	99th	130	131	132	133	135	136	136	88	88	89	90	90	91	92
15	50th	107	108	109	110	111	113	113	64	64	64	65	66	67	67
	90th	120	121	122	123	125	126	127	78	78	78	79	80	81	81
	95th	124	125	126	127	129	130	131	82	82	82	83	84	85	85
	99th	131	132	133	134	136	137	138	89	89	90	91	91	92	93
16	50th	108	108	110	111	112	114	114	64	64	65	66	66	67	68
	90th	121	122	123	124	126	127	128	78	78	79	80	81	81	82
	95th	125	126	127	128	130	131	132	82	82	83	84	85	85	86
	99th	132	133	134	135	137	138	139	90	90	90	91	92	93	93
17	50th	108	109	110	111	113	114	115	64	65	65	66	67	67	68
	90th	122	122	123	125	126	127	128	78	79	79	80	81	81	82
	95th	125	126	127	129	130	131	132	82	83	83	84	85	85	86
	99th	133	133	134	136	137	138	139	90	90	91	91	92	93	93

[a] The 90th percentile is 1.28 SD, the 95th percentile is 1.645 SD, and the 99th percentile is 2.326 SD over the mean.

Insulin Resistance As Central Pathomechanism of MetS

The central pathogenetic feature to the MetS is insulin resistance. The term insulin resistance refers to a reduced glucose uptake of the whole body in response to physiological insulin levels. Insulin resistance has an inverse relation to insulin sensitivity. Euglycemic hyperinsulinemia clamp studies showed that insulin resistance is especially determinated by the insulin response of the skeletal muscle (more than 75% of the amount of glucose during the test is absorbed from muscle and only 2%–3% from adipose tissue). Nevertheless, the adipose tissue has an important function especially with increasing fat mass (see below). Insulin resistance typically occurs in obesity with abdominal body fat distribution, where the visceral fat mass is increased.

In pediatrics and adolescent medicine, it is important to note that during adolescence a physiological insulin resistance of the body is observed. During adolescence, you will find a 25%–50% decrease in insulin sensitivity of the body to return to normal after the end of adolescence.

From epidemiological studies, it is also known that individuals who were born to mothers with poorly controlled diabetes mellitus or gestational diabetes or who were born as small-for-gestational-age babies showing increased weight gain during infancy tend to develop insulin resistance later in life. Epigenetic mechanisms are very likely to come into play (the principle of fetal programming) (see Chapter 27).

The Molecular Basis of Insulin Resistance in MetS

Insulin mediates its effect on the target organs via specific membrane receptors.[9] The insulin receptor consists in an extracellular α-subunit and two transmembrane β-subunits, which are linked by disulfide bonds. The binding of insulin to the α-subunit results in a change of three-dimensional receptor structure (conformation), which causes autophosphorylation of β-subunit. Receptor activation leads to phosphorylation of insulin receptor substrate proteins (IRS-proteins), which are in turn coupled to another kinase, phosphoinositide-3-kinase (PI3-kinase, PI3K). This lipid kinase phosphorylates the signal substance phosphatidylinositol biphosphate into phosphatidylinositol triphosphate, which ultimately enables the central kinase of the insulin signal transmission Akt. The insulin signal is now

passed on to the various effector cells. Thus, the uptake of glucose into the cell through the glucose transporter-4 (Glut-4), or glycogen synthesis, is activated.

TABLE 38-4 Classification of Hypertension in Children and Adolescents With Measurement Frequency and Therapy Recommendations

	SBP or DBP Percentile[a]	Frequency of BP Measurement	Therapeutic Lifestyle Changes	Pharmacolc Therapy
Normal	<90th	Recheck at next scheduled physical examination	Encourage healthy diet, sleep, and physical activity	—
Prehypertension	90th to <95th or if BP exceeds 120/80 even if <90th percentile up to <95th percentile[b]	Recheck in 6 mo	Weight-management counseling if overweight; introduce physical activity and diet management[c]	None unless compelling indications suc chronic kidney disease, diabet mellitus, heart failure, or left ventricular hypertrophy (L exist
Stage 1 hypertension	95th–99th percentile plus 5 mm Hg	Recheck in 1–2 wk or sooner if the patient is symptomatic; if persistently elevated on 2 additional occasions, evaluate or refer to source of care within 1 mo	Weight-management counseling if overweight; introduce physical activity and diet management[c]	Initiate therapy hypertension persists despite trial of four to s months of nonpharmacolc therapy or if compelling indications (as shown above)
Stage 2 hypertension	>99th percentile plus 5 mm Hg	Evaluate or refer to source of care within 1 wk or immediately if the patient is symptomatic	Weight-management counseling if overweight; introduce physical activity and diet	Initiate therapy

	management[c]

[a]For gender, age, and height measured on at least three separate occasions; if systolic and diastolic categories are different, categorize by the higher value.

[b]This occurs typically at 12 years old for SBP and at 16 years old for DBP.

[c]Parents and children trying to modify the eating plan to the Dietary Approaches to Stop Hypertension Study eating plan could benefit from consultation with a registered or licensed nutritionist to get them started.

[d]More than one drug may be required.



Figure 38-2 Typical presentation of axillary skin with acanthosis nigricans.

Although insulin stimulates glucose storage in muscle, it inhibits the glycogenesis and glycogenolysis in the liver (Figure 38-3).

In the MetS—or “insulin resistance syndrome”—the number of insulin

receptors is reduced. This downregulation is triggered by the existing hyperinsulinemia. Moreover, the tyrosine kinase activity of the insulin receptor is lowered, resulting in a further disturbance of the proximal signal transmission.

Insulin is an anabolic hormone and thus a negative regulator of the β-adrenergic-stimulated lipolysis.[9] With insulin resistance, there is a reduced insulin-mediated inhibition of basal lipolysis. Together with the increased body fat mass, this contributes to an increase in the level of circulating free fatty acids. They are in turn causally related to insulin resistance. For the development or strengthening of insulin resistance and reactive hyperinsulinemia, free fatty acids are responsible in three ways. They reduce the insulin binding to the receptor and decrease—depending on concentration—the intake of insulin and the degradation in the liver. A second mechanism is that the glucose utilization in the muscle is significantly impaired by the presence of high levels of free fatty acids. Free fatty acids inhibit the tyrosine phosphorylation of IRS-1. As a result, the activity of PI3-kinase is reduced and the transport of glucose is impaired. The resulting discrete hyperglycemia causes a stimulation of insulin secretion, with the consequence of hyperinsulinemia.

TABLE 38-5 Estimate of the Hereditability of Individual Components of MetS[8]

Phenotype	Estimated Hereditability (%)
Body composition	25–60
• BMI	25–40
• Body fat	40–55
• Abdominal obesity	
Insulin/glucose	10–75
• Fasting glucose	20–55
• Fasting insulin	46–90
• Insulin resistance or T2DM	
Lipids	25–60
• Triglycerides	25–60
• LDL-cholesterol	30–80
• HDL-cholesterol	50–60
• Total cholesterol	
Blood pressure	20–70
	10–50

• Systolic	50
• Diastolic	20–60
• Hypertension	30
• Hemostasis-related features	
• Microalbuminuria	

The third mechanism is shown in a stimulation of insulin secretion by the β-cell by high levels of free fatty acids.

The Role of Adipose Tissue in the Pathogenesis of Insulin Resistance and the MetS

The components of the MetS hypertension, dyslipidemia, and insulin resistance show a significant dependence on the manifestation of obesity and with that on the fat mass.

An analysis of 26,000 datasets of children and adolescents of the obesity patient registry (APV-registry) documented this in an impressive way (Figure 38-4). With increasing relative BMI, there is an increase in the values of the measured cardiovascular risk factors and the clustering of risk factors.

The described correlation between the measure of obesity and the manifestation of the individual components of the MetS implies a causal relationship between increased fat mass and metabolic changes. Recent studies have shown that the adipose tissue in particular the visceral fat mass contributes to the development of insulin resistance as well as to the development of dyslipidemia and the impaired β-cell function.[12] The increased levels of circulating fatty acids pass through the portal vein to the liver. They act as a substrate there for the endogenous synthesis of very-low-density lipoproteins, whose secretion is increased under these conditions.

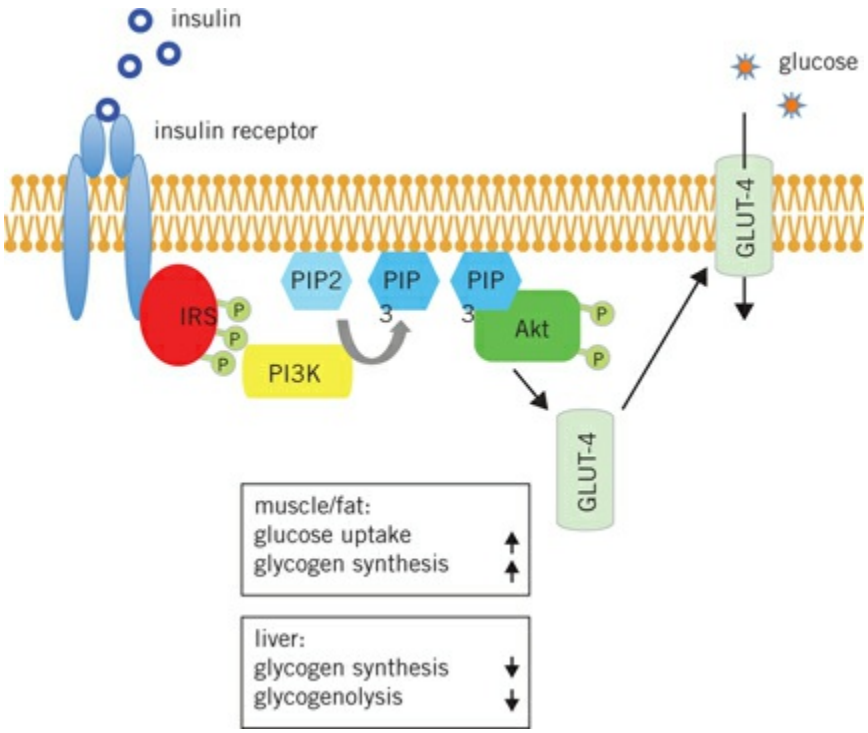


Figure 38-3 Insulin-mediated signal transmission (Wabitsch et al[10]). Insulin binds to the extracellular α-subunit of the insulin receptor. This leads to the autophosphorylation of the intracellular β-subunit of the receptor and the add-on phosphorylation of insulin receptor substrate proteins (IRS). This in turn results in the activation of phosphoinositide 3-kinase (PI3-K), which catalyzes the conversion of phosphatidylinositol diphosphate (PIP2) into phosphatidylinositol triphosphate (PIP3). The lipid molecule activates the key kinase Akt, which forwards the insulin signal now to the various cell effector systems. In adipose tissue as well as in muscle, the glucose transporter Glut-4 is translocated to the cell surface and glucose is added to the cell. Glycogen synthesis is stimulated, whereas hepatic gluconeogenesis and glycogenolysis are inhibited by insulin.

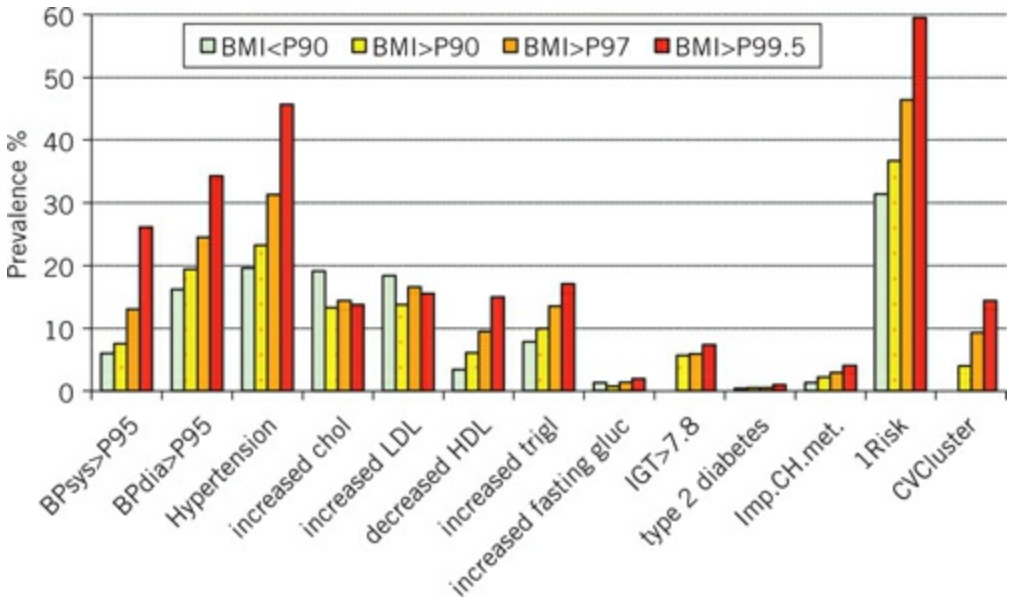


Figure 38-4 Cardiovascular risk factors in relation to BMI categories in 26,008 overweight children and adolescents—an analysis of the APV-registry.[11] Abbreviations: BP, blood pressure; BPdia, diastolic BP; BPsys, systolic BP; CH, carbohydrate; two or three cardiovascular (CV) cluster; accumulation of two or three or more than cardiovascular risk factors; Chol, cholesterol; DM2, diabetes mellitus type 2, impaired fasting glucose (IFG) >7 and/or 2 h OGTT >11.1 mmol/L; Fast.Glu, fasting glucose; HDL, high-density lipoprotein; IGT, impaired glucose tolerance >7.8 mmol/L after two hours of oral glucose-tolerance test (OGTT); Imp.CH.met, impaired CH metabolism; LDL, low-density lipoprotein; P, percentile; Trigl, triglyceride Modified from I'Allemand et al.[11]

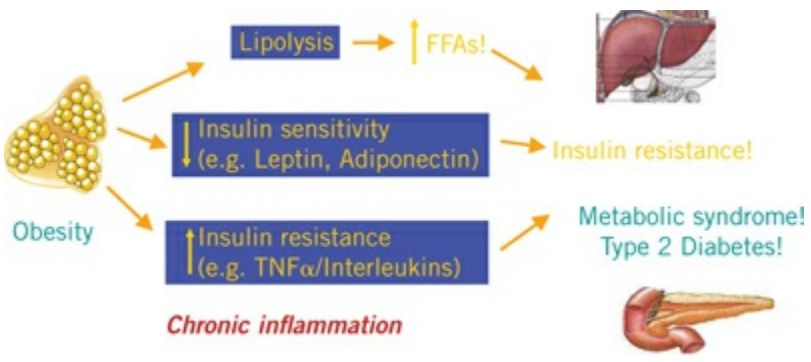


Figure 38-5 The role of adipose tissue in the pathogenesis of insulin

resistance and the metabolic syndrome.

The increased supply of glycerol and lactate in the portal circulation leads to increased gluconeogenesis and hepatic glucose production as a result of the substrate pressure.

Figure 38-5 shows the relationship between an increased fat mass and the development of insulin resistance, dysfunction of glucose regulation, and impaired β-cell function.

The adipose tissue produces many secretion products, which reduce insulin sensitivity and increase insulin resistance. Relatively large amounts of adipokines, which promote insulin resistance, are produced in visceral adipose tissue. In addition to the free fatty acids, these adipokines also result in the liver in an increased gluconeogenesis, in skeletal muscles to an impaired glucose absorption, and in pancreatic β-cells in an impaired insulin secretion.

A pathognomic feature of insulin resistance in obesity is known as **chronic inflammation** of the adipose tissue. This is characterized by the accumulation of macrophages that surround the adipocytes in a crown-like structure (Figure 38-6) and the expression of inflammation genes. These findings are observed very early in the adipose tissue, that is, before a significant increase in circulating insulin concentration. They also refer to an early pathophysiological event in the development of MetS.

ECTOPIC FAT STORAGE IN MUSCLE AND LIVER

Numerous experimental results support the idea that the adipose tissue may reach its capacity limits as the fat storage depot in obesity. In that case, the adipose tissue is characterized by hypertrophy of the adipocyte. These hypertrophic adipocytes are insulin resistant and produce typically high levels of insulin resistance–promoting adipokines. Interestingly, individuals with insulin resistance show ectopic fat accumulation in muscle and liver probably due to adipose tissue that has exceeded its capacity. Both by fat measurements on muscle biopsies and by modern methods of magnetic resonance spectroscopy, an increased ^{1}H-intramyocellular and intrahepatocellular triglyceride concentration was measured in patients with MetS. Increased muscle fat concentration is closely and inversely correlated with the insulin sensitivity of the body. The accumulation of intramyocellular fat is triggered by a reduced fat

oxidation in insulin resistance as well as by promoting the availability of fatty acids by increased lipolysis in adipose tissue. It has also been shown a direct link between visceral fat mass and the occurrence of nonalcoholic steatohepatitis, fat accumulation in hepatocytes. The findings of a fat accumulation in skeletal muscle and liver are typical findings in patients in which a MetS is present.

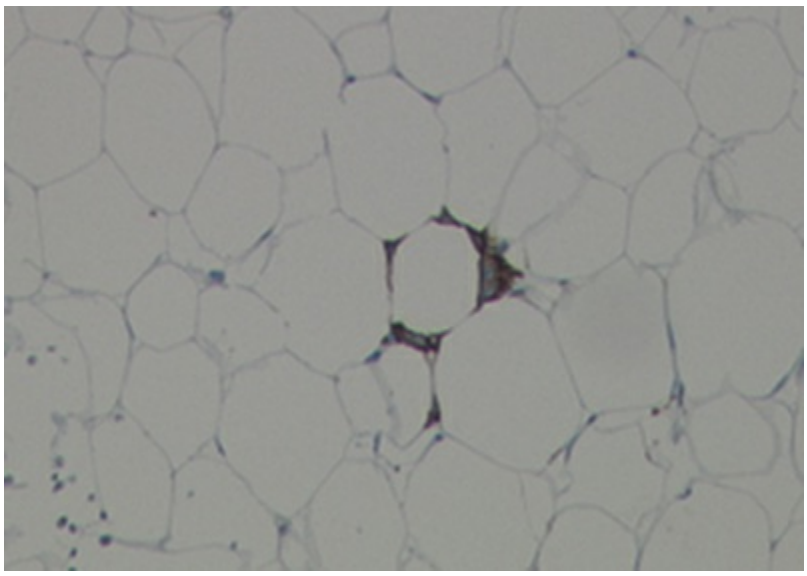


Figure 38-6 Obesity associated inflammation of adipose tissue. In obesity, increased macrophages migrate into the adipose tissue. In the figure, CD11c-positive macrophages are visible in visceral adipose tissue of an obese patient. Macrophages accumulate to a fat cell and form a crown-like structure. This inflammation of adipose tissue plays an important role for the pathogenesis of obesity-associated insulin resistance.

In this context, it is also interesting to see that patients without or with a lack of adipose tissue (e.g., with connatal or acquired lipodystophy), in which there also the maximum capacity of adipose tissue has been reached, have similar metabolic changes in the sense of MetS, as can be observed in obesity with insulin resistance. As in patients with obesity and MetS, in patients with lipodystrophies, there is also an accumulation of fat in the skeletal muscle and in the liver. According to that, a functionally healthy adipose tissue, which is characterized by a high insulin sensitivity and a high storage capacity, is essential for a healthy metabolism and helps to prevent the development of MetS.

PREVALENCE

In a study of 1205 obese children and adolescents aged between 4 and 16 years (965 of them were obese) Reinehr et al[13] could show that the prevalence of MetS differs significantly, depending upon which of the different published definitions were used. In the industrial nations, it can be assumed that the prevalence of MetS, depending on the definition of normal weight children is about 0.1%, of overweight children about 5%–8%, and of obese children about 20%–35%. According to a report by the International Obesity Task Force, cautious estimates indicate that about 20,000 young people in the European Union have an adult onset diabetes (mostly without knowing it); 400,000 have impaired glucose tolerance; 1 million adolescents show different indicators for cardiovascular diseases; and 1.4 million have a fatty liver disease (www.iaso.org/iotf/).

THERAPY

Although now there are good insights into the pathophysiology of insulin resistance and MetS available, there are still no causal therapies for MetS. Because of an insufficient number of longitudinal studies, which allow a reliable conclusion for the long-term prospective risk of MetS in childhood and adolescence, it is unclear whether MetS represents a distinct indication for treatment routinely. Undisputed, however, is the indication for therapy for components of MetS, hypertension, and impaired glucose tolerance, for which reliable risk evaluations exist.

In adolescents, it can be shown that the amount of daily physical activity is inversely associated with the occurrence of MetS. Furthermore, the prevalence of MetS in adolescents was lower, if the affected persons had a healthy diet including a lot of fruits.

Finally, it was convincingly shown that weight loss in children and adolescents leads to a significant improvement of the contributing factors of MetS.[14,15] These observations form the basis for today's behavioral treatment approaches, which primarily have the aim to enforce a change in the behavior of the affected persons. They have only limited effectiveness, however, and are generally not applicable for treating children and adolescents who do not find substantial support from their families and their social environment. Lack of compliance on the part of the affected persons and their families is often the essential problem.

If there is an indication of MetS in children and adolescents, intensive training with the aim to change the lifestyle, especially nutrition and

physical activity behavior, should be used alongside preventive measures (see below). By currently established obesity-training programs (e.g., Obeldicks, Babeluga, Freiburg Intervention Trial for Obese Children (FITOC), and others; see also www.a-g-a.de) these objectives can be achieved.

As an example for the effectiveness of simple behavioral changes, it should be mentioned that by the reduction of the daily fat intake and by the increase of fiber intake, a significant improvement of insulin resistance can be achieved. Increasing physical activity and muscle work is particularly significant.[16] Intensive training programs containing a physical activity program can lead—even after only two weeks—to a significant improvement in insulin sensitivity. This improvement is normally recognizable before a measurable weight loss. Physical activity increases glucose uptake in muscle, especially during intense physical activity. Muscle contractions during physical activity stimulate glucose transport additively to the effect of insulin. Physical activity improves the systematic effect of insulin with a lasting effect up to 72 hours. Physical training leads to an increase in cellular insulin receptors as well as to an increase in PI3-kinase activity. Physical training also reduces the systolic blood pressure, the muscular fat oxidation, and the LDL-cholesterol level.

If the components of MetS are insufficiently influenced by behavioral training and if there is an indication for therapy of associated diseases existing (hypertension, impaired glucose tolerance, dyslipidemia, see above), then drug treatment for these secondary diseases should be implemented.

PREVENTION

Prospective examinations suggest that effective behavioral preventive measures in primary prevention of MetS can be achieved through daycare centers and schools (involving parents). The measures should induce a change in nutrition and physical activity of children and their families. Thereby, a few specific behavioral changes are effective:

1. Reduction or waiver of intake of drinks containing sugar
2. Avoiding energy-dense snacks
3. Ensuring one to two shared meals at home per day
4. Controlled handling of fast food
5. Reduction of television and media time (screen time)
6. Increasing physical activity in everyday life

A consistent, negative relationship exists between the level of physical activity and manifestation of MetS. Interestingly, this relationship is independent of the degree of obesity and can be found to operate in the same way in lean and obese children. This finding indicates that adequate physical activity— *independent* of obesity—has a protective effect on cardiovascular risk factors.

The physical activity of children and adolescents has decreased in recent years. Increasing the time spent in physical activity is therefore an important target for prevention programs. It is still unclear how much physical activity a child needs to build optimal protection against MetS. It is recommended that children and adolescents integrate at least 60 minutes a day of moderate physical activity firmly into the daily plan.[7] The implementation of such projects in schools and daycare centers should have a high priority in addition to the dietary recommendations.

In view of the high and increasing prevalence rates for MetS, it is obvious that the causes of MetS can be found not only in individual behaviors of children and their families but also in the significantly changed living conditions in which today's children grow up. These have a strong and hardly controllable influence on dietary and physical activity behaviors. It follows that in the future, behavioral measures for the prevention of MetS, cardiovascular disease, and type 2 diabetes mellitus must be supported on various social and political levels (see also network causal factors http://www.a-g-a.de).

CONCLUSION FOR PRACTICAL EXPERIENCE

MetS is the pacemaker for the development of cardiovascular diseases and type 2 diabetes mellitus. The components of MetS should be evaluated in children with obesity. If there is evidence of the presence of MetS, special efforts should be made to decrease physical inactivity, to increase physical activity, and to avoid high-calorie and sugar-rich foods. With manifest hypertension or impaired glucose tolerance, early pharmacological treatment is necessary.

ACKNOWLEDGMENTS

The author thanks Bettina Wabitsch for expert secretarial assistance. This chapter was written on the basis of an article in German in the *Journal of the Germany Society for Pediatrics and Adolescent Medicine* (*Monatssch. Kinderheilkunde.* 160:277–292; 2012).

REFERENCES

. Denzer C, Wabitsch M. Das mets bei kindern und jugendlichen. In: Wirth A, Hauner, H, eds. *Das Metabolische Syndrom*. Germany: Urban & Vogel GmbH München; 2007:30–41.
. Wiegand S, l'Allemand D, Holl R, et al. Das metabolische Syndrom bei Kindern und Jugendlichen. In: *Konsensbasierte Leitlinie zur Diagnostik, Therapie und Prävention der Adipositas im Kindes- und Jugendalter*. Arbeitsgemeinschaft Adipositas im Kindes- und Jugendalter (AGA), www.a-g-a.de; 2011.
. Wabitsch M, Hauner H, Heinze E, et al. The relationship between body fat distribution, fasting insulin levels and blood pressure in 79 obese adolescent girls. (abstract). Paper presented at: 30th Annual Meeting European Society for Pediatric Endocrinology; 1991; Berlin, Germany.
. Wabitsch M, Kunze D, (Federführend für die AGA) et al. *(Version 2011) Konsensbasierte Leitlinie zur Diagnostik, Therapie und Prävention der Adipositas im Kindes- und Jugendalter*. Arbeitsgemeinschaft Adipositas im Kindes- und Jugendalter www.a-g-a.de; 2013.
. Kromeyer-Hauschild K, Gläßer N, Zellner K. Perzentile für den Taillenumfang von Jenaer Kindern im Alter von 6 bis 18 Jahren. *Akt Ernaehr Med*. 2008;33:116–122.
. Alberti K, Zimmet P, Shaw J; IDF Epidemiological Task Force Consensus Group. The metabolic syndrome: a new worldwide definition. *Lancet*. 2005;366:1059–1062.
. Wabitsch M, Moss A, (Federführend für die AGA), et al. *(Version 2009) Evidenzbasierte Leitlinie Therapie der Adipositas im Kindes- und Jugendalter (S3-Leitlinie). Arbeitsgemeinschaft Adipositas im Kindes- und Jugendalter*. www.a-g-a.de
. Teran-Garcia M, Bouchard C. Genetics of the metabolic syndrome. *Appl Physiol Nutr Metab*. 2007;32:89–114.
. Saltiel AR, Kahn CR. Insulin signalling and the regulation of glucose and lipid metabolism. *Nature*. 2001;414:799–806.
. Wabitsch M, Moss A, Denzer C, Fischer-Posovszky P. Das metabolische Syndrom *Monatsschr. Kinderheilkunde*. 2012;160:277–292. (in German).
. l'Allemand D, Wiegand S, Reinehr T, et al. Cardiovascular risk in 26,008 European overweight children as established by a multicenter database. *Obesity (Silver Spring)*. 2008;16:1672–1679.
. Wirth A, Hauner H, Jacob S. Metabolisches syndrom und insulinresistenz: ätiologie und mechanismen. In: Wirth A, Hauner, H,

eds. Germany: *Das Metabolische Syndrom*. Urban & Vogel GmbH, München; 2007: 56–80.

. Reinehr T, De Sousa G, Toschke AM, et al. Comparison of metabolic syndrome prevalence using eight different definitions: a critical approach. *Arch Dis Child*. 2007;92:1067–1072.

. Reinehr T, Andler W. Changes in the atherogenic risk factor profile according to degree of weight loss. *Arch Dis Child*. 2004;89:419–422.

. Wabitsch M, Hauner H, Heinze E, et al. Body-fat distribution and changes in the atherogenic risk-factor profile in obese adolescent girls during weight reduction. *Am J Clin Nutr*. 1994;60:54–60.

. Colberg SR, Sigal RJ, Fernhall B, et al. Exercise and type 2 diabetes: the American College of Sports Medicine and the American Diabetes Association: joint position statement. *Diabetes Care*. 2010;33: e147–e167.

Additional Resources

De Man SA, Andre JL, Bachmann H, et al. Blood pressure in childhood: pooled findings of six European studies. *J Hypertens*. 1991;9: 109–114.

Haak T, Kellerer, M. et al. () *Diagnostik, Therapie und Verlaufskontrolle des Diabetes mellitus im Kindes- und Jugendalter. Arbeitsgemeinschaft für Pädiatrische Diabetologie e.V.* (AGPD) und Deutsche Diabetes Gesellschaft (DDG). www.diabetes-kinder.de. Updated 2009.

Kiess W, Hauner H, Wabitsch M, Reinehr T. *Das Metabolische Syndrom im Kindes- und Jugendalter. Diagnose -Therapie -Prävention*. Germany: Elsevier GmbH, Urban & Fischer, München; 2009.

National high blood pressure education program. Working group on high blood pressure in children and adolescents. The fourth report on the diagnosis, evaluation, and treatment of high blood pressure in children and adolescents. *Pediatrics*. 2004;114:555–576.

Povel CM, Boer JM, Reiling E, et al. Genetic variants and the metabolic syndrome: a systematic review. *Obes Rev*. 2011;12:952–967.

Soergel M, Kirschstein M, Busch C et al. Oscillometric twenty-four-hour ambulatory blood pressure values in healthy children and adolescents: a multicenter trial including 1141 subjects. *J Pediatr*. 1997;130: 178–184.

Wuhl E, Witte K, Soergel M, et al. Distribution of 24-h ambulatory blood pressure in children: normalized reference values and role of body dimensions. *J Hypertens*. 2002;20:1995–2007.

Childhood Obesity Prevention

39

John J. Reilly, BSc, PhD and Adrienne H. Hughes, BSc, PhD

CHILDHOOD OBESITY PREVALENCE AND TRENDS

In most high-income countries the prevalence of childhood obesity increased dramatically over the past few decades.[1] Recently, the rate of increase in the prevalence of childhood obesity appears to have slowed down in high-income countries,[2] but prevalence remains high and obesity rates have generally not slowed among children of lower socioeconomic status. Moreover, obesity prevalence estimates usually define obesity as a high body mass index (BMI) for age and sex, but such definitions are conservative, with a relatively high proportion of excessively fat children not identified as obese by BMI.[3] This means that most national estimates of childhood obesity prevalence underestimate the scale of the problem. Estimates of obesity prevalence based on the widely used "Cole-International Obesity Task Force" BMI-based definition of obesity are even more conservative.[3]

ARGUMENTS FOR EARLY PREVENTION OF CHILDHOOD OBESITY

There is now general agreement that childhood obesity prevention is important, and that attempts to prevent obesity should include the infancy–early childhood period. In brief, the arguments in favor of focusing obesity

prevention in early life are as follows. First, the obesity epidemic affected young children—they have not been immune from the societal changes that have affected the rest of the population. Second, there is a strong tendency for obesity to persist at any age, and it is more persistent when more severe[4,5]; many obese children will not simply "grow out of their obesity" without an intervention that helps them to do so. The longer term consequences of childhood obesity are generally more severe when obesity has its onset in early life.[5] There may also be critical periods in early life when children may be set on a path to later obesity, and even relatively short-term interventions in early life may therefore provide long-term protection against obesity. It is believed that obesity prevention in early life may be more effective than attempting to prevent obesity later in life, when obesity-promoting behaviors have become well-established. Finally, recent evidence suggests that attempting to prevent obesity in early life in the current generation of children may have beneficial effects on future generations of children via epigenetic mechanisms.[6]

ORIGINS OF CHILDHOOD OBESITY: ENERGY IMBALANCE

Obesity results from a chronic positive energy balance, a sustained excess of energy intake over the amount of energy expended. Although children need a small positive energy balance to grow (of less than about 50 kcal/d after the period of rapid growth in the first six months of infancy), the size of the energy imbalance in modern children who become obese is usually much larger.[7,8] Since energy imbalance depends only on the amount of energy being consumed and expended, the causes of obesity can seem very simple. This apparent simplicity can lead the public, health professionals, and health policymakers to the view that prevention or treatment of obesity should be simple tasks. In fact, it has become clear that children and their families live in a much more complex "ecology" and the "causes of the causes" of excessive energy intake or inadequate energy expenditure are very much more complex.[9] The UK government Foresight Report on tackling obesity,[9] for example, recognized over 200 factors that influence obesity, and these factors operate at different levels in an "ecological model": at the level of the child, the family, the local neighborhood, the school, national policy environment, and the international policy environment. In addition, the Foresight Report pointed out that many of these factors interact with each other in multiple and complex ways. In

summary, the apparent simplicity of the development of obesity disguises a marked complexity.

ESTABLISHED RISK FACTORS FOR CHILDHOOD OBESITY

Modifying the main risk factors for childhood obesity is the key to prevention. The complexity of childhood obesity has hindered our understanding of etiology, and other complexities arise from gaps in the evidence base on etiology, and from limitations arising from observational study designs. Most etiological studies to date have been observational, with the inherent limitations of observational studies, in particular difficulties in attributing cause and effect with certainty. High quality intervention studies which inform our understanding of etiology have been scarce.[8] A full understanding of the etiology and prevention of childhood obesity is therefore not available at present, but there is little doubt that obesity etiology is sufficiently well-understood to justify preventive interventions. Recent systematic reviews and meta-analyses of etiology have demonstrated that there is a fairly large body of fairly consistent and high-quality evidence.[10–14] Moreover, recent systematic reviews of preventive interventions have demonstrated a reasonably sizeable body of evidence upon which to design future research, clinical, or policy interventions, for example.[15]

Recent systematic reviews and meta-analyses of etiological studies have established a number of modifiable obesity risk factors in early life: rapid early growth, formula feeding, screen time, lack of sleep/disrupted sleep, low physical activity, and sugar-sweetened drinks. These risk factors promote an excessive energy intake, a reduced energy expenditure, or both, and are summarized in brief below.

Rapid Early Growth

Growing rapidly in utero, in infancy, or in early childhood strongly and consistently predicts increased risk of childhood obesity.[10–14,16] In the large Avon Longitudinal Study of Parents and Children (ALSPAC) cohort study in England, for example, the adjusted odds ratio (AOR) for obesity at age 7 years for those with catch-up growth (defined as gain in weight Z score of >0.67 from birth to 24 months[16]) was 2.6 compared to those with no catch-up growth.

One aspect of rapid early growth that promotes increased risk of

childhood obesity, but is not widely known, is early "adiposity rebound." The adiposity rebound (AR) is the period in childhood (typically between around age 3–6 years in western populations) when BMI begins to increase after reaching a nadir. In the ALSPAC cohort study, early AR (before age 3.5 years) was the risk factor that was most strongly associated with later obesity (AOR 15 vs. later AR after age 5 years), stronger even than both parents being obese (AOR 10 vs. neither parent obese).[16] Identifying the timing of AR in individual children is problematic because of the need to obtain a number of measures of BMI and plot them on a BMI for age reference chart, and so using the timing of rebound in clinical practice may be impractical. For public health applications, such as the planning of obesity prevention interventions, knowledge of the precise timing of AR is less important, and such applications simply need to focus on modifiable behaviors (discussed below) before the AR takes place.

The mechanisms that underlie the association between rapid early growth and later obesity are still not fully understood. Intervention studies that aim to reduce risk of obesity via modification of growth trajectory can be very informative of underlying mechanisms, and can provide potential clinical or public health strategies for preventing obesity.[17] For example, the recent European Childhood Obesity Trial suggests that modifying the protein content of infant formula may reduce the risk of developing "obesogenic" growth trajectories in early life.[17]

Formula Feeding

Formula feeding is a well-established risk factor for childhood obesity,[10,12] and this may be mediated in part by the impact of formula composition on growth trajectory, as suggested by the European Childhood Obesity Trial.[17] The concept that formula-feeding increases risk of childhood obesity remains controversial, but promotion of breast-feeding as a means of preventing obesity is relatively uncontroversial because it has so many other benefits.

Sedentary Behavior/Screen Time

High exposure to screen/media (particularly TV) is a well-established early risk factor for childhood obesity,[10,16] and many national and international guidelines on obesity prevention recommend that exposure to screens should be restricted in early life[18–21]: to less than 1 h/d for children aged 2–4 years and no more than 2 h/d for children aged 5 and over. Promotion of a reduction in screen time/TV viewing should have a number

of benefits for child health and development beyond obesity prevention and screen-time behavior seems to be modifiable, so this is a logical target of clinical or public health actions aimed at obesity prevention.

Lack of Sleep/Disrupted Sleep

Lack of sleep/disrupted sleep are increasingly well-established early risk factors for childhood obesity.[10,16] As with most of the well-established risk factors for childhood obesity, the mechanisms underlying the impact of sleep deficit on obesity risk are not fully understood. However, as with most of these risk factors, clinical or public health action aimed at promoting sleep should have a number of benefits for child health and development beyond obesity prevention, and sleep behavior seems modifiable, so this seems to be a justifiable target for obesity prevention efforts.

Low Physical Activity

Low physical activity predisposes children to increased risk of obesity,[10,22] and so is a justifiable target of clinical or public health efforts directed at obesity prevention. Low physical activity also increases the risk of a range of other health problems: physical activity is therefore worth promoting in its own right. National and international guidelines recommend accumulating at least 180 min/d of physical activity at any intensity for young children (aged 1–4 years) and 60 min/d of moderate to vigorous intensity physical activity for children aged 5 and over.

Sugar-Sweetened Drink Consumption

Consumption of sugar-sweetened drinks increases risk of childhood obesity,[1] and a number of other health problems, and appears to be modifiable,[23] so represents a logical target of clinical or public health efforts that aim to prevent childhood obesity.

Other Risk Factors for Childhood Obesity

A number of nonmodifiable risk factors (socioeconomic status, ethnicity, age, and gender) and less modifiable factors (notably parental obesity) have been associated with risk of childhood obesity in various studies. Although nonmodifiable or less-modifiable risk factors are of limited value in designing interventions, they can help identify families at high risk of obesity, as discussed below.

USING THE ESTABLISHED RISK FACTORS FOR CHILDHOOD OBESITY IN CLINICAL AND PUBLIC HEALTH PRACTICE

Planning Preventive Interventions

Having a substantial body of evidence on early life risk factors for childhood obesity provides health professionals and policymakers with a suite of "candidate" behavioral targets of obesity prevention interventions. The well-established candidates at present are rapid early growth, excess screen time, lack of sleep, low physical activity, formula feeding, sugar-sweetened drink consumption, as described above. Novel risk factors will emerge as new evidence comes to light.

There are so many well-established behavioral targets of obesity prevention interventions that it is helpful to prioritize them when planning research, clinical practice, or policy interventions. Logic frameworks should be helpful in prioritizing obesity prevention interventions. One of the earliest and most useful logic frameworks was that proposed by Whitaker,[24] who suggested that the highest priority clinical and public health preventive interventions for childhood obesity should meet the following criteria: there should be good evidence that the behavior or target is implicated in causing obesity, the behavior should be modifiable, modifying the behavior should do no harm, modifying the behavior should promote child health and/or development for reasons other than obesity. Applying these "Whitaker Criteria" should be useful to guide decisions on the behaviors to target in obesity prevention, and should "future-proof" such decisions, as the framework should be applicable when novel risk factors for obesity also emerge.

It is striking that the well-established early life behavioral risk factors for childhood obesity outlined above meet the "Whitaker criteria" well. A recent critique of research interventions, and clinical practice and policy interventions, to prevent obesity in the United Kingdom concluded that many such interventions do not meet the "Whitaker criteria" well.[25] In the UK preventive interventions are often not well-informed either by the evidence on etiology and interventions, or by the application of any logic model.[25]

The evidence summarized above has focused on *why* obesity develops during childhood. The issue of *when* obesity is most likely to develop during infancy, childhood, and adolescence, is important in the planning of population-based strategies for the prevention of childhood obesity, but

has been somewhat neglected. Many such interventions have failed to provide a rationale as to *when* (at what age) interventions should take place, for example, as highlighted in the review by Nichols and Swinburn.[26]

Preventive interventions in populations might be best targeted at stages of the life cycle when excess positive energy balance (or excess weight gain) is at its peak, when the incidence (development of new cases of obesity) is at its highest, and/or the stage at which obesity is least likely to resolve spontaneously. Cohort studies can be very informative of such issues. In the ALSPAC birth cohort in England, for example, one study attempted to identify at what point(s) in infancy, childhood, and adolescence excess weight gain was at its highest: this was in mid-late childhood (7–11 years).[27] A subsequent study in the ALSPAC cohort[28] also attempted to identify peak periods for the development of new cases of obesity, and again incident obesity was most likely during mid-late childhood (age 7–11 years). The generalizability of these ALSPAC findings is unclear, but obtaining similar evidence in other societies could be very helpful in making population-based obesity prevention more evidence-informed in future, and may increase the efficacy, effectiveness, and cost-effectiveness of future interventions.

Identifying Children and Families at High Risk of Developing Obesity

Knowledge of the suite of well-established risk factors for childhood obesity, and the magnitude of the risk which they provide, can permit health professionals to identify infants or children at high risk of obesity. This information could then be used to observe changes in growth more often as a result of a child being identified at high risk (e.g., by annual monitoring of weight and height in those deemed to be at high risk[29]), or to inform a decision to begin a weight management intervention. This sort of intervention could be based on an informal use of information on the presence or absence of obesity risk factors in each case, or could be more formal, for example, a screening tool for identifying those at high risk of obesity. The literature contains a number of formal screening tools that aim to quantify the risk of childhood obesity on the basis of the presence or absence of early life obesity risk factors. One recent research-based example is that from Morandi et al.[30] In England, an example of a clinical practice-based screening and management tool for early obesity prevention is provided by the "HENRY" program.[31] One basic question is whether or

not any such screening tools add value to the information which is both readily available in clinical practice and which strongly predicts risk of later obesity, that is, parental obesity.

Knowledge of the *magnitude* of the risk factors for childhood obesity is also likely to be helpful to health professionals and policymakers. Comparing the magnitude of increased risk associated with different obesity risk factors is problematic, but in the large ALSPAC cohort study in England[16] a number of risk factors were investigated within the same sample, and so the relative impact of the different risk factors can be assessed. Table 39-1 summarizes the AOR for each of the early life risk factors that were associated with obesity at age 7 years in the ALSPAC cohort study. It should be noted that if we are considering targeting preventive efforts at the population, rather than for the individual family, the population exposure will be important as well as the magnitude of the risk associated with each risk factor. Risk factors that are more prevalent in the population will usually have a greater impact on obesity than rare risk factors, even if they have only modest effects.

IMPLEMENTING INTERVENTIONS TO PREVENT CHILDHOOD OBESITY: EVIDENCE FOR OPTIMISM

The most recent Cochrane review on interventions for the prevention of childhood obesity in 2011[15] was very positive about the prospects for preventive interventions. It concluded that interventions that aimed to prevent childhood obesity should be successful so long as they implement changes in diet, physical activity, and sedentary behavior. The review even argued that most future research effort in childhood obesity prevention should focus on the question of *how* to implement such changes, as the question of whether such changes would be efficacious was now redundant.[15] This sort of evidence should help dispel pessimism about the prospects for obesity prevention interventions, and make it easier for health professionals and policymakers to argue for the development and implementation of interventions.

TABLE 39-1 Early Life Risk Factors for Obesity From the ALSPAC Study[16]

Risk Factor From Conception–Age 3	Magnitude of Obesity

Years	Risk at 7 Years
Rapid Growth • Very early AR (before age 3.5 y) • Every 100 g increase in birth weight • Every 100 g increase in weight from 0 to 12 m • Catch up growth (weight gain of >0.67 standard deviation scores in first two years)	AOR[a] 15 vs. later AR (after age 5) 5% increase in risk (AOR 1.05/100 g increase) 6% increase in risk (AOR 1.06/100 g increase) AOR 2.6 vs. no catch up
Parental obesity (both parents obese)	AOR 10.0 vs. neither parent obese
>8 h/wk of TV viewing at age 3	AOR 1.6 vs. < 4 h/wk
<10.5 h/night of sleep at age 3	AOR 1.5 vs. ≥ 12 h/night of sleep

[a]AOR, adjusted odds ratio.

TABLE 39-2 Promising Recent Early Life Obesity Prevention Trials

Age Group, Location	Trial/Intervention Name/Reference	Target of Intervention
Infants, Australia	Nourish[36]	Infant feeding
Antenatal–24 mo, Australia	Healthy Beginnings[37]	Infant feeding, active play
Infants, Europe	European Childhood Obesity Trial[17]	Growth modification via protein content of infant formula
2–5-y-olds, USA	Healthy Habits, Happy Homes[38]	Increased sleep, reduced TV, family meals
Birth–5 y, Australia	Infant[39]	Reduced sugar-sweetened drinks and TV, increased active play
	Romp and Chomp[40]	

A number of recent obesity prevention interventions, summarized in Table 39-2, have focused on modifying the well-established early life (infancy–early childhood) risk factors for childhood obesity discussed above, and they show great promise. These interventions may therefore be considered as a high priority for adoption in other settings, perhaps with modifications as appropriate to the circumstances. Alternatively, individual elements of these recent early life interventions might be generalizable to other settings.

GAPS IN THE EVIDENCE ON CHILDHOOD OBESITY PREVENTION

Although much evidence has accumulated on how to prevent childhood obesity, some notable gaps in the evidence remain. Recent systematic reviews have found very little evidence on preventive interventions outside the western world, in low-middle-income countries, for example, where the prevalence of childhood obesity is increasing markedly[15,32] and the capacity to deal with this increased prevalence is limited at present. The generalizability of much of the current research evidence on obesity prevention interventions to non-Western cultures in general, and to low-middle-income countries in particular, is unclear.

An additional notable gap in the evidence is on "upstream" interventions. In socioecological models of health the child is seen as being at the center- or base-of a complex hierarchy of influences on child health behavior and health outcomes. To date, systematic reviews on childhood obesity prevention have found evidence largely from studies that target the "micro-environment" of the child, family, and/or school, and there is much less evidence from interventions that target higher level influences in the hierarchy (e.g., changes in the national and international policy environment), despite the likely high degree of importance of such "upstream" influences.[33] Research that addresses the gaps in the evidence base on childhood obesity prevention should be an urgent priority.

One other gap in the evidence base that has been emerging as an important issue is the question of what are the main barriers to obesity prevention interventions in clinical practice and public health? Qualitative studies have been very informative in understanding the barriers to lifestyle change among groups who would be high priority targets of such interventions. For example, Lakshman et al[34] reported in a qualitative study of English mothers of infants that there was strong resistance to the types of preventive interventions that are indicated by the etiological evidence summarized above. Lakshman et al[34] found that their sample of mothers commonly and strongly believed that it was not possible to overfeed a baby, that infants crying always signaled hunger, that babies "can't be overfed," and that infancy was "too early" to intervene to prevent later obesity and related diseases, beliefs that are all at odds with the research evidence and the beliefs of most obesity prevention researchers. The possible need to overcome such barriers should be considered by those working in clinical practice. For those developing research-based preventive interventions, the most appropriate way to deal with barriers of this kind is to use a framework for the development of the intervention that takes specific account of them, and makes study participant beliefs central to intervention development, such as the UK Medical Research Council

Framework for the Development and Evaluation of Complex Interventions in Public Health.[35]

CONCLUSIONS

The evidence on the etiology of childhood obesity is incomplete, but more than sufficient to develop and implement evidence-informed obesity prevention interventions that target infancy and/or early childhood. Numerous modifiable and nonmodifiable early life risk factors for childhood obesity are now well established, and these have been summarized in this chapter. Interventions that target the modifiable risk factors would generally do no harm and would also improve child health and development in many other respects, beyond obesity. A number of promising "model" interventions exist and these should be considered as priorities for translation beyond their original research settings and populations.

The well-established early life risk factors for childhood obesity should be the basis of future evidence-informed preventive efforts in research, clinical practice, and health policy, and used for identifying families and population groups at particularly high obesity risk. As novel obesity risk factors come to light these should be prioritized for action if they are based on good evidence of causality, and if their modification is likely to do no harm and benefit child health and development in other ways.

REFERENCES

1. Wang Y, Lobstein T. Worldwide trends in childhood overweight and obesity. *Int J Pediatr Obes*. 2006;1:11–25.
2. Rokholm B, Baker JL, Sorensen TI. The levelling off of the obesity epidemic since the year 1999: a review of evidence and perspectives. *Obes Rev*. 2010;11:835–846.
3. Reilly JJ, Kelly J, DC Wilson DC. Accuracy of simple clinical and epidemiological definitions of childhood obesity: systematic review and evidence appraisal. *Obes Rev*. 2010;11:645–655.
4. Singh AS, Mulder C, Twisk JWR, et al. Tracking of childhood overweight into adulthood: a systematic review of the literature. *Obes Rev*. 2008;9:474–488.
5. Reilly JJ, Kelly J. Long-term impact of overweight and obesity in childhood and adolescence on morbidity and mortality in adulthood: systematic review. *Int J Obes*. 2011;35:891–898.
6. Martinez JA, Cordero P, Campion J, Milagro FI. Interplay of early life nutritional programming on obesity, inflammation, and epigenetic outcomes. *Proc Nutr Soc*. 2012;71:276–283.
7. Butte NF, Ellis KJ. Obesity and the environment: where do we go from here? *Science*. 2003;299:853–855.

8. Reilly JJ, Ness AR, Sherriff A. Epidemiologic and physiologic approaches to understanding the etiology of pediatric obesity: finding the needle in the haystack. *Pediatr Res*. 2007;61:646–652.
9. Government Office for Science. *Foresight Report Tackling Obesities: Future Choices,* 2nd ed. http://www.bis.gov/assets/foresight/docs/obesity/17.pdf. Updated 2007.
10. Monasta L, Batty GD, Cattaneo A, et al. Early-life determinants of overweight and obesity: a review of systematic reviews. *Obes Rev*. 2010;11:695–708.
11. Baird J, Fisher D, Lucas P, et al. Being big or growing fast: systematic review of size and growth in infancy and later obesity. *Br Med J*. 2005;331:929–935.
12. Weng SF, Redsell SA, Swift JA, et al. Systematic review and meta-analyses of risk factors for childhood overweight identifiable during infancy. *Arch Dis Child*. 2012;97:1019–1026.
13. Horta BL, Bahl R, Martines JC, Victora CG. *Evidence on the Long-Term Effects of Breastfeeding*. Geneva, Switzerland: WHO Library; 2007.
14. Ip S, Chung M, Raman G, et al. Breastfeeding and maternal and infant health outcomes in developed countries. *Evid Rep Technol Assess*. 2007;153:1–186.
15. Waters E, de Silva-Sanigorski A, Burford BJ, et al. *Interventions for Preventing Obesity in Children*. Oxford, UK: Cochrane Library; 2011.
16. Reilly JJ, Armstrong J, Sherriff A, et al. Early life risk factors for obesity in contemporary children: cohort study. *Br Med J*. 2005;330: 1357–1362.
17. Koletzko B, von Kries R, Closa R, et al. Lower protein in infant formula is associated with lower weight up to age 2 years: a randomized clinical trial. *Am J Clin Nutr*. 2009;89:1836–1845.
18. Scottish Intercollegiate Guidelines Network. *SIGN Guideline 115, Managing Obesity: A National Clinical Guideline* http://www.sign.ac.uk. Updated 2010.
19. Canadian Society for Exercise Physiology. *Canadian Physical Activity and Sedentary Behaviour Guidelines.* http://www.csep.ca/CMFiles/Guidelines/CSEP Guidelines Handbook.pdf. Updated 2012.
20. Get Up and Grow. *Healthy Eating and Physical Activity for Early Childhood Resources.* http://www.health.gov.au. Updated 2013.
21. Children, Adolescents, and the Media. American Academy of Pediatrics Policy Statement. *Pediatrics*. doi: 10.1542/peds.2013–2656.
22. Jimenez-Pavon D, Kelly J, Reilly JJ. Associations between objectively measured habitual physical activity and adiposity in children and adolescents: systematic review. *Int J Pediatr Obes*. 2010;5:3–18.
23. James J, Thomas P, Cavan D, Kerr D. Preventing childhood obesity by reducing consumption of carbonated drinks: cluster randomised controlled trial. *Br Med J*. 2004;328:1237.
24. Whitaker RC. Obesity in childhood and adolescence: four behaviors to target. *Arch Pediatr Adolesc Med*. 2003;151:725–727.
25. Reilly JJ. Evidence-based obesity prevention in childhood and adolescence: critique of recent etiological studies, preventive interventions, and policies. *Adv Nutr*. 2012;3:636S–641S.
26. Nichols MS, Swinburn BA. Selection of priority groups for obesity prevention: current approaches and development of an evidence-informed approach. *Obes Rev*. 2010;11:731–739.
27. Hughes AR, Sherriff A, Ness A, et al. Timing of excess weight gain in a large cohort of English children growing up in the 1990's (ALSPAC). *Pediatrics*. 2011;127:e730–e736.
28. Hughes AR, Sherriff A, Ness A, et al. Incidence of obesity during childhood and

adolescence in a large contemporary cohort. *Prev Med*. 2011;52:300–304.
29. Spear BA, Barlow SE, Ervin C, et al. Recommendations for treatment of child and adolescent overweight and obesity. *Pediatrics*. 2007;120 (suppl 4):S254–S288.
30. Morandi A, Meyre D, Lobbans S, et al. Estimation of newborn risk for child or adolescent obesity: lessons from longitudinal birth cohorts. *PLoS One*. 2012,7:e49919.
31. Rudolf MC, Hunt C, George J, et al. HENRY: development, pilot, and long-term evaluation of a programme to help practitioners work more effectively with parents of babies and pre-school children to prevent obesity. *Child Care Health Dev*. 2010;36:850–857.
32. Verstraeten R, Roberfroid D, Lachat C, et al. Effectiveness of preventive school-based obesity interventions in low- and middle-income countries: a systematic review. *Am J Clin Nutr*. 2012;96:415–438.
33. Lawlor DA, Pearce N. The Vienna declaration on nutrition and non-communicable diseases. *Br Med J*. 2013;347:f4417.
34. Lakshman R, Landsbaugh JR, Schiff A, et al. Developing a programme for healthy growth and nutrition during infancy: understanding user perspectives. *Child Care Health Dev*. 2012;38:675–682.
35. Craig P, Dieppe P, Macintyre S, et al. Developing and evaluating complex interventions: the new Medical Research Council Guidance. *Br Med J*. 2008;337:a1655.
36. Daniels LA, Mallan KM, Battistutta D, et al. Evaluation of an intervention to promote protective infant feeding practices to prevent childhood obesity: outcomes of the NOURISH RCT. *Int J Obes*. 2012;36:1292–1298.
37. Ming Wen L, Baur LA, Simpson JM, et al. Effectiveness of home based early intervention on children's BMI at age 2: RCT. *Br Med J*. 2012;344:e3732.
38. Haines J, McDonald J, O Brian A, et al. Healthy Habits, Happy Homes: randomized trial to improve household routines for obesity prevention among pre-school children. *JAMA Pediatr*. 2013;67(11):1072–1079.
39. Campbell KJ, Lioret S, McNaughton SA, et al. A parent-focused intervention to reduce infant obesity risk behaviors: a randomized trial. *Pediatrics*. 2013;13:652–660.
40. De Silva Sanigorski AM, Bell AC, Kremer P, et al. Reducing obesity in early childhood: results from Romp & Chomp, an Australian community wide intervention program. *Am J Clin Nutr*. 2010;91:831–840.

The Nutrition Transition and Global Child Health

40

Benjamin Caballero, MD, PhD

Health professionals in today's world must keep current on the global health situation, since some of the major domestic health issues, such as HIV/AIDS and obesity cannot be addressed without a global framework. Furthermore, the dramatic expansion of global trade, travel, and communications have torn physical and cultural barriers, exposing most of the world's population not only to communicable diseases but also to dietary and lifestyle practices carrying common risk factors for chronic disease. In this chapter we present an update of current trends on these risk factors, and discuss their impact on children and on the general population.

The world population reached its seven billion mark sometime in October 2011. As expected, the continuing reduction in child mortality over the past several decades led to declining fertility, and by the end of the 20th century all but 16 countries had experienced this demographic transition.[1] This relative slowing in population growth over the past 30 years is one of the important elements of the global nutrition transition. The other, discussed below, is that the vast majority of predicted growth in world population will take place in urban environments of developing countries.

GLOBAL TRENDS IN DISEASE BURDEN

Global disease burden continues its transition from communicable diseases

and predominance of children's diseases to chronic, noncommunicable diseases affecting primarily adults.[2] This epidemiological transition is driven by a combination of reduction in childhood mortality, ageing of the world population, and emergence of chronic noncommunicable diseases of the adult, such as obesity, diabetes, and cardiovascular diseases.[3] These trends are documented in the most recent Global Burden of Disease Study,[4] which compared changes in disability-adjusted life years (DALYs, a metric that reflects the number of life years lost to premature death or disability) between 1990 and 2010. For children under 5 years, overall DALYs declined from 41% in 1990 to 25% in 2010. Over the same period, the DALYs attributed to diarrheal diseases, one of the most common causes of illness in children, decreased by 51%. Conversely, the leading cause of DALYs worldwide was ischemic heart disease, a typical disease of the adult. These shifts are also reflected in the changing composition of DALYs, with a reduction in the contribution of premature mortality (dominated by infant and childhood mortality) and an increase in disability years (driven by chronic diseases of the adult).

In another report, the Global Burden study expanded the analysis of DALYs to assess attributable risk factors for leading disease burdens.[3] From this analysis, the most evident shift among the leading 10 risk factors is childhood underweight, which moved from first place in 1990 to eighth place in 2010. Conversely, high body mass index, which was not in the top 10 in 1990, became the sixth leading risk factor in 2010. Risk factors affecting primarily children, such as water sanitation and micronutrient deficiencies, also declined during that period. A comparison of the top 10 risk factors for global disease burden in 1990 and 2010 is shown in Table 40-1.

TABLE 40-1 Top 10 Causes of Global Disease Burden in 1990 and in 2010, Both Sexes

	1990	2010
1	Childhood underweight	High blood pressure
2	Household air pollution from solid fuels	Tobacco smoking
3	Tobacco smoking	Alcohol use
4	High blood pressure	Household air pollution from solid fuels
5	Suboptimal breast feeding	Diet low in fruits
6	Alcohol use	High body-mass index
7	Diet low in fruits	High blood glucose

8	Ambient particulate matter pollution	Childhood underweight
9	High blood glucose	Ambient particulate matter pollution
10	Iron deficiency	Physical inactivity and low physical activity

Source: From Lim et al.[3]

TABLE 40-2 Regional and Global Trends in Childhood Mortality, 1990–2010

	Infant (1 year) Mortality			Under 5 Years Mortality		
	1990	2000	2010	1990	2000	2010
Northern Africa	62.0	38.2	22.6	82.1	46.9	26.6
Sub-Saharan Africa	104.8	94.1	76.2	173.9	154.3	121.0
Latin America and Caribbean	42.6	28.5	18.1	53.9	34.5	23.4
Caucasus and Central Asia	62.8	51.8	38.6	77.4	62.3	45.3
Eastern Asia	37.6	27.1	15.7	47.6	32.9	18.3
Southern Asia	84.0	64.7	50.7	117.4	87.2	65.5
Southeastern Asia	49.2	35.7	25.3	71.5	48.4	32.2
Western Asia	52.3	35.2	25.4	66.6	45.2	32.2
Oceania	55.1	47.8	40.5	74.6	63.1	52.2
Developed regions	7.0	5.0	3.7	12.1	8.1	5.7
Developing regions	35.6	30.7	25.1	67.1	55.7	44.3
World	60.9	50.9	40.2	87.6	72.8	56.7

Data are number of deaths per 1000 live births.

Source: Adapted from Hill et al.[8]

Behind these global trends are dramatic regional differences in disease burden and risk factors. In sub-Saharan Africa, childhood diseases and mortality are still predominant public health problems, and communicable diseases continue to be major contributors to DALYs.[4] Similarly, leading risk factors in that region are childhood underweight, household air pollution, and inadequate breast-feeding.[3]

GLOBAL TRENDS IN CHILD MORTALITY

Childhood deaths are the end-result of the complex interaction between health, nutritional and environmental factors. In the developing world,

underweight, micronutrient deficiencies (vitamin A, zinc, iron), and suboptimal breast-feeding lead to aggravated morbidity and higher risk of death from diseases such as pneumonia, diarrhea, measles, HIV/AIDS, and malaria.[5] A contaminated environment (unsafe sanitation, air pollution) is also a common contributor. Other determinants include access and utilization of prenatal care, immunizations, and maternal education.[6]

Global rates of under-five child mortality (U5MR) have been declining modestly but consistently for the past two decades, albeit at a pace too low to meet the targets defined by the Millennium Development Goals (MDG).[7] Still, an important milestone was reached by 2010, when for the first time the number of under-five deaths fell below the 10 million/year mark. A revised methodology for mortality estimates was recently introduced by UN agencies.[8] Using this new approach, the data show that between 1990 and 2010 U5MR has declined from 88 to 57 per 1000 live births, equivalent to a reduction from 12 million deaths in 1990 to 7.6 million in 2010.[8] During these 20 years, the rate of decline in mortality has slightly accelerated from the first decade to the second (1.9%–2.5%), but remains well below the MDG target of 4.4% per year. Infant mortality rate (deaths in the first 12 months of life), which accounts for almost 40% of U5MR, has also declined, from 32.5/1000 in 1990 to 22.8/1000 in 2010.[8] There is a marked difference in mortality rates between developed and developing countries. For U5MR, the 2010 figures are 6.8/1000 for developed countries and 62.7/1000 for developing countries. Similarly, there are clear disparities in mortality rate within the developing world, as shown on Table 40-2.

Major factors driving the continuing reduction in child mortality are improved sanitation, expanded immunization and micronutrient supplementation programs, and breast-feeding promotion. Maternal education and income, and social empowerment of women of reproductive age have also been found to favorably impact on child mortality. The continuing reduction in child mortality usually leads to a reduction in population fertility, which in turn favors child survival.[1]

UNDERNUTRITION

Undernutrition in all its forms, including fetal growth restriction, are major factors determining mortality risk in children under 5 years of age, and are responsible for 2.2 million deaths of children under 5 years of age every year.[5] Childhood underweight continues to decline globally, but with

important disparities across countries and regions. Global geographic distribution of under- and overweight in children is presented in Figures 40-1A and B.

The most common definition of undernutrition is low body weight for age, based on reference standards such those of the WHO.[9] Chronic undernutrition or *stunting* defines a deficit in longitudinal growth, and is assessed by height-for-age. *Wasting* is usually an acute or subacute form of undernutrition, in which there is a loss of body mass relative to height that occurs in a relatively short time, and is diagnosed using weight-for-height. Each of these conditions can be defined as moderate or severe, based on the degree of deficit of the corresponding indicator. Data from UNICEF/WHO/World Bank for children under 5 years[10] show that stunting has declined globally from 32.5% in 1990 to 25.7% in 2011, and is projected to fall further to 22% by 2025. By 2011, the prevalence rates in developed and developing regions were 7.2% and 28%, respectively. Since the pace of decline is substantially faster in developing countries, it is expected that this gap will continue to decrease, as has been the case for the past two decades. East Africa (42.1%) and Oceania (35.5%) are the regions with the highest stunting prevalence.[10]

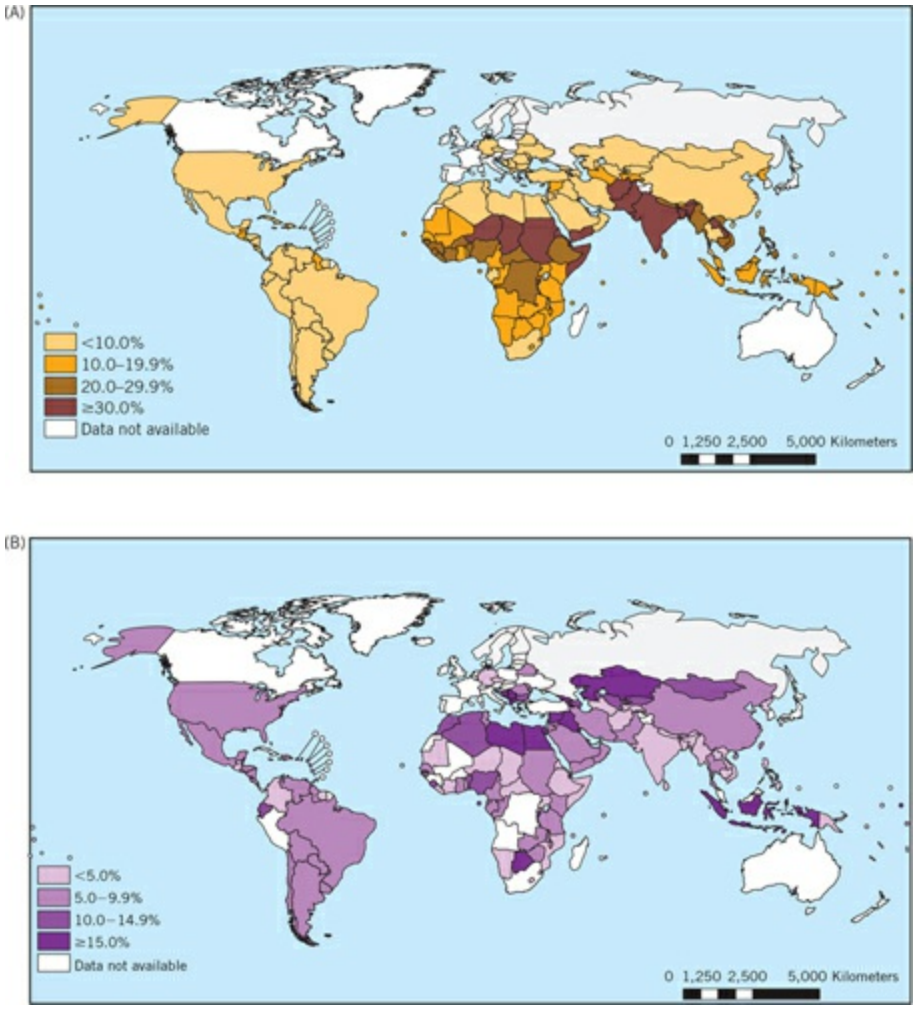


Figure 40-1 A, Global distribution of under-five prevalence of underweight (below –2 SD of the median weight-for-age). Reproduced from UNICEF-WHO-World Bank.[10] B, Global distribution of under-five prevalence of overweight (above +2 SD of the median weight-for-height). Reproduced from UNICEF-WHO-World Bank.[10]

Several studies have demonstrated an inverse correlation between stunting and cognitive and physical development.[11] Childhood stunting also leads to reduced physical work capacity in the adult[12] and has an important transgenerational effect that perpetuates the cycle of suboptimal growth and reduced productivity in a population.[13]

Wasting can occur in a previously healthy child, but most commonly occurs in children who are already stunted or that have experienced previous acute episodes of growth faltering. Acute weight loss may be caused by inadequate dietary intake due to illness or simply to

unavailability of food. A rapid drop in food availability can be caused by natural or human-made disasters, including war and forced population displacements. Wasting is less common than stunting, and while it has experienced modest but consistent declines over the past two decades, it still affects 8% of children worldwide, which represents almost 50 million children (Table 40-3), essentially all of them residing in the developing world.[14]

FETAL AND EARLY POSTNATAL GROWTH

Birth weight is a commonly used indicator of fetal growth, particularly during the last trimester of pregnancy. Low-birth-weight (LBW) infants have a 10-fold higher risk of neonatal mortality compared with newborns weighing 3–3.5 kg.[15] The prevalence of LBW in the developing world ranges from 8% in Latin America to 27% in South Asia,[14] and is on a gradual downward trend. Although prematurity is the main contributor to LBW in the developed world, the vast majority of LBW infants in developing countries are small for gestational age, reflecting intrauterine growth retardation (IUGR). The more common type of IUGR in the developing world is symmetric IUGR, indicating a sustained energy restriction throughout pregnancy.

Maternal prepregnancy weight is strongly correlated with birth weight. A low prepregnancy body mass index is associated with higher morbidity and mortality for both mother and fetus.[16] Thus, there is an intergenerational effect of stunting, because stunted girls reaching their reproductive age will have an increased risk of producing LBW babies.[13] Maternal behaviors and cultural factors determining level of physical activity and food intake also influence energy balance during pregnancy.

TABLE 40-3 Estimated Regional and Global Prevalence (%) of Undernutrition in Children Under 5 Years of Age, 2011

Region	Underweight[1]	Stunted[2]	Wasted[3]
AFRICA	17.7	35.6	8.5
Eastern	19.3	42.1	6.7
Middle	17.4	35.0	9.3
Northern	6.5	21.3	8.2
Southern	11.9	30.8	5.7

Western	21.8	36.4	10.4
ASIA	19.3	26.8	10.1
Eastern	3.1	8.5	2.3
South-Central	29.8	36.4	14.8
South-Eastern	16.6	27.4	9.7
Western	4.8	18.0	3.5
LATIN AMERICA & CARIBBEAN	3.4	13.4	1.4
Caribbean	3.5	6.7	3.4
Central America	3.9	18.6	1.0
South America	3.1	11.5	1.3
OCEANIA	14.0	35.5	4.3
Developing countries	17.4	28.0	8.8
Developed countries	2.4	7.2	1.7
World	15.7	25.8	8.0

[1] Moderate or severe underweight: below -2 standard deviations (SD) of the median for weight-for-age of the WHO reference.

[2] Moderate or severe stunting: below -2 SD of the median for height-for-age of the WHO reference.

[3] Moderate or severe wasting: below -2 SD of the median for weight-for-height of the WHO reference.

Data does not include Japan, Australia, or New Zealand.

Source: Adapted from UNCEF-WHO-World Bank.[10]

Micronutrient deficiencies can also affect fetal growth, as well as impact on obstetric mortality. Vitamin A deficiency appears to have a significant effect on maternal mortality, as suggested by studies in Nepal where vitamin A supplementation reduced mortality by 30%.[17] Other studies in Nepal reported that prenatal folic acid and iron supplementation had also a modest positive effect on birth weight.[18] Data are less clear on the effect of zinc on birth weight, although studies in Peru found significant effects of prenatal zinc supplementation on fetal neurobehavioral development.[19] Additional maternal factors increasing the risk of LBW are low maternal age (<18 years), chronic infections, such as malaria, gastrointestinal parasites, sexually transmitted diseases (STDs), and first-time births.[15]

IUGR has long-term effects beyond birth weight. Studies in the developing world have reported lower Bayley scores and lower mental development index scores in LBW infants.[20] These findings are associated with environmental factors as well, such as standard of living, housing, and parental education. The trends in these studies would suggest that

cognitive development and deficits change over time, and many of the conclusions drawn are dependent at the age at which they are actually followed up, and may actually be transient.[21]

Under the appropriate conditions, LBW infants may be able to recover their weight through "catch-up" growth during the first few years of life. The Guatemala longitudinal study[22] documented some degree of catch-up growth of infants with IUGR during the first two years of life, with subsequent stabilization at the centiles achieved by that age. Nevertheless, body weight and height deficits in 17- to 19-year-olds were evident, averaging 5 kg and 5 cm below non-IUGR children.[23]

In the postnatal period, growth performance during the first 12 months of life appears to be critical for conditioning long-term body size and vulnerability to disease. Data from Latin America, sub-Saharan Africa, and South Asia (Figure 40-2) show the dramatic decline in height-for-age over the first 12 months of life, resulting in a sustained deficit (stunting) over the following years.[24] This evidence underscores the need for early intervention to prevent growth faltering. This window of opportunity to prevent stunting is commonly known as "the 1000 days."

FETAL AND EARLY POSTNATAL GROWTH AND RISK OF ADULT DISEASE

The "Barker hypothesis" originated from Barker's observation of an association between LBW and adult cardiovascular disease.[25] These original observations evolved into an important field of research, known as the developmental origins of health and disease. Its aim is to define the role of fetal and early postnatal growth patterns on long-term susceptibility to chronic, noncommunicable chronic diseases. There is now a consistent body of evidence, including cohort studies and mechanistic experiments, documenting the impact of early growth and metabolic differentiation on long-term risk of disease. Chapter 27, Human Growth and Disease Later in Life, provides further discussion of this concept. A compelling evidence emerged from cohort studies of the Dutch famine in World War II, showing increased adiposity in adults born to mothers who experienced energy restriction during gestation.[26,27] Similarly, cohort studies in India[28] and Guatemala[29] found associations between birth weight or early postnatal growth retardation and increased adiposity, blood pressure, or insulin resistance later in life. Taken together these observations suggest that the increased risk stems from periods of accelerated growth, such as

catch-up growth, perhaps when it occurs at critical times of maturation. This interpretation has raised concern on the potential long-term adverse consequences of rehabilitation of malnourished children, which usually involve at some point high rates of growth. Some studies in nutritionally rehabilitated LBW infants have shown significantly higher rates of excess adiposity, insulin resistance, and elevated blood pressure at 3–5 years of age.[28,30,31]

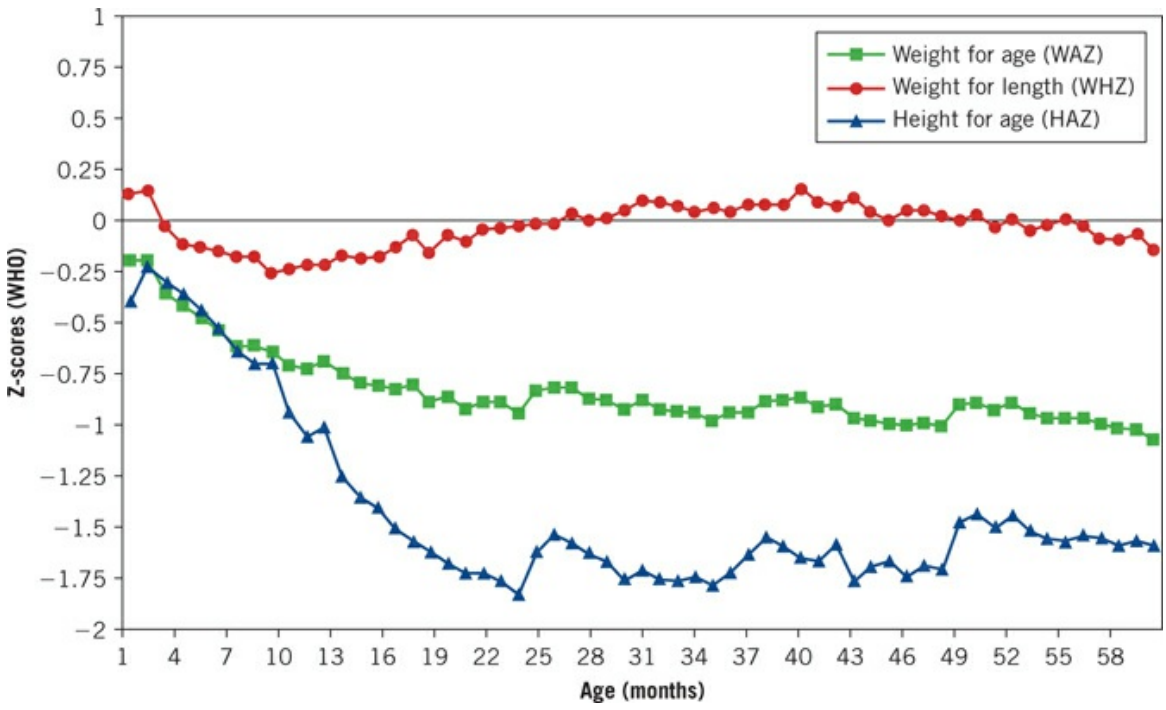


Figure 40-2 Mean anthropometric z-scores for age, relative to the WHO growth reference standard. Data from 54 countries in the WHO Global Database on Child Growth and Malnutrition. Reproduced from Victora et al.[24]

Beyond the potential risks associated with growth restriction and recovery, neonatal illness in general carries a heavy lifelong burden in developing countries. A study in Bangladesh estimated that neonatal conditions accounts for 195 years lost to disability for each 1000 neonates.[32]

UNDERNUTRITION AND SUSCEPTIBILITY TO

COMMUNICABLE DISEASES

The vicious circle of protein-energy malnutrition, impaired immune response, increased infections, and decreased food intake are well recognized since the seminal work of Scrimshaw et al.[33] In spite of the global epidemiological transition toward chronic, noncommunicable diseases, infectious diseases continue to be important disease burdens for children in developing countries. For example, a recent analysis of data from 84 countries estimated that diarrheal disease was responsible for 30% of deaths in children <5 years of age.[34]

Malnutrition affects most of the host defense systems against infection, including epithelial and mucosal integrity, mucociliary clearance, gastric acid production, immunoglobulin synthesis, and lymphocyte differentiation.[35] Micronutrient deficiencies, particularly of vitamin A and zinc, also result in severe impairment of host resistance mechanisms. The role of vitamin A on the immune system and on infectious disease morbidity and mortality has been extensively studied.[36–38] Evidence from field trials led to the policy of routinely administering high dose vitamin A supplements to children at risk of hypovitaminosis A. The role of zinc deficiency on susceptibility to infection and the efficacy of supplementation to reduce infection morbidity and mortality were also explored in several field trials, which showed that zinc supplementation significantly reduces the incidence of pneumonia and diarrhea, with a modest but significant effect on mortality as well.[39,40] See also Chapter 14 (Protein Energy Malnutrition).

Repeated, nonlife threatening infectious episodes can have a devastating effect on growth during the first years of life. The landmark work of L. Mata in the village of Santa María Cauqué in Guatemala[41] documented the slow but progressive growth delay during the first year of life in children suffering from minor but frequent episodes of acute infections, such as gastroenteritis, urinary tract infection, and upper respiratory infection. These episodes result in decreased dietary intake because of anorexia or withholding of food by caregivers, excess nutrient losses, and increased nutrient requirements.[42] In addition, the immune response to infection induces a catabolic state that may persist beyond the clinical course of the infectious episode, as shown by the pioneering studies of Beisel.[43]

MICRONUTRIENT MALNUTRITION

Single- and multiple-nutrient deficiencies are widespread, affecting over two billion persons worldwide, mostly but not only in developing countries. Iron deficiency is considered the most prevalent single-nutrient deficiency, affecting over one billion persons, and is perhaps the only micronutrient deficiency that has public health significance for developed countries as well. See Chapter 9 (Iron). We will only point out that about 40% of children in developing countries are anemic, largely because of iron deficiency.[44] Although marginal iron intake is a common factor, many times anemia is triggered by acute infectious diseases that may increase losses or reduce food intake.

Other major micronutrient deficiencies are hypovitaminosis A and iodine deficiency.[16] Although data on zinc deficiency is more limited, extensive regions with moderate or high risk for deficiency have been identified as well.[5] Along with inadequate dietary intake, infectious diseases are an important contributor to micronutrient deficiencies. They increase gastrointestinal losses and may impair effective utilization of dietary sources as well. Conversely, micronutrient deficiencies may increase susceptibility to infection, as noted above.

Vitamin A deficiency is one of the major single-nutrient deficiencies in the world. The WHO estimates that currently there are over 250 million children <5 years of age who are vitamin A deficient, and up to half-million of them will become blind every year.[44] In addition, vitamin A deficiency increases the risk of maternal mortality and may cause night blindness during pregnancy.[17,45] Among the main factors contributing to this deficiency are low consumption of animal protein sources, reduced bioavailability and bioconversion of dietary provitamin A (beta-carotene), and excess losses during episodes of acute infection.

Traditionally linked to blindness, the role of vitamin A deficiency on child mortality was demonstrated by the landmark studies of Sommer et al in the early 1980s,[46] showing that children who exhibited mild xerophthalmia had significantly higher mortality that those without signs of vitamin A deficiency. He subsequently showed that a single large dose of vitamin A resulted in a 30% reduction in child mortality.[47] Numerous subsequent trials in different countries have confirmed those findings, yielding on average a reduction of about 20% in mortality.[48] Supplementation is now a national policy in most countries with endemic hypovitaminosis A. The effect of vitamin A on mortality is likely due to its positive effects on integrity of epithelia, cell differentiation, and the immune system, thus reducing the severity of many infectious diseases,

particularly gastrointestinal and respiratory.[49,50]

Zinc deficiency is an emerging concern in the developing world, although there are less survey data than for other micronutrients, in part because of the difficulty in defining a standardized, reliable biomarker of zinc status in children. At-risk populations may be identified by assessing their dietary intake, alone or combined with other biochemical indices of zinc status. A geographic distribution of zinc deficiency was reported by Black et al.[5] Zinc supplementation trials have shown that it reduces the incidence of acute lower respiratory infections,[51] limits the severity of severe pneumonia,[39] and reduces the morbidity and mortality because of diarrhea.[52] Although most trials have administered the zinc supplement daily, weekly dosing appears to be also effective, assuming adequate compliance.[40]

Iodine deficiency continues to be the most common nutritional cause of brain damage worldwide. Fortunately, salt iodization has progressively reduced the incidence and prevalence of iodine-deficiency–related disorders, including cognitive and mental impairment, growth failure, and cretinism.[44] Although the number of countries where iodine deficiency is a public health problem has dropped by half over the past decades, the WHO estimates that there are still 54 countries with iodine deficiency in part or all of their territory. But it is the hope that with continuing efforts to promote and sustain iodine fortification, iodine deficiency may be confined to a few remote regions in selected countries.

Multiple-nutrient deficiencies are receiving increasing attention, particularly during reproductive age. The role of folic acid on pregnancy is well recognized in developed countries, but evidence reflects conditions that differ from those of developing country populations, who are likely to present folic acid deficiency combined with other micronutrient deficiencies. Field trials evaluating the impact of prenatal supplementation with multiple micronutrients on infant mortality[17] and birth weight[18] have yielded inconclusive results. Long-term follow-up (up to age 7 years) of offsprings, however, showed that the combination of folic acid–iron supplementation during pregnancy reduced mortality by 31% between birth and 7 years of age.[53] Similarly, other field trial demonstrated that antenatal supplementation with a combination of iron, folic acid, and zinc improved linear growth in schoolchildren.[54]

COMBATING MICRONUTRIENT MALNUTRITION

Based on the evidence from numerous field trials showing significant benefits from micronutrient supplementation, several countries have implemented regional or national supplement distribution programs. Although the supplement themselves are usually inexpensive, the logistics of mass distribution (transportation, coverage, etc.) are complex and difficult to sustain over time. Distribution programs, therefore, are considered an interim response to endemic deficiencies, with the ultimate goal of ensuring adequate intake through locally available foods, with or without fortification. Nevertheless, given the complexity of factors affecting food availability and intake in different populations, it is likely that micronutrient supplementation will continue to be a cost-effective option for the immediate future. See Chapter 45 (Interventions in Global Child Health) for more discussion on this topic.

Food fortification is a powerful approach to combat micronutrient malnutrition. Although its implementation and sustainability is more complex than for supplement distribution, it can take advantage of the market forces and food distribution channels, requiring little direct involvement from the target population. The requirements for a successful fortification program have been well defined.[55] A number of countries have implemented fortification programs with varied degree of success. Similarly, folic acid fortification had a significant impact on the prevalence of neural tube defects. Agricultural policies can provide incentive to rotate crops and to enhance the diversity and yield of foods grown and available. Agricultural technology, selective crop breeding, and transfer of technology may aid in increasing the micronutrient content of particular foods. Subsidization of certain seed types and of fertilizers may aid in encouraging the horticulture of certain crops as well. Finally, sustainability will ultimately depend on progress in educating consumers and on the overall behavioral change of the population.

THE NUTRITION TRANSITION

The dramatic advances in communications and transportation and the globalization of economic markets have resulted in rapid changes in diet and lifestyle in developing countries. This phenomenon, often called the nutrition transition, is a key factor in the changing disease patterns for developing countries.[56,57] Other factors are the increase in number and the aging of the world population. In turn, these changes led to a preeminent role of chronic, noncommunicable diseases as global cause of death, which by 2010 accounted for two of every three deaths worldwide.[2] The majority

of these deaths occur in low- and middle-income countries.

One of the critical consequences of the nutrition transition is the continuing rise on childhood obesity prevalence. It is estimated that there are over 42 million children <5 years of age who are overweight or obese (Table 40-4), more than 80% of them residing in the developing world.[58] By 2010, obesity prevalence in children <5 years reached 6.7%, up from 4.2 in 1990, and is expected to continue to increase in the coming years.[10] A more detailed discussion of this topic is available elsewhere,[59] and in Chapters 37, 38, 39, 40, and 47.

The three major factors driving the nutrition transition are food type and availability, urbanization, and lifestyle changes leading to reduced physical activity and increased sedentary time. Dietary changes are dominated by an overall increase in energy availability (except in sub-Saharan Africa), mostly because of higher consumption of vegetable oils and refined sugars, with modest reductions in cereals and pulses.[60] A particular phenomenon is the global increase in consumption of caloric sweeteners, primarily in beverages.[61,62] Although much debate on the role of energy-dense, low-nutrient foods (sometimes called “fast” or “junk” foods) have centered in the United States, over half the revenues of United States fast-food companies come from overseas markets, driven by their rapid growth in low- and middle-income countries.[63]

Urbanization, which occurs at a faster pace in the developing world, is having a profound effect on dietary patterns and lifestyle. The UN predicts that almost 90% of the projected population growth for the next 20 years will occur in urban areas of the developing world.[64] The urban environment has important effects on dietary and lifestyle patterns. Consumption of processed foods, usually of higher fat content, and total dietary energy intake tends to increase.[56] Even the modest improvement in income that may be associated with urban migration may initially have a detrimental effect on diet quality, by providing access to processed, energy-dense foods, as is suggested by data from Brazil[65] and China.[66] Urban families become dependent on the cash food market, which exhibit high price elasticity, thus having the potential for driving food choices in low-income populations.

TABLE 40-4 Estimated Regional and Global Prevalence (%) of Overweight in Children Under 5 Years of Age in 1990 and 2011

Region	1990	2011
AFRICA	4.2	7.3
Eastern	4.4	5.0
Middle	3.5	5.8
Northern	7.3	13.1
Southern	6.1	16.3
Western	1.9	6.6
ASIA	3.7	4.7
Eastern	6.8	5.5
South-Central	2.0	3.1
South-Eastern	1.8	6.1
Western	4.4	11.3
LATIN AMERICA & CARIBBEAN	6.5	7.1
Caribbean	4.0	7.5
Central America	5.1	6.4
South America	7.3	7.4
OCEANIA	2.6	3.7
Developing countries	4.1	5.7
Developed countries	7.4	14.5
World	4.5	6.6

Overweight: above +2 SD of the median weight-for-height of the WHO reference.

Source: Adapted from UNICEF.[14]

Daily energy expenditure in the urban setting is usually substantially lower than in rural areas. The intense physical labor of rural work in the fields is replaced by sedentary, low-energy work.[67,68] Television viewing is also a well-documented factor in increasing physical inactivity in children,[69] and can also negatively affect consumption of healthy foods.[70] An increased energy intake and a reduced energy output lead to excess weight gain and obesity.

THE NUTRITION TRANSITION, POVERTY, AND CHILD HEALTH

The nutrition transition, particularly urbanization, has an important impact on child health. Over one billion children already reside in urban areas, and this figure will continue to increase in the foreseeable future.[14] While

urban dwelling may bring some benefits, such as better sanitation, access to health services, and job opportunities, at the same time it frequently magnifies social and economic disparities between rich and poor. Unemployment, insecure housing, street violence, and social isolation draw a sharper contrast between those who benefit from urbanization and those who are negatively affected by it. Indeed, some health indicators of child health show a significantly larger gap between rich and poor in urban than in rural areas.[71,72] Furthermore, some of the potential benefits, such as sanitation, are not keeping up with population growth in urban areas, and may remain a privilege for the few.[14]

The health effects (specifically on obesity prevalence) of socioeconomic development are the result of multiple factors, and are by no means linear. Low- and middle-income countries show different associations between gross domestic product and obesity prevalence.[73] An analysis by Jones-Smith et al[74] showed that the differential effects of increases in GDP on obesity rates may be mediated by differences in income inequalities within country.

Although the epicenter of the global crisis of 2007–2008 was in the developed economies, it has caused a worldwide recession affecting virtually every country. According to the International Labor Organization, in 2012 there were 25 million more unemployed than in 2007, with rates twice as high in the younger population.[75] The potential effect of these changes on health outcomes is still not clear, but major economic readjustments under way in many LMIC may result in further constraints in basic health care services for the poor.

CONFRONTING THE EFFECTS OF THE NUTRITION TRANSITION

There is compelling evidence of the impact that socioeconomic development and the nutrition transition are having on global health. A handful of modifiable risk factors, including tobacco and alcohol use, low consumption of fruits and vegetables, and physical inactivity, are having a disproportionate impact on disease burden. The WHO and other organizations have developed a global framework to combat these risk factors, endorsed by all UN countries.[76,77] Along with the MDG,[7] they provide a consistent action plan. Indeed, a number of countries have made significant progress toward the MDG, and practical experiences on preventing obesity in children have been disseminated.[78] But the MDG do

not address directly the problem of chronic, noncommunicable diseases. To confront these, the environment favoring obesity, predominant today in many countries, need to be changed. Policy decisions are necessary to increase the availability and affordability of healthy foods, limit access and promotion of unhealthy foods to children, create opportunities for safe recreational physical activity and reduce sedentary time at home and school. Some countries that are at an advanced stage of the nutrition transition, such as Brazil and Mexico, have begun to implement some of these policies, and can offer a template for others to follow.

REFERENCES

1. Caballero B, Popkin BM. *The Nutrition Transition: Diet and Disease in the Developing World*. London, UK: Academic Press; 2002.
2. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet*. 2013;380(9859):2095–2128.
3. Lim SS, Vos T, Flaxman AD, et al. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet*. 2013;380(9859):2224–2260.
4. Murray CJ, Vos T, Lozano R, et al. Disability-adjusted life years (DALYs) for 291 diseases and injuries in 21 regions, 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. *Lancet*. 2013;380(9859):2197–2223.
5. Black RE, Allen LH, Bhutta ZA, et al. Maternal and child undernutrition: global and regional exposures and health consequences. *Lancet*. 2008;371(9608):243–260.
6. Schofield C, Ashworth A. Why have mortality rates for severe malnutrition remained so high? *Bull World Health Organ*. 1996;74(2):223–229.
7. United Nations. *Millennium Development Goals* http://www.un.org/millenniumgoals/. Accessed March 4, 2013.
8. Hill K, You D, Inoue M, et al. Child mortality estimation: accelerated progress in reducing global child mortality, 1990-2010. *PLoS Med*. 2012;9(8):e1001303.
9. WHO. *WHO Child Growth Standards*. Geneva, Switzerland: WHO; 2006.
10. UNICEF-WHO-World Bank. *Joint Child Malnutrition Estimates*. New York, NY; Geneva, Switzerland; Washington, DC: Bank U-W-W; 2012.
11. Pollitt E, Gorman K, Engle PL, et al. Early supplementary feeding and cognition - effects over 2 decades. *Monogr Soc Res Child Dev*. 1993;58(7):R5–R98.
12. Haas JD, Murdoch S, Rivera J, et al. Early nutrition and later physical work capacity. *Nutr Rev*. 1996;54(2 pt 2):S41–S48.
13. Ramakrishnan U, Martorell R, Schroeder DG, et al. Role of intergenerational effects on linear growth. *J Nutr*. 1999;129(suppl 2):544S–549S.
14. UNICEF. *The State of the World's Children 2012*. New York, NY: UNICEF; 2012.
15. Sub-Committee on Nutrition, U.N. Low birth weight: report of a meeting. Dhaka, Bangladesh. *Bull World Health Organ*. 2000;(18).
16. West KP Jr, Caballero B, Black RE. Nutrition. In: Merson M, Black RE, Mills AJ, eds. *International Public Health: Diseases, Programs and Policies*. Burlington,

MA: Jones & Bartlett; 2011:187–237.
17. Christian P, West KP, Khatry SK, et al. Effects of maternal micronutrient supplementation on fetal loss and infant mortality: a cluster-randomized trial in Nepal. *Am J Clin Nutr*. 2003;**78**(6):1194–1202.
18. Christian P, Khatry SK, Katz J, et al. Effects of alternative maternal micronutrient supplements on low birth weight in rural Nepal: double blind randomised community trial. *BMJ*. 2003;326(7389):571.
19. Caulfield LE, Zavaleta N, Chen P, et al. Maternal zinc supplementation during pregnancy affects autonomic function of Peruvian children assessed at 54 months of age. *J Nutr*. 2011;141(2):327–332.
20. Goldenberg R, Hack M, Grantham M, et al. Report of the IDECG/IUNS working group on IUGR effects on neurological, sensory, cognitive, and behavioral function. *Eur J Clin Nutr*. 1998;52:S100–S101.
21. Hack M. Effects of intrauterine growth retardation on mental performance and behavior, outcomes during adolescence and adulthood. *Eur J Clin Nutr*. 1998;52:S65–S71.
22. Martorell R. Results and implications of the INCAP follow-up study. *J Nutr*. 1995;125(suppl 4):1127S–1138S.
23. Martorell R, Ramakrishnan U, Schroeder DG, et al. Intrauterine growth retardation, body size, body composition and physical performance in adolescence. *Eur J Clin Nutr*. 1998;52(suppl 1):S43–S52.
24. Victora CG, de Onis M, Hallal PC, et al. Worldwide timing of growth faltering: revisiting implications for interventions. *Pediatrics*. 2010;125(3):e473–e480.
25. Barker DJ, Osmond C, Law CM. The intrauterine and early postnatal origins of cardiovascular disease and chronic bronchitis. *J Epidemiol Community Health*. 1989;43(3):237–240.
26. Ravelli GP, Stein ZA, Susser MW. Obesity in young men after famine exposure in utero and early infancy. *N EnglJ Med*. 1976;295(7):349–353.
27. Ravelli AC, Der Meulen JH, Osmond C, et al. Obesity at the age of 50 y in men and women exposed to famine prenatally. *Am J Clin Nutr*. 1999;70(5):811–816.
28. Joglekar CV, Fall CH, Deshpande VU, et al. Newborn size, infant and childhood growth, and body composition and cardiovascular disease risk factors at the age of 6 years: the Pune Maternal Nutrition Study. *Int J Obes (Lond)*. 2007;31(10):1534–1544.
29. Martorell R, Stein AD, Schroeder DG. Early nutrition and later adiposity. *J Nutr*. 2001;131(3):874S–880S.
30. Ibanez L, Ong K, Dunger DB, et al. Early development of adiposity and insulin resistance after catch-up weight gain in small-for-gestational-age children. *J Clin Endocrinol Metab*. 2006;91(6):2153–2158.
31. Ibanez L, Suarez L, Lopez-Bermejo A, et al. Early development of visceral fat excess after spontaneous catch-up growth in children with low birth weight. *J Clin Endocrinol Metab*. 2008;**93**(3):925–928.
32. Shillcutt SD, Lefevre AE, Lee AC, et al. Forecasting burden of long-term disability from neonatal conditions: results from the Projahnmo I trial, Sylhet, Bangladesh. *Health Policy Plan.* 2013;28:435–452.
33. Scrimshaw NS, Taylor CE, Gordon JE. *Interactions of Nutrition and Infection*. Geneva, Switzerland: WHO; 1968.
34. Walker CL, Aryee MJ, Boschi-Pinto C, et al. Estimating diarrhea mortality among young children in low and middle income countries. *PLoS One*. 2012;7(1):e29151.
35. Rice AL, Sacco L, Hyder A, et al. Malnutrition as an underlying cause of childhood deaths associated with infectious diseases in developing countries. *Bull*

World Health Organ. 2000;78(10):1207–1221.
36. Sommer A. Vitamin A status, resistance to infection, and childhood mortality. *Ann N Y Acad Sci*. 1990;587:17–23.
37. Sommer A, Katz J, Tarwotjo I. Increased risk of respiratory disease and diarrhea in children with pre-existing mild vitamin A deficiency. *Am J Clin Nutr*. 1984;40:1090–1095.
38. Humphrey JH, Agoestina T, Wu L, et al. Impact of neonatal vitamin A supplementation on infant morbidity and mortality. *J Pediatr*. 1996;128(4):489–496.
39. Brooks WA, Yunus M, Santosham M, et al. Zinc for severe pneumonia in very young children: double-blind placebo-controlled trial. *Lancet*. 2004;363(9422):1683–1688.
40. Brooks WA, Santosham M, Naheed A, et al. Effect of weekly zinc supplements on incidence of pneumonia and diarrhoea in children younger than 2 years in an urban, low-income population in Bangladesh: randomised controlled trial. *Lancet*. 2005;366(9490):999–1004.
41. Mata LJ. *The Children of Santa Maria Cauque: A Prospective Field Study of Health and Growth*. Cambridge, MA: MIT Press; 1978.
42. Mata LJ, Urrutia JJ, Lechtig A. Infection and nutrition of children of a low socioeconomic rural community. *Am J Clin Nutr*. 1971;35:249–259.
43. Beisel WR. Metabolic response to infection. *Ann Rev Med*. 1975;26:9–20.
44. World Health Organization. *Micronutrient Deficiencies* http://www.who.int/nutrition/topics/vad/en/index.html. Accessed March 4, 2013.
45. Christian P, West KP Jr, Khatry SK, et al. Maternal night blindness increases risk of mortality in the first 6 months of life among infants in Nepal. *J Nutr*. 2001;131(5):1510–1512.
46. Sommer A. Mortality associated with mild, untreated xerophthalmia. Trans. *Am Ophthalmol Soc*. 1983;81:825–853.
47. Sommer A, Tarwotjo I, Djunaedi E, et al. Impact of vitamin A supplementation on childhood mortality. *Lancet*. 1986;1:1169–1173.
48. Beaton GH, Martorell R, Aronson KJ, et al. *Effectiveness of Vitamin A Supplementation in the Control of Young Child Morbidity & Mortality in Developing Countries*. Toronto, Ontario, Canada: International Nutrition Program; 1993.
49. Villamor E, Fawzi WW. Vitamin A supplementation: implications for morbidity and mortality in children. *J Infect Dis*. 2000;182(suppl 1):S122–S133.
50. Sommer A, West Keith P Jr. *Vitamin A Deficiency: Health, Survival and Vision*. New York, NY: Oxford University Press; 1996.
51. Sazawal S, Black RE, Jalla S, et al. Zinc supplementation reduces the incidence of acute lower respiratory infections in infants and preschool children: a double-blind, controlled trial. *Pediatrics*. 1998;102(1 pt 1):1–5.
52. Baqui AH, Black RE, El Arifeen S, et al. Effect of zinc supplementation started during diarrhoea on morbidity and mortality in Bangladeshi children: community randomised trial. *BMJ*. 2002;325(7372):1059.
53. Christian P, Stewart CP, LeClerq SC, et al. Antenatal and postnatal iron supplementation and childhood mortality in rural Nepal: a prospective follow-up in a randomized, controlled community trial. *Am J Epidemiol*. 2009;170(9):1127–1136.
54. Stewart CP, Christian P, LeClerq SC, et al. Antenatal supplementation with folic acid + iron + zinc improves linear growth and reduces peripheral adiposity in school-age children in rural Nepal. *Am J Clin Nutr*. 2009;90(1):132–140.

55. Institute of Medicine. *Prevention of Micronutrient Deficiencies: Tools for Policy Makers and Public Health Workers*. Washington DC: National Academy Press; 1998.
56. Popkin BM, Adair LS, Ng SW. Global nutrition transition and the pandemic of obesity in developing countries. *Nutr Rev*. 2012;70(1):3–21.
57. Caballero B. The nutrition transition: global trends in diet, lifestyle, and non-communicable diseases. In: Ross C, Caballero B, Cousins RJ, et al., eds. *Modern Nutrition in Health and Disease*. 11th ed. Philadelphia, PA: LWW; 2013:1511–1517.
58. de Onis M, Blossner M, Borghi E. Global prevalence and trends of overweight and obesity among preschool children. *Am J Clin Nutr*. 2010;92(5):1257–1264.
59. Caballero B. Childhood obesity in low- and middle-income countries. *BMC Med*. In press.
60. Food and Agriculture Organization. FAOSTAT. Food Production. http://faostat.fao.org/site/339/default.aspx. Accessed March 4, 2013.
61. Barquera S, Campirano F, Bonvecchio A, et al. Caloric beverage consumption patterns in Mexican children. *Nutr J*. 2010;9:47.
62. Popkin BM. Contemporary nutritional transition: determinants of diet and its impact on body composition. *Proc Nutr Soc*. 2011;70(1):82–91.
63. Tandon, S, Landes M, Woolverton A: The Expansion of Modern Grocery Retailing and Trade in Developing Countries. *Economic Research Service Report No*. 122. Washington, DC: USDA; 2011;2–20.
64. United Nations Population Division. *World Population Prospects, the 2010 Revision* http://esa.un.org/unpd/wpp/index.htm. Accessed March 4, 2013.
65. Monteiro CA, Conde WL, Lu B, et al. Obesity and inequities in health in the developing world. *Int J Obes Relat Metab Disord*. 2004;28:1181–1186
66. Du S, Mroz TA, Zhai F, et al. Rapid income growth adversely affects diet quality in China—particularly for the poor! *Soc Sci Med*. 2004;59(7):1505–1515.
67. Ng SW, Norton EC, Popkin BM. Why have physical activity levels declined among Chinese adults? Findings from the 1991-2006 China Health and Nutrition Surveys. *Soc Sci Med*. 2009;68(7):1305–1314.
68. Monda KL, Gordon-Larsen P, Stevens J, et al. China's transition: the effect of rapid urbanization on adult occupational physical activity. *Soc Sci Med*. 2007;64(4):858–870.
69. Dietz WH Jr, Gortmaker SL. Do we fatten our children at the television set? Obesity and television viewing in children and adolescents. *Pediatrics*. 1985;75(5):807–812.
70. Boynton-Jarrett R, Thomas TN, Peterson KE, et al. Impact of television viewing patterns on fruit and vegetable consumption among adolescents. *Pediatrics*. 2003;112(6 pt 1):1321–1326.
71. Menon P, Ruel M, Morris SS. *Socioeconomic Differentials in Child Stunting Are Consistently Larger in Urban Than in Rural Areas* http://www.ifpri.org/publication/socio-economic-differentials-child-stunting-are-consistently-larger-urban-rural-areas-0. Accessed March 4, 2013.
72. Gwatkin DR, Guillot M, Heuveline P. The burden of disease among the urban poor. *Lancet*. 1999;354:586–589.
73. Subramanian SV, Perkins JM, Ozaltin E, et al. Weight of nations: a socioeconomic analysis of women in low- to middle-income countries. *Am J Clin Nutr*. 2011;93(2):413–421.
74. Jones-Smith JC, Gordon-Larsen P, Siddiqi A, et al. Cross-national comparisons of time trends in overweight inequality by socioeconomic status among women using

repeated cross-sectional surveys from 37 developing countries, 1989-2007. *Am J Epidemiol*. 2011;173(6):667–675.
75. International Labour Organization. *Global Employment Trends 2013*. Geneva, Switzerland: Organization IL; 2013.
76. WHO. *Framework Convention for Tobacco Control* http://www.who.int/tobacco/framework/en/. Accessed March 4, 2013.
77. WHO. *Diet, Nutrition and the Prevention of Chronic Diseases: Report of a Joint WHO/FAO Expert Consultation*. Geneva, Switzerland: WHO; 2002.
78. WHO. *Population-Based Approaches to Childhood Obesity Prevention*. Geneva, Switzerland: WHO; 2012.

Infant and Young Child Feeding in Low- and Middle-Income Countries

41

Souheila Abbeddou, MSc, PhD and Kenneth H. Brown, MD

INTRODUCTION

Breast-feeding and complementary feeding are key contributors to children's nutrition and health during the first two years of life.[1] In this chapter, we provide an overview of (1) current recommendations regarding optimal breast-feeding practices and the appropriate timing of introduction of complementary feeding, (2) estimated energy and nutrient requirements from complementary foods for children of different ages, and (3) available intervention strategies to encourage the recommended practices and ensure that young children consume adequate amounts of energy and nutrients during this particularly vulnerable period. We briefly describe the evidence supporting current feeding recommendations, the prevalence of recommended practices in low- and middle-income countries (LMICs), and examples of the potential impact of interventions to improve these practices.

Throughout the chapter, we adopt the infant and young child feeding

definitions proposed by the World Health Organization (WHO). In particular, “early initiation of breast-feeding” refers to breast-feeding within the first hour postpartum and “exclusive breast-feeding” refers to the consumption of no foods or liquids other than breast milk and possibly small amounts of medicines or vitamin–mineral supplements.[2] Infants are considered “predominantly breast-fed” when they occasionally receive small amounts of other liquids in addition to breast milk. The period of “complementary feeding” refers to the stage of infant and child feeding when other foods and/or energy-containing liquids are provided to the young child along with breast milk,[3] and “complementary foods” are the non-breast-milk components of mixed diets consumed by young children while they are still breast-feeding. In some situations, where breast-feeding is either temporarily interrupted or replaced due to pathological conditions of the mother or the infant or because the mother elects not to breast-feed, other dietary practices are necessary. Alternative feeding practices employing breast-milk substitutes are discussed in a separate chapter.

BREAST-FEEDING

Timing of Initiation

During the first 2–4 days of early lactogenesis, the mammary gland secretes colostrum, which is a concentrated mixture of essential nutrients, immunoglobulins, and other protective factors.[4] A recent observational study of more than 10,000 Ghanaian newborns who survived beyond the first day postpartum found a direct relationship between delays in the initiation of breast-feeding and the risk of newborn deaths during the period from 2 to 28 days postpartum. Initiation of breast-feeding after the first day was associated with a 2.4-fold increased risk of neonatal mortality compared with introduction on the first day, regardless of the type of breast-feeding practices (exclusive or nonexclusive). Introduction of prelacteal feeds (fluids or foods) before initiating breast-feeding was also associated with a significantly increased mortality risk; nonexclusively breast-fed infants had higher neonatal mortality rates than those who were breast-fed exclusively, even when controlling for the time of introduction of breast-feeding. These results indicate that both the time of initiation of breast-feeding and the type of breast-feeding practices affect neonatal mortality risk. The authors estimated that in this study population, in which approximately 40% of mothers started breast-feeding within one hour of delivery, the overall neonatal mortality rate from day 2 to 28

postpartum could be reduced by as much as 22% if all women would adopt the recommended practice of early initiation of breast-feeding.[5,6]

Early initiation of breast-feeding may protect the newborn via several possible mechanisms, including (1) enhanced milk production by the mammary gland and greater subsequent sustainability of exclusive breast-feeding; (2) accelerated maturation of the infant digestive tract and colonization by protective microbiota, thereby enhancing resistance to enteric infections; (3) protection from exposure to potentially contaminated liquids or breast-milk substitutes; and (4) reduced risk of hypothermia afforded by the close contact between mother and child.[4,5]

Statistics on the global prevalence of early initiation of breast-feeding indicate that fewer than half the infants born each year (42% globally in 2011, 52% in LMICs) are put to the breast within the first hour. The lowest rates of early breast-feeding initiation occur in South Asia (39%) and the highest in Eastern and Southern Africa (56%).[4]

Intervention strategies to promote early breast-feeding include (1) informing mothers about the benefits of early breast-feeding well before the anticipated delivery date, (2) assisting mothers to recognize when newborns are ready to start breast-feeding, (3) ensuring that health workers communicate and encourage mothers to start breast-feeding within the first hour, and (4) providing the same guidance regardless of whether the mother gives birth in a health facility or at home.

Key point

- Increases in early initiation of breast-feeding within the first hour postpartum can reduce neonatal mortality.

Duration of Exclusive Breast-Feeding

Compared with other forms of infant feeding, exclusive breast-feeding during the first half of infancy is associated with reduced risks of gastrointestinal, respiratory and other infections.[7–9] Consistent with these results from observational studies, a randomized trial to promote exclusive breast-feeding in India found that the prevalence of diarrhea was significantly less (relative risk = 0.64 [confidence interval: 0.44–0.95] at three months and 0.85 [CI: 0.72–0.99] at six months) in the intervention communities, where 79% of infants were exclusively or predominantly breast-fed at three months, compared to the control communities, where

only 48% of infants adhered to these feeding practices.[10] A pooled analysis of the relationships between infant feeding practices and mortality risk in six countries (Brazil, The Gambia, Ghana, Pakistan, the Philippines, and Senegal) found that infants who were not exclusively or predominantly breast-fed had an increased risk of dying, especially at younger ages (odds ratio = 5.8 [95% CI: 3.4–9.8] from 0 to 2 months, 4.1 [2.7 – 6.4] from 2 to 3 months, 2.6 [1.6 – 3.9] from 4 to 5 months, and 1.8 [1.2 – 2.8] from 6 to 8 months).[11]

During the first six months of life, exclusive breast-feeding provides sufficient water, energy, and macro- and micronutrients to satisfy the growth requirements of normal birth weight, term infants.[12,13] Several randomized trials have examined whether introduction of complementary foods before six months enhances the growth of infants from four to six months compared with exclusive breast-feeding for six months. According to studies conducted both in low-income countries (LICs; Honduras,[14,15] India[10]) and more affluent settings (Belarus;[16] Iceland[17]), earlier introduction of complementary foods does not promote greater growth rates. These results, combined with the aforementioned information on the beneficial effects of exclusive breast-feeding for reducing the risks of infant morbidity and mortality, provide the rationale for recommending exclusive breast-feeding until six months.[8]

Infants born to HIV-infected mothers face an increased risk of transmission of HIV infection through breast milk unless their mothers are taking antiretroviral therapy.[18] At the same time, because exclusive breast-feeding protects the infant from other infections, breast-feeding may still be more favorable for infant survival in many LMIC settings. In view of these trade-offs, in countries with high infant mortality rates WHO recommends “exclusive breast-feeding for HIV-infected mothers for the first six months of life unless replacement feeding is acceptable, feasible, affordable, sustainable and safe for them and their infants before that time.”[18] More information on feeding infants of HIV-infected mothers is presented in Chapter 46.

Prevalence of Exclusive Breast-Feeding in Low- and Middle-Income Countries

Despite the convincing scientific evidence on the protective role of exclusive breast-feeding, and the related WHO recommendations on infant and young child feeding practices, the prevalence of exclusive breast-feeding remains disappointingly low. The global prevalence of exclusive

breast-feeding until six months was 39% for the period from 2006 to 2010,[4] ranging from 25% in Western and Central Africa to 52% in Eastern and Southern Africa.[4] In many LMICs, administration of water and other non-milk liquids is common,[19] indicating that the rates of exclusive breast-feeding could be substantially improved just by eliminating these nonnutritive beverages. On the basis of the increased mortality risk of suboptimal infant-feeding practices and the current prevalence of these practices, the recent *Lancet* series on maternal and child nutrition estimated that approximately 800,000 child deaths a year (11.6% of total child deaths) are attributable to poor breast-feeding practices.[20]

Strategies to Promote Exclusive Breast-Feeding

Promotion of appropriate breast-feeding practices is possible through health system contacts, such as with the Baby-Friendly Hospital Initiative,[21] and community-based interventions, such as peer counseling groups.[22,23] A recent systematic review of the effects of breast-feeding promotion concluded that it is possible to achieve substantial increases in any breast-feeding and in exclusive breast-feeding at both 4–6 weeks and six months.[24] Both prenatal and postnatal counseling were deemed to be important to achieve maximum impact. Another review of this topic similarly concluded that using multiple channels of communication to reach caregivers and those who influence them yields greater impact than single channel approaches.[25] As one example, an effectiveness trial in India used multiple communication channels before and after delivery to promote appropriate breast-feeding practices, including monthly contacts by a nurse midwife, quarterly contacts by community health agents, and monthly meetings convened by a community representative.[10] The prevalence of exclusive breast-feeding reached 79% in the intervention communities, compared with 48% in the nonintervention communities, and was accompanied by reductions in diarrhea, as described above.

National policies need to be formulated to protect breast-feeding, for example, through adoption and implementation of the International Code of Marketing of Breast-milk Substitutes.[26] Protection of breast-feeding is particularly challenging in LMICs because of limited systematic oversight of private sector marketing practices. Regrettably, studies continue to show that health workers accept free samples of milk and other gifts from unscrupulous companies that inappropriately market breast-milk substitutes and/or help disseminate misleading or incorrect infant feeding information produced by these companies.[27–29] It is also important to

recognize the critical roles of government and civil society in supporting appropriate infant and young child feeding practices. A recent systemic review indicated that there are failures at multiple levels of the monitoring and control process, which need to be addressed by more effective government activities and participation of key stakeholders.[30,31]

Effect of Maternal Nutritional Status and Dietary Intake on Breast-Milk Volume and Composition

Studies show that unless a woman is severely wasted (i.e., chronically energy deficient with a body mass index [BMI] <18.5 kg/m^2),[32] she is generally able to produce an adequate volume of milk. For example, studies of low BMI Bangladeshi women (mean BMI 17.8 kg/m^2) found that they produced an average of 750 mL/d of breast milk when their infants were 5–7 months of age,[33] which is similar to reports from more affluent countries (Sweden,[34] USA[35]). Protein and carbohydrate contents of mature human milk are largely unaffected by maternal nutritional status and diet.[33,36] Milk fat content (and, thus, milk energy content) is lower in thin women; however, infants who are allowed to nurse on demand generally compensate for this by consuming a higher volume of milk.[3] Although the protein content of human milk is considerably less than in cow milk, protein is not a limiting factor in the growth of exclusively breast-fed infants. In a randomized trial in Honduras, infants who continued to be exclusively breast-fed from four to six months had the same growth pattern as infants who were given additional foods that provided 20% more total daily protein intake, thus demonstrating that the lower protein intake associated with exclusive breast-feeding does not limit infant growth.[3,37]

Whereas some vitamins and minerals are excreted in mature breast milk in relation to maternal nutritional status and dietary (or supplement) intake, others are secreted in regulated amounts, regardless of maternal nutritional conditions. The former set of nutrients, that is, those that are affected by maternal body stores and dietary intake, are vitamin A, thiamin, riboflavin, vitamin B6, vitamin B12, iodine, and selenium. Maternal dietary deficiency of these nutrients reduces their concentrations in milk and may adversely affect infant nutritional status and health (e.g., resulting in infant beriberi in case of maternal thiamin deficiency[38] or increased risk of night blindness and xerophthalmia in case of vitamin A deficiency[39]). The concentration of these nutrients in breast milk can be rapidly restored by increasing maternal intake. The second set of nutrients,

which are not affected by maternal intake or stores, are calcium, copper, iron, zinc, folate, and vitamin D. Although maternal deficiency of these micronutrients does not affect their excretion in breast milk, continued breast-feeding may contribute further to the woman's poor nutritional status. Thus, maternal supplementation with these nutrients is more important for the mother's nutritional adequacy than for her infant's.[3]

Summary information on average breast milk nutrient composition of presumably well-nourished U.S. women is presented in Table 41-1, based on a report prepared previously by the U.S. Institute of Medicine.[40] As indicated in the table, the iron content of mature human milk is very low. Nevertheless, infants are able to draw on tissue iron reserves accumulated during fetal life, so normal-weight term infants generally have adequate iron status at six months. However, small-for-gestational-age and premature infants often require iron supplements before six months to prevent iron deficiency anemia.[41] Another approach to preventing iron deficiency in young infants is to avoid premature clamping of the umbilical cord at the time of delivery.[42]

The zinc content of human milk is relatively high during the first few months postpartum, but declines to fairly low levels thereafter.[43] Nevertheless, the combination of breast milk and newborn hepatic zinc reserves supplies all of the theoretical zinc needs for at least the first several months of life. Recent WHO guidelines no longer recommend early postpartum maternal vitamin A supplementation for the prevention of maternal and infant morbidity and mortality because of weak evidence of efficacy.[44] However, these guidelines do not address the effect of maternal vitamin A supplementation on breast milk vitamin A content, which may benefit infant vitamin A status.[39,45]

Exclusively breast-fed infants of mothers who are complete vegetarians or whose diet is very low in animal-source foods may exhibit signs of vitamin B12 deficiency. Direct supplementation of lactating women does not seem to increase breast milk vitamin B12 content substantially,[46] so it may be necessary to initiate maternal supplementation before or during pregnancy or provide parenteral vitamin B12 if the deficiency is pronounced. Information on the risk of other specific micronutrient deficiencies during the period of exclusive breast-feeding is generally limited; and, as described above, may depend in part on maternal nutritional status and related aspects of breast milk composition.

TABLE 41-1 Average Protein and Micronutrient

Composition of Mature Human Milk[3]

Nutrient	Amount in Mature Human Milk
Protein (g/L)	10.5 ± 2.0
Folate (µg/L)	85 ± 37
Niacin (mg/L)	1.50 ± 0.20
Riboflavin (mg/L)	0.35 ± 0.025
Thiamin (mg/L)	0.21 ± 0.03
Vitamin B_6 (µg/L)	93 ± 8
Vitamin B_{12} (µg/L)	0.97
Vitamin A (µg retinol equivalents/L)[47]	500
Vitamin D (µg/L)	0.55 ± 0.10
Calcium (mg/L)	280 ± 26
Iron (mg/L)	0.30 ± 0.10
Zinc (mg/L)[a]	1.2 ± 0.2

[a] The figure 1.2 mg Zn/L refers to early lactation. A recent review concluded that the zinc concentration of human milk declines during the first few weeks and months following delivery, with average values of 0.93 ± 0.58 mg/L at 6-8 months and 0.77 ± 0.51 mg/L at 9 months post-partum.[43]

Key points

- In LMICs, exclusive breast-feeding for six months decreases the risk of diarrhea and other infections compared to earlier introduction of complementary feeding or discontinuation of breast-feeding.
- Exclusive breast-feeding during the first 6 months of life and continued breast-feeding up to 2 years of age is associated with a decreased risk of under-5 mortality.
- There is no evidence of enhanced growth when complementary foods are introduced before six months.
- These combined sets of information indicate that exclusive breast-feeding during the first six months postpartum and continued breast-feeding thereafter should continue to be supported as public health policy in both low-income and more affluent countries.
- Pre- and postnatal counseling via multiple channels of communication can increase appropriate breast-feeding practices.
- In undernourished mothers, maternal dietary supplementation

may be advisable, both to optimize breast milk nutrient concentrations and to prevent further maternal depletion.

COMPLEMENTARY FEEDING IN LOW-INCOME COUNTRIES

Complementary Feeding

As children reach approximately 6 months of age, breast milk alone may no longer satisfy energy and nutrient needs of the growing infant, so complementary feeding needs to be initiated. Complementary feeding refers to the consumption of both breast milk and other foods, usually during the period from 6 to 24 months of age, until the child ceases to nurse at the breast, at which time the child is usually consuming the same foods as the rest of the family. Complementary food is defined as any non-breast milk foods or nutritive liquids that are given to young children during this period of complementary feeding.[3]

Despite current WHO recommendations that infants should begin receiving nutritionally adequate and safe complementary foods at six months of age,[3] global data collected from 2007 to 2011 indicate that only 60% of children had started receiving soft, semisolid, or solid food by 6–8 months of age.[4] For example, Figure 41-1 portrays the actual child feeding practices in Nigeria compared with the scenario recommended by WHO. The Nigerian data indicate that a sizeable proportion of infants fail to receive adequate complementary foods from 6 to 11 months of age, and a large number are weaned from the breast prematurely.

Energy Requirements From Complementary Foods

The average amount of energy that should be provided from complementary foods has been calculated for children of different ages based on differences between their average energy requirements and the average amount of energy provided by breast milk. Energy requirements of children of different ages have been estimated from direct measurements of energy expenditure and the amount of energy contained in newly accrued tissue. The average amounts of energy provided by breast milk were determined using pooled data from breast-fed children in low-income countries.[3] The amounts of energy provided by breast milk were then subtracted from the amounts of energy required by children in each age group to determine the average amounts of energy needed from

complementary food. According to these calculations, the average amounts of energy required from complementary food are approximately 200, 300, and 550 kcal/d for infants 6–8, 9–11, and 12–23 months of age, respectively (Table 41-2).[48]

Several caveats must be recognized in using these estimates. First, there is considerable interindividual variation in young children's energy requirements, so the needs of specific children may differ substantially from the average value. Second, the volume and composition of breast milk vary among women. Thus, the total amount of energy provided to the child from complementary foods may need to be adjusted according to the child's usual level of consumption and pattern of growth, although the average figures can be used as a first approximation of the child's needs.

Energy Density and Frequency of Feeding Complementary Foods

The total daily amount of energy consumed from complementary foods is a product of the frequency of feeding (number of servings per day), the amount of food provided and the energy density of these foods (kcal/g prepared food weight) (Figure 41-2).[3] Using this combined set of information, it is possible to formulate appropriate complementary foods to satisfy daily energy requirements of children of different ages (Table 41-3). As indicated in the table, when a child is served complementary foods fewer times per day, the foods must be of greater energy density to achieve desired levels of energy intake, and vice versa. Guidelines are available on the appropriate frequency of feeding complementary foods for children of different ages.[1]

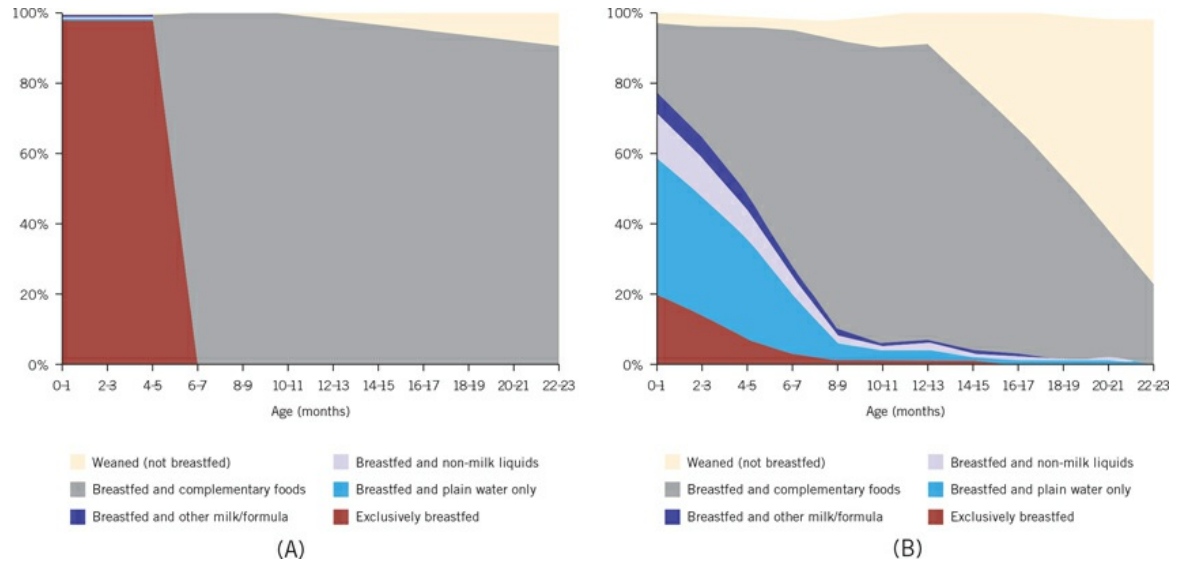


Figure 41-1 Timing of introducing complementary feeding. A, The scenario recommended by WHO. B, Suboptimal scenario (case of Nigeria).[4]

TABLE 41-2 Average Amount of Energy (kcal) Needed From Complementary Food as Function of Energy Content of Breast Milk Fed to Children in Low-Income Countries, by Age Group

	Age Group (Months)		
Energy requirements and energy sources	**6–8**	**9–11**	**12–23**
Average total energy requirement (kcal/d)	615	686	894
Average amount of energy transferred from mother to infant in breast milk (kcal/d)	403	379	346
Average amount of energy required from complementary foods (kcal/d)	212	307	548

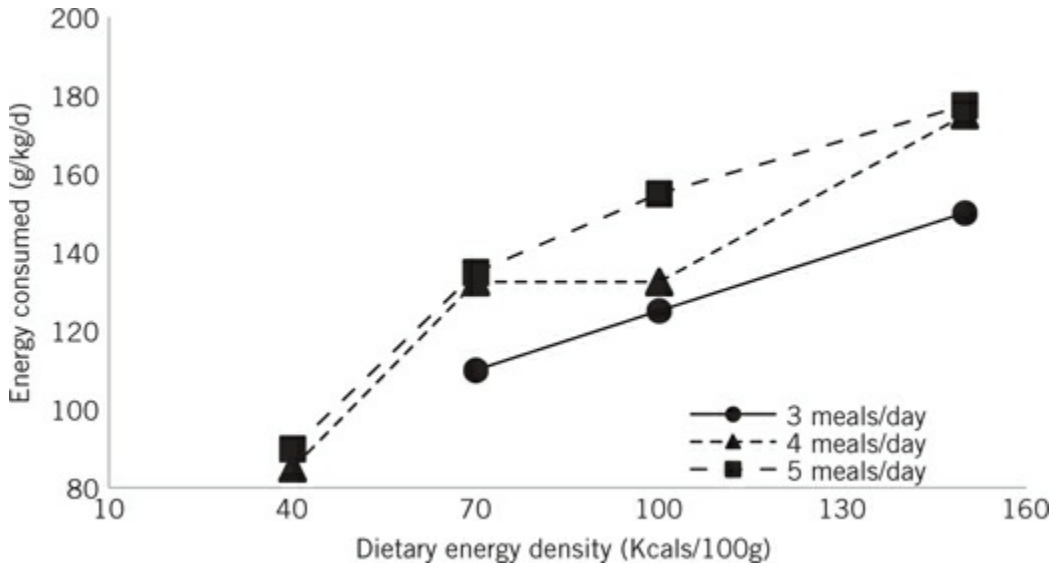


Figure 41-2 Effects of dietary energy density and frequency of feeding on total daily energy intakes by young children (6–18 months of age) recovering from severe malnutrition.[3]

TABLE 41-3 Minimum Dietary Energy Density Needed to Satisfy Energy Requirements from Complementary Food at Different Ages According to the Number of Meals Served Per Day[a]

	Energy Density (kcal/g Food) by Age Group (Months)		
Number of Meals	**6–8**	**9–11**	**12–23**
2	0.71	0.84	1.12
3	0.48	0.56	0.75
4	0.36	0.42	0.56
5	0.29	0.34	0.45

[a] Values estimated based on average level of breast milk intake and assumed gastric capacity of 30 g/kg body weight in each age group.

Key point

- Complementary food of sufficient energy density should be offered an age-appropriate number of times per day, beginning at 6 months of age.

Concerns have been raised that providing high-energy-density complementary foods could displace breast milk intake or reduce the duration of breast-feeding. For example, a study carried out among healthy breast-fed children in Bangladesh found slightly lower breast milk intakes when high-energy-density foods were provided, although total energy intake was greater.[49] By contrast, a recent study, which provided small quantity lipid-based nutrient supplements to breast-fed children, found no effect on breast milk intake.[50] Thus, more information is needed on this issue.

Nutrient Requirements From Complementary Foods

The same approach used for calculating the energy needs from complementary foods can be used for estimating the requirements for individual nutrients. In particular, the average amount of each nutrient that is, provided by breast milk is subtracted from the total dietary requirement to determine the amount needed from complementary foods at different ages, as described in the following sections. For these analyses, nutrient intakes from breast milk were estimated based on breast milk content for women in affluent countries (nutrient concentration of mature milk multiplied by the appropriate breast-milk volume), with the exception of vitamin A, where estimates are available from low-income countries.[3]

Nutrient Requirements of Infants and Young Children

Several expert committees of the United Nations and the U.S. Institute of Medicine (IOM), among others, have developed estimates of young children's nutrient requirements, as summarized in Table 41-4. Regrettably, there are discrepancies in the methods used by the different committees to derive these requirements and in their resulting recommendations, so it is not yet possible to achieve full consensus on the appropriate nutrient contents of complementary foods. The IOM report generally estimated adequate intakes (AIs) for infants between 7 and 12 months of age, based on mean observed intakes of healthy U.S. infants.[48] This approach is not ideal, however, because many of the infants whose intakes were observed may have been consuming considerably more than

their true requirements, so the estimates may be unnecessarily high. For children between 13 and 23 months, the IOM generally estimated dietary requirements based on extrapolation from values for older children or adults or by using a factorial method. Because of the inconsistent methods used for different age groups, there are some counterintuitive reductions in estimated requirements as children become older. The most recent WHO estimates published in 2002 adopted many, but not all, of the recommendations published by the IOM, so some discrepancies in the different estimates still remain. The reasons for these discrepancies and implications for estimated nutrient requirements from complementary foods have been discussed in more detail by Dewey and Brown.[48]

Nutrient Requirements From Complementary Foods

As indicated above, nutrient requirements from complementary foods can be calculated as the difference between the total nutrient requirement and the amounts of these nutrients transferred from the mother to the child in breast milk.[51] These requirements can then be expressed in terms of desirable nutrient densities of complementary foods (amount/100 kcal), by dividing the age-specific amounts of each nutrient required in complementary foods by the respective average amount of energy required from these foods. Examples of these recommended nutrient densities of complementary foods for children of different ages are shown in Table 41-5, by age group and source of estimated nutrient requirements. Despite the aforementioned discrepancies in estimated nutrient requirements, desirable nutrient densities based on the IOM and WHO recommendations are reasonably consistent for protein, iron, riboflavin, thiamine, niacin, folate, and pyridoxine. However, there are large discrepancies for vitamin A and C, calcium and zinc, mainly because of the different approaches used to estimate these requirements.

Problem Nutrients in Complementary Foods and Strategies to Achieve Adequate Nutrient Intakes

Problem nutrients are those for which there is the greatest discrepancy between the estimated amount required by the child and their content in complementary foods.[3] The problem nutrients can be identified by comparing the estimates of desirable nutrient density of complementary foods (amount of nutrient/100 kcal) with the actual nutrient densities of the foods consumed by breast-fed children in various populations.[48] Analyses of dietary intake data from multiple LICs indicate that iron, zinc, calcium, selected B vitamins, and sometimes vitamin A may be present in

inadequate amounts in traditional, home-available complementary foods.[48,52]

Consistent with the data on micronutrient inadequacy of traditional complementary feeding regimens, limited global data on the prevalence of micronutrient deficiencies indicate that deficiencies of zinc, iron, vitamin A, and others are common among young children in LICs.[53,54] Thus, more effort is needed to improve the micronutrient quality of complementary foods or supply the missing nutrients in other forms. Available strategies to supply "problem nutrients" include promotion of a diversified diet, with generous intake of animal-source foods; provision of fortified, processed complementary foods; delivery of nutrient supplements through home-based fortification or supplementation or through biofortification of commonly consumed foods. The following sections briefly describe each of these approaches.

DIETARY MODIFICATION AND DIVERSIFICATION

Methods have been described to identify locally available, affordable foods that can supply problem nutrients, based on the results of quantitative dietary intake data and application of linear programming techniques. Relevant information on the data needs and modeling procedures for this approach is available through the PROPAN website.[55] In general, adequate intake of many of the problem nutrients described above can be achieved only by incorporating animal-source foods, such as animal flesh and organs, dairy products, eggs and fish, into the diet. Animal flesh foods are excellent sources of highly bioavailable heme iron and zinc, along with a number of B vitamins, so inclusion of these foods in young children's diets should be encouraged. These foods are also rich in high-quality protein, although traditional complementary feeding regimens generally contain adequate amounts of protein, unless they are composed mainly of low-protein tubers, such as cassava or sweet potato, or refined starches.

The availability of animal-source foods may be very limited in the poorest countries,[56] and their relatively high cost frequently limits the amounts consumed in poorer households. Moreover, religious proscriptions or cultural taboos may prevent their use in some societies. Nevertheless, if these foods are adequately cooked and finely minced, experience shows that they can be consumed without difficulty by young

children. Recent studies in both the United States and a number of LICs have demonstrated the feasibility of including meat in the feeding regimens of young infants.[57] Another study carried out in slum communities in Peru, found that, following an intensive educational campaign, even poor households were able to increase children's consumption of animal-source foods, and the linear growth of children in the intervention communities was significantly greater than in the control areas, reducing the rate of stunting at 12 months by two-thirds.[58] Thus, it may be possible to increase children's consumption of these foods in some settings, by using appropriately targeted educational interventions.

TABLE 41-4 Nutrient Requirements, by Age Group Based on the WHO/United Nations Children's Fund (UNICEF) 1998 Complementary Feeding Report[3], the IOM Dietary Reference Intakes (DRI) and WHO/Food and Agriculture Organization 2002 Recommendations[48]

Nutrient	Age 6–8 Months			Age 9–11 Months			Age 12–23 Months		
	WHO/ UNICEF 1998	New DRI	WHO 2002	WHO/ UNICEF 1998	New DRI	WHO 2002	WHO/ UNICEF 1998	New DRI	WHO 2002
Protein (g/d)	9.1	NA	NA	9.6	NA	NA	10.9	NA	NA
Vitamin A (μg RE/d)	350	500[a]	400	350	500[a]	400	400	300	400
Folate (μg/d)	32	80[a]	80	32	80[a]	80	50	150	160
Niacin (mg/d)	4	4[a]	4	5	4[a]	4	8	6	6
Pantothenic acid (mg/d)	1.7[b]	1.8[a]	1.8	1.7[b]	1.8[a]	1.8	1.7[b]	2.0[a]	2.0
Riboflavin (mg/d)	0.4	0.4[a]	0.4	0.4	0.4[a]	0.4	0.6	0.5	0.5
Thiamine (mg/d)	0.2	0.3[a]	0.3	0.3	0.3[a]	0.3	0.5	0.5	0.5
Vitamin B_6 (mg/d)	0.3	0.3[a]	0.3	0.4	0.3[a]	0.3	0.7	0.5	0.5
Vitamin B_{12} (μg/d)	0.4	0.5[a]	0.5	0.4	0.5[a]	0.5	0.5	0.9	0.9
Vitamin C (mg/d)	25	50[a]	30	25	50[a]	30	30	15	30
Vitamin D (μg/d)	7	5[a]	5	7	5[a]	5	7	5[a]	5
Vitamin K (μg/d)	10[b]	2.5[a]	10	10[b]	2.5[a]	10	10[b]	30[a]	15
Calcium (mg/d)	525	270[a]	400	525	270[a]	400	350	500[a]	500
Chloride (mg/d)	500	NA	NA	500	NA	NA	800	NA	NA
Copper (mg/d)	0.3	0.2[a]	NA	0.3	0.2[a]	NA	0.4	0.3	NA
Fluoride (μg/d)	0.05[b]	0.5[a]	NA	0.05[b]	0.5[a]	NA	0.05[b]	0.7[a]	NA
Iodine (μg/d)	21	130[a]	90	21	130[a]	90	12	90	90
Iron (mg/d)[c]	11	11	9.3	11	11	9.3	6	7	5.8
Magnesium (mg/d)	75	75[a]	54	80	75[a]	54	85	80	60
Manganese (mg/d)	0.02[b]	0.6[a]	NA	0.02[b]	0.6[a]	NA	0.02[b]	1.2[a]	NA
Phosphorus (mg/d)	400	275[a]	NA	400	275[a]	NA	270	460	NA
Potassium (mg/d)	700	NA	NA	700	NA	NA	800	NA	NA
Selenium (μg/d)	10	20[a]	10	10	20[a]	10	15	20	17
Sodium (mg/d)	320	NA	NA	350	NA	NA	500	NA	NA
Zinc (mg/d)	2.8[d]	3	4.1[e]	2.8[d]	3	4.1[e]	2.8[d]	3	4.1[e]

[a]Based on adequate intake estimates.

[b]Based on safe nutrient intake from British dietary reference values.

[c]Assumes medium iron availability (10%).

[d]Based on Annex III of the 1998 report.[3]

[e]Assumes medium zinc availability (30%).

NA, not yet available.

TABLE 41-5 Recommended Nutrient Densities (Amount per 100 kcal) of Complementary Foods for Children 6–8, 9–11, and 12–23 Months of Age, Based on the WHO/UNICEF 1998 Complementary Feeding Report[3], the IOM Dietary

Reference Intakes (DRI) and WHO/Food and Agriculture Organization 2002 Recommendations[48]

Nutrient	Age 6–8 Months			Age 9–11 Months			Age 12–23 Months		
	WHO/ UNICEF 1998	New DRI	WHO 2002	WHO/ UNICEF 1998	New DRI	WHO 2002	WHO/ UNICEF 1998	New DRI	WHO 2002
Protein (g/100 kcal)	0.7	1.0	1.0	0.7	1.0	1.0	0.7	0.9	0.9
Vitamin A (μg RE/100 kcal)	5	81	31	9	63	30	17	5	23
Calcium (mg/100 kcal)	125	40	105	78	32	74	26	63	63
Iron (mg/100 kcal)	4.0[a]	5.3[a]	4.5	2.4[a]	3.5[a]	3	0.8[a]	1.2[a]	1.0
Zinc (mg/100 kcal)	0.8	1.1	1.6	0.5	0.7	1.1	0.3	0.4	0.6
Riboflavin (mg/100 kcal)	0.07	0.08	0.08	0.04	0.06	0.06	0.05	0.06	0.06
Thiamine (mg/100 kcal)	0.04	0.08	0.08	0.04	0.06	0.06	0.05	0.07	0.07
Niacin (mg/100 kcal)[b]	1.1	1.5	1.5	0.9	1.0	1.0	0.9	0.9	0.9
Folate (μg/100 kcal)	0	11	11	0	9	9	0	19	21
Vitamin B_6 (mg/100 kcal)	0.09[c]	0.12	0.12	0.08[c]	0.08	0.08	0.09[c]	0.08	0.08
Vitamin C (mg/100 kcal)	0	11	1.5	0	8	1.7	1.1	0	1.5

[a]Assumes medium iron availability (10%).

[b]Excluding the contribution of dietary tryptophan to niacin synthesis.

[c]Corrected value.

Nonhuman animal milks and related dairy products, such as yogurt and some cheeses, are alternative sources of selected problem nutrients. These foods are rich in calcium and several B vitamins, but low in iron unless fortified. Moreover, if milk products are not properly heat-treated, they can cause gastrointestinal bleeding in infants, causing a further threat to their iron status.[59] Furthermore, because of the relatively high total fat and saturated fatty acid contents of these products, some authorities recommend using partially skimmed milk.[60] However, there is little evidence of any long-term adverse effects of milk fat intake in early childhood on the subsequent risk of cardiovascular disease, so some child feeding guidelines suggest that reduced fat milks should not be introduced before 2–3 years of age to limit the possibility of inadequate energy intakes.[61]

CEREAL PRODUCTS

Cereal products are commonly used as a major component of complementary feeding regimens. Such foods provide energy, mostly as carbohydrate, and they contribute some protein. However, cereal protein is

limited in certain amino acids (e.g., lysine in wheat and rice and lysine and tryptophan in maize), compared with reference protein (egg albumin or milk protein); the digestibility of cereal proteins is generally less than that of proteins derived from animal-source foods. For these reasons, total protein intakes from cereals must be adjusted for their equivalent amount of reference protein, as described by the Food and Agriculture Organization (FAO).[62] Nevertheless, as indicated above, protein intake is usually sufficient from most complementary feeding regimens that have been examined. Although cereals provide essential minerals, especially when consumed as whole-grain products, concerns have been raised about mineral bioavailability from these foods because their high phytate content can inhibit mineral absorption. However, a recent study of Swedish infants who were randomly assigned to receive cereal-based diets with and without phytate reduction from 6 to 12 months of age found no long-term differences in their iron or zinc status.[63]

PROCESSED COMPLEMENTARY FOODS

Distribution of processed, fortified complementary foods, either through public sector programs or through commercial channels, offers an alternative food-based approach to supply problem nutrients.[64] Although such foods can successfully cover current nutrient shortfalls of the home diet, the rapid changes in young children's nutrient requirements during the period of complementary feeding make it difficult to design a single product that meets the nutrient needs of all children in this age range without imposing the risk of excessive intakes by some children.[65] Moreover, the impact of these foods on children's growth has been inconsistent, possibly related to the timing, amount, and composition of these foods, as well as the underlying nutritional status and frequency of infections of the study populations.[66–68] Despite the inconsistent growth responses observed in these earlier studies, several recent studies have found positive effects on iron status and/or developmental milestones among children who received these products.[69–71]

MICRONUTRIENT SUPPLEMENTS

Addition of micronutrients to current feeding regimens, either through home-based food fortification or direct child supplementation, offers a lower cost strategy than providing a processed, nutritionally complete

complementary food. Home-based fortification involves the addition of a prepackaged micronutrient supplement to the child's food one or more times daily at the time of serving. One such formulation, based on a micronutrient powder containing iron as microencapsulated ferrous fumarate (to avoid adverse organoleptic effects of iron on food) and vitamins A and D, folic acid, ascorbic acid, and zinc, has been shown to reduce rates of anemia among consumers, although evidence of its positive impact on other indicators of micronutrient status and growth are still lacking.[72] WHO concluded that there is strong evidence on the effects of home fortification of complementary foods with micronutrient powders (MNPs) for children aged 6–23 months on their nutritional, health, and development outcomes.[73] Thus, WHO recommends the use of MNPs to improve iron status and reduce anemia among infants and children 6–23 months of age.[74]

Another approach to home fortification uses small-quantity lipid-based supplements, which are usually prepared with peanut paste, vegetable oil, protein concentrate (milk powder or soy), and multiple micronutrients.[75] Recent studies in West Africa have found greater linear growth and earlier achievement of developmental milestones among young children who received these products.[76,77] Micronutrient supplements also may be supplied as liquid formulations or chewable, crushable, or dispersible tablets.[78] A complete review of these products and related issues is beyond the scope of the present chapter. Nevertheless, each of these formulations has been found to yield positive impacts on selected indicators of micronutrient status, depending on the specific formulation, the preexisting nutritional status of the target population and the presence of conditions that may interfere with nutrient absorption or utilization; but there is only limited evidence of any positive effects of these products on child growth. On the other hand, some studies have detected adverse effects of iron supplements and iron-containing micronutrient powders on risk of infectious diseases and related hospitalizations,[79–81] especially among iron-replete individuals, so WHO recommends that these products should be used only where iron deficiency is common and in the context of infection control programs.[82] Additional research is required to define optimal dosing regimens of such multiple micronutrient formulations added to complementary foods, interactions among nutrients in the various preparations, and any possible adverse effects on child growth and other health outcomes in particular settings.

BIOFORTIFICATION

Biofortification is a process whereby the density of minerals and vitamins of individual crops can be increased through conventional plant breeding or the use of transgenic techniques. To date, most biofortification programs have focused on increasing the iron, zinc, or provitamin A contents of staple foods. One of the most successful programs is currently promoting greater production and consumption of orange-fleshed sweet potato, which is a rich source of the vitamin A precursor beta carotene. Introduction of orange-fleshed sweet potato in rural Mozambique, along with feeding advice, resulted in a considerable increase in vitamin A intakes compared to the nonintervention group and a reduction in the prevalence of vitamin A deficiency.[83] More information on the scope and future potential of biofortification is available through existing reviews of this topic.[83–85]

SAFE PREPARATION OF COMPLEMENTARY FOODS

An important contributor to growth faltering among children 6–23 months of age in LICs is the high incidence of enteric infections and environmental enteropathy,[86,87] some of which is due to food-borne infections. Thus, improving the hygienic quality of complementary foods may be just as important as focusing on their adequate nutritional quality. A study examining the microbiological quality of complementary food in rural and urban areas in Bangladesh, for example, found that 40% of the complementary foods contained *Escherichia coli*, an indicator of fecal contamination.[88] Poor hygienic conditions during preparation and serving were highly related to food contamination. Moreover, storing the food at ambient temperatures between meals and consumption of contaminated complementary foods were associated with a higher frequency of diarrhea. Contaminated feeding bottles are a particularly important source of pathogen transmission. In periurban Peru, for example, 35% of bottle nipples were contaminated with *E. coli*, and 31% of teas served in bottles were contaminated compared to just 2% when teas were served in cups.[1]

The Pan American Health Organization/WHO recommends the following food hygiene practices when preparing, serving, and storing complementary foods: (1) hand washing of both caregiver and child before food preparation and feeding; (2) serving food immediately after preparation, and/or storing the food properly at sufficiently low or high

temperatures to prevent microbial proliferation; (3) using clean serving utensils; and (4) avoiding feeding bottles in settings where quality of water is poor and chemical and/or thermal treatments are not easily accomplished. Several interventions that have proved to be effective in improving the microbiological quality of complementary food are caregiver education on good hygienic practices, and improving traditional food storage and processing methods (e.g., by using food fermentation).[89]

INTERVENTIONS TO IMPROVE COMPLEMENTARY FEEDING PRACTICES

The range of intervention programs to promote enhanced complementary feeding practices and information on their impact on children's growth have been described in several systematic reviews.[67,90,91] Intervention strategies that were examined include behavior change/communication interventions to encourage improved feeding practices and/or energy density, provision of processed complementary foods and distribution of different forms of added micronutrients. In general, the average effect sizes of these interventions on children's growth were fairly small (effect sizes ~0.2–0.3 SD units) and did not differ by intervention strategy. These results suggest that enhanced growth is possible through improved complementary feeding, but feeding alone may not fully ameliorate growth restriction unless prenatal factors contributing to growth restriction[92,93] and postnatal infections are also addressed.[94]

Key points

- Despite uncertainty regarding the nutrient requirements of infants and young children, recommendations have been developed on the minimum adequate nutrient density of complementary foods for children of different ages.
- Iron, zinc, calcium, selected B vitamins, and vitamin A are often present in inadequate amounts in traditional complementary feeding regimens.
- Feeding a variety of foods, including animal-source foods, particular fruits and vegetables, and biofortified foods can improve micronutrient intake from complementary foods. When suitable low-cost local diets cannot be prepared from acceptable, locally available foods, home fortification and

nutrient supplementation provide other options to achieve adequate nutrient intakes.

- A variety of intervention strategies can be used to enhance complementary feeding practices; but additional interventions, including postnatal infection control, may be necessary to allow children to meet their growth potential during the period of complementary feeding.

SUMMARY

Increased information on the multiple advantages of exclusive breast-feeding during the early months of life, the optimal timing of introduction of complementary foods, and the appropriate energy density, feeding frequency, and micronutrient composition of these foods has become available in recent years. Relevant feeding guidelines have been disseminated and intervention strategies tested and promoted. However, complete understanding of the effects of different complementary feeding regimens on breast milk intakes and the ultimate duration of breast-feeding is still lacking. Likewise, more information is needed on the ideal micronutrient composition of fortified complementary foods, the design of integrated nutrition and infection control programs and the impact of alternative micronutrient delivery systems on young children's nutritional status, physical growth, and neurobehavioral development.

REFERENCES

1. PAN American Health Organisation/World Health Organization. *Guiding Principles for Complementary Feeding of the Breastfed Child*. Washington, DC: Division of Health Promotion and Protection, Food Nutrition Program; 2004.
2. World Health Organisation. *Indicators for Assessing Breast-Feeding Practices*. Geneva, Switzerland: WHO; 1991.
3. World Health Organisation. *Complementary Feeding of Young Children in Developing Countries: A Review of Current Scientific Knowledge*. Geneva, Switzerland: WHO; 1998.
4. UNICEF. *Improving Child Nutrition: The Achievable Imperative for Global Progress*. New York, NY: United Nations Publication; 2013. http://www.childinfo.org/files/NutritionReport_April2013_Final.pdf.
5. Edmond KM, Zandoh C, Quigley MA, et al. Delayed breastfeeding initiation increases risk of neonatal mortality. *Pediatrics*. 2006;117:e380–e386.
6. Debes AK, Kohli A, Walker N, et al. Time to initiation of breastfeeding and neonatal mortality and morbidity: a systematic review. *BMC Public Health*. 2013;13:S19.
7. Brown KH, Black RE, de Romaña GL, et al. Infant-feeding practices and their

relationship with diarrheal and other diseases in Huascar (Lima), Peru. *Pediatrics*. 1989;83:31–40.
8. Kramer MS, Kakuma R. *The Optimal Duration of Exclusive Breastfeeding: A Systematic Review*. Geneva, Switzerland: WHO; 2002.
9. Popkin BM, Adair L, Akin JS, et al. Breast-feeding and diarrheal morbidity. *Pediatrics*. 1990;86:874–882.
10. Bhandari N, Bahl R, Mazumdar S, et al. Effect of community-based promotion of exclusive breastfeeding on diarrhoeal illness and growth: a cluster randomised controlled trial. *Lancet*. 2003;361:1418–1423.
11. WHO Collaborative Study Team on the Role of Breastfeeding on the Prevention of Infant Mortality. Effect of breastfeeding on infant and child mortality due to infectious diseases in less developed countries: a pooled analysis. *Lancet*. 2000;355:451–455.
12. Butte NF, Lopez-Alarcon MG, Garza C. *Nutrient Adequacy of Exclusive Breastfeeing for the Term Infant During the First Six Months of Life*. Geneva, Switzerland: WHO; 2002.
13. Brown KH, Creed de Kanashiro H, del Aguila R, et al. Milk consumption and hydration status of exclusively breast-fed infants in a warm climate. *J Pediatr*. 1986;108:677–680.
14. Cohen RJ, Brown KH, Canahuati J, et al. Effects of age of introduction of complementary foods on infant breast milk intake, total energy intake, and growth: a randomised intervention study in Honduras. *Lancet*. 1994;344:288–293.
15. Dewey KG, Cohen RJ, Brown KH, et al. Age of introduction of complementary foods and growth of term, low-birth-weight, breast-fed infants: a randomized intervention study in Honduras. *Am J Clin Nutr*. 1999;69:679–686.
16. Kramer MS, Guo T, Platt RW, et al. Breastfeeding and infant growth: biology or bias? *Pediatrics*. 2002;110:343–347.
17. Jonsdottir OH, Thorsdottir I, Hibberd PL, et al. Timing of the introduction of complementary foods in infancy: a randomized controlled trial. *Pediatrics*. 2012; 130:1038–45.
18. WHO, UNICEF, UNAIDS. *Guidelines on HIV and Infant Feeding 2010: Principles and Recommendations for Infant Feeding in the Context of HIV and a Summary of Evidence*. Geneva, Switzerland; 2010.
19. Cai X, Wardlaw T, Brown DW. Global trends in exclusive breastfeeding. *Int Breastfeed J*. 2012;7:12.
20. Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet*. 2013;382:427–451.
21. Kramer MS, Chalmers B, Hodnett ED, et al. Promotion of Breastfeeding Intervention Trial (PROBIT): a randomized trial in the Republic of Belarus. *JAMA*. 2001;285:413–420.
22. Morrow AL, Guerrero ML, Shults J, et al. Efficacy of home-based peer counselling to promote exclusive breastfeeding: a randomised controlled trial. *Lancet*. 1999;353:1226–1231.
23. Haider R, Ashworth A, Kabir I, et al. Effect of community-based peer counsellors on exclusive breastfeeding practices in Dhaka, Bangladesh: a randomised controlled trial. *Lancet*. 2000;356:1643–1647.
24. Imdad A, Yakoob MY, Bhutta ZA. Effect of breastfeeding promotion interventions on breastfeeding rates, with special focus on developing countries. *BMC Public Health*. 2011;11:S24.
25. Bhandari N, Kabir AK, Salam MA. Mainstreaming nutrition into maternal and

child health programmes: scaling up of exclusive breastfeeding. *Matern Child Nutr*. 2008;4:S5–S23.
26. Cattaneo A, Quintero-Romero S. Protection, promotion and support of breastfeeding in low-income countries. *Semin Fetal Neonatal Med*. 2006;11:48–53.
27. Sobel HL, Iellamo A, Raya RR, et al. Is unimpeded marketing for breast milk substitutes responsible for the decline in breastfeeding in the Philippines? An exploratory survey and focus group analysis. *Soc Sci Med*. 2011;73:1445–1448.
28. Brady JP. Marketing breast milk substitutes: problems and perils throughout the world. *Arch Dis Child*. 2012;97:529–532.
29. Waterston T, Tumwine J. Monitoring the marketing of infant formula feeds: Manufacturers of breast milk substitutes violate the WHO code—again. *BMJ*. 2003;326:113–114.
30. Forsyth S. Non-compliance with the International Code of Marketing of Breast Milk Substitutes is not confined to the infant formula industry. *J Public Health*. 2013;35:185–190.
31. Hardy BJ. Commentary: getting to compliance. Multisector dialogue, collaboration and the International Code of Marketing of Breast Milk Substitutes. *J Public Health*. 2013;35:191–192.
32. James WP, Ferro-Luzzi A, Waterlow JC. Definition of chronic energy deficiency in adults. Report of a working party of the International Dietary Energy Consultative Group. *Eur J Clin Nutr*. 1988;42:969–981.
33. Brown KH, Akhtar NA, Robertson AD, et al. Lactational capacity of marginally nourished mothers: relationships between maternal nutritional status and quantity and proximate composition of milk. *Pediatrics*. 1986;78:909–919.
34. Lönnerdal B, Forsum E, Hambraeus L. A longitudinal study of the protein, nitrogen, and lactose contents of human milk from Swedish well-nourished mothers. *Am J Clin Nutr*. 1976;29:1127–1133.
35. Butte NF, Garza C, Smith EO, et al. Human milk intake and growth in exclusively breast-fed infants. *J Pediatr*. 1984;104:187–195.
36. Prentice AM, Roberts SB, Prentice A, et al. Dietary supplementation of lactating Gambian women. I. Effect on breast-milk volume and quality. *Hum Nutr Clin Nutr*. 1983;37:53–64.
37. Dewey KG, Beaton G, Fjeld C, et al. Protein requirements of infants and children. *Eur J Clin Nutr*. 1996;50:S119–S147; discussion S147–S150.
38. McGready R, Simpson JA, Cho T, et al. Postpartum thiamine deficiency in a Karen displaced population. *Am J Clin Nutr*. 2001;74:808–813.
39. Rice AL, Stoltzfus RJ, de Francisco A, et al. Evaluation of serum retinol, the modified-relative-dose-response ratio, and breast-milk vitamin A as indicators of response to postpartum maternal vitamin A supplementation. *Am J Clin Nutr*. 2000;71:799–806.
40. Institute of Medicine. *Nutrition During Lactation*. Washington, DC: The National Academies Press; 1991.
41. UNICEF/WHO Joint Committee on Health Policy. *Strategic Approach to Operationalizing Selected End-Decade Goals: Reduction of Iron Deficiency Anemia, JCHP 30/95/4.5*. Geneva, Switzerland: World Health Organization; 1995.
42. McDonald SJ, Middleton P, Dowswell T, et al. Effect of timing of umbilical cord clamping of term infants on maternal and neonatal outcomes. *Cochrane Database Syst Rev*. 2013;7:CD004074.
43. Brown KH, Engle-Stone R, Krebs NF, et al. Dietary intervention strategies to enhance zinc nutrition: promotion and support of breastfeeding for infants and

young children. *Food Nutr Bull*. 2009;30:S144–S171.
44. World Health Organisation. *Guideline: Vitamin A Supplementation in Postpartum Women*. Geneva, Switzerland: WHO; 2011.
45. Stoltzfus RJ, Humphrey JH. Vitamin A and the nursing mother-infant dyad: evidence for intervention. *Adv Exp Med Biol*. 2002;503:39–47.
46. Allen LH. B vitamins in breast milk: relative importance of maternal status and intake, and effects on infant status and function. *Adv Nutr*. 2012;3:362–369.
47. Underwood BA. Maternal vitamin A status and its importance in infancy and early childhood. *Am J Clin Nutr*. 1994;59:517S–522S.
48. Dewey KG, Brown KH. Update on technical issues concerning complementary feeding of young children in developing countries and implications for intervention programs. *Food Nutr Bull*. 2003;24:5–28.
49. Islam MM, Khatun M, Peerson JM, et al. Effects of energy density and feeding frequency of complementary foods on total daily energy intakes and consumption of breast milk by healthy breastfed Bangladeshi children. *Am J Clin Nutr*. 2008;88:84–94.
50. Kumwenda C, Dewey KG, Hemsworth J, et al. Lipid-based nutrient supplements do not decrease breast milk intake of Malawian infants. *Am J Clin Nutr*. 2014;99:617–623.
51. Brown KH. Breastfeeding and complementary feeding of children up to 2 years of age. *Nestle Nutr Workshop Ser Pediatr Program*. 2007;60:1–10.
52. Gibson RS, Ferguson EL, Lehrfeld J. Complementary foods for infant feeding in developing countries: Their nutrient adequacy and improvement. *Eur J Clin Nutr*. 1998;52:764–770.
53. Vitamin and Mineral Nutrition Information System (VMNIS). *Micronutrients Database*. Geneva, Switzerland: World Health Organisation; 2007. http://www.who.int/vmnis/database/en/. Accessed May 4, 2015.
54. Muthayya S, Rah JH, Sugimoto JD, et al. The global hidden hunger indices and maps: an advocacy tool for action. *PLoS One*. 2013;8:e67860.
55. PAHO, UNICEF. *ProPAN: Process for the Promotion of Child Feeding*. Washington, DC: PAHO; 2013.
56. Ferguson EL, Darmon N, Fahmida U, et al. Design of optimal food-based complementary feeding recommendations and identification of key "problem nutrients" using goal programming. *J Nutr*. 2006;136:2399–2404.
57. Krebs NF, Westcott JE, Butler N, et al. Meat as a first complementary food for breastfed infants: feasibility and impact on zinc intake and status. *J Pediatr Gastroenterol Nutr*. 2006;42:207–214.
58. Penny ME, Creed-Kanashiro HM, Robert RC, et al. Effectiveness of an educational intervention delivered through the health services to improve nutrition in young children: a cluster-randomised controlled trial. *Lancet*. 2005;365:1863–1872.
59. Fomon SJ, Ziegler EE, Nelson SE, et al. Cow milk feeding in infancy: gastrointestinal blood loss and iron nutritional status. *J Pediatr*. 1981;98:540–545.
60. Simell O, Niinikoski H, Rönnemaa T, et al. Special Turku Coronary Risk Factor Intervention Project for Babies (STRIP). *Am J Clin Nutr*. 2000;72:1316S–1331S.
61. Department of Health. *Weaning and the Weaning Diet. Report of the Working Group on the Weaning Diet. Committee on Medical Aspects of Food Policy*. London, UK: HMSO; 1994:1–113.
62. Food and Agriculture Organization: Report of an FAO Expert Consultation. *Dietary Protein Quality Evaluation in Human Nutrition* Rome, Italy. FAO; 2013
63. Lind T, Lönnerdal B, Persson LA, et al. Effects of weaning cereals with different

phytate contents on hemoglobin, iron stores, and serum zinc: a randomized intervention in infants from 6 to 12 mo of age. *Am J Clin Nutr*. 2003;78:168–175.
64. Brown KH, Lutter CK. Potential role of processed complementary foods in the improvement of early childhood nutrition in Latin America. *Food Nutr Bull*. 2000;21:5–11.
65. Dewey KG. Nutrient composition of fortified complementary foods: should age-specific micronutrient content and ration sizes be recommended? *J Nutr*. 2003;133:2950S–2952S.
66. Simondon KB, Gartner A, Berger J, et al. Effect of early, short-term supplementation on weight and linear growth of 4-7-mo-old infants in developing countries: a four-country randomized trial. *Am J Clin Nutr*. 1996;64:537–545.
67. Caulfield LE, Huffman SL, Piwoz EG. Interventions to improve intake of complementary foods by infants 6 to 12 months of age in developing countries : impact on growth and on the prevalence of malnutrition and potential contribution to child survival. *Food Nutr Bull*. 1999;20:183–200.
68. Dewey KG. Success of intervention programs to promote complementary feeding. In: Black RE, Fleischer Michalesen K, eds. *Public Health Issues in Infant and Child Nutrition*. 1st ed. Philadelphia, PA: Lippincott Williams & Wilkins; 2002:199–216.
69. Lartey A, Manu A, Brown KH, et al. A randomized, community-based trial of the effects of improved, centrally processed complementary foods on growth and micronutrient status of Ghanaian infants from 6 to 12 mo of age. *Am J Clin Nutr*. 1999;70:391–404.
70. Rivera JA, Sotres-Alvarez D, Habicht JP, et al. Impact of the Mexican program for education, health, and nutrition (Progresa) on rates of growth and anemia in infants and young children: a randomized effectiveness study. *JAMA*. 2004;291:2563–2570.
71. Faber M, Kvalsvig JD, Lombard CJ, et al. Effect of a fortified maize-meal porridge on anemia, micronutrient status, and motor development of infants. *Am J Clin Nutr*. 2005;82:1032–1039.
72. Zlotkin SH, Christofides AL, Hyder SM, et al. Controlling iron deficiency anemia through the use of home-fortified complementary foods. *Indian J Pediatr*. 2004;71:1015–1019.
73. De-Regil LM, Suchdev PS, Vist GE, et al. Home fortification of foods with multiple micronutrient powders for health and nutrition in children under two years of age. *Cochrane Database Syst Rev*. 2011;9:CD008959.
74. World Health Organisation. *Guideline: Use of Multiple Micronutrient Powders for Home Fortification of Foods Consumed by Infants and Children 6–23 Months of Age*. Geneva, Switzerland: WHO; 2011.
75. Arimond M, Zeilani M, Jungjohann S, et al. Considerations in developing lipid-based nutrient supplements for prevention of undernutrition: experience from the International Lipid-Based Nutrient Supplements (iLiNS) Project. *Matern Child Nutr*. 2013;6:12049.
76. Adu-Afarwuah S, Lartey A, Brown KH, et al. Randomized comparison of 3 types of micronutrient supplements for home fortification of complementary foods in Ghana: effects on growth and motor development. *Am J Clin Nutr*. 2007;86:412–420.
77. Hess SY, Abbeddou S, Yakes EJ, et al. Small-quantity lipid-based nutrient supplements, regardless of their zinc content, increase growth and reduce the prevalence of stunting and wasting in young Burkinabe children: A cluster-randomized trial. Plos One. 2015;10:e0122242.

78. Smuts CM, Lombard CJ, Benadé AJ, et al. Efficacy of a foodlet-based multiple micronutrient supplement for preventing growth faltering, anemia, and micronutrient deficiency of infants: the four country IRIS trial pooled data analysis. *J Nutr*. 2005;135:631S–638S.
79. Sazawal S, Black RE, Ramsan M, et al. Effects of routine prophylactic supplementation with iron and folic acid on admission to hospital and mortality in preschool children in a high malaria transmission setting: community-based, randomised, placebo-controlled trial. *Lancet*. 2006;367:133–143.
80. Veenemans J, Milligan P, Prentice AM, et al. Effect of supplementation with zinc and other micronutrients on malaria in Tanzanian children: a randomised trial. *PLoS Med*. 2011;8:e1001125.
81. Soofi S, Cousens S, Iqbal SP, et al. Effect of provision of daily zinc and iron with several micronutrients on growth and morbidity among young children in Pakistan: a cluster-randomised trial. *Lancet*. 2013;382:29–40.
82. Report of the World Health Organization Technical Consultation on Prevention and Control of Iron Deficiency in Infants and Young Children in Malaria-Endemic Areas, Lyon, France, 12–14 June 2006. *Food Nutr Bull*. 2007;28:S489–S631.
83. The World Bank and IFPRI. *From Agriculture to Nutrition: Pathways, Synergies, and Outcomes*. Washington, DC: The International Bank for Reconstruction and Development/The World Bank; 2007.
84. Bouis HE, Hotz C, McClafferty B, et al. Biofortification: a new tool to reduce micronutrient malnutrition. *Food Nutr Bull*. 2011;32:S31–S40.
85. Ruel MT, Alderman H. Nutrition-sensitive interventions and programmes: how can they help to accelerate progress in improving maternal and child nutrition? *Lancet*. 2013;382:536–551.
86. Checkley W, Epstein LD, Gilman RH, et al. Effects of acute diarrhea on linear growth in Peruvian children. *Am J Epidemiol*. 2003;157:166–175.
87. Kosek M, Haque R, Lima A, et al. Fecal markers of intestinal inflammation and permeability associated with the subsequent acquisition of linear growth deficits in infants. *Am J Trop Med Hyg*. 2013;88:390–396.
88. Islam MA, Ahmed T, Faruque ASG, et al. Microbiological quality of complementary foods and its association with diarrhoeal morbidity and nutritional status of Bangladeshi children. *Eur J Clin Nutr*. 2012;66:1242–1246.
89. Keusch GT, Fontaine O, Bhargava A, et al. Diarrheal diseases. In: Jamison DT, Breman JG, Measham AR, et al., eds. *Disease Control Priorities in Developing Countries*. 2nd ed. Washington, DC: World Bank; 2006:371–387.
90. Dewey KG, Adu-Afarwuah S. Systematic review of the efficacy and effectiveness of complementary feeding interventions in developing countries. *Matern Child Nutr*. 2008;4: S24–S85.
91. Bhutta ZA, Das JK, Rizvi A, et al. Evidence-based interventions for improvement of maternal and child nutrition: what can be done and at what cost? *Lancet*. 2013;382:452–477.
92. Christian P. Prenatal origins of undernutrition. In: Kalhan SC, Prentice AM, Yajnik CS, eds. *Emerging Societies—Coexistence of Childhood Malnutrition and Obesity*. Basel, Switzerland: Nestec Ltd. Vevey/S. Karger AG; 2009:59–77.
93. Christian P, Lee SE, Donahue Angel M, et al. Risk of childhood undernutrition related to small-for-gestational age and preterm birth in low- and middle-income countries. *Int J Epidemiol*. 2013;42:1340–1355.
94. Humphrey JH. Child undernutrition, tropical enteropathy, toilets, and handwashing. *Lancet*. 2009; 374:1032–1035.

42

Child Nutrition and Economic Development

William A. Masters, PhD

Children's nutrition is both cause and consequence of broader health conditions, household income, and living standards. This chapter describes the principal changes in child nutrition that have been associated with economic development across countries in recent decades, summarizing newly available data on anthropometric status, breast-feeding, and food consumption, as well as other influences on nutritional balances such as sanitation and disease prevention. Some of these changes are mediated by national or community-level policies and programs as well as medical interventions described elsewhere in this text, but most of the patterns described in this chapter occur autonomously as individuals respond to changing circumstances associated with economic development. These changes often occur slowly, without the conscious knowledge of those involved, and have only recently been documented thanks to the accumulation of evidence from a wide range of social environments.

IMPROVEMENTS OVER TIME AND COMPARISONS ACROSS COUNTRIES

Leo Tolstoy's classic novel *Anna Karenina* begins: "All happy families are alike; each unhappy family is unhappy in its own way." This claim starts an epic, questioning search for whatever it might be that successes have in common. For economic development, a large social science literature documents wide variation in peoples' living conditions. These

circumstances have often stagnated for centuries, with almost all people experiencing relatively poor health, low income, and less preferred living standards, until new opportunities and forms of social organization trigger the onset of sustained improvement.[1] Rising income is eventually associated with large gains in adult height, weight, and many measures of health[2] and even measureable increases in subjective well-being.[3] It turns out that success in one dimension is typically accompanied by successes in other areas. Many different obstacles might slow development, but successful societies often move in similar directions as people gradually achieve similar objectives.

In this chapter, we focus on changing child nutrition, which is among the most universal human goals that economic development can help people pursue.[4,5] We describe how child nutrition alters during economic development, defined very broadly to mean households' overall purchasing power over real goods and services. Households vary in how they acquire that purchasing power, which comes from labor earnings, home production, and wealth of all kinds. Household purchasing power is measured as the sum of all income sources, adjusted for local prices in each year. For this calculation, we use the Penn World Tables (PWT), which offers the world's broadest and most complete set of real income data, updated regularly by the Center for International Comparisons of Production, Income and Prices at the University of Pennsylvania. We use PWT version 7.1, based on national accounts and price data from 146 countries between 1950 and 2010.[6]

Many aspects of child nutrition are likely to change as their families acquire more purchasing power; here, we focus only on what is most widely measured. Surveys of several million children from more than a hundred countries over multiple decades allow us to look systematically at changes in child height, stunting, and wasting, as well as various contributors to nutritional status including food availability and diet quality, sanitation and water supplies, breast-feeding behavior and treatment of childhood diarrhea, vaccination and vitamin A supplementation. Each of these measures reflects a different aspect of child nutrition that is discussed only briefly here, and analyzed at greater length in other chapters.

To describe changes during economic development, we compare the various child nutrition indicators to average real income per person in each country, at the time of observation. National income is an aggregate measure of all economic activity, but structural changes within society also matter. We end the chapter with a discussion of two structural changes that

are particularly important for child nutrition: agricultural transformation (a shift from food production to other enterprises) and demographic transition (a shift from large to small families). These are closely related to each other and to average national income, and to a multitude of other social and institutional changes in gender relations, education, and child care practices.

Our focus in this chapter is on changes over long periods and differences among very diverse countries, using national averages and prevalence rates as indicators of how populations differ from each other. Variation within a given country is itself of interest but is addressed primarily elsewhere. The one distributional issue of great concern here is the fraction of children in the country who fall below a clinical threshold of severe malnutrition. The economic-development counterpart of this is the fraction of households that fall below a poverty line. A long literature in economics concerns how increases in average income affect the prevalence of poverty, and the same methods have now been applied to changes in malnutrition.[7]

Many factors other than economic development clearly matter for child nutrition. Ethnographic studies describe in detail how children are raised, finding that social norms and beliefs can lead to a variety of choices regarding complementary feeding and other nutritional practices.[8,9] These norms may eventually adjust to income changes, or may introduce variation that is unrelated to economic circumstances. Either way, by assembling data from millions of children around the world, we can look over a wide enough range of circumstances to see patterns that would otherwise be hidden. This chapter is descriptive, aimed at revealing stylized facts about long-term relationships. Once those facts are known, other techniques would be needed to address causality. In particular, for this chapter, we are looking at contemporaneous correlation, recognizing that this misses the time delays that play a major role in any underlying causal mechanisms.[10]

For visual clarity, our description of the evidence uses scatter plots and regression lines of each child nutrition indicator against the natural log of real income. This approach transforms exponential relationships into linear ones, and allows us to span the full range of purchasing power which varies from US$180 per person per year (a log value of 5.2) in Liberia and the Democratic Republic of Congo during their years of civil war, to above US$60,000 (a log value of 11) in Qatar and the United Arab Emirates during their years of highest oil revenue. The regression line traces the average change in each nutrition indicator associated with a constant 1%

change in purchasing power, while the scatter of individual observations reveals the pattern of variation around that central tendency. Systematic nonlinearities are often visible, for example, if observations cluster below or above particular parts of the regression line, and the overall variance or precision is shown by a 95% confidence interval around the regression's estimated mean at each income level. The resulting figures reveal long-term, cross-country patterns that could not be seen within individual countries or shorter time periods.

ANTHROPOMETRIC FINDINGS AND ECONOMIC DEVELOPMENT

Child Height, Stunting, and Wasting

Until very recently, the anthropometric data used to study nutrition in economic history referred mainly to adolescents and adults, whose heights were often measured for military or other institutional purposes.[2] Only since the 1990s have large-scale anthropometric surveys of children in developing countries obtained sufficient sample sizes to identify variation in the first years of life. Among the first striking results was that the growth faltering associated with undernutrition actually occurs very early in infancy and early childhood, mostly prior to age 2.[11] The most widely reported data typically refer to children from birth to age 5, and have been assembled from the pioneering Demographic and Health Surveys (DHS) funded by United States Agency for International Development (USAID), as well as the Multiple Indicator Cluster Surveys (MICS) of United Nations Children's Fund (UNICEF) and other sources.

We begin our description of the data with the broadest possible coverage of countries and years, using estimates provided by the Nutrition Impact Model Study (NIMS).[12] They compute nationally representative height and weight distributions for children under 5 across 141 developing countries, interpolating between surveys for annual data from 1985 to 2011 to measure progress over time.[13] Here, we extend their analysis to correlations with income, by combining these estimates with the PWT 7.1 measure of each country's real purchasing power. We begin with average heights, and then consider the prevalence of stunting and wasting using the smaller sample size of actual surveys starting in 1966 from the WHO Global Database on Child Growth and Malnutrition.[14]

For each anthropometric measure, we show a scatter plot of each observation against the log of real income per person. The observations are

drawn in two different shadings, one for the most recent decade (2000–2010) and one for earlier years, with separate regression lines that show a 95% confidence interval. This presentation is in the tradition of Samuel Preston who used a somewhat similar diagram to show how life expectancy at each income level changed over successive decades.[15] A shift in such Preston curves would be associated with variables other than contemporaneous income, and could include the cumulative effect of past income, new technologies, or changed policies and institutions.

Figure 42-1 shows the correlation between average real income per person and average child height, for the 137 countries that are in both the NIMS and PWT datasets, from 1985 (when NIMS data begin) to 2010 (when PWT data end). We have a total of 3485 country-year observations, based on underlying data from over 7.7 million survey subjects.[13] On this scatter plot, a few outlier countries can be seen trending over time. For example, the series of observations that crosses the -2 line at a log income level of about 9 is Guatemala, a country with famously poor nutrition outcomes given its income level.[16] This chart reveals that Guatemala has recently been converging toward the levels achieved elsewhere, as both income and height have risen sharply and moved closer to the regression line.

The regression lines in Figure 42-1 suggest that a very poor country with a log income around 6 (US$400 per person, per year) can be expected to have a mean height-for-age z score around –2. In other words, their average child would be shorter than 97.5% of children in the WHO's global reference population. At the highest income levels in this sample (Qatar and UAE), the average child is still shorter than the WHO reference median, which could be because this sample includes only "developing" countries, all of whom had relatively low incomes in the recent past. Comparing the two regression lines, average child heights at each income level are significantly higher in the most recent decade than in previous years. This could be because of a cumulative lagged effect of income itself, or to omitted variables that improve over time, such as innovations in health care, improved access to medical care, child health norms, and nutritional innovations of all kinds.

Figure 42-2 shows the prevalence of stunting, as the fraction of children with HAZ scores below –2. The regression line shows that at the world's lowest income levels, roughly half of children can be expected to be stunted. As with Figure 42-1, the scatter plot shows a clear association and also an improvement over time in the position of the regression line: at low income levels, fewer children were stunted in the 2000s than in

previous decades. Like Figure 42-1, the regression lines converge and are not significantly different from each other at high income levels. Also, nonlinearity in the scatter plot data is readily apparent in these data, as the richest of these developing countries have brought their stunting rates down close to the level seen in the WHO's reference population itself. Curvature of the scatter plots around the linear regression line is consistent with the result found elsewhere that a given percentage increase in income is most powerful in reducing malnutrition in poor countries with widespread undernutrition, while similar improvements in countries that have already made progress would require more targeted programs.[7]

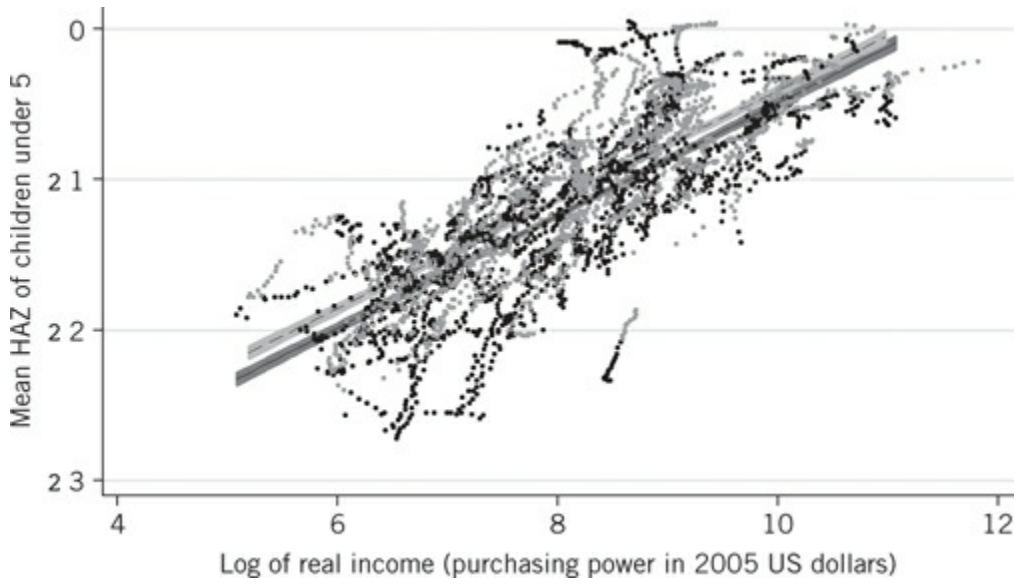


Figure 42-1 Child height and real income, 1985–1999 and 2000–2010 (Ezzati et al estimates, *N* = 3485 from 137 countries).

Note: Data shown are for 1985–1999 (darker shading) and 2000–2010 (lighter shading), with 95% confidence intervals around linear regressions. Sample sizes are *N* = 1978 across 137 countries for 1985–1999 and *N* = 1507 across 138 countries for 2000–2010.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Height estimates are courtesy of Prof. Majid Ezzati, as detailed in Stevens et al,[13] downloaded December 30, 2012 from www1.imperial.ac.uk/publichealth/departments/ebs/projects/eresh/majidez

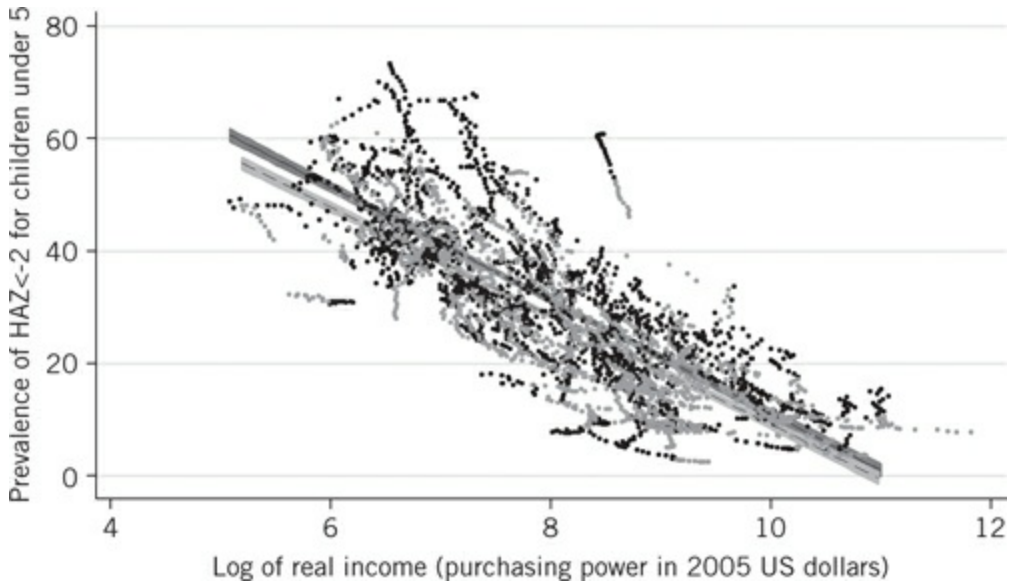


Figure 42-2 Prevalence of stunting and real income, 1985–1999 and 2000–2010 (Ezzati et al estimates, **N** = 3485 from 137 countries).

Note: Data shown are for 1985–1999 (darker shading) and 2000–2010 (lighter shading), with 95% confidence intervals around linear regressions. Sample sizes are *N* = 1978 across 137 countries for 1985–1999 and *N* = 1507 across 137 countries for 2000–2010.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Stunting estimates are courtesy of Prof. Majid Ezzati, as detailed in Stevens et al,[13] downloaded December 30, 2012 from www1.imperial.ac.uk/publichealth/departments/ebs/projects/eresh/majidez

The height-for-age data shown in Figures 42-1 and 42-2 are constructed by the NIMS project, imputing values for the countries and years without survey data.[13] If we limit the picture to actual surveys that are in the WHO's Global Database on Child Growth and Malnutrition,[14] we have a smaller sample size but broader coverage, since we can include a few observations made earlier in time and in industrialized countries. Figures 42-3 and 42-4 use the actual surveys compiled by the WHO to show both stunting and wasting prevalence. Looking first at stunting, Figure 42-3 confirms the result found in Figure 42-2. The regression lines have similar slope and somewhat wider confidence intervals due to a smaller sample size, and yet confirm that there was a significant improvement over time, with less stunting in poor countries for their

income level. Figure 42-4 shows the prevalence of wasting, defined as weight for height z scores below -2. This also has a strong association with income, with the average very poor country having a prevalence rate above 10%, but many countries near the lower bound for this type of malnutrition.

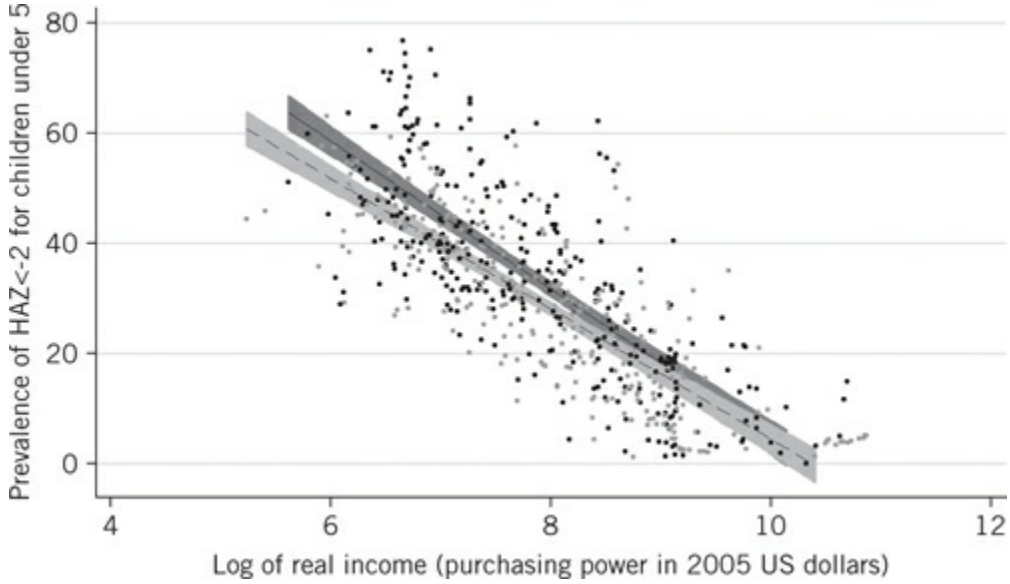


Figure 42-3 Prevalence of stunting and real income, 1966–1999 and 2000–2010 (WHO data, *N* = 608 from 146 countries).

Note: Data shown are for 1966–1999 (darker shading) and 2000–2010 (lighter shading), with 95% confidence intervals around linear regressions. Sample sizes are *N* = 320 across 128 countries for 1966–1999 and *N* = 288 for 127 countries for 2000–2010.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Stunting data are from WHO, Global Database on Child Growth Malnutrition, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statistics (http://databank.worldbank.org).

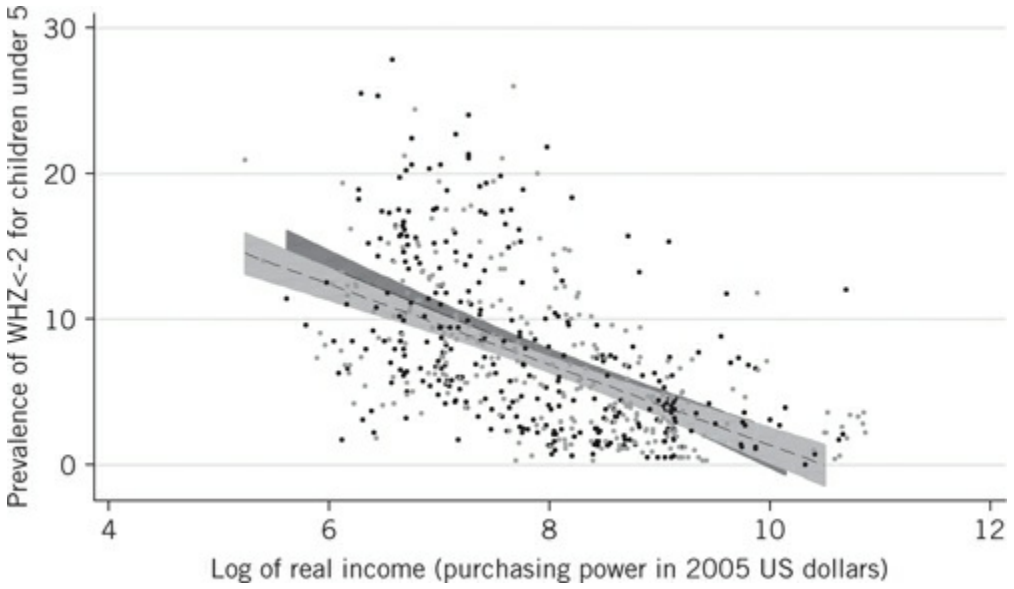


Figure 42-4 Prevalence of wasting and real income, 1966–1999 and 2000–2010 (WHO data, *N* = 601 from 146 countries).

Note: Data shown are for 1966–1999 (darker shading) and 2000–2010 (lighter shading), with 95% confidence intervals around linear regressions. Sample sizes are *N* = 316 across 127 countries for 1966–1999 and *N* = 285 across 126 countries for 2000–2010.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Stunting data are from WHO, Global Database on Child Growth Malnutrition, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statistics (http://databank.worldbank.org).

Overall, the anthropometric evidence shows a clear positive association between income and child nutritional status, with additional improvement in the 2000s relative to previous decades. This applies both to the prevalence of extreme undernutrition below a z score of -2 as shown in Figures 42-2, 42-3, 42-4, and also to national average child heights as shown in Figure 42-1. These changes in stunting, wasting, and average stature convey different kinds of information. Prevalence rates of extreme conditions are particularly important for targeting interventions to the most needy, whereas changes in means or medians provide more sensitive measures of population-wide changes.[17]

FOOD AVAILABILITY

Diet Quality and Dietary Energy

Child nutrition is not merely a matter of anthropometrics. Dietary patterns are important for many other reasons, including health and activity levels as well as cultural values. Figure 42-5 shows the major changes in food availability associated with per capita income, distinguishing between one aspect of diet quality (on the left axis) and total energy availability (on the right).

The data shown in Figure 42-5 refer to a country's national average family diet. Direct surveys of what children eat have not yet had sufficient precision, over a large enough sample size and range of incomes, to capture the economic-development effects described in this chapter. In fact, even the national-average data shown here are estimates computed by Food and Agriculture Organization (FAO) staff from other observations, using an accounting approach to balance consumption against total production, waste and seed use, international trade, and stock changes. These "Food Balance Sheets" infer the national-average diet from other more readily observed flows in food markets.[18]

The energy availability shown in Figure 42-5 includes all kind of calories. A corresponding measure of food quality could be computed in various dimensions, such as dietary diversity (e.g., the number of distinct types of food consumed on a given day) or food security (e.g., the fraction of time in which households are unable to access needed foods). Here, we introduce the simplest and most direct possible measure of dietary composition, namely the fraction of calories that are derived from animal sources including dairy, eggs, and fish, as well as animal meats.

Figure 42-5 illustrates how many of the world's poorest countries have a nearly vegan diet, while higher income countries get 20% or more of their total calories from animal sources. Various kinds of anthropological and contemporary evidence suggests that availability of these foods at the low levels of consumption observed historically and in today's low-income countries could play a causal role in stature,[19] although that association could also be because of reverse causality, if people who are better nourished and also have higher incomes prefer more animal-sourced foods for taste or other reasons.

DISEASE EXPOSURE AND TREATMENT

Sanitation and Water Supplies, Breast-feeding and

Diarrhea Treatment

Nutritional status depends on the balance between intake and needs, which in turn depends on disease exposure and treatment. Figures 42-6 and 42-7 illustrate some key dimensions of how these factors change during economic development, using datasets assembled by WHO, UNICEF, and the DHS.[14]

Figure 42-6 shows the interrelated issues of access to sanitation and access to improved water sources, compiled by the WHO/UNICEF Joint Monitoring Program (JMP) for Water Supply and Sanitation. For our data, access to sanitation is defined as households using any kind of improved facility such as a protected pit latrine or composting toilet as well as flush toilets, piped sewage and septic tanks, as opposed to open pits, buckets, hanging toilets, or flat land. Access to improved water is defined as any improved source such as protected wells and springs as well as piped water sources, as opposed to open springs, wells, and tanks or surface water. As shown by the two scatter plots and regression lines, in the poorest countries under 20% of the population has access to sanitation, whereas around 50% has access to improved water supplies, and both converge to near 100% at higher income levels. The improvement in sanitation is clearly one of the most dramatic changes in children's nutrition-related environment, being closely associated with diseases discussed elsewhere in this volume including environmental enteropathy.[20]

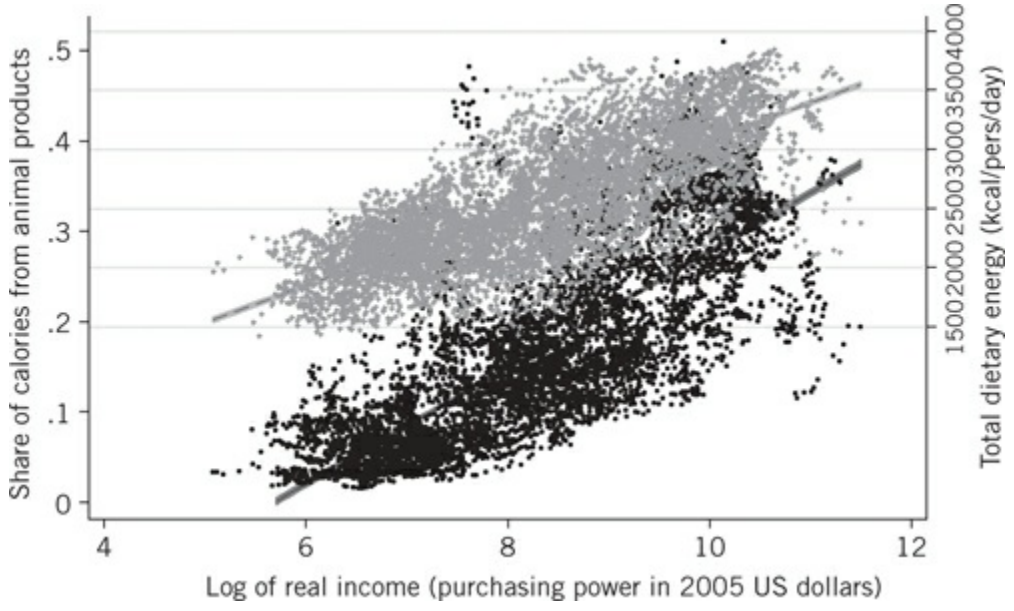


Figure 42-5 Food supply and real income, 1961–2009 (FAO

estimates, *N* = 7031 from 169 countries).

Note: Animal product share is shown in darker shading on the left scale; total energy is shown in lighter shading on the right (higher) scale. Both variables have the same sample size, and linear regression lines with 95% confidence intervals.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Food supply data are Food Balance Sheet estimates, downloaded December 30, 2012 from http://faostat3.fao.org.

Figure 42-7 addresses two other, very different aspects of nutrition-related disease exposure and treatment, namely the fraction of infants under 6 months who are exclusively breast-fed (as opposed to other drinks or solid food), and the fraction of children under 5 for whom diarrhea is treated with oral rehydration and continued feeding (as opposed to suspension of feeding, or other liquids). These appear to have almost no significant correlation with real income across the countries surveyed. The fact that higher income people do not pursue these practices more often does not imply that they are undesirable. Richer people may be able to afford more breast milk substitutes and have less exposure to diarrheal diseases, or may be surveyed differently regarding these practices.

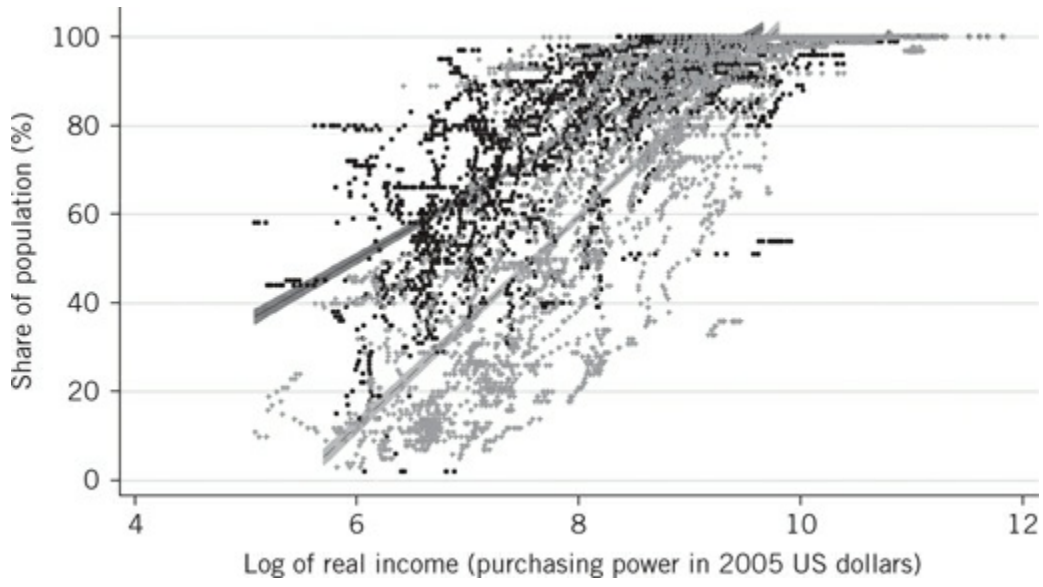


Figure 42-6 Access to sanitation, improved water supplies, and real income, 1990–2010 (WHO and UNICEF data).

Note: Access to improved water (*N* = 3556 from 178 countries) is shown in darker shading; access to sanitation (*N* = 3485 from countries) is shown in lighter shading. Both variables have linear regression lines showing 95% confidence intervals.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Water and sanitation data are from WHO and UNICEF Joint Measurement Program, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statics (http://databank.worldbank.org).

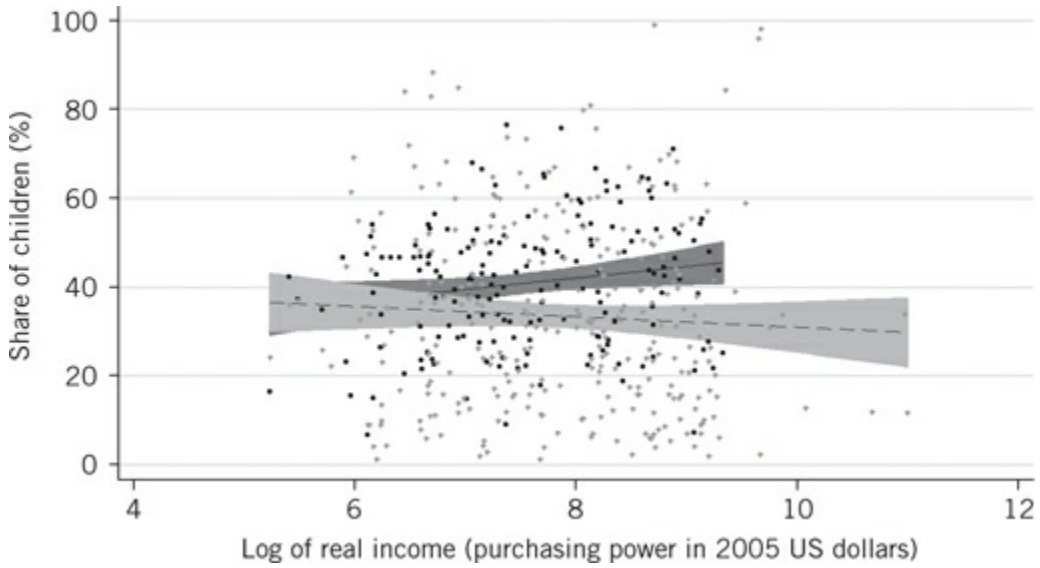


Figure 42-7 Exclusive breast-feeding, diarrhea treatment, and real income, 1990–2010 (UNICEF and DHS data).

Note: Treatment of diarrhea (% of children under 5 receiving oral rehydration and continued feeding) is shown in darker shading, with *N* = 174 from 96 countries. Exclusive breast-feeding (% of children under 6 months) is shown in lighter shading, with *N* = 297 from 134 countries). Both variables have linear regression lines with 95% confidence intervals.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Breast-feeding and diarrhea treatment data are from UNICEF and the Demographic and Health Surveys (DHS) by Macro International, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statics ((http://databank.worldbank.org).

VACCINATION AND SUPPLEMENTATION

Immunization and Vitamin A Campaigns

The data presented so far are the aggregate result of decentralized decisions taken by individual households. None of the variables shown in Figures 42-1, 42-2, 42-3, 42-4, 42-5, 42-6, 42-7 are directly controlled by government or philanthropic agencies, although these organizations' policies, programs, and interventions do have some influence on individual household behavior. Centralized agencies are relatively more important for the vaccination and supplementation campaigns presented in Figures 42-8 and 42-9, which are typically undertaken on a regional scale by a small number of organizations, to supplement whatever households can do for themselves. Figure 42-8 shows coverage data for vaccination against tuberculosis (BCG) and against diphtheria, pertussis, and tetanus (DPT) among children surveyed between their first and second birthdays, and Figure 42-9 shows coverage of measles vaccination and vitamin A supplementation. Vaccination data are from WHO and UNICEF, whereas vitamin supplementation data are from UNICEF.[14]

The vertical axes in Figures 42-8 and 42-9 show the proportion of children receiving vaccination and supplementation, so the scaling of these scatter plots and regression lines is directly comparable to the population shares shown in Figures 42-6 and 42-7. Visual comparison reveals that vaccination coverage (Figures 42-8 and 42-9) has wider variation at each income level, and less steep increase at higher incomes, than household access to improved sanitation (Figure 42-6). One reason could be that vaccination requires less investment by households themselves, and so can more readily be provided in low-income settings. Vitamin A supplementation coverage (Figure 42-9) is like diarrhea treatment in its relation to economic development, with very wide variation at each income level and less use at higher incomes. Supplementation may be less widely needed at higher incomes, or less likely to attract the support of public and philanthropic agencies.

STRUCTURAL CHANGES AFFECTING CHILD NUTRITION

Agricultural Transformation and Demographic Transition

The figures presented above describe how various dimensions of child nutrition vary with the growth of national income, showing strong trends as well as variation across countries and over time. Two other important influences on child nutrition during economic development involve

structural change within society: the agricultural transformation from farming to other occupations, and the demographic transition from short to long life expectancy. These changes are described in detail elsewhere,[21] and are briefly summarized here in respect to their influence on child nutrition.

Agricultural transformation is a shift in employment and expenditure from food to other enterprises. In the world's poorest societies, most people have no choice but to live in rural areas, relying on natural resources and household labor to obtain food, fuel, and water. If opportunities arise to improve productivity, either within agriculture or in other activities, then people can save and invest, specialize and trade, ultimately moving from into a wide array of services and manufacturing. This transformation occurs within rural areas, and also allows migration to towns and cities, which sustains further economic development. The transformation out of agriculture as incomes rise explains why the poorest people and most malnourished children are typically found in the most agriculturally oriented places, both across and within countries. Because of agricultural transformation, the apparent paradox in economic development that food production is associated with being malnourished can typically be resolved by controlling for real income.

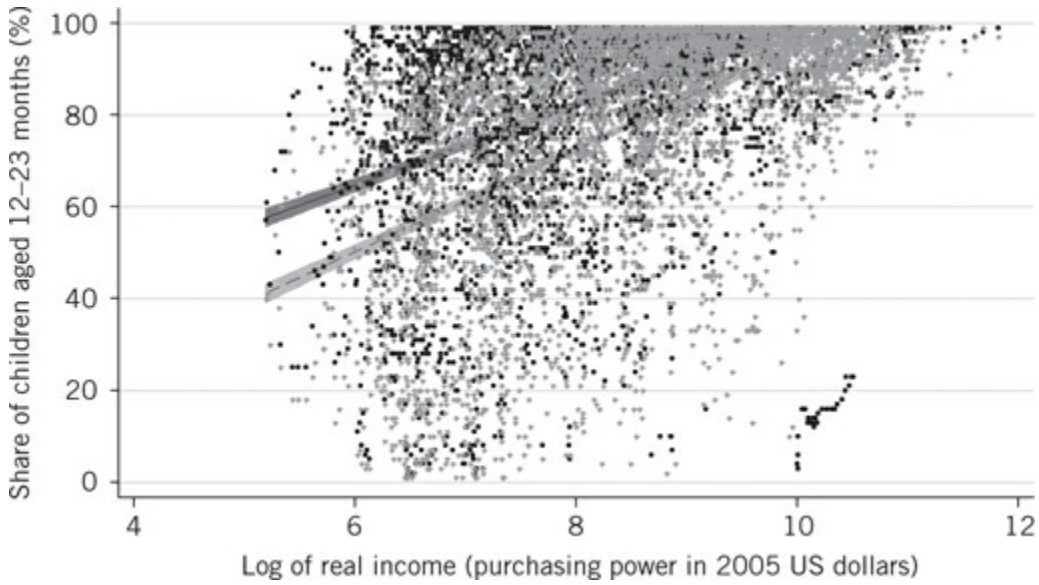


Figure 42-8 BCG and DPT vaccination rates and real income, 1980–2010 (WHO and UNICEF data).

Note: BCG vaccination against diphtheria, pertussis, and tetanus (% of

children, 3 doses) is in lighter shading, with *N* = 5096 from 183 countries. Both variables have linear regression lines with 95% confidence intervals.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Vaccination is from WHO and UNICEF, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statics ((http://databank.worldbank.org).

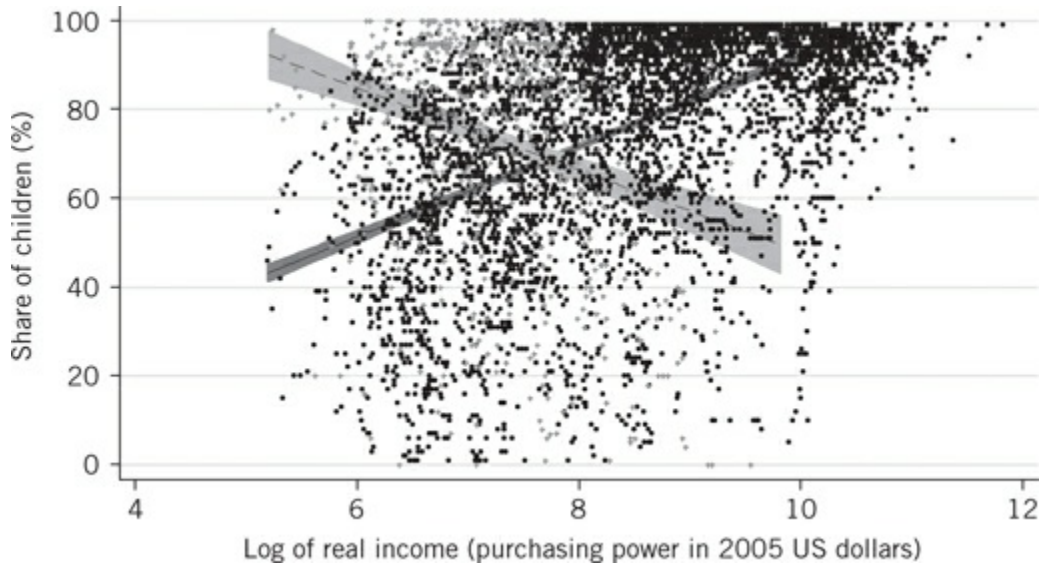


Figure 42-9 Measles vaccination, vitamin A supplementation, and real income, 1980–2010 (WHO and UNCEF data).

Note: Measles vaccination (% of children aged 12–23 mo, single dose) is shown in darker shading, with *N* = 5005 from 183 countries. Vitamin A supplementation (% of children aged 6–59 mo, at least one capsule in the 6 mo before survey) is shown in lighter shading, with *N* = 602 from 86 countries. Both variables have linear regression lines with 95% confidence intervals.

Source: Author's calculation. Income data are from Penn World Table Version 7.1 (November 2012), online at https://pwt.sas.upenn.edu. Vitamin A data begin in 1999 and are from UNICEF. Vaccination data are from WHO and UNICEF, downloaded December 30, 2012 from World Bank, Health Nutrition and Population Statics ((http://databank.worldbank.org).

Demographic transition is another kind of structural change with surprising implications for child nutrition. This familiar shift toward longer life and then also lower fertility almost always starts with lower child

mortality, often associated with an improvement in nutrition. That can lead quickly to a decline in birth rates, particularly if accompanied by opportunities for education, women's employment, and access to contraception, but in the meantime there is a burst of population growth and a demographic burden of more children per household. The rise in child dependency during the demographic transition helps explain why child malnutrition often worsens over time, before it improves. The demographic burden of child care falls disproportionately on young women, until declining fertility rates help the age structure swing back toward more working-age adults per child. The eventual rise in the workforce as a fraction of the population then offers a demographic gift that can fuel rapid economic development, particularly when combined with agricultural transformation and off-farm employment growth.

Demographic transition interacts with agricultural transformation in a way that often makes nutrition improvement harder in low-income countries, and then easier at higher income levels. In poor countries, the nonagricultural sector is initially so small that even very rapid rates of off-farm employment growth cannot absorb all of the increasing number of working-age people. Despite successful year-to-year economic development and rapid urbanization, there will be a rising number of workers who have no choice but to farm, typically with less and less land and other natural resources per worker. That burden of declining land area per farmer in poor countries can be offset only by raising their crop yields, until the size of their country's nonagricultural sector has become large enough its annual growth to absorb each year's increase in the adult workforce.

In summary, these two structural changes associated with economic development impose temporary burdens on child nutrition in low-income countries. By the arithmetic of year-to-year change, they serve to raise the number of children per adult and lower the area of land per farmer in the early stages of structural change, making progress harder before it gets easier. At later stages of economic development, child dependency rates and area per farmer can increase, both of which help the poorest and most malnourished escape more quickly from poverty. The development process may start earlier or later, varies widely in speed and occasionally stalls altogether. But to the extent that economic development proceeds, it is characterized by remarkable similarities that help explain the patterns we see across countries and over time. In that sense, successful societies are alike even as their progress toward that shared destination involves a variety of diverse forces.

CONCLUSIONS

This chapter describes changes in child nutrition that have been associated with economic development over several decades in more than 100 countries. We use data recently assembled from millions of children to compare changes in purchasing power with observed child height, stunting, and wasting, as well as various contributors to nutritional status including food availability and diet quality, sanitation and water supplies, breast-feeding behavior and treatment of childhood diarrhea, vaccination, and vitamin A supplementation. Each aspect of child nutrition has wide variation across countries, but some are closely linked to national income.

The most notable links between nutrition and income are for child height and stunting, both of which improve sharply with economic development especially among the poorest countries. We also find strong links for access to sanitation and diet quality, as measured here by the fraction of calories obtained from animal products. Total food availability, as measured by total calories per person, and access to improved water as well as child vaccination rates are also associated with higher income, although vaccination rates have wide variance across countries at each income level.

Nutrition practices that have almost no correlation with income across the available data are the rates at which children are exclusively breast-fed, or are treated for diarrhea with oral rehydration and continued feeding. Vitamin A supplementation rates are actually lower in more developed countries. These low or negative correlations with income could be because of diminished need for these practices as income rises, or to a lack of public support in richer countries, or perhaps to less frequent observation even when they are practiced.

The links between child nutrition and economic development described in this chapter are accompanied by very large changes within countries, including the agricultural transformation from a focus on food production to more other activities, and the demographic transition from large to small families. Together these impose headwinds against nutrition improvements in low-income countries that help explain the cross-country patterns we see, and make success in those settings all the more remarkable. The extraordinary gains in nutrition associated with economic development are driven by deep social and institutional changes involving gender relations, educational and child care practices as well as medical and nutritional innovations described elsewhere in this volume, all of which offer opportunities for health practitioners and policy officials to achieve even greater improvements in the future.

REFERENCES

1. Maddison A. *The World Economy: A Millennial Perspective*. Vol. 1. and *Historical Statistics*. Vol. 2. Paris, France: OECD; 2006.
2. Floud R, Fogel RW, Harris B, Hong SC. *The Changing Body: Health, Nutrition, and Human Development in the Western World Since 1700*. New York, NY: Cambridge University Press; 2011.
3. Sacks DW, Stevenson B, Wolfers J. The new stylized facts about income and subjective well-being. *Emotion.* 2012;12(6):1181–1187.
4. Dasgupta P. *An Inquiry into Well-Being and Destitution*. New York, NY: Oxford University Press; 1995.
5. Sen AK. *Development as Freedom*. New York, NY: Oxford University Press; 1999.
6. Heston A, Summers R, Aten B. Penn World Table. https://pwt.sas.upenn.edu. Accessed December 30, 2012.
7. Block SA, Masters WA, Bhagowalia P. Does child undernutrition persist despite poverty reduction in developing countries? Quantile regression results. *J Dev Studies.* 2012;48:1699–1715.
8. Pelto GH, Armar-Klemesu M. Balancing nurturance, cost and time: complementary feeding in Accra, Ghana. *Mat Child Nutr*. 2011;7(suppl 3):66–81.
9. Arabi M, Frongillo EA, Avula R, Mangasaryan N. Infant and young child feeding in developing countries. *Child Dev.* 2012;83:32–45.
10. Alderman H. The response of child nutrition to changes in income: linking biology with economics. *CESifo Econ Stud*. 2012;58:256–273.
11. Victora CG, de Onis M, Hallal PC, et al. Worldwide timing of growth faltering: revisiting implications for interventions. *Pediatrics*. 2010;125(3):e473–e480.
12. Ezzati M, et al. Nutrition Impact Model Study (NIMS) Data. http://www1.imperial.ac.uk/publichealth/departments/ebs/projects/eresh/majidezza Accessed December 30, 2012.
13. Stevens GA, Finucane MM, Paciorek CJ, et al. Trends in mild, moderate, and severe stunting and underweight, and progress towards MDG 1 in 141 developing countries: a systematic analysis of population representative data. *Lancet.* 2012;9844:824–834.
14. World Bank. World DataBank. http://databank.worldbank.org. Accessed December 30, 2012.
15. Preston SH. The changing relation between mortality and level of economic development. *Popul Stud.* 1975;29:231–248.
16. The Economist. Malnutrition in Guatemala: a national shame. *The Economist* Magazine. August 27, 2009. Available at www.economist.com/node/14313735. Accessed December 30, 2012.
17. Bhagowalia P, Chen SW, Masters WA. Effects and determinants of mild underweight among preschool children across countries and over time. *Econ Hum Biol.* 2011;9:66–77.
18. FAO (Food and Agricultural Organization of the United Nations). Food balance sheets. FAOSTAT http://faostat3.fao.org. Accessed December 30, 2012.
19. Larsen CS. Animal source foods and human health during evolution. *J Nutr.* 2003;133(11):3893S–3897S.
20. Dewey KG, Mayers DR. Early child growth: how do nutrition and infection interact? *Matern Child Nutr.* 2011;7(suppl 3):129–142.
21. Norton G, Alwang J, Masters WA. *Economics of Agricultural Development: World Food Systems and Resource Use*. 3rd ed. New York, NY: Routledge; 2014.

Global Interventions in Child Health

43

Zulfiqar A. Bhutta, MBBS, FRCP, FRCPCH, FCPS, PhD and Jai K. Das, MD, MBA

The Millennium Summit in the year 2000 led to the formulation of the Millennium Declaration that advocated the provision of basic human rights to all.[1,2] It led to the constitution of Millennium Development Goals (MDGs), which consisted of eight core goals that marked the culmination of years of exhaustive research and consultations, and was approved by 193 United Nations member states.[1,2] Of these goals was the MDG 1; which was to eradicate extreme poverty and hunger and halve, between 1990 and 2015, the proportion of people who suffer from hunger and MDG 4; which was to reduce the under-5 child mortality by two-thirds (baseline of 1990) by the year 2015. Although the role of nutrition in these two MDGs is direct and obvious, nutritional status, dietary intake, food security, and production are integral components of the other MDG's as well (Table 43-1).[3] Malnutrition hinders child education and school performance (MDG 2) and is also risk factors for infection transmission including HIV, malaria, and TB (MDG 6). Gender biases impact women's access to food and health care, consequently leading to maternal undernutrition (MDG 3), hence resulting in compromised maternal health (MDG 5).

Child mortality has been a focus of public health interventions since the early 1980s, and much progress has been made from the early 1990s, when under-5 mortality rates were approximately 12.5 million deaths a year to 2011 when child deaths are approximately 6.9 million.[4] This reduction is notable as the total child population has grown during this time, but has unfortunately not been adequate to achieve the end-points for

2015. The acceleration is slower than desired and now would require a reduction of 13.5% per year as opposed to current global rates of fall of 2.2% per year[5] to be within the reach of MDG 4

In addition, this burden is spread disproportionately across the world and regions of the world with the highest mortality in the Central African states followed by South East Asia, Eastern Mediterranean, and Western Pacific regions, all of which account for around 92%–99% of all child mortality.[6] Unfortunately, most of these deaths are preventable or treatable with existing interventions that are currently available and recommended for universal implementation. About one-third of these deaths in children under 5 years of age are due to underlying undernutrition, which includes stunting, severe wasting, deficiencies of vitamin A and zinc, and suboptimum breast-feeding (BF).[7] An estimated 178 million children under five suffer from stunting, the vast majority in sub-Saharan Africa and South-central Asia (Figures 43-1 and 43-2).[8] Of these, 160 million (90%) live in just 36 countries, representing almost half (46%) of the 348 million children in those countries.[8] An estimated 55 million children are wasted, 19 million of these are severely affected and at high risk of premature death.[8] Altogether, more than 3.5 million mothers and children under five die unnecessarily each year in poor countries due to the underlying cause of undernutrition.[9]

TABLE 43-1 Relationship of Nutrition and MDGs[3]

Goal	Nutrition Effect
Goal 1: Eradicate extreme poverty and hunger.	Malnutrition erodes human capital through irreversible and intergenerational effects on cognitive and physical development.
Goal 2: Achieve universal primary education.	Malnutrition affects the chances that a child will go to school, stay in school, and perform well.
Goal 3: Promote gender equality and empower women.	Anti-female biases in access to food, health, and care resources may result in malnutrition, possibly reducing women's access to assets. Addressing malnutrition empowers women more than men.
Goal 4:	Malnutrition is directly or indirectly associated with most child

Reduce child mortality.	deaths, and it is the main contributor to the burden of disease in the developing world.
Goal 5: Improve maternal health.	Maternal health is compromised by malnutrition, which is associated with most major risk factors for maternal mortality. Maternal stunting and iron and iodine deficiencies particularly pose serious problems.
Goal 6: Combat HIV/AIDS, malaria, and other diseases.	Malnutrition may increase risk of HIV transmission, compromise antiretroviral therapy, and hasten the onset of full-blown AIDS and premature death. It increases the chances of tuberculosis infection, resulting in disease, and it also reduces malarial survival rates.

Source: Adapted from Gillespie and Haddad.[3]

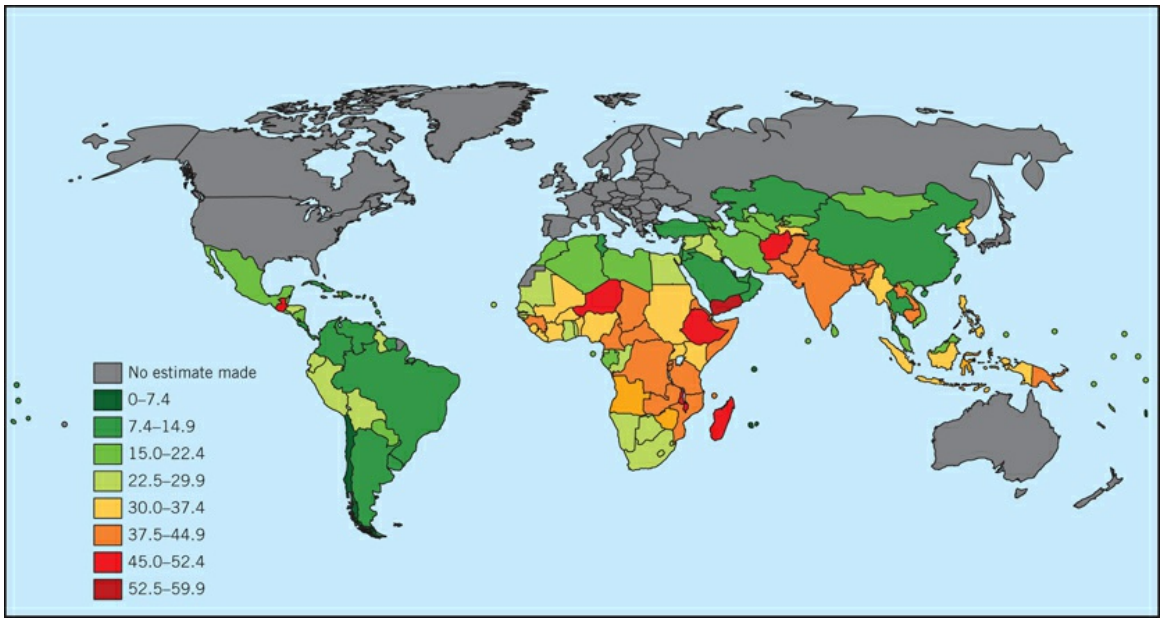


Figure 43-1 Prevalence of stunting in 2011. (Modified from Stevens et al.[79])

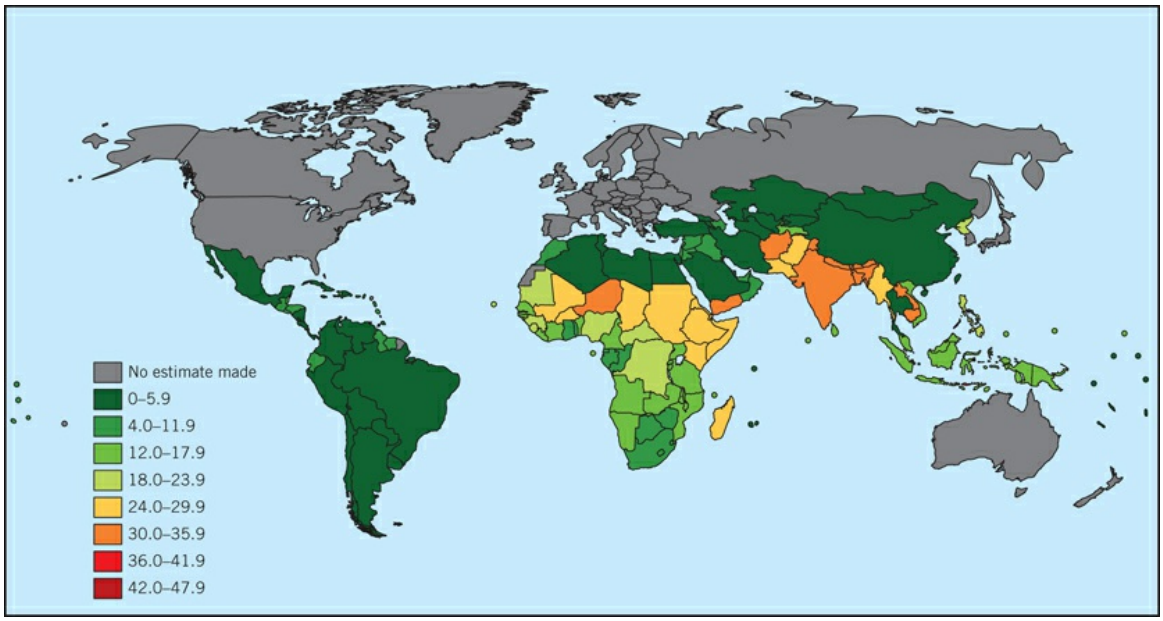


Figure 43-2 Prevalence of underweight in 2011. (Modified from Stevens et al.[79])

Evidence-based systematic reviews suggest that substantial improvements in child health can be achieved by making existing, proven, key and affordable interventions available to children who need them the most. Table 43-2 summarizes the maternal directed as well as the child health interventions to address childhood health and undernutrition. These are some of the key steps toward achieving the targets set forth in the MDG 4, especially in the 75 developing countries that have some of the highest child mortality rates as established by United Nations Children's Fund (UNICEF). Knowing the magnitude of the task at hand and the success stories that have lit the way after years of hardwork is extremely important in planning ahead. Any future strategies in reducing excessive child deaths will depend on the availability of local data, and clearly marked evidence of successful and unsuccessful interventions. Our discussion here will give an in-depth account of these interventions along with their effectiveness. We will also discuss the various delivery strategies that can act as catalyst and help ensure that these amenities reach the most needed and at the same time ensuring equity.

WHAT WORKS: INTERVENTIONS TARGETED

TOWARD WOMEN

Poor maternal nutrition contributes to at least 20% of maternal deaths, and increases the probability of other poor pregnancy outcomes including newborn deaths.[10] Prevalence of low body mass index (BMI) among women of reproductive age is an important risk factor for intrauterine growth restriction and low birth weight (LBW), as well as for neonatal mortality. Low BMI is not the only indicator; key indicators include low maternal stature, anemia, and other micronutrient deficiencies. In the following section, we discuss the evidence from maternal-directed nutrition interventions, which impact not only maternal undernutrition but also have subsequent impact on birth outcomes and child health.

Maternal Micronutrient Supplementation

Women in developing countries often consume inadequate levels of micronutrients due to limited intake of animal products, fruits, vegetables, and fortified foods. The resulting micronutrient deficiencies are exacerbated in pregnancy leading to potentially adverse effects on the mother, such as anemia, hypertension, complications of labor, and even death.

Maternal iron deficiency is widespread and globally about 42% pregnant women are anemic. Iron deficiency is the most common cause of anemia and is a risk factor for perinatal complications such as preeclampsia, LBW, prematurity, and perinatal mortality.[11,12] Studies have provided strong evidence to suggest that iron supplementation with or without folic acid results in a significant reduction in the incidence of anemia during pregnancy.[13,14] It has also been reported that iron supplementation during pregnancy can reduce perinatal mortality.[15] A recent review by Imdad et al[16] shows that daily iron supplementation in pregnant women results in a 70% reduction (Relative Risk [RR]: 0.30, 95% Confidence Interval [CI]: 0.19–0.46) in the incidence of anemia at term, a 67% reduction (RR: 0.33, 95% CI: 0.16–0.69) in iron deficiency anemia at term and a 19% reduction (RR: 0.81, 95% CI: 0.68–0.97) in the incidence of LBW.[16] Iron supplementation also has a significant effect on mean birth weight (Mean Difference [MD]: 30.81g, 95% CI: 5.94–55.68). There was however no effect reported on the incidence of preterm or small-for-gestational-age births (SGA).

Another essential micronutrient during pregnancy is calcium, and studies have shown that an inverse relationship exists between calcium intake and the development of hypertension in pregnancy.[17,18]

Preeclamptic women have 2.7 times increased risk of delivering infants with intrauterine growth restriction.[19] Calcium supplementation during pregnancy is associated with a reduction in the risk of gestational hypertensive disorders and has also shown a 52% reduction (RR: 0.48, 95% CI: 0.34–0.67) in the incidence of preeclampsia and 24% reduction (RR: 0.76, 95% CI: 0.60–0.97) in preterm birth accompanied with an increase in birth weight of 85 g.[20] However, there was no effect observed for calcium supplementation on LBW, perinatal mortality, and neonatal mortality.

TABLE 43-2 Maternal and Child Nutrition Interventions

Maternal Interventions	Child Interventions
Maternal balanced energy protein supplementation	*BF*
Micronutrient Supplementation:	Micronutrient Supplementation:
Iron/Iron-folate supplementation	Iron supplementation
Maternal calcium supplementation	VAS
Maternal iodine supplementation	Zinc Supplementation
Maternal MMN supplementation	MMN supplementation/Micronutrient Powder (MNP)
	Complementary Feeding
Disease Prevention:	Disease Prevention:
Deworming	Deworming
Malaria prevention and treatment	Malaria prevention and treatment
WASH strategies	WASH strategies
Delivery Strategies	
IMCI, CHDs, School feeding programs, Community platforms	

Animal and human studies have shown that iodine deficiency has multiple adverse effects on growth and development that are collectively termed the iodine deficiency disorders. Iodine deficiency during pregnancy impairs the neurological development of the fetus as thyroid hormone is required for the normal neuronal migration and myelination of the brain during fetal and early postnatal life, and hypothyroxinemia during these critical periods causes irreversible brain damage, with mental retardation and neurological abnormalities.[21] A review by Zimmermann et al[22]

reported that maternal iodine supplementation in moderate-to-severe iodine-deficient areas before or during early pregnancy eliminates new cases of cretinism at 4 years of age by 73% (RR: 0.27, 95% CI: 0.12–0.60), increases birth weight (3.82%–6.30% higher), reduces rates of infant mortality (25%–60% reduction), and increases developmental scores in young children by 10%–20%.[22]

Since multiple-micronutrient (MMN) deficiencies often coexist in low- to middle-income countries, MMN supplementation could be one intervention to limit the potentially adverse newborn health outcomes. A recent Cochrane review[23] shows that MMN supplementation during pregnancy significantly decreased the number of LBW infants by 11% (RR: 0.89, 95% CI: 0.83–0.94) and SGA by 13% (RR: 0.87, 95% CI: 0.81–0.95). MMN supplementation, however, was not associated with lower rates of preterm birth, miscarriage, preeclampsia, maternal mortality, perinatal mortality, still birth, or neonatal mortality.

Maternal Balanced Protein Energy Supplementation

Macronutrient interventions have also been proposed and evaluated in accordance with the maternal needs during pregnancy. Some of the macronutrient interventions include dietary advice to pregnant women, balanced protein energy supplementation, high protein, isocaloric protein supplementation, and prescribing a low-energy diet to pregnant women who are either overweight or who exhibit high weight gain earlier in gestation.[24,25] Balanced protein energy supplementation is defined as nutritional supplementation during pregnancy in which proteins provide less than 25% of the total energy content (ranging from 417 to 1017 kcal). A review by Imdad and Bhuta[26] shows that providing pregnant women with balanced protein energy supplementation results in a significant reduction of 34% (RR: 0.66, 95% CI: 0.49–0.89) in the risk of giving birth to SGA infants and a 38% reduction (RR: 0.62, 95% CI: 0.40–0.98) in stillbirth as well as an increase in birth weight (MD: 73 g, 95% CI: 30–117). These effects were more pronounced in malnourished women when compared to adequately nourished women.

WHAT WORKS: INTERVENTIONS TARGETED TOWARD NEWBORNS/CHILDREN

Factors responsible for childhood undernutrition include lack of early initiation of BF and exclusive breast-feeding (EBF) in the first six months

after birth, lack of sufficient macronutrients and calorie intake, and micronutrient deficiencies (especially vitamin A, iron, iodine, and zinc).

Breast-Feeding Promotion

Breast milk provides numerous immunologic, psychologic, social, economic, and environmental benefits; and is a natural first food and an ideal nutrition for the newborn.[27] BF is therefore recommended as the optimal strategy for feeding newborns and young infants[28–31] (see Chapters 30–32). BF has a significant positive impact on child growth and development and decreases the risk for many acute and chronic diseases,[32–37] including infections such as diarrhea and respiratory tract infections during infancy.[38] EBF rates in infants less than 6 months of age vary from as low as 20% in central and eastern European countries to 44% in South Asia.[39] The reasons for low prevalence of EBF could be the lack of support for BF by social workers and health-care providers, emotional stress in mothers and their perception of not having enough breast milk, and pressure from close relatives to introduce other liquid and solid foods, unsupportive hospital practices that delay early initiation of BF, maternal employment, and lack of commercial advertising.[40] For reasons like these, effective programs of education, counseling, and support are considered necessary to not only promote BF but also to prolong the duration of EBF to up to 6 months of age. BF promotion, therefore, is a global priority with benefits for maternal and child health, especially in low-/middle-income countries where its relevance for child survival is undisputed.[41]

World Health Organization (WHO) recommends EBF the infant for the first six months of life to achieve optimal growth, development, and health.[42] A review by Imdad et al[43] evaluated the effect of BF education and promotion interventions on BF rates and concluded that EBF at 4–6 weeks postpartum improved significantly by 43% with 89% and 20% significant increases in developing and developed countries respectively when compared to no intervention. EBF at six months improved by 137% with a significant six times increase in EBF in developing countries, compared to 1.3-fold increase in developed country studies. The review also showed that prenatal counseling had a significant impact on BF outcomes at 4–6 weeks, although both prenatal and postnatal counseling were important for EBF at six months.

Complementary Feeding

Complementary feeding for infants refers to the timely introduction of safe

and nutritional foods in addition to BF, that is, clean and nutritionally rich additional foods introduced at about 6 months of infant age[35] (see Chapters 36 and 43). These foods are typically provided to children from 6 to 24 months of age.[44] It has been suggested that in addition to disease prevention strategies, complementary feeding interventions targeting this "critical window" are most efficient in reducing malnutrition and promoting adequate growth and development.[45] A review by Imdad et al[46] evaluated the impact of nutrition counseling alone and of provision of complementary foods (with or without nutrition counseling) and showed that these interventions resulted in a significant increases in weight (Weighted Mean Difference [WMD]: 0.30 Standard Deviation [SD], 95 % CI: 0.05–0.54 and WMD: 0.34 SD, 95% CI: 0.11–0.56, respectively) and linear growth (WMD: 0.21 SD, 95 % CI: 0.01–0.41 and 0.26 SD, 95 % CI: 0.08–0.43, respectively).

Micronutrient Supplementation

It is estimated that approximately 600 million preschool and school-aged children are anemic worldwide, and at least half of the cases are due to iron deficiency.[47] A review by De-Regil et al[48] evaluating the impact of iron supplementation in children showed that children receiving intermittent iron supplements have a 49% lower risk of anemia (RR: 0.51, 95% CI: 0.37–0.72) and a 74% lower risk of iron deficiency (RR: 0.24, 95% CI: 0.06–0.91) and have higher hemoglobin (MD: 5.20 g/L, 95% CI: 2.51–7.88) and ferritin concentrations (MD: 14.17 µg/L, 95% CI: 3.53–24.81). Nonsignificant effects were observed for height-for-age and weight-for-age. Evidence on mental development, motor skill development, school performance, and physical capacity was evaluated by very few studies and indicated no clear effect of iron supplementation.

A review by Haider and Bhutta[49] evaluating the effectiveness of neonatal vitamin A supplementation (VAS) showed that for term neonates, infant mortality at six months was reduced significantly by 18% in the vitamin A group compared with the control group (RR: 0.82, 95% CI: 0.68–0.99) and among all infants a 14% significant reduction in the risk of infant mortality at six months was observed (RR: 0.86, 95% CI: 0.77–0.97). A review by Imdad et al[50] on the effectiveness of VAS for preventing morbidity and mortality in children aged 6 months to 5 years showed a 24% reduction in the risk of all-cause mortality (RR: 0.76, 95% CI: 0.69–0.83) and a 28% overall reduction in diarrhea mortality (RR: 0.72, 95% CI: 0.57–0.91). There was no significant effect observed of

VAS on cause-specific mortality due to measles, respiratory disease, and meningitis. VAS also reduced incidence of diarrhea by 15% (RR: 0.85, 95% CI: 0.82–0.87) and measles morbidity by 50% (RR: 0.50, 95% CI: 0.37–0.67); however, there was no significant effect on the incidence of respiratory disease or hospitalizations due to diarrhea or pneumonia.

Zinc is a trace element that serves greater than 300 bodily functions, and deficiency of this micronutrient is said to play a major role in the pathogenesis of diarrhea.[51] Current evidence collected over the last decade suggests that zinc can potentially confer a protective effect to two particular systems, which are the gastrointestinal and respiratory systems.[52,53] Adequate zinc is a prerequisite to maintaining normal development, growth, and also contributes substantially to maintaining the immune system, therefore, its importance cannot be stressed enough in children under the age of 5, which suffer a large burden of diarrheal illnesses across the globe[54,55] (see Chapter 62). Current evidence suggests that the provision of zinc on a weekly basis for a period of one year among young children in developing countries can help lower mortality levels, by either reducing the severity of diarrhea, its duration, or by way of prevention.[51,56–58] A review by Imdad et al[59] of zinc supplementation showed that a dose of 10 mg zinc/d for duration of 24 weeks could lead to a net a gain of 0.37 (±0.25) cm in zinc-supplemented group compared to placebo. A separate review by Yakoob et al[60] evaluated 18 studies from developing countries showed that zinc supplementation decreased the incidence of diarrhea by 13% (RR: 0.87; 95% CI: 0.81, 0.94) and reduced pneumonia morbidity by 19% (RR: 0.81; 95% CI: 0.73, 0.90) but had nonsignificant effects on mortality. The impact of therapeutic zinc supplementation for the management of diarrhea was evaluated by Walker et al.[61] The zinc dose included was 10–40 mg/d, which aligns with the WHO recommendations of 20 mg/d (for children aged 6–59 months) and 10 mg/d (for those 1–5 months). The review concluded that zinc supplementation reduced all-cause mortality by 46% and diarrhea-related hospitalization by 23%, while there were nonsignificant impacts on diarrhea-specific mortality and diarrhea prevalence. In another review, therapeutic zinc supplementation as an adjunct in the treatment of diarrhea has shown to reduce the duration of acute diarrhea by 0.5 days and that of persistent diarrhea by 0.68 days.[62] These analyses support the inclusion of preventive zinc supplementation in public health programs to improve child health and survival.

Providing multiple micronutrients via supplements, powders, or

fortified ready-to-use foods is increasingly becoming a strategy for simultaneously addressing multiple nutrient deficiencies in developing countries. A review[63] on MMN supplementation with three or more micronutrients for children showed small effect sizes of (MD: 0.13, 95% CI: 0.06, 0.21) for length/height and (MD: 0.14, 95% CI: 0.03, 0.25) for weight, and there was limited evidence for an impact on outcomes such as morbidity and cognitive function. A review concluded that home fortification with micronutrient powder reduced anemia by 31% (RR: 0.69, 95% CI: 0.60–0.78) and iron deficiency by 51% (RR: 0.49, 95% CI: 0.35–0.67) in infants and young children, however, no effect was observed on growth. The micronutrient powders containing multiple nutrients are well accepted but adherence is variable and in some cases comparable to that achieved in infants and young children receiving standard iron supplements as drops or syrups.

WHAT WORKS: INTERVENTIONS FOR DISEASE PREVENTION

Recurrent infectious illnesses contribute to the undernutrition burden, and strategies devised at the prevention and treatment of recurrent infections can help reduce this burden. Among these include the water, sanitation, and hygiene (WASH) interventions, as well as interventions, such as vaccines, for the prevention and treatment of diarrhea and malaria.

There are serious adverse health consequences of poor access to WASH, particularly in children, including diarrhea, respiratory illness, trachoma, and death. A review drawing upon three systematic reviews focused on the effect of hand washing with soap on diarrhea, of water quality improvement and of excreta disposal. It concluded that hand washing with soap significantly reduces diarrhea morbidity by 47% (RR: 0.53, 95%CI: 0.37–0.76) though it depends on access to water. The effect of water treatment on diarrhea morbidity also appears similarly large with a 42% reduction (RR: 0.58, 95% CI: 0.46–0.72). There is very little rigorous evidence for the health benefit of sanitation.

WHO recommends that all school children should be treated at regular intervals with deworming drugs in helminthic prevalent areas. A recent Cochrane review[64] showed that single dose deworming had a nonsignificant effect on hemoglobin while multiple doses at one year follow up, also had a nonsignificant effect on weight, hemoglobin, cognition, and school attendance while the results were same for greater

than one year follow-up. But treating children after screening for worms showed that a single dose of deworming drugs may increase weight (MD: 0.58 kg, 95% CI: 0.40–0.76) and hemoglobin (MD: 0.37 g/dL, 95% CI: 0.1–0.64), while evidence on cognition was inconclusive. Administration of a single dose of antihelminthics in the second trimester of pregnancy also failed to show a statistically significant impact on maternal anemia (RR: 0.88, 95% CI: 0.69–1.13). A single dose of antihelminthics in the second trimester of pregnancy was not associated with an impact on LBW, preterm births, and perinatal mortality.

Effective interventions to prevent malaria morbidity and mortality include insecticide-treated mosquito nets (ITNs), indoor residual spraying, and intermittent preventive therapy (IPTc). A Cochrane review[65] concluded that antimalarials, when given to prevent malaria in pregnant women, increased mean birth weight significantly (MD: 126.70 g, 95% CI: 88.64–164.75 g) and reduced the incidence of LBW by 43% and severe antenatal anemia by 38%.[65] Another Cochrane review[66] evaluated the impact of ITNs with no nets and concluded that ITNs in pregnancy reduced LBW by 23% (RR: 0.77, 95% CI: 0.61–0.98) and fetal loss in the first to fourth pregnancy by 33% (RR: 0.67, 95% CI: 0.47–0.97), while effects were nonsignificant for anemia.

TABLE 43-3 Median National Coverage of Countdown Intervention (%) Based on Most Recent Measurement Since 2006, With Number of Countries Reporting and Range[68,69]

Countdown Indicator	No. of Countries With Data	Median Coverage (%)	Range (%)
Prepregnancy			
Demand for family planning satisfied	46	56	17–97
Pregnancy			
Antenatal care (at least one visit)	69	88	26–100
Antenatal care (four or more visits)	49	55	6–97
IPTp[a]	39	13	0–69
Neonatal tetanus protection	66	85	60–94

Birth			
Skilled attendant at birth	67	57	10–100
Postnatal			
Postnatal visit for mother	22	41	22–87
Postnatal visit for baby	4	50	8–77
Early initiation of BF	55	46	18–81
Infancy			
EBF	57	37	1–74
Introduction of semisolid/solid foods	39	73	16–94
Vitamin A (two dose)	56	92	0–100
Measles	73	84	46–99
DTP3	74	85	33–99
HiB3	58	83	45–99
Childhood			
ORT	53	45	7–68
ORS	57	33	10–77
Care seeking for pneumonia	57	55	13–83
Antibiotic treatment for pneumonia	45	39	3–88
ITN use[a]	36	34	3–70
Antimalarial treatment[a]	31	25	0–91
Water and sanitation			
Improved drinking water sources (total)	70	76	29–99
Improved sanitation facilities (total)	71	40	9–100

Abbreviation: ORT, Oral rehydration therapy.

[a]For the malaria indicators, the number of countries is based on those countries with 75% or more of the population at risk of *Plasmodium falciparum* transmission with an estimate during the time period 2006–2010.

Source: Adapted from Bhutta et al[68] and Countdown to 2015.[69]

Evidence-Based Intervention's Coverage Trends

Existing proven strategies to improve child health include promotion of BF, dietary supplementation of micronutrients, prevention of protein-energy malnutrition as far as possible, and improvement in the standard of preparation and hygiene of available weaning foods.[67] The median national coverage of these interventions in countdown countries since

2006 is summarized in Table 43-3.[68,69] All four vaccines (measles, DTP3, HiB3, neonatal tetanus protection) and vitamin A (two doses) have reached a median coverage level of 80% or more in the Countdown countries with available data. In most Countdown countries, vaccines and vitamin A are provided in fixed health facilities as well as during campaigns such as national child health days (CHDs) when outreach teams are able to reach high proportions of the population.

The current intervention coverage trends show that substantial progress is still needed. The median coverage levels for several interventions hover at or below 50% including interventions related to early initiation and EBF for at least six months, case management of childhood illnesses, and demand for family planning satisfied.[68,69] In absolute terms, the slowest absolute gains have been observed for oral rehydration solution (ORS) use and early initiation of BF, both with 4%. However, absolute gains need to be interpreted with caution because increases are harder to achieve if baseline levels are already high.[68,69]

Table 43-4 shows progress in intervention coverage from 2000–2005 to 2006–2011.[68,69] Coverages for the interventions requiring strong health systems (skilled attendant at birth, ORS) or those requiring behavior change (BF, care seeking for pneumonia) appear to be stalled between 30% and 50%, suggesting that what we have been doing for these interventions over the past decade is not working and needs to be reexamined, to find more effective ways of reaching the women and children who need to receive them.

WHAT CAN WORK: DELIVERY STRATEGIES AND PLATFORMS

Although individual interventions such as EBF, micronutrient supplementation, deworming, and malaria treatment have shown to be effective, an integrated approach is needed to achieve optimal results. The coverage of various proven interventions discussed above is below the mark and various delivery strategies have been undertaken to increase the uptake of these interventions. The measures discussed below not only ensure effective delivery but also ensure equity.

Integrated Management of Childhood Illnesses

In the mid-1990s, the WHO, in collaboration with UNICEF and other agencies, developed a strategy known as the Integrated Management of

Childhood Illness (IMCI). It includes both curative and preventive interventions targeted at improving the health practices at health facilities, at home, and in the community. Evaluations of IMCI in Uganda, Tanzania, Bangladesh, Brazil, Peru, South Africa, China, Armenia, Nigeria, and Morocco have shown various benefits in health service quality, mortality reduction, and health-care cost savings.[70] In Bangladesh, in the last two years of the study, the mortality rate was 13.4% (95% CI: 14.2–34.3) lower in IMCI than in comparison areas, corresponding to 4.2 fewer deaths per 1000 live births (95% CI:4.1–12.4, *P* = 0.30). It also reported an increase in exclusively breast-fed children (76% vs. 65%, difference of differences 10.1%, 95% CI: 2.65–17.62), and the prevalence of stunting in children aged 24–59 months decreased more rapidly than in comparison areas.[71]

TABLE 43-4 Trends in Countdown Indicators, Countries With Data From at Least Two Surveys; 2000–2005 and 2006–2011[68,69]

Countdown Indicator	No. of Countries With Data	Median Coverage, 2000–2005 (%)	Range, 2000–2005 (%)	Median Coverage, 2006–2011 (%)	Range, 2006–2010 (%)	Ch in Pe Po
Antenatal care (at least one visit)	61	76	27–98	88	34–100	
Skilled attendant at birth	61	49	6–99	57	10–100	
Early initiation of BF	21	49	32–72	53	20–81	
EBF (<6 mo)	48	26	1–68	40	3–74	
Measles[a]	73	71	17–98	79	27–99	
DTP3[a]	73	71	25–99	83	19–99	
ORS	46	29	10–66	33	10–77	
Care seeking for pneumonia	45	44	16–93	51	23–83	
ITN[b]	26	2	0–23	37	3–64	

[a]The immunization data, measles and DTP3, are based on the interagency estimates from 2002 to 200 average reference years for calculating trends for the nonvaccine indicators included in Table 43-3.

[b]For the malaria indicators, analyses are restricted to countries with 75% or more of the population at ris *falciparum* transmission, and with trend data available.

Source: Adapted from Bhutta et al[68] and Countdown to 2015.[69]

School Feeding Programs

Many countries have introduced school feeding programs designed to improve attendance, achievement, growth, and other health outcomes. A Cochrane review[72] on the effectiveness of school feeding programs reported an average of 0.39 kg (95% CI: 0.11–0.67) gain in weight over an average of 19 months and 0.71 kg (95% CI: 0.48–0.95) over 11.3 months. For height, results were mixed and height gain was greater for younger children. Attendance also increased by 4–6 days a year. In short-term studies, small improvements in some cognitive tasks were also found. School meals may have some small benefits for disadvantaged children, while further well-designed studies on the effectiveness of school meals should be undertaken and results reported according to socioeconomic status.

Child Health Days

CHDs have been introduced in many settings in response to the declining coverage of many essential child survival interventions as a result of weakening health systems. However, formal large-scale evaluations considering both positive and negative effects on health-related outcomes are required. A review of CHDs from 1999 to 2009[73] presents a broad picture of the scale-up of CHDs for the delivery of key child health and nutrition interventions. It confirms that CHDs have become a popular mode of delivery for a wide range of services. The authors suggest that CHDs are generally more effective than stand-alone campaigns, but if the number of interventions exceeds four, they may have a negative impact on coverage. Still CHDs may not be reaching those who need it most, so countries need to prioritize a specific number of interventions to be delivered through CHDs and also determine how to integrate this routine health care to impart effective and equitable delivery of services.

Community Platforms

Employing local semiskilled labor, and giving them the necessary training and resources to identify and treat certain common conditions, will increase the outreach of preventive and therapeutic strategies, as well as increase equity.[74–76] The concept of community case management, where community health workers have been employed to impart cost-effective treatment for conditions such as diarrhea and pneumonia, have also been

initiated. This is a strategy that has the potential to accelerate the pace toward achieving the goals set for MDG 4.[77]

A review on the effectiveness of community-based intervention packages in reducing maternal and neonatal morbidity and mortality and improving neonatal outcomes reported significant reductions in maternal morbidity (RR: 0.75, 95% CI: 0.61–0.92), neonatal mortality (RR: 0.76, 95% CI: 0.68–0.84), stillbirths (RR: 0.84, 95% CI: 0.74–0.97), and perinatal mortality (RR: 0.80, 95% CI: 0.71–0.91).[33] These interventions also increased referrals to health facility for pregnancy-related complication by 40% (RR: 1.40, 95% CI: 1.19–1.65), and improved the rates of early BF by 94% (RR: 1.94, 95% CI: 1.56–2.42).

CONTEXTUAL FACTORS: FOOD INSECURITY AND INEQUITIES

Despite the above-mentioned existing proven interventions, progress in improving child health and reducing undernutrition has been painfully slow. Though many countries are on track in reducing income poverty (MDG 1a), less than a quarter of developing countries are on track for achieving the goal of halving undernutrition (MDG 1c). The global burden of undernutrition remains high with little evidence of change in many countries despite economic growth. Millions of people are faced with starvation and malnutrition. The State of Food Insecurity in the World 2012 estimates that about 870 million people have been undernourished (in terms of dietary energy supply) in the period 2010–2012.[78] The vast majority of these, 852 million, live in developing countries, where the prevalence of undernourishment is around 14.9% of the population.[78] Food security is a complex issue resulting from a mix of climate change, rural poverty, inadequate stress on agricultural growth, urban development, population growth, oil price shifts, general inflationary trends, political influences, power balances, internal and external displacement of people, poor governance, and inequitable distribution of wealth and other resources. Paradoxically, there’s enough food in the world to feed everyone. Despite global food sufficiency, its inequitable distribution within countries and within households affects individuals at every level with too much for some and too little for the others. Women and children are among the highly affected population groups due to the discriminatory unequal and inequitable power relationships and practices.

WAY FORWARD

With these data in mind, it is important to note that currently global child mortality stands at 6.9 million deaths per year and to achieve the aims set forth for the MDG-2015, there is still much progress to achieve. However, to realize those aims, current preventive practices need to be formulated into appropriate unified guidelines. Given the wide disparities in the coverage of existing interventions, the challenge is to implement appropriately packaged interventions targeting the ones in need. This would require addressing the various contextual factors simultaneously including poverty, agricultural growth, education, and political commitment. Emerging data from community intervention trials now provide evidence that such efforts are both feasible and can lead to alleviation of childhood undernutrition. The latest Countdown Report[69] suggests that countries should implement national health plans that emphasize service integration, strengthen health information systems, increase domestic funding allocations on health, improve the health of the workforce, and identify gaps and inequities to increase coverage of essential health interventions across the full continuum of care, especially for the poor.

REFERENCES

1. Organisation for Economic Co-operation and Development. *The OECD and the Millennium Development Goals*. Paris, France: OECD; 2011.
2. Hulme D. *The Making of the Millennium Development Goals: Human Development Meets Results-Based Management in an Imperfect World*. Manchester, UK: Brooks World Poverty Institute. Working Paper No. 16. 2007.
3. Gillespie SR, Haddad L. *The Relationship Between Nutrition and the Millennium Development Goals: A Strategic Review of the Scope for DFID's Influencing Role*. London, UK: Department for International Development; 2003.
4. UNICEF. *Levels & Trends in Child Mortality, Estimates Developed by the UN Inter-agency Group for Child Mortality Estimation*. New York, NY: UNICEF; 2012.
5. *Child info Monitoring the Situation of Children and Women* http://www.childinfo.org/mortality_underfive.php. Updated 2011. Accessed February 25, 2013.
6. Walker CLF, Aryee MJ, Boschi-Pinto C, Black RE. Estimating diarrhea mortality among young children in low and middle income countries. *PloS one*. 2012;7(1):e29151.
7. Dewey KG, Adu-Afarwuah S. Systematic review of the efficacy and effectiveness of complementary feeding interventions in developing countries. *Matern Child Nutr*. 2008;4(suppl 1):24–85.
8. Bhutta ZA. Micronutrient needs of malnourished children. *Curr Opin Clin Nutr Metab Care*. 2008;11(3):309–314.
9. World Health Organization. *Severe Malnutrition: Report of a Consultation to*

Review Current Literature. Geneva, Switzerland: World Health Organization; 2005.
10. Black RE, Allen LH, Bhutta ZA, et al. Maternal and child undernutrition: global and regional exposures and health consequences. *Lancet*. 2008;371(9608):243–260.
11. Holly RG. Anemia in pregnancy. *Obstet Gynecol*. 1955;5(4):562–568.
12. Murphy JF, O'Riordan J, Newcombe RG, et al. Relation of haemoglobin levels in first and second trimesters to outcome of pregnancy. *Lancet*. 1986;1(8488):992–995.
13. Pena-Rosas JP, Viteri FE. Effects and safety of preventive oral iron or iron+folic acid supplementation for women during pregnancy. *Cochrane Database Syst Rev*. 2009;(4):CD004736.
14. Villar J, Merialdi M, Gulmezoglu AM, et al. Nutritional interventions during pregnancy for the prevention or treatment of maternal morbidity and preterm delivery: an overview of randomized controlled trials. *J Nutr*. 2003;133(5)(suppl 2):1606S–1625S.
15. Zeng L, Dibley MJ, Cheng Y, et al. Impact of micronutrient supplementation during pregnancy on birth weight, duration of gestation, and perinatal mortality in rural western China: double blind cluster randomised controlled trial. *BMJ*. 2008;337:a2001.
16. Imdad A, Bhutta ZA. Routine Iron/Folate Supplementation during Pregnancy: effect on maternal anaemia and birth outcomes. *Paediatr Perinat Epidemiol*. 2012;26(suppl 1):168–177.
17. Villar J, Belizan JM, Fischer PJ. Epidemiologic observations on the relationship between calcium intake and eclampsia. *Int J Gynaecol Obstet*. 1983;21(4):271–278.
18. Ortega RM, Martinez RM, Lopez-Sobaler AM, et al. Influence of calcium intake on gestational hypertension. *Ann Nutr Metab*. 1999;43(1):37–46.
19. Srinivas SK, Edlow AG, Neff PM, et al. Rethinking IUGR in preeclampsia: dependent or independent of maternal hypertension?. *J Perinatol*. 2009;29(10):680–684.
20. Imdad A, Bhutta ZA. Effects of calcium supplementation during pregnancy on maternal, fetal and birth outcomes. *Paediatr Perinat Epidemiol*. 2012;26(suppl 1):138–152.
21. Morreale de Escobar G, Obregon MJ, Escobar del Rey F. Role of thyroid hormone during early brain development. *Eur J Endocrinol*. 2004;151(suppl 3):U25–U37.
22. Zimmermann MB. The effects of iodine deficiency in pregnancy and infancy. *Paediatr Perinat Epidemiol*. 2012;26(suppl 1):108–117.
23. Haider BA, Bhutta ZA. Multiple-micronutrient supplementation for women during pregnancy. *Cochrane Database Syst Rev*. 2012; 11:CD004905.
24. De Onis M, Villar J, Gülmezoglu M. Nutritional interventions to prevent intrauterine growth retardation: evidence from randomized controlled trials. *Eur J Clin Nutr*. 1998;52:S83.
25. Kulier R, de Onis M, Gülmezoglu AM, Villar J. Nutritional interventions for the prevention of maternal morbidity. *Int J Gynaecol Obst*. 1998;63(3):231–246.
26. Imdad A, Bhutta ZA. Effect of balanced protein energy supplementation during pregnancy on birth outcomes. *BMC Public Health*. 2011;11(suppl 3):S17.
27. Dewey KG, Heinig MJ, Nommsen LA, et al. Growth of breast-fed and formula-fed infants from 0 to 18 months: the DARLING study. *Pediatrics*. 1992;89(6):1035–1041.
28. Breastfeeding and the use of human milk. American Academy of Pediatrics. Work

Group on Breastfeeding. *Pediatrics*. 1997;100(6): 1035–1039.
29. The optimal duration of exclusive breastfeeding: results of a WHO systematic review. *Indian Pediatr*. 2001;38(5):565–567.
30. Institute of Medicine. *Nutrition During Lactation*. Washington, DC: National Academy Press; 1991.
31. US Department of Health and Human Services. *Healthy People 2010*. Washington, DC: US Government Printing Office; 2000.
32. American Academy of Pediatrics. Breastfeeding and the use of human milk. American Academy of Pediatrics, Work Group on breastfeeding. *Breastfeed Rev*. 1998;6(1):31–36.
33. Birch E, Birch D, Hoffman D, et al. Breast-feeding and optimal visual development. *J Pediatr Ophthalmol Strabismus*. 1993;30(1):33.
34. Dewey KG, Heinig MJ, Nommsen-Rivers LA. Differences in morbidity between breast-fed and formula-fed infants. *J Pediatr*. 1995;126(5):696–702.
35. Florey CV, Leech AM, Blackhall A. Infant feeding and mental and motor development at 18 months of age in first born singletons. *Int J Epidemiol*. 1995;24(suppl 1):S21–S26.
36. Hanson LA, Wiedermann U, Ashraf R, et al. Effects of breastfeeding on the baby and on its immune system. *Food Nutr Bull*. 1996;17(4):384.
37. Lanting CI, Huisman M, Boersma ER, et al. Neurological differences between 9-year-old children fed breast-milk or formula-milk as babies. *Lancet*. 1994;344(8933):1319–1322.
38. Jalil F, Karlberg J, Hanson LÃ, Lindblad BS. Growth Disturbance in an Urban Area of Lahore, Pakistan Related to Feeding Patterns, Infections and Age, Sex, Socio-Economic Factors and Seasons. *Acta Paediatr*. 1989;78:44–54.
39. Infant and Young Child Feeding (2000–2007). http://www.childinfo.org/breastfeeding_countrydata.php. Accessed February 27, 2013.
40. Froozani MD, Permehzadeh K, Dorosty Motlagh AR, Golestan B. Effect of breastfeeding education on the feeding pattern and health of infants in their first 4 months in the Islamic Republic of Iran. *Bull World Health Organ*. 1999;77:381–385.
41. Jones G, Steketee RW, Black RE, et al. How many child deaths can we prevent this year? *Lancet*. 2003;362(9377):65–71.
42. *Infant and Young Child Nutrition: Global Strategy on Infant and Young Child Feeding*. http://apps.who.int/gb/archive/pdf_files/WHA55/ea5515.pdf. Accessed February 27, 2013.
43. Imdad A, Yakoob MY, Bhutta ZA. Effect of breastfeeding promotion interventions on breastfeeding rates, with special focus on developing countries. *BMC Public Health*. 2011;11(suppl 3):S24.
44. World Health Organization. *Report of Informal Meeting to Review and Develop Indicators for Complementary Feeding; 2002 December 3–5Washington, DC*. Washington, DC: World Health Organization; 2002.
45. Martorell R, Khan LK, Schroeder DG. Reversibility of stunting: epidemiological findings in children from developing countries. *Eur J Clin Nutr*. 1994;48:S45.
46. Imdad A, Yakoob MY, Bhutta ZA. Impact of maternal education about complementary feeding and provision of complementary foods on child growth in developing countries. *BMC Public Health*. 2011;11 (suppl 3):S25.
47. World Health Organization. *Worldwide Prevalence of Anaemia 1993–2005. World Health Organization Global Database on Anaemia* http://whqlibdoc.who.int/publications/2008/9789241596657_eng.pdf. Updated

2008. Accessed March 26, 2013.
48. De-Regil LM, Jefferds ME, Sylvetsky AC, Dowswell T. Intermittent iron supplementation for improving nutrition and development in children under 12 years of age. *Cochrane Database Syst Rev*. 2011;(12):CD009085.
49. Haider BA, Bhutta ZA. Neonatal vitamin A supplementation for the prevention of mortality and morbidity in term neonates in developing countries. *Cochrane Database Syst Rev*. 2011;(10):CD006980.
50. Imdad A, Herzer K, Mayo-Wilson E, et al. Vitamin A supplementation for preventing morbidity and mortality in children from 6 months to 5 years of age. *Cochrane Database Syst Rev*. 2010;(12):CD008524.
51. Larson CP, Roy S, Khan AI, et al. Zinc treatment to under-five children: applications to improve child survival and reduce burden of disease. *J Health Popul Nutr*. 2008;26(3):356.
52. Patel AB, Mamtani M, Badhoniya N, Kulkarni H. What zinc supplementation does and does not achieve in diarrhea prevention: a systematic review and meta-analysis. *BMC Infect Dis*. 2011;11(1):122.
53. Salvatore S, Hauser B, Devreker T, et al. Probiotics and zinc in acute infectious gastroenteritis in children: are they effective? *Nutrition*. 2007;23(6):498–506.
54. Rudan I, Boschi-Pinto C, Biloglav Z, et al. Epidemiology and etiology of childhood pneumonia. *Bull World Health Organ*. 2008;86(5):408–416B.
55. Williams BG, Gouws E, Boschi-Pinto C, et al. Estimates of world-wide distribution of child deaths from acute respiratory infections. *Lancet Infect Dis*. 2002;2(1):25–32.
56. Aggarwal R, Sentz J, Miller MA. Role of zinc administration in prevention of childhood diarrhea and respiratory illnesses: a meta-analysis. *Pediatrics*. 2007;119(6):1120–1130.
57. Bhandari N, Bahl R, Taneja S, et al. Effect of routine zinc supplementation on pneumonia in children aged 6 months to 3 years: randomised controlled trial in an urban slum. *BMJ*. 2002;324(7350):1358.
58. Bhutta Z, Black R, Brown K, et al. Prevention of diarrhea and pneumonia by zinc supplementation in children in developing countries: pooled analysis of randomized controlled trials. *J Pediatr*. 1999;135(6):689–697.
59. Imdad A, Bhutta Z. Effect of preventive zinc supplementation on linear growth in children under 5 years of age in developing countries: a meta-analysis of studies for input to the lives saved tool. *BMC Public Health*. 2011;11(suppl 3):S22.
60. Yakoob MY, Theodoratou E, Jabeen A, et al. Preventive zinc supplementation in developing countries: impact on mortality and morbidity due to diarrhea, pneumonia and malaria. *BMC Public Health*. 2011;11(suppl 3):S23.
61. Walker CLF, Black RE. Zinc for the treatment of diarrhoea: effect on diarrhoea morbidity, mortality and incidence of future episodes. *Int J Epidemiol*. 2010;39(suppl 1):i63–i69.
62. Haider BA, Bhutta ZA. The effect of therapeutic zinc supplementation among young children with selected infections: a review of the evidence. *Food Nutr Bull*. 2009;30(1 suppl):S41–S59.
63. Christian P, Tielsch JM. Evidence for multiple micronutrient effects based on randomized controlled trials and meta-analyses in developing countries. *J Nutr*. 2011;142(1):173S–177S.
64. Taylor-Robinson D, Maayan N, Soares-Weiser K, et al. Deworming drugs for treating soil-transmitted intestinal worms in children: effects on growth and school performance. *Cochrane Database Syst Rev*. 2012;11:CD000371.
65. Garner P, Golmezoglu AM. Drugs for preventing malaria in pregnant women.

Cochrane Database Syst Rev. 2006;4:CD000169.
66. Gamble C, Ekwaru JP, ter Kuile FO. Insecticide-treated nets for preventing malaria in pregnancy. *Cochrane Database Syst Rev*. 2006;2(2):CD003755.
67. Torun B, Chew F. Recent developments in the nutritional management of diarrhoea. 3. Practical approaches towards dietary management of acute diarrhoea in developing communities. *Trans R Soc Trop Med Hyg*. 1991;85(1):12.
68. Bhutta ZA, Chopra M, Axelson H, et al. Countdown to 2015 decade report (2000–10): taking stock of maternal, newborn, and child survival. *Lancet (London, England)*. 2010;375(9730):2032.
69. Countdown to 2015. *Building a Future for Women and Children: The 2012 Report* http://www.countdown2015mnch.org/reports-and-articles /2012-report. Updated 2012. Accessed February 22, 2013.
70. Ahmed HM, Mitchell M, Hedt B. National implementation of Integrated Management of Childhood Illness (IMCI): Policy constraints and strategies. *Health Policy*. 2010;96(2):128–133.
71. Arifeen SE, Hoque DM, Akter T, et al. Effect of the Integrated Management of Childhood Illness strategy on childhood mortality and nutrition in a rural area in Bangladesh: a cluster randomised trial. *Lancet*. 2009;374(9687):393–403.
72. Kristjansson B, Petticrew M, MacDonald B, et al. *School feeding for improving the physical and psychosocial health of disadvantaged students. Cochrane Database of Systematic Reviews. 2007; Issue 1. Art. No.: CD004676. DOI: 10.1002/14651858.CD004676.pub2*.
73. UNICEF. *Child Health Days 1999–2009: Key Achievements and the Way Forward, A report prepared for the UNICEF Joint Working Group on Child Health Days.* New York, NY: UNICEF Headquarters; 2011.
74. Reidpath DD, Morel CM, Mecaskey JW, Allotey P. The Millennium Development Goals fail poor children: the case for equity-adjusted measures. *PLoS Med*. 2009;6(4):e1000062.
75. Hotez PJ, Molyneux DH, Fenwick A, et al. Control of neglected tropical diseases. *N Engl J Med*. 2007;357(10):1018–1027.
76. Haines A, Sanders D, Lehmann U, et al. Achieving child survival goals: potential contribution of community health workers. *Lancet*. 2007;369(9579):2121–2131.
77. *Millennium Development Goals: A Compact Among Nations to End Human Poverty*. New York, NY: United Nations Development Programme; 2003.
78. FAO. *The State of Food Insecurity in the World 2012*. Rome, Italy: Food and Agriculture Organization of the United Nations. http://www.fao.org/docrep/016/i3027e/i3027e.pdf. Updated 2012. Accessed March 26, 2013.
79. Stevens GA, Finucane MM, Paciorek CJ, et al. Trends in mild, moderate, and severe stunting and underweight, and progress towards MDG 1 in 141 developing countries: a systematic analysis of population representative data. *Lancet*. 2012;380:824–834.

Pediatric HIV Infection

44

Colleen Hadigan, MD, MPH and Anna Coutsoudis, PhD

There are a number of nutritional challenges that clinicians face in the care of children infected with human immunodeficiency virus (HIV). Important interactions between the immune system and nutritional status, which are discussed in this and other chapters, have led to the recognition that treatment of nutritional problems can play a critical role in the health and well-being of HIV-infected children. Careful attention to the nutritional needs of children with HIV can help optimize growth, and more importantly may improve clinical outcomes including hospitalization rates and survival. This chapter will provide current background information on pediatric HIV infection and the associations between nutrition and HIV. Focus is also given to nutritional management in resource-limited settings where the HIV epidemic continues to affect large numbers of children. Furthermore, the growth patterns of HIV-infected children, metabolic complications of HIV and its therapies, and current nutritional diagnostic and therapeutic interventions are presented. It is hoped that this chapter provides practical advice to clinicians caring for HIV-infected children as well as stimulates much needed scientific research in this area.

HIV AND THE PEDIATRIC HIV/AIDS EPIDEMIC

HIV is an RNA virus that belongs to the lentivirus family and was first isolated in 1983. HIV is a virus with a particular tropism for the CD4 surface antigen of cells, and the binding of HIV to the CD4 receptor initiates the viral life cycle, which includes the replication of the virus within the host cell. Human T lymphocytes and monocyte or macrophages are the primary cells that are infected with HIV. The net biological effect of HIV infection is impairment of the immune system through the

progressive decline in CD4-positive T lymphocytes, leaving those infected with HIV susceptible to opportunistic and recurrent bacterial infections. Treatment of HIV with antiretroviral (ARV) medications suppresses HIV viral replication and permits reconstitution of the immune system with a compensatory rise in the CD4-positive T cells. While there are significant clinical and immune benefits of therapy, it is important to note, however, that even among treated patients with undetectable HIV viral RNA in the blood, virtually all patients develop detectable virus after withdrawal of therapy.

There has been dramatic progress made over the past two decades regarding the prevention, detection, diagnosis, and treatment of HIV infection, but HIV continues to be a global epidemic. The World Health Organization and the UNAIDS report on the global AIDS Epidemic estimated that 34 million people worldwide were living with HIV/AIDS in 2011.[1] Efforts to expand access to ARV therapy (ART) and to provide prenatal counseling, testing, and therapy are ongoing with impressive improvements seen in regions of the world with high HIV prevalence. The UNAIDS 2011 report documented encouraging global declines in the number of cases of mother-to-child transmission and of deaths in persons under age 15 years attributable to HIV. In high-income countries such as the United States, new infections among children dropped by 93% between 1992 and 2005, and now similar progress is being detected in lower income regions. Between 2002 and 2010, new infections among children declined approximately 30% with the majority (86%) of averted infections occurring in the region of sub-Saharan Africa. Nonetheless, as of 2011, over 2 million children were living with HIV infection. The greatest burden of pediatric HIV infection remains concentrated in regions of the world where endemic problems of protracted diarrhea and severe malnutrition often co-exist, emphasizing the importance of nutrition in the approach and management of HIV infection in children.

NUTRITION AND IMMUNITY

Nutritional status and immunity have long been linked, and malnutrition is the most common cause of immunodeficiency worldwide. Long before HIV was described, opportunistic infections such as *Pneumocystis carinii* pneumonia were described in otherwise healthy but malnourished individuals in developing nations.[2] This association led investigators to conclude that nutrition alone can impact on the immunologic response of an individual. In malnourished children, for example, there is a profound

involution of lymphoid tissues. In young infants and children, protein-energy malnutrition increases the risk of death several fold by increasing susceptibility to infection.[3] Historically, mortality rates in many countries increase from 0.5% in children whose weight-for-height percent of standard is greater than 80%, to a mortality rate of 18% in children whose weight-for-height percent of standard is less than 60%.[4]

Biochemically, protein-energy malnutrition leads to changes in several aspects of the immune system that are similar to the immunologic changes in HIV infection. Cell-mediated immunity, antimicrobial function of phagocytes, complement systems, secretory antibodies, and antibody affinity are consistently impaired in patients with significant malnutrition. Additionally, deficiencies of micronutrients, especially zinc and vitamin A, as well as many others, may also have deleterious effects on the immune system. Other aspects of immunity that are altered by protein-energy malnutrition include impaired chemotaxis of neutrophils, decreased lysozyme levels in serum and secretions, and decreased interferon production in antibody response to T-cell-dependent antigens. A child with protein-energy malnutrition may also have impaired mucosal immunity with lowered concentrations of secretory IgA in saliva, nasopharynx, and tears compared with well-nourished children.

Because many of the immunologic changes in malnutrition are similar to those found in children infected with HIV, malnutrition has been identified as a cofactor of immune dysfunction by influencing both susceptibility to HIV infection and progression of the disease. Therefore, improving the nutritional status of an HIV-infected child may improve the function of an already compromised immune system and subsequently prolong health and survival.

WASTING IN HIV-INFECTED CHILDREN: PATHOGENESIS

Globally, severe weight loss, growth failure, and wasting remain a central component of HIV infection in children, even in the era of more widely accessible ART. The pathogenesis of malnutrition or the wasting syndrome in pediatric HIV disease is most likely multifactorial. Identified risk factors include fever, diarrhea, acute and opportunistic infections, anorexia, as well as social factors such as ill parents or caregivers, food insecurity and lack of clean water. For example, in a recent cross-sectional study of undernutrition in Dar es Salaam Tanzania, Sunguya, and

colleagues[5] showed that 13.6% of HIV-infected children treated with ART met criteria for wasting (weight-for-height Z-score < −2 SD). Important factors associated with wasting in this study were the presence of diarrhea and low feeding frequency. The four key mechanisms for weight loss and wasting in HIV infection, which are considered in this section, are inadequate oral intake, impaired gastrointestinal absorption, abnormal or increased energy utilization, and psychosocial influences. In many cases, however, several factors may be operating simultaneously.

Energy Intake

A variety of potential factors may lead to abnormal energy intake, as outlined in Table 44-1. For example, inflammation and ulceration of the upper gastrointestinal tract can lead to anorexia owing to odynophagia, dysphagia, or abdominal pain associated with eating. These lesions may be due to infectious agents such as *Candida albicans*, cytomegalovirus, or herpes simplex virus, as well as acid-related injury, all of which may cause inflammation and pain with swallowing or after eating, and thus diminished energy intake.

Pancreatic and biliary tract diseases can also cause vomiting and abdominal pain in HIV-infected children leading to poor oral intake. Pancreatic dysfunction has been linked to medications used in HIV (e.g., nucleotide reverse transcriptase inhibitors, protease inhibitors, sulfonamides) and opportunistic infections (e.g., cytomegalovirus and mycobacterial disease). Biliary tract disease, such as sclerosing cholangitis and papillary stenosis, while less common, has been associated with *Cryptosporidium*, cytomegalovirus and *Microsporidia* infection. Elevations in circulating cytokines such as interleukin-6 and tumor necrosis factor (TNF)-α in association with HIV infection and other opportunistic infections are believed to contribute directly to anorexia. Lettow and colleagues found, for example, that interleukin-6 levels and HIV viral burden were closely associated with severe wasting and loss of appetite among a cohort of HIV-infected adult patients with pulmonary tuberculosis in Malawi.[6] Side effects of ART may also contribute to decreased energy intake. There are currently more than 25 agents approved by the US Food and Drug Administration for the treatment of HIV and many of these as well as other medications frequently used in HIV-infected children may result in gastric irritation, vomiting, and nausea (See Table 44-2).

TABLE 44-1 Causes of Malnutrition in HIV-infected Children

Decreased Nutrient Intake	Mechanism
Peptic disease	Pain
Opportunistic infections of upper gastrointestinal tract (Candida, CMV, HSV)	Pain, dysphagia
Pancreatic/hepatobiliary disease	Pain
Encephalopathy	Uncoordinated swallow
Aphthous ulcers	Pain
Primary anorexia	Nausea, CNS, cytokines
Gastrointestinal Malabsorption	
Mucosal disease	
Infectious	Bacterial, parasitic, viral
Inflammatory	HIV enteropathy
Disaccharidase deficiency	Infectious, inflammatory
Protein-losing enteropathy	Infectious, inflammatory
Fat malabsorption	Infectious, inflammatory
Hepatobiliary	
Sclerosing cholangitis	Infectious
Chronic pancreatitis	Infectious, Drug-induced
Cirrhosis	HBV, HCV
Increased nutritional requirements or tissue catabolism	
Protein wasting	Fever, infections, sepsis
Hypermetabolism	Neoplasms (Kaposi's
Futile metabolic cycling	sarcoma, lymphoma)
	Medications
	Release of catabolic factors
	(cytokines, tumor necrosis factor)
Psychosocial Factors	
Poverty	
Illness in biological family members	
Limited access to health care	
Substance abuse	

CMV, cytomegalovirus; CNS, central nervous system; HBV, hepatitis B virus; HCV, hepatitis C virus; HSV, herpes simplex virus.

There are increasing data suggesting decreased energy intake among HIV-infected children who have more symptomatic infection and/or growth failure. For example, Arpadi and colleagues[7] found, in the absence of active secondary infections or other illnesses, HIV-infected children with growth failure had significantly lower mean energy intake (77% of the RDA) compared with HIV-infected children without growth failure (103% of the RDA for mean energy intake). In this study, when energy balance was assessed as the difference between energy intake and total energy expenditure, HIV-infected children with growth failure had a 674 kJ/d deficit whereas those without growth failure had a surplus intake of 1448 kJ/d. Therefore, while energy intake below the RDA is not common in HIV-infected children during stable periods of disease, inadequate energy intake occurs during symptomatic periods of disease and may contribute to growth failure.

Gastrointestinal Absorption

Adequate nutrient absorption is essential to maintain growth and nutrition in infants and children. HIV-infected patients are at risk for developing malabsorptive disorders, which can potentially impact on their growth and nutritional status. Reasons for gastrointestinal malabsorption in HIV-infected children include infections of the gastrointestinal tract and/or direct and indirect effects of HIV on the intestinal epithelial cell. Infections that are known to affect the small bowel and that may promote malabsorption of nutrients include *Cryptosporidium*, *Isospora belli*, *Microsporidia*, *Mycobacterium avium-intracellulare* complex (MAC), *Giardia lamblia*, and enteric viruses. These pathogens can injure the intestinal epithelial cell where critical digestive enzymes are present to digest essential nutrients. Enteric infections may also decrease bowel transit time, thus shortening nutrient contact with the epithelial cell and causing malabsorption. Malnutrition alone can cause villous blunting and secondary malabsorption of nutrients, which would further impact on nutritional status. Malabsorption has also been linked to an “HIV enteropathy” where the villi are blunted with a hypercellular lamina propria and absence of a detectable enteric pathogen.[8] There is evidence of enterocyte cell death or apoptosis. In addition, in vitro studies of enterocytes show that HIV exposure and/or exposure to HIV gp120 may evoke disruption of microtubules, and tight junctions as well as impaired ion flux, suggesting a pathophysiologic mechanism for HIV enteropathy and malabsorption.[9,10] The fact that many HIV-infected children live in

areas of the world with a high prevalence of tropical or environmental enteropathy make studies of this entity difficult.

In the pre-HAART era, a few studies provided evidence of malabsorption in HIV-infected children.[11,12] For example, in one Italian study,[11] intestinal function of 47 symptomatically infected children was compared with 50 uninfected children with diarrhea and 48 healthy children. Intestinal absorption was measured using the steatocrit method to determine fat absorption, fecal concentration of α-1-antitrypsin as a measure of protein absorption, and d-xylose absorption. Fecal fat loss was detected in 30%, carbohydrate malabsorption in 32%, and protein loss in 17% of HIV-infected children. Investigators found intestinal dysfunction was not associated with diarrhea, poor growth, enteric infections, or degree of immune dysfunction, but was significantly different than healthy control children. In a similar series, Sentongo et al.[12] identified fat malabsorption in 39% of 44 HIV-infected children, but no patient had exocrine pancreatic insufficiency and there was no relationship between steatorrhea, growth, HIV viral load or CD4 count. Thus, although malabsorption is fairly common, it cannot completely account for the gastrointestinal symptoms or malnutrition in HIV-infected children.

TABLE 44-2 Common Gastrointestinal Side Effects Associated with Medications Used in HIV Care

Side Effects	Medications
Nausea	NRTI: abacavir, didanosine, emtricitabine, lamivudine, stavudine, zidovudine
	NNRTI: efavirenz, nevirapine
	PI: atazanavir, darunavir, indinavir, nelfinavir, ritonavir, saquinavir, tipranavir
	Integrase Inhibitor: raltegravir, elvitegravir
	Antiviral: acyclovir, ganciclovir
	Antibacterial: macrolides, sulfonamides, rifampin
	Antiparasitic: pentamidine, atovaquone
Abdominal pain	NRTI: abacavir, didanosine, emtricitabine, lamivudine, stavudine, tenofovir, zidovudine
	NNRTI: nevirapine
	PI: amprenavir, atazanavir, darunavir, indinavir, nelfinavir, ritonavir, saquinavir

	Integrase Inhibitor: raltegravir, elvitegravir
	Antibacterial: macrolides, sulfonamides, ciprofloxacin, rifampin
	Antiparasitic: pentamadine
Vomiting	NRTI: abacavir, didanosine, emtricitabine, lamivudine, stavudine, zidovudine
	NNRTI: efavirenz
	PI: amprenavir, atazanavir, indinavir, nelfinavir, ritonavir
	Integrase Inhibitor: raltegravir, elvitegravir
	Antiviral: ganciclovir
	Antibacterial: macrolides, sulfonamides, ciprofloxacin, rifampin
	Antiparasitic: pentamadine, atovaquone
Diarrhea	NRTI: emtricitabine, lamivudine
	PI: amprenavir, indinavir, lopinavir/ritonavir, nelfinavir, ritonavir, saquinavir, tipranavir
	Integrase Inhibitor: elvitegravir
	Antibacterial: macrolides, ciprofloxacin, rifampin
	Antiviral: acyclovir, ganciclovir
Pancreatitis	NRTI: abacavir, didanosine, lamivudine, stavudine, tenofovir
	PI: lopinavir/ritonavir, ritonavir, saquinavir, tipranavir
	Antibacterial: sulfonamides, ciprofloxacin
	Antiparasitic: pentamadine

NNRTI, non-nucleoside reverse transcriptase inhibitor; NRTI, nucleoside reverse transcriptase inhibitor; PI, protease inhibitor.

Metabolism

Increased energy metabolism of HIV-infected children can contribute to malnutrition by increasing the child's caloric demands to maintain weight and sustain growth. Total daily energy expenditure is the sum of energy needed to maintain a child at rest, the energy needed for activities of daily living, the thermic effect of food, energy needs for growth, and obligatory energy losses. Measurements of resting energy expenditure through indirect calorimetry and total daily energy expenditure through techniques such as doubly-labeled water studies enable investigators to determine metabolic rates of HIV-infected patients.

There are several studies in the pediatric HIV population which delineate resting or total daily energy expenditure, although more studies have been performed in the adult HIV population. The literature in children with HIV infection generally shows no differences in resting energy expenditure (REE)[7,13,14] or total energy expenditure (TEE)[7,15] between children with growth failure and those with normal rates of

growth. However, these studies have been performed in clinically stable children; metabolism in sicker children is likely to be much higher. One study determined whole body protein turnover in children with HIV and found increased protein turnover is associated with low weight and height.[16] Interestingly, Hardin et al.[17] showed the use of a protease inhibitor in the treatment of HIV resulted in decreased protein catabolism and was associated with improvement in lean tissue mass in HIV-infected children tested pre- and post- therapy.

Cytokines

Elevated cytokine expression may contribute to altered metabolic rates and weight regulation abnormalities in HIV and this hypothesis is supported by experimental and animal studies. The cachectin hypothesis was first described by Beutler and Cerami[18] who noticed that patients with chronic infections have significant weight loss that is associated with hypertriglyceridemia and reduced clearance of triglycerides by lipoproteins. They postulated that a decrease in triglyceride clearance could lead to a decrease in fat storage and wasting of lean body mass as a result of the inability to utilize fats as a reserve source for energy. In animal studies, supernatants from activated macrophages induced weight loss when given daily to rodents.[19] This factor in the supernatant was subsequently isolated and named cachetin; later discovered to be the cytokine, TNF. Other cytokines, such as interleukin-1 (IL-1) and the interferons, have similar effects.

Cytokines are known to alter metabolic processes in non-HIV populations. In patients with HIV, cytokines can cause ineffective use of energy substrates. Hellerstein[20] studied hepatic lipogenesis in HIV-infected adult patients and correlated it with peripheral cytokine levels. Both symptomatic HIV-infected patients with normal CD4 counts and HIV-infected patients with weight loss had elevated hepatic lipogenesis, compared with non-infected controls. Hepatic lipogenesis correlated with interferon-α levels in the fasted state in the patients with HIV infection and weight loss. Other cytokines were not correlated with increased hepatic lipogenesis. In addition to increasing the hepatic synthesis of fatty acids, TNF also mobilizes free fatty acids by stimulating peripheral lipolysis. The net result, “futile cycling”, the process by which fatty acids are shuttled from adipose tissue to liver and back to adipose tissue, utilizes energy inefficiently. The process results in increased metabolic rates, with greater caloric needs to maintain nutrition. The net effect of cytokines for patients

infected with HIV is nonproductive use of energy substrates. Approximately 28% of calories are lost if lipogenesis from carbohydrate precedes its oxidation. Roubenoff et al.[21] found in HIV-infected men that loss of lean body mass was driven by catabolic cytokines and not by inadequate dietary intake or hypogonadism. Other investigators have found similar associations, independent of viral load.[22] Limited studies in children showed increased IL-6 activity, as well as low IGF-1 and increased viral load to be associated with growth impairment.[15] The net effect of elevated cytokine levels may contribute to the relative hypermetabolism seen in HIV-infected patients. Since depression of immune function secondary to malnutrition is potentially reversible, nutritional rehabilitation in HIV-infected children may significantly lower serum cytokine levels by positively improving immunologic function, decreasing opportunistic infection and diminishing HIV replication. This, in turn, would decrease cytokine production. With lower TNF and IL-1 production, promotion of HIV replication would be decreased and the systemic effects of these cytokines such as the wasting and malabsorptive symptoms would be reduced.

PSYCHOSOCIAL FACTORS

Psychosocial factors are also important contributors to suboptimal nutrition of HIV-infected children. An unstable home environment and inadequate emotional and social support have been shown to be a factor for growth problems in both HIV-1-infected[23] and non-HIV-infected children by many mechanisms, including growth hormone deficiencies.[24,25] Children with HIV infection are at risk for living with parents who are ill, who have limited access to social services and support, and who may have ongoing problems with drug and substance abuse. Investigators have found maternal crack and cocaine use during pregnancy to be a predictor of growth and nutritional problems for the child.[26,27] This finding is not unique for HIV, as it has been reported in other non-HIV cohorts.[28] Children born to drug-using women are often small, suggesting that drugs have a prenatal effect, but the postnatal home environment is likely to influence growth as well. Current studies have also found that home care providers can both positively or negatively influence functional status of HIV-infected children.[23]

GROWTH AND BODY COMPOSITION OF HIV-INFECTED CHILDREN

Growth

Impaired growth and malnutrition are common among infants and children with HIV infection and are closely associated with morbidity and mortality in this population.[27,29–34] Even in the era of ART, failure to thrive, defined as weight for age z-score ≤ −2.0 S.D. was common in HIV-infected children in the U.S. and was found in 37% of perinatally infected children at one year of age.[26] In a large cohort of almost 18,000 children in 12 ART programs in sub-Saharan Africa, it was observed that although there was a sustained growth response and catch-up growth in children, after three years of ART treatment not all children achieved normal weights and heights.[35] Furthermore, since even with ART impaired growth has been shown to be associated with increased mortality, independent of age or HIV viral load,[36,37] nutritional status and growth assessment remains vital in the management of HIV-infected children.

Weight loss and gastrointestinal symptoms are one of the most common presenting symptoms of HIV disease in developing countries. In Tanzania, 200 children with severe malnutrition and non-malnourished controls who were matched for age, sex and area of residence, were screened for evidence of HIV infection.[38] Twenty-five percent of the malnourished group was HIV-positive compared with 1.5% of the control group. Since these are common observations routine screening for HIV infection in malnourished children is recommended as part of IMCI guidelines in developing nations.

In developing nations, other confounding factors may exist which can explain different outcomes relating to growth. In one study of 822 HIV-infected pregnant women from Tanzania, low birth weight was associated with maternal advanced stage HIV, however malaria, helmintic infections, and intestinal parasites were also strong predictors of low birth weight.[39] Maternal nutritional status of HIV-infected women receiving ART was associated with low birth weight and stunting in their infants.[40] These data demonstrate the importance of maternal health in fetal growth and birth weight. Interventions to improve maternal health include timely initiation of ARVs when CD4 counts drop below 500 or 350; micronutrient supplementation; good nutrition and psychosocial support. However, with the increased use of highly active or combination antiretroviral therapy (HAART) to prevent mother–to-child transmission, new data on the

increased risk of low birth weight and prematurity associated with HAART has created a concern.[41,42]

In recent years, the importance of breastfeeding for promoting growth in HIV-infected infants has become evident. A South African study[43] which examined growth of HIV-infected and uninfected infants according to infant feeding mode showed that in HIV-infected infants especially, breastfeeding was associated with consistently higher weight z scores than was seen in those infants fed formula milk. This was more pronounced in the first four months and particularly in the first six weeks, where the mean difference was 130 g higher for male infants and 110 g higher for the female infants. These data underscore the importance of promoting WHO recommendations for exclusive breastfeeding in the first six months and continued breastfeeding for at least two years in order to ameliorate the growth deficits associated with HIV infection. Furthermore, these data together with data showing that breastfeeding improves health and survival of HIV-infected infants[44–47] encourage the promotion of community-based human breastmilk banks to supply breastmilk for HIV-infected AIDS orphans.

Body Weight and Composition

Accurate assessment of weight, linear growth, and evaluation of body composition are key components in determining nutritional status in all children. Weight loss or a decrease in growth velocity are simple markers used to diagnose the wasting syndrome in children with HIV infection. However, using weight as the only marker of nutritional status can be misleading, since weight can be confounded by symptoms such as vomiting and diarrhea, which affect hydration. In addition, hydration status and edema can influence weight measurements and obscure the true nutritional condition. Mid-upper arm circumference (MUAC) is a useful and simple tool used globally to determine nutritional status in adults and children. When available, a more accurate measure of nutritional status is body-cell-mass, which refers to the nonadipose cellular mass and is equivalent roughly to the intracellular water volume and can be determined using bioelectrical impedance. The usefulness of bioelectrical impedance measurements is however limited, since it is affected by hydration status of the individual.

Anthropometry and body composition studies can provide insight into the potential etiology of a nutritional disorder. The body's first (and normal) response to starvation (inadequate food intake) is the preferential

loss of body fat, which can be reflected in diminished triceps skinfold thickness measurements or other measurements of body fat. When fat stores are depleted, wasting of lean body mass occurs. In contrast, cachexia or wasting refers to the preferential and inappropriate loss of lean body mass over fat mass, with or without concomitant weight loss. Cachexia has been described previously in patients with cancer, but more recently has been found in patients with HIV infection, including children. Cachexia may be mediated by chemical intermediates such as cytokines and is discussed in the section on pathogenesis.

Earlier studies utilizing various methods to assess body composition have been performed in HIV-infected children. Body composition measurements of 89 children born to HIV-infected women, (52 children were HIV infected) found lean body mass measured by arm muscle circumference was significantly less in the HIV-infected group by 2 years of age. There were no differences in fat mass between the two groups of children.[48] In a cross-sectional study of HIV-infected children treated with nucleoside reverse transcriptase inhibitors, Fontana and colleagues compared bioelectrical impedance measurements of fat free mass (FFM) in 86 HIV-infected children (mean age 6.9 years) with 113 healthy controls (mean age 7.7 years).[49] These investigators found that FFM was lower in HIV-infected children compared with controls, however, when FFM was evaluated as a percentage of total body weight, there was no difference between groups. Weight for age and FFM z-scores <−2.0 were associated with increased risk of death (RR 11.4 and 5.1, respectively), with weight for age serving as a stronger prognostic indicator than FFM. Arpadi and colleagues used dual energy x-ray absorptiometry (DXA) and also found reduced FFM but normal fat stores among HIV-infected children compared with uninfected controls, regardless of whether or not growth failure was also present.[50] These findings indicate that poor growth and decreased FFM are considerable problems for children living with HIV infection and that these parameters have a significant deleterious effect on survival. The introduction of HAART has revolutionized the management of HIV infection in children. Much recent attention has focused on the potential impact of HAART on growth and body composition as well as immune function and survival. Early data from Europe showed clearly that HIV-infected infants were born with lower birth weights compared with exposed-uninfected infants and that their weights continued to diverge until they were commenced on HAART whereafter their growth improves.[51–53]

Nachman and colleagues[54] reported that, in addition to a significant

relationship between HIV viral RNA levels and indices of poor height and weight, the introduction of HAART was associated with improved weight gain. Importantly, height growth achieved essentially normal levels after two years of therapy. Peacock-Villada et al. examining outcomes after HAART treatment of children in both developed and developing countries reported that mortality rates were much lower compared with that before HAART in both settings.[55] However the mean weight for age z-score (WAZ) was significantly lower in children in developing vs developed countries (−2.2 vs −0.4, $p < 0.001$); not surprisingly the mortality rate was higher in children from developing countries. In studies conducted in Zambia,[56] Kenya,[57] and Lesotho[58] of HIV-infected children treated with HAART, lower WAZ and weight for height Z-scores (WHZ) were predictors of mortality. Studies in developing countries also show that although treatment with HAART results in improvements in underweight and stunting the values often do not reach normal values and therefore attention needs to be given to other means of improving these parameters.

There is further evidence that abnormalities in growth parameters may be linked to virologic and immune response to ART. Verweel and colleagues found that both weight and height improved after initiation of HAART, but only in a subset of children who demonstrated virologic response defined as 1.5 or greater log reduction in HIV RNA.[59] Although data show convincing evidence that effective viral suppression and immune reconstitution with HAART can substantially ameliorate the growth deficits in HIV-infected children, unanticipated effects of HAART have been identified including metabolic alterations and abnormal patterns of body fat distribution or lipodystrophy. As will be covered in the next section, further longitudinal information is needed to determine the potential long-term benefits of ART and its effects on growth and body composition.

HIV-ASSOCIATED METABOLIC DISTURBANCES AND LIPODYSTROPHY

Shortly after the widespread introduction of highly active ART (HAART), a constellation of metabolic abnormalities and alterations in body fat distribution were recognized among many HIV-infected patients (Table 44-3). The loss of subcutaneous fat as well as increased adiposity of the abdomen and hypertrophy of the dorsocervical fat pad were reminiscent of congenital and acquired forms of lipodystrophy and this term has been

used in the HIV population. There is increasing evidence that ARV medications, in particular some protease inhibitors and nucleoside reverse transcriptase inhibitors, contribute directly to lipodystrophy and other identified metabolic complications.[60,61]

In addition to significant and often disturbing changes in fat distribution, patients may develop hyperlipidemia, insulin resistance and diabetes.[60,62] The long-term effects of insulin resistance, hyperlipidemia and fat redistribution may require many years to be fully realized, but there is growing evidence of increased cardiovascular disease in HIV-infected patients. The Data Collection on Adverse Events of anti-HIV Drugs (D.A.D.) study evaluated 17,852 HIV-infected adults and found that hypertriglyceridemia and total cholesterol increased with exposure to ARV medications. The study also established increased rates of myocardial infarction with increasing years of exposure to ART.[63]

TABLE 44-3 Clinical and Biochemical Abnormalities Associated With HIV and Its Therapy in Children

Clinical Features	Laboratory Features
Increased abdominal (visceral) fat	Dyslipidemia
Increased waist-to-hip ratio (not reliable in growing children)	Increased triglycerides Increased total cholesterol
Hypertrophy of the dorsocervical fat pad ("buffalo hump")	Increased LDL cholesterol
Fat atrophy	Decreased HDL cholesterol
Loss of fat in extremities	Insulin resistance
Loss of fat of buttocks	Normal to increased serum glucose
Loss or thinning of facial fat, prominence of nasolabial fold	Increased insulin Increased C peptide
No change to increased weight	Decreased glucose tolerance
Fatigue and weakness	Elevated lactate levels

Several studies have been conducted to evaluate whether pediatric patients with HIV infection receiving HAART are also experiencing changes in body fat distribution and metabolic abnormalities. For example, Arpadi and colleagues[64] assessed body fat distribution in 28 prepubertal HIV-infected children with DEXA scans and 29% had evidence of lipodystrophy (defined as fat atrophy and trunk fat accumulation). The

presence of lipodystrophy was positively associated with protease inhibitor use (OR 7.0) and stavudine use (OR 9.0) in this population. A multicenter two-year prospective evaluation of 130 HIV-infected children on ART was completed in Paris, France.[65] Nearly one quarter of the participants had evidence of lipodystrophy. Other metabolic disturbances were frequently identified in this cohort including low HDL cholesterol, increased total cholesterol, and triglycerides. Insulin resistance, defined as a fasting insulin > 95% for normoglycemic children matched for pubertal status, doubled in frequency over the two-year study period from 13.2% to 25.6%. Important predictors of hyperinsulinemia and dyslipidemia were development of puberty, exposure to protease inhibitors and pretreatment severity of HIV disease. More research is needed to determine the effects of HAART on fat redistribution and lipids and glucose homeostasis in children, particularly given the potential life-long exposure to HAART, which may be required for HIV viral suppression. In addition to the potential increased risk of diabetes and cardiovascular disease, children may also experience as yet unappreciated abnormalities in pubertal development, bone health, and kidney function in relation to metabolic complications of HIV and its therapy.

MICRONUTRIENTS

Several micronutrients have been linked to immunomodulation, both in vivo and in vitro. For example, deficiencies of pyridoxine, folic acid, vitamin A, vitamin C, and vitamin E result in impaired cell-mediated immunity and reduced antibody responses. Vitamin B6 deficiency results in decreased lymphocyte response to mitogens, yet a moderate increase in β-carotene intake may enhance immune responses. With zinc deficiency, there is lymphoid atrophy and decreased delayed cutaneous hypersensitivity responses. Likewise, zinc deficiency in animals can reduce the number of antibody-forming cells in the spleen and impair natural killer activity. Clinically, wound healing is impaired in zinc deficiency, whereas, excessive zinc can depress neutrophil function and lymphocyte responses. Iron is important for bacterial growth as well as neutrophil and lymphocytes function. Deficiencies of iron may reduce bacterial multiplication, but may also impair lymphocyte proliferation response to mitogens and antigens. In patients with iron deficiency, lymphocyte response to tetanus toxoid and herpes simplex antigen is low. Overall, in animal models, it is clear that selected vitamin and micronutrient deficiencies impact on immune function in many ways. See

Chapters 8, 9, and 10 for full discussions of trace elements, iron, and vitamins.

Because of the close link between immunologic function and micronutrients, it is logical to expect that low micronutrient levels can affect susceptibility to infection. For HIV-infected children, whose immunologic function is already compromised, further alterations of immunologic function, through micronutrient deficiencies or excesses, may synergistically decrease resistance to infections. Available studies of micronutrients and HIV infection, particularly among children, are outlined in the following sections.

General Studies

There is increasing data available on the micronutrient status of children with HIV infection. Eley and colleagues evaluated 60 HIV-infected children in Cape Town, South Africa and, using blood nutrient concentrations, vitamin deficiencies were frequently identified: vitamin A (80%), retinol-binding protein (85%), vitamin B6 (37%), vitamin E (37%), zinc (20%), and copper (25%).[66] Sixty-two percent of children had two or more deficiencies, and deficiencies were more common and more severe in children older than 24 months. Only 5% of patients were found to have low Vitamin B12 levels.

Anemia is prevalent in HIV-infected children and associated with a poor prognosis.[67] Among Indian HIV-infected children aged 2 to 12 years, the prevalence of anemia was 52.5%.[68] Iron deficiency alone was ascribed as the etiology in 38% of the children and inflammation as the etiology in another 38% of children. Compared with nonanemic children, anemic children were more likely to be underweight and stunted. Children who commenced ART had a significant increase in hemoglobin and if iron supplements were received in addition to ART, children experienced an even greater increase in hemoglobin.

In a large cohort of HIV-infected children and adolescents in the United States (n = 264), there were no differences in intake of vitamins A, C, iron, or zinc, compared with uninfected controls (n = 127).[69] Approximately, one-third of the children studied reported use of a daily vitamin/mineral supplement. The investigators noted intake below the estimated average requirement of the dietary reference intakes (DRI) in 40% of children for vitamin E, 19% for vitamin A, and 10% for zinc. However, there were no identified differences in the prevalence of micronutrient inadequacy when evaluated according to HIV status or CD4

T-cell strata. Therefore, there appears to be important regional variation in the prevalence of micronutrient deficiencies in children.

Vitamin A

Vitamin A is arguably one of the most frequently studied vitamins in relationship to immune function and HIV. In adults, Tang and colleagues[70] showed a relationship between total vitamin A intake and progression to AIDS, but the relationship was U-shaped, the lowest and highest quartiles of intake did less well than the two middle quartiles in terms of progression to AIDS. Data available from HIV-infected children in North America,[71] found vitamin A deficiency to be relatively uncommon and vitamin A concentrations did not appear to be related to mortality in this population.

Vitamin A deficiency and supplementation, however, may have a significant role in HIV disease progression in areas of the world where vitamin A deficiency is more prevalent.[72–77] In a trial of vitamin A supplementation in Tanzania, HIV-infected children experienced a 63% reduction in all-cause mortality with vitamin A supplementation compared with placebo.[72] In a longitudinal observational study of 194 HIV-infected children in Uganda, low plasma β-carotene levels, but not vitamin A levels, were predictive of increased mortality. This and other studies, however, show a relationship between low vitamin A levels and poor growth in HIV-infected children.[72,75] For example, in a placebo-controlled trial of vitamin A supplementation in Tanzania, including 50 HIV-infected children, vitamin A–treated children under 18 months of age experienced a 2.8-cm greater height increase compared with placebo.[77] One potential mechanism for this beneficial effect on growth may be improved gut permeability (i.e., reduced permeability with vitamin A), which was shown with perinatal vitamin A supplementation in HIV-infected infants.[73]

B Vitamins and Folate

Baum and colleagues[78] evaluated vitamin B6 status in HIV-infected adults in the United States. In this study, vitamin B6 deficiency was associated with decreased lymphocyte responsiveness to mitogens and suggested as a cofactor in reduced natural killer cell cytotoxicity. However, vitamin B6 was not related to CD4 T-cell number. In a study of 210 ART-naive HIV-infected adults in Uganda, 10% were identified as vitamin B12 deficient, and among those with a CD4 count ≤ 350 cells/μl, suboptimal B12 levels were associated with a higher mean rate of CD4 decline, compared with

those with normal B12 levels.[79] In an HIV outpatient clinic setting in Nigeria, plasma folate levels were significantly lower in HIV-infected compared with uninfected controls, and folate correlated positively with CD4 count and inversely with HIV viral burden.[80] Intestinal absorption of folate and other nutrients may be responsible for abnormal blood levels. Folic acid absorption was studied in 25 HIV-infected patients after an oral folic acid dose, with blood samples taken sequentially over three hours.[81] Absorption of folic acid was significantly impaired in patients, regardless of the stage of the disease. Thus, intestinal absorption of folate and other nutrients may be responsible for abnormal blood levels.

Supplementation with B vitamins, vitamin C, and vitamin E was evaluated in a large clinical trial of pregnant HIV-infected women in Tanzania. In addition to improved birth outcomes (lower risk of both preterm delivery and low birth weight), maternal supplementation was associated with lowered infant morbidity and higher CD4 counts[82] as well as a reduced rate of mortality and AIDS progression in mothers.[83]

Antioxidants

Oxygen-free radicals are generated during normal metabolism, yet their production may increase during infection, malnutrition, stress, or drug therapy. Endogenous antioxidant activity such as catalase production and exogenous antioxidants (e.g., vitamin E and selenium) may be important in HIV. Selenium is a potent antioxidant and is also an important cofactor for immune function in humans. In vivo studies have shown the importance of this element in protecting cells from HIV activation and the effects of TNF, through increasing glutathione peroxidase activity.[84] In one cross-sectional study, HIV-infected children had low glutathione levels which correlated with CD4 cell count.[85] Low plasma selenium levels were associated with increased risk of mortality of children born to HIV-infected mothers in Tanzania,[86] supporting a potential relationship with nutritional antioxidant and immunity.

Trace Metals

Zinc's role in metabolic function has been well studied. It is important in the stabilization and function of metabolic enzymes including those involved in protein synthesis, catabolism, energy metabolism and RNA and DNA synthesis. Regeneration of gut epithelium, stabilization of brush border enzymes and improved immune function are all proposed mechanisms for zinc's beneficial effects in acute diarrhea.[87] A study by

Graham and colleagues showed that serum copper levels were higher and zinc levels lower in HIV-infected adult patients who progressed to AIDS than in seropositive patients who did not progress.[88] Using logistic regression, they found that higher serum copper levels (odds ratio 2.23 [95% CI 1.02, 4.87]) and lower serum zinc levels (odds ratio 0.3 [95% CI 0.14, 0.66]) predicted progression to AIDS independent of baseline CD4 lymphocyte levels, age, and energy adjusted dietary intake of both of these nutrients. In an open treatment study of HIV-infected children with a CD4 count < 500 cells/mm^3, supplemental zinc was administered for one month.[89] Although 69% of the children had low zinc levels at baseline, supplementation did not have an effect on p24 antigen or CD4 count. The prevalence of zinc deficiency among HIV-infected children, which was estimated at 20% in one South African study,[66] combined with the observed reduction in episodes of watery diarrhea in HIV-infected children who received zinc supplementation compared with a placebo,[90] points to a potential benefit for zinc supplementation in children with HIV.

While there is some evidence of benefit of individual micronutrients, there is no conclusive evidence of benefit for routine multiple micronutrient supplementation. For instance, a recent randomized controlled trial of multiple micronutrient supplementation in Ugandan HIV-infected children showed no reduction of diarrheal morbidity associated with supplementation.[91]

Another trial among 2387 HIV-exposed Tanzanian infants (all of whose mothers received micronutrient supplementation during pregnancy and lactation) showed that infant micronutrients did not reduce all causes of mortality such as episodes of diarrhea, but did reduce episodes of fever and vomiting. Among 429 children whose mothers received ART, multivitamin use was associated with a significant reduction in hospitalizations (P = .035), episodes of fever (P = .005), and episodes of fever and cough (P = .019) compared with placebo.[92]

DIAGNOSTIC EVALUATION

Malnutrition and wasting are common in HIV-infected children, and therefore routine nutritional assessment should be performed at regular intervals, particularly under 24 months of age, in every child who is at risk for HIV infection. Careful consideration of the pattern of growth will lead to an investigation of nutritional versus endocrine dysfunction. Subtle changes in growth and altered nutrient intake can be detected. The

nutritional assessment should include measures of weight, height, head circumference, triceps skinfold thickness (to measure fat mass) and MUAC (to assess muscle mass). A detailed dietary history and recall should be ascertained regardless of the child's nutritional condition. Dietary advice or encouragement should be reinforced at each visit. Figure 44-1 outlines a clinical algorithm of diagnosis and management of HIV-infected children with nutritional abnormalities.

If a child shows evidence of malnutrition (Table 44-4), nutrient losses should be evaluated in addition to nutrient intake. Clinical symptoms should be determined with the focus on gastrointestinal problems, such as vomiting, abdominal pain, and diarrhea. A complete physical examination will provide additional important clues. The child should be evaluated for oral thrush, abdominal pain, and gross or occult blood in the stool. Laboratory studies may include a complete blood count, liver function studies, pancreatic enzyme levels, and a complete evaluation for enteric pathogens. When resources are available, studies such as a radiographic upper gastrointestinal contrast series, upper gastrointestinal endoscopy, and an abdominal ultrasound may provide a definitive diagnosis. If clinical symptoms warrant, a gastrointestinal absorption evaluation may be performed; including a lactose hydrogen breath test, d-xylose absorption study, and/or 72-hour fecal fat collection.

Serum micronutrients may be assessed in malnourished or wasted HIV-infected children. Consider measurements of vitamins A, E, and D, especially if there is liver, biliary tract or pancreatic disease. Serum zinc and selenium can also be measured and may be low; if so, they should be replaced to optimize immune function.

It may also be useful, where able, to evaluate a child's energy needs. Although the use of doubly labeled water as a measure of total energy expenditure remains a research test, clinical centers are increasingly able to perform indirect calorimetry as a bedside test of resting energy expenditure in children. In a practical sense, the best estimation of energy needs for an HIV-infected child is the child's response to a diet enriched in calories, with accurate documentation of caloric intake and type of food.

Among HIV-infected children receiving chronic ART, it is also important to monitor for potential changes in body fat distribution and abnormalities in metabolic parameters that have been linked to the use of HAART. For example, skinfold thickness measurements obtained using standardized calipers by trained bionutritionist can be effective in identifying and monitoring peripheral lipoatrophy.[93] This combined with annual anthropometric assessment of waist and hip circumference with

calculation of a waist-to-hip ratio may be very useful in detecting alterations in body fat distribution that develop over time with exposure to HAART. Noninvasive measures of body composition (bioelectrical impedance, air displacement plethysmography) may also be helpful in some settings.

It is currently recommended that HIV-infected adults have fasting serum glucose and lipids checked prior to initiation of ART and then repeated biannually after initiation of therapy as well as following any changes in ART. While there are no HIV-specific recommendations for performing more detailed screening for diabetes, such as on oral glucose tolerance testing, this may be beneficial in cases where there is concern for insulin resistance or documented impaired fasting glucose (fasting glucose > 100 mg/dL).

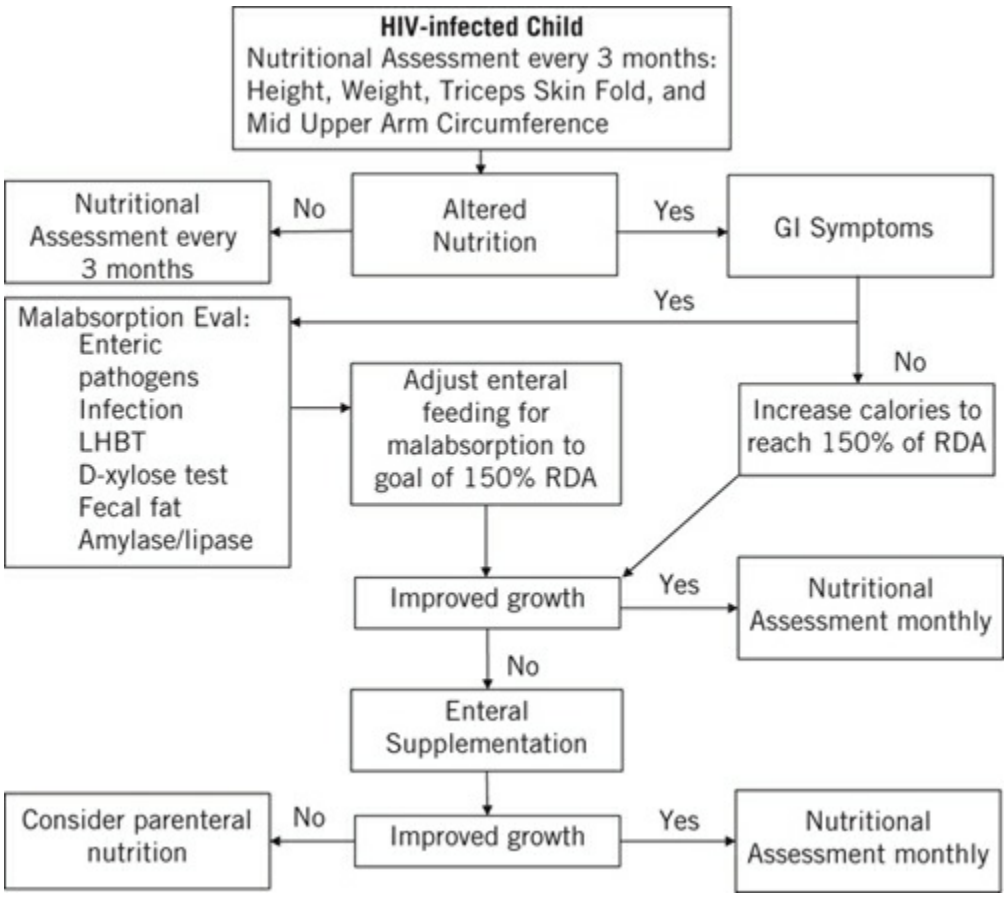


Figure 44-1 Algorithm for monitoring, diagnosing, and treating malnutrition associated with human immunodeficiency (HIV) disease in infants and children. GI, gastrointestinal; LHBT, lactose hydrogen breath test; RDA, recommended dietary allowance.

TABLE 44-4 Clinical Indicators of Malnutrition in HIV-infected Children

Measurements
Weight growth velocity < 5% for more than 2 months
Decrease in 1 major growth percentile for weight
Weight or weight-for-height percent standard < 90%
Weight-for-height < 5th percentile
Loss of > 5% of lean body mass
Serum albumin < 3 g/dL

Nutritional Interventions

Oral Supplementation

Nutritional interventions should be instituted when the child fails to meet growth standards as defined in Table 44-4. In general, a proactive approach to nutritional guidance is preferred when early nutritional interventions are more effective than during the later stages of HIV disease. Nutritional therapies should be instituted in parallel with treatment for other ongoing systemic or gastrointestinal diseases. Oral nutritional therapies, in most patients, should be tried initially. Formula caloric density should be increased by concentration or adding sunflower oil. If the child is eating solids, adding extra fat in the form of butter, margarine, peanut butter, or sunflower oil may be helpful. Since a precise estimation of caloric needs is usually not available, the caloric goal, in general, should be targeted at 50% above the RDA for age and sex for a child with growth failure. Other nutritional requirements are shown in Table 44-5, although these requirements are considered minimal, and more may be required if the child has clinical and biochemical evidence of nutritional deficiencies. Formula composition should be prescribed according to the extent of nutrient malabsorption. If a child has clinical symptoms indicating malabsorption of lactose (e.g., diarrhea, bloating, flatulence), a lactose-free diet is indicated. Caution is advised, however, in prescribing a lactose-free diet if the child has no clinical symptoms of lactose malabsorption, as limiting lactose may also restrict the foods that are the most enjoyable and richest in energy. In addition, an oral lactase supplement may be added in some cases of lactose intolerance. Yogurt and other fermented milks may also be better tolerated as they have less lactose and are also often more culturally acceptable in developing countries. A protein hydrolysate

formula should be considered if the child has evidence of gastrointestinal mucosal dysfunction with malabsorption of fats and proteins.

A wide variety of food supplements have been tested in studies in HIV-infected children and generally those children that receive supplements have shown better weight gain than those receiving standard nutritional support. A study in South Africa showed that protein-supplemented milk formula improved WAZ and HAZ scores in HIV-infected children with prolonged diarrhea.[94] A Zambian study using an amino acid–based elemental diet rather than the standard skim milk/soy diet showed the intervention diet resulted in HIV-infected children having better weight gain.[95] Ready-to-use-therapeutic food (RUTF) was shown to result in more rapid weight gain in Malawian HIV-infected children compared with maize-soy flour.[96] In Tanzanian children, the provision of RUTF for at least four months was associated with a lower incidence of underweight and wasting compared with children who did not receive RUTF.[97] Although RUTF has gained a wide acceptance and is becoming standard of care for HIV-infected children, there is a dearth of clinical trials justifying it's use, and insufficient clinical evidence showing that RUTF is a better option than ready to use supplementary food or fortified blended foods or other options.[98]

TABLE 44-5 Nutritional Requirements for HIV-infected Children

Nutrient	Requirements Per Day*
Total calories	100%–150% RDA for age and sex
Protein	100%–150% RDA for age and sex
Vitamin A	400–1000 µg retinol
Vitamin D	400 IU
Vitamin E	3–10 mg d-alpha tocopherol
Vitamin K	5–80 µg
Vitamin C	30–60 mg
Thiamin	0.3–1.1 mg
Riboflavin	0.4–1.3 mg
Niacin	5–15 mg
Vitamin B6	0.3–1.6 mg
Folate	25–180 µg
Vitamin B12	0.3–2.0 µg

Iron	6–15 mg
Zinc	5–12 mg
Selenium	10–55 µg

* Requirements reflect the clinical nutritional status of the child; more or less may be needed depending on evidence of deficiency. Ranges reflect variations in needs based on age and sex.

For infants and young children in particular, breast milk is considered the best nutrition since it contains factors that protect the infant against pathogens. Breastfeeding also fosters the maternal-infant bond. Unfortunately, women with HIV infection can transmit the virus to their infant through breast milk and therefore breastfeeding in this circumstance is discouraged in countries with low infant mortality rates where other safe feeding options exist. In developing countries where the majority of HIV-infected women live, WHO recommendations[99] encourage women to practice exclusive breastfeeding during the first 6 months of the infant's life and then to introduce complementary foods while continuing breastfeeding for 12 months or longer as indicated. Breastfeeding women should be provided with ARV treatment at least for the duration of the breastfeeding period and their infants should receive daily nevirapine prophylaxis for 6 weeks post-delivery.[100,101]

Enteral Supplementation

If oral interventions fail, because the child cannot consume adequate nutrients to sustain growth, enteral supplementation should be considered. Nasogastric tube feedings should be instituted first, to document the child's ability to gain weight with supplemental enteral feedings. Complications relating to nasogastric tube feedings include sinusitis and the technical inability of the caretaker to administer the feedings or to correctly position the tube. If delivery of feedings through a nasogastric tube improves growth and feeding is required beyond six to eight weeks, then placement of a more permanent device, such as a gastrostomy tube, should be considered. A gastrostomy tube provides easy access to the stomach with less physical and emotional trauma to the child. In addition, studies show that gastrostomy tube placement results in significant improvement in medication compliance and reductions in medication administration time in children with HIV infection.[102,103]

Prior to the widespread use of ART in the United States, a study of 23 HIV-infected children found that weight and WHZ scores increased with gastrostomy-tube feedings.[104] This increase in weight and weight-for-height was coincident with an increase in caloric intake. Additionally,

children who gained weight with gastrostomy-tube feedings were hospitalized less in the six months after gastrostomy-tube placement and had a 2.8-fold reduction in risk of dying. Guarino and colleagues evaluated the effects of enteral (n = 16) and parenteral (n = 46) nutritional therapy for HIV-infected children with malnutrition and evidence of diarrhea or malabsorption.[105] More than 80% of patients receiving enteral therapy gained >5% of their baseline weight, and enteral therapy was associated with significant increases in CD4 cell count. In addition, enteral therapy improved gut absorption with 75% of patients experiencing normalization in the xylose absorption test. Combined, these findings suggest that early nutritional intervention is indicated in children with HIV infection and may improve morbidity and survival.[106]

Parenteral Nutrition

Parenteral nutrition should be reserved for HIV-infected children who continue to lose weight on an aggressive enteral program, who have severe recurrent or chronic pancreatic or biliary tract disease, or have intractable diarrhea with weight loss. The placement of a central venous catheter can provide venous access that is often difficult to obtain in terminal stages. Central venous catheters present an additional risk for sepsis, and studies in adults and children with HIV infection present conflicting results regarding the risk of infection.

Limited data are available on the benefits of parenteral nutrition (PN) in children with HIV infection. In the series reported by Guarino et al., 46 HIV-infected children with malnutrition and malabsorption received a median of 157 days of PN.[105] While CD4 cell count, weight, and xylose absorption all improved significantly in the patients receiving enteral therapy (n = 16), these measures improved on PN, but not significantly. One possible explanation for the difference in findings between PN and enteral nutritional support is that this was an observational study and patients on PN had more advanced disease as indicated by a lower baseline CD4 count (154 cells/mm^3 for enteral patients vs. 55 cells/mm^3 for PN patients). In addition, 58.7% of PN patients vs. 18.8% of enteral patients died during the follow-up period, which may be related to increased risk of infection with central lines and PN, but may also reflect the practice of reserving PN for late and end-stage patients. In resource-poor settings with more advanced HIV disease, the availability of PN is usually limited as well.

Other Nutritional Therapies

Appetite Stimulants

Megestrol acetate, a synthetic orally active progesterone, widely used to treat breast cancer, has shown promising results as an appetite stimulant in adults with HIV infection. The mechanism of weight gain with megestrol acetate is under investigation, but research suggests that it has an effect both on behavior (i.e., food intake) and metabolism, by the induction of lipogenic enzymes and fat synthesis. There are few reports on the use of megestrol acetate in children with HIV infection. Clarick et al. evaluated the effects of megestrol acetate in 19 HIV-infected children for a median of seven months at doses between 4.0 and 8.5 mg/kg/d and found increased WAZ scores with continued therapy, but no improvement in linear growth.[107] The authors also reported that poor weight gain and/or weight loss occurred after megestrol acetate was withdrawn. Adrenal suppression was also noted in 7/10 HIV-infected children treated with megestrol acetate compared with 0/10 HIV-infected children not on megestrol acetate.[108] Therefore, the use of megestrol acetate may be beneficial for weight gain and improved appetite in HIV-infected children, but caution is warranted particularly when discontinuing or withdrawing therapy.

Dronabinol (delta-9-tetrahydrocannabinol) is another appetite stimulant that has been used in adult HIV-infected patients. It was approved for use in the United States in 1986 for treatment of chemotherapy-induced nausea and vomiting, although it was found to stimulate appetite as well. A randomized, placebo- controlled study of dronabinol was performed on 139 HIV patients. Appetite stimulation, measured by visual analog scale, was maintained or increased in patients receiving dronabinol.[109] After at least 2 months of treatment with dronabinol, 38% of patients gained greater than or equal to 2 kg of weight. Altered mental status was the primary adverse effect for this medication. Currently, there are no dosing recommendations for children.

Numerous other therapies have been employed to initiate weight gain in patients with HIV infection. These include essential fatty acid supplementation (n-3 fatty acids), specific vitamin and antioxidant replacements, pancreatic enzymes, testosterone replacement, and cytokine inhibitors such as pentoxifylline and thalidomide. Additionally, studies are underway evaluating the effects of exercise and aerobic training in patients with HIV infection. Unfortunately, there are no therapies discovered to date that universally prevent or reverse nutritional derangements in HIV-

infected patients. However, the improved nutritional status observed with the use of ART supports the importance of suppression of viral replication and avoidance of opportunistic infections in the maintenance of nutritional health.

There are several ancillary interventions, which should be considered to support nutritional interventions, some of these are listed below:

1. Management of Diarrhea:
   Since diarrhea (especially in children under 2 years of age) is integrally associated with malnutrition, it is important to have adequate diarrheal management programs. Starting with preventative strategies, there should be promotion of improved sanitation and hygiene practices including the use of clean water, handwashing, and use of latrines. Since vitamin A supplementation has been shown to reduce the morbidity associated with diarrheal disease,[110] it is important that HIV-infected children receive periodic supplementation to prevent diarrheal disease and to lessen its severity. WHO recommendations suggest that children should be supplemented with vitamin A twice a year (100,000 IU for infants aged 6–12 months and 200,000 IU for children aged 1–5 years).[111] Furthermore, adherence to Integrated Management of Childhood Illness (IMCI) guidelines with prompt treatment of diarrhea will have an important effect on preserving the nutritional status of the child. WHO recommendations for treatment include low osmolarity oral rehydration fluids and oral zinc supplementation for 10 to 14 days (10 mg/d for infants <6 months and 20 mg/d for those ≥6 months. Children with persistent diarrhea should receive daily multiple micronutrients for 14 days.
2. Deworming of young children in areas where worm infestations are common:
   Since worm infestations often results in poor appetite, and anemia, six monthly deworming is recommended using oral Albendazole (single dose of 400 mg).
3. Improved care and stimulation of children:
   Improved care with appropriate stimulation of children including physical activity and play can help develop and maintain adequate muscle development. This, in turn, improves their sense of well-being, which impacts positively on their appetite and growth.

4. Following nutritional food guidelines:
   Most countries have adapted WHO recommendations and developed locally appropriate guidelines. The WHO has encouraged an integrated approach to the nutritional care of HIV-infected children and have developed guidelines[112] to assist countries to implement such integrated programs, as well as a guide for local adaption of these guidelines at country level in order to reflect local dietary practices.

CONCLUSION

Great strides have been made over the past two decades regarding the detection, prevention, and virologic treatment of HIV in childhood, with the basic understanding of the interaction between nutritional status and HIV becoming better understood. The cause of nutritional problems in patients with HIV infection is complex and likely multifactorial. It is clear, however, that malnutrition of HIV-infected children is among the most serious, chronic, and difficult to treat problems confronting clinicians caring for HIV-infected children, especially in developing countries. Clinicians should be aware of nutritional problems, and close surveillance of nutrition should be conducted on all children who are at risk for HIV infection. Early, aggressive nutritional interventions are indicated in cases of malnutrition. As a treatment strategy, improving nutritional status of HIV-infected children may improve clinical outcome.

While the incidence of malnutrition in developed countries has decreased with HAART therapy, other metabolic problems can arise. The long-term risks of HAART on HIV-infected children are unknown. Close surveillance for the emergence of insulin resistance, dyslipidemias, and premature cardiovascular disease will be important as children with perinatally acquired HIV age into adulthood. Further investigations on the short- and long-term nutritional and metabolic effects of HAART therapy are still underway. Investigators should continue to study the effects of optimal nutrition, lipid management, and healthy life style changes for HIV-infected children.

ACKNOWLEDGMENT

This chapter is a revised and updated version of the previous chapter by Miller T. and Hadigan C., Nutritional aspects of pediatric HIV infection, which appeared in *Nutrition in Pediatrics: Basic Science and Clinical*

Applications. 3rd ed. Eds. Watkins JB, Duggan C, and Walker WA. BC Decker Europe Inc.

REFERENCES

1. WHO. *Global report: UNAIDS report on the global AIDS epidemic 2012*. Geneva, Switzerland: WHO; 2012.
2. Hughes WT, Price RA, Sisko F, et al. Protein-calorie malnutrition. A host determinant for Pneumocystis carinii infection. *Am J Dis Child*. 1974;128(1):44–52.
3. Chandra RK, Newbuerne PM. *Nutrition, Immunity and Infection: Mechanisms of Interactions*. New York, NY: Plenum; 1977.
4. Scrimshaw NS, Taylor CE, Gordon JE. Interactions of nutrition and infection. *Monogr Ser World Health Organ*. 1968;57:3–329.
5. Sunguya BF, Poudel KC, Otsuka K, et al. Undernutrition among HIV-positive children in Dar es Salaam, Tanzania: antiretroviral therapy alone is not enough. *BMC Public Health*. 2011;11:869.
6. van Lettow M, van der Meer JW, West CE, et al. Interleukin-6 and human immunodeficiency virus load, but not plasma leptin concentration, predict anorexia and wasting in adults with pulmonary tuberculosis in Malawi. *J Clin Endocrinol Metab*. 2005;90(8):4771–4776.
7. Arpadi SM, Cuff PA, Kotler DP, et al. Growth velocity, fat-free mass and energy intake are inversely related to viral load in HIV-infected children. *J Nutr*. 2000;130(10):2498–2502.
8. Cello JP, Day LW. Idiopathic AIDS enteropathy and treatment of gastrointestinal opportunistic pathogens. *Gastroenterology*. 2009;136(6):1952–1965.
9. Kotler DP. HIV infection and the gastrointestinal tract. *AIDS*. 2005;19(2):107–117.
10. Delezay O, Yahi N, Tamalet C, et al. Direct effect of type 1 human immunodeficiency virus (HIV-1) on intestinal epithelial cell differentiation: relationship to HIV-1 enteropathy. *Virology*. 1997;238(2):231–242.
11. Intestinal malabsorption of HIV-infected children: relationship to diarrhoea, failure to thrive, enteric micro-organisms and immune impairment. The Italian Paediatric Intestinal/HIV Study Group. *AIDS*. 1993;7(11):1435–1440.
12. Sentongo TA, Rutstein RM, Stettler N, et al. Association between steatorrhea, growth, and immunologic status in children with perinatally acquired HIV infection. *Arch Pediatr Adolesc Med*. 2001;155(2):149–153.
13. Alfaro MP, Siegel RM, Baker RC, Heubi JE. Resting energy expenditure and body composition in pediatric HIV infection. *Pediatr AIDS HIV Infect*. 1995;6(5):276–280.
14. Henderson RA, Talusan K, Hutton N, et al. Resting energy expenditure and body composition in children with HIV infection. *J Acquir Immune Defic Syndromes Hum Retrovirol*. 1998;19(2):150–157.
15. Johann-Liang R, O'Neill L, Cervia J, et al. Energy balance, viral burden, insulin-like growth factor-1, interleukin-6 and growth impairment in children infected with human immunodeficiency virus. *AIDS*. 2000;14(6):683–690.
16. Henderson RA, Talusan K, Hutton N, et al. Whole body protein turnover in children with human immunodeficiency virus (HIV) infection. *Nutrition*. 1999;15(3):189–194.
17. Hardin DS, Ellis KJ, Rice J, Doyle ME. Protease inhibitor therapy improves

protein catabolism in prepubertal children with HIV infection. *J Pediatr Endocrinol Metab*. 2004;17(3):321–325.
18. Beutler B, Cerami A. Cachectin: more than a tumor necrosis factor. *N Engl J Med*. 1987;316(7):379–385.
19. Beutler B. Cytokines and cancer cachexia. *Hosp Pract*. 1993;28(4):45–52.
20. Hellerstein MK, Grunfeld C, Wu K, et al. Increased de novo hepatic lipogenesis in human immunodeficiency virus infection. *J Clin Endocrinol Metab*. 1993;76(3):559–565.
21. Roubenoff R, Grinspoon S, Skolnik PR, et al. Role of cytokines and testosterone in regulating lean body mass and resting energy expenditure in HIV-infected men. *Am J Physiol Endocrinol Metab*. 2002;283(1):E138-E145.
22. Galhardo MC, de Carvalho MG, Georg I, et al. Nutritional status impairments in HIV-infected patients are associated with increased TNF-alpha and IL-6 serum levels but not with viral load. *Infection*. 2001;29(5):257–261.
23. Missmer SA, Spiegelman D, Gorbach SL, Miller TL. Predictors of change in the functional status of children with human immunodeficiency virus infection. *Pediatrics*. 2000;106(2):E24.
24. Boulton TJ, Smith R, Single T. Psychosocial growth failure: a positive response to growth hormone and placebo. *Acta Paediatr*. 1992;81(4):322–325.
25. Money J. The syndrome of abuse dwarfism (psychosocial dwarfism or reversible hyposomatotropism). *Am J Dis Child*. 1977;131(5):508–513.
26. Miller TL, Easley KA, Zhang W, et al. Maternal and infant factors associated with failure to thrive in children with vertically transmitted human immunodeficiency virus-1 infection: the prospective, P2C2 human immunodeficiency virus multicenter study. *Pediatrics*. 2001;108(6):1287–1296.
27. Moye J Jr, Rich KC, Kalish LA, et al. Natural history of somatic growth in infants born to women infected by human immunodeficiency virus. Women and Infants Transmission Study Group. *J Pediatr*. 1996;128(1):58–69.
28. Lifschitz MH, Wilson GS, Smith EO, Desmond MM. Fetal and postnatal growth of children born to narcotic-dependent women. *J Pediatr*. 1983;102(5):686–691.
29. Agostoni C, Zuccotti GV, Gianni ML, et al. Body mass index development during the first 6 months of life in infants born to human immunodeficiency virus-seropositive mothers. *Acta Paediatr*. 1998;87(4):378–380.
30. Arpadi SM. Growth failure in children with HIV infection. *J Acquir Immune Defic Syndr*. 2000;25 (suppl 1):S37-S42.
31. Bobat R, Coovadia H, Moodley D, et al. Growth in early childhood in a cohort of children born to HIV-1-infected women from Durban, South Africa. *Ann Trop Paediatr*. 2001;21(3):203–210.
32. Chiaba A, Lebela J, Kobela M, et al. The frequency and magnitude of growth failure in a group of HIV-infected children in Cameroon. *Pan Afr Med J*. 2012;11:15. Epub 2012 March 01.
33. Newell ML, Borja MC, Peckham C; European Collaborative Study. Height, weight, and growth in children born to mothers with HIV-1 infection in Europe. *Pediatrics*. 2003;111(1):e52–e60.
34. Saavedra JM, Henderson RA, Perman JA, et al. Longitudinal assessment of growth in children born to mothers with human immunodeficiency virus infection. *Arch Pediatr Adolesc Med*. 1995;149(5):497–502.
35. Gsponer T, Weigel R, Davies MA, et al. Variability of growth in children starting antiretroviral treatment in southern Africa. *Pediatrics*. 2012;130(4):e966–e977.
36. Chantry CJ, Byrd RS, Englund JA, et al; Pediatric ACTGPST. Growth, survival and viral load in symptomatic childhood human immunodeficiency virus

infection. *Pediatr Infect Dis J*. 2003;22(12):1033–1039.
37. Rainwater-Lovett K, Nkamba HC, Mubiana-Mbewe M, et al. Immunologic risk factors for early mortality after starting antiretroviral therapy in HIV-infected zambian children. *AIDS Res Hum Retroviruses*. 2013;29:479–487.
38. Mgone CS, Mhalu FS, Shao JF, et al. Prevalence of HIV-1 infection and symptomatology of AIDS in severely malnourished children in Dar Es Salaam, Tanzania. *J Acquir Immune Defic Syndr*. 1991;4(9):910–913.
39. Dreyfuss ML, Msamanga GI, Spiegelman D, et al. Determinants of low birth weight among HIV-infected pregnant women in Tanzania. *Am J Clin Nutr*. 2001;74(6):814–826.
40. Young S, Murray K, Mwesigwa J, et al. Maternal nutritional status predicts adverse birth outcomes among HIV-infected Rural Ugandan women receiving combination antiretroviral therapy. *PloS One*. 2012;7(8):e41934.
41. Chen JY, Ribaudo HJ, Souda S, et al. Highly active antiretroviral therapy and adverse birth outcomes among HIV-infected women in Botswana. *J Infect Dis*. 2012;206(11):1695–1705.
42. Sibiude J, Warszawski J, Tubiana R, et al. Premature delivery in HIV-infected women starting protease inhibitor therapy during pregnancy: role of the ritonavir boost? *Clin Infect Dise.* 2012;54(9):1348–1360.
43. Patel D, Bland R, Coovadia H, et al. Breastfeeding, HIV status and weights in South African children: a comparison of HIV-exposed and unexposed children. *AIDS*. 2010;24(3):437–445.
44. Kuhn L, Aldrovandi GM, Sinkala M, et al. Effects of early, abrupt weaning on HIV-free survival of children in Zambia. *N Engl J Med*. 2008;359(2):130–141.
45. Obimbo EM, Mbori-Ngacha DA, Ochieng JO, et al. Predictors of early mortality in a cohort of human immunodeficiency virus type 1-infected african children. *Pediatr Infect Dis J*. 2004;23(6):536–543.
46. Taha TE, Kumwenda NI, Hoover DR, et al. The impact of breastfeeding on the health of HIV-positive mothers and their children in sub-Saharan Africa. *Bull World Health Organ*. 2006;84(7):546–554.
47. Thior I, Lockman S, Smeaton LM, et al. Breastfeeding plus infant zidovudine prophylaxis for 6 months vs formula feeding plus infant zidovudine for 1 month to reduce mother-to-child HIV transmission in Botswana: a randomized trial: the Mashi Study. *JAMA*. 2006;296(7):794–805.
48. Miller TL, Evans SJ, Orav EJ, et al. Growth and body composition in children infected with the human immunodeficiency virus-1. *Am J Clin Nutr*. 1993;57(4):588–592.
49. Fontana M, Zuin G, Plebani A, et al. Body composition in HIV-infected children: relations with disease progression and survival. *Am J Clin Nutr*. 1999;69(6):1282–1286.
50. Arpadi SM, Horlick MN, Wang J, et al. Body composition in prepubertal children with human immunodeficiency virus type 1 infection. *Arch Pediatr Adolesc Med*. 1998;152(7):688–693.
51. Weight, height and human immunodeficiency virus infection in young children of infected mothers. The European Collaborative Study. *Pediatr Infect Dis J*. 1995;14(8):685–690.
52. Nadal D, Steiner F, Cheseaux JJ, Rudin C. Ritonavir promotes increased growth in HIV-infected children. Paediatric AIDS Group of Switzerland. *AIDS*. 1998;12(17):2356–2357.
53. Steiner F, Kind C, Aebi C, et al. Growth in human immunodeficiency virus type 1-infected children treated with protease inhibitors. *Eur J Pediatr*.

2001;160(10):611–616.
54. Nachman SA, Lindsey JC, Moye J, et al. Growth of human immunodeficiency virus-infected children receiving highly active antiretroviral therapy. *Pediatr Infect Dis J*. 2005;24(4):352–357.
55. Peacock-Villada E, Richardson BA, John-Stewart GC. Post-HAART outcomes in pediatric populations: comparison of resource-limited and developed countries. *Pediatrics*. 2011;127(2):e423–e441.
56. Bolton-Moore C, Mubiana-Mbewe M, Cantrell RA, et al. Clinical outcomes and CD4 cell response in children receiving antiretroviral therapy at primary health care facilities in Zambia. *JAMA*. 2007;298(16):1888–1899.
57. Wamalwa DC, Obimbo EM, Farquhar C, et al. Predictors of mortality in HIV-1 infected children on antiretroviral therapy in Kenya: a prospective cohort. *BMC Pediatr*. 2010;10:33.
58. Leyenaar JK, Novosad PM, Ferrer KT, et al. Early clinical outcomes in children enrolled in human immunodeficiency virus infection care and treatment in lesotho. *Pediatr Infect Dis J*. 2010;29(4):340–345.
59. Verweel G, van Rossum AM, Hartwig NG, et al. Treatment with highly active antiretroviral therapy in human immunodeficiency virus type 1-infected children is associated with a sustained effect on growth. *Pediatrics*. 2002;109(2):E25.
60. Grinspoon S, Carr A. Cardiovascular risk and body-fat abnormalities in HIV-infected adults. *N Engl J Med*. 2005;352(1):48–62.
61. Kotler DP. HIV and antiretroviral therapy: lipid abnormalities and associated cardiovascular risk in HIV-infected patients. *J Acquir Immune Defic Syndr*. 2008;49 (suppl 2):S79-S85.
62. Brown TT, Cole SR, Li X, et al. Antiretroviral therapy and the prevalence and incidence of diabetes mellitus in the multicenter AIDS cohort study. *Arch Intern Med*. 2005;165(10):1179–1184.
63. Friis-Moller N, Sabin CA, Weber R, et al. Combination antiretroviral therapy and the risk of myocardial infarction. *N Engl J Med*. 2003;349(21):1993–2003.
64. Arpadi SM, Cuff PA, Horlick M, et al. Lipodystrophy in HIV-infected children is associated with high viral load and low CD4+ -lymphocyte count and CD4+ -lymphocyte percentage at baseline and use of protease inhibitors and stavudine. *J Acquir Immune Defic Syndr*. 2001;27(1):30–34.
65. Beregszaszi M, Dollfus C, Levine M, et al. Longitudinal evaluation and risk factors of lipodystrophy and associated metabolic changes in HIV-infected children. *J Acquir Immune Defic Syndr*. 2005;40(2):161–168.
66. Eley BS, Sive AA, Abelse L, et al. Growth and micronutrient disturbances in stable, HIV-infected children in Cape Town. *Ann Trop Paediatr*. 2002;22(1):19–23.
67. Calis JC, van Hensbroek MB, de Haan RJ, et al. HIV-associated anemia in children: a systematic review from a global perspective. *AIDS*. 2008;22(10):1099–1112.
68. Shet A, Arumugam K, Rajagopalan N, et al. The prevalence and etiology of anemia among HIV-infected children in India. *Eur J Pediatr*. 2012;171(3):531–540.
69. Kruzich LA, Marquis GS, Carriquiry AL, et al. US youths in the early stages of HIV disease have low intakes of some micronutrients important for optimal immune function. *J Am Diet Assoc*. 2004;104(7):1095–1101.
70. Tang AM, Graham NM, Kirby AJ, et al. Dietary micronutrient intake and risk of progression to acquired immunodeficiency syndrome (AIDS) in human immunodeficiency virus type 1 (HIV-1)-infected homosexual men. *Am J*

Epidemiol. 1993;138(11):937–951.
71. Read JS, Bethel J, Harris DR, et al. Serum vitamin A concentrations in a North American cohort of human immunodeficiency virus type 1-infected children. National Institute of Child Health and Human Development Intravenous Immunoglobulin Clinical Trial Study Group. *Pediatr Infect Dis J*. 1999;18(2):134–142.
72. Fawzi WW, Mbise RL, Hertzmark E, et al. A randomized trial of vitamin A supplements in relation to mortality among human immunodeficiency virus-infected and uninfected children in Tanzania. *Pediatr Infect Dis J*. 1999;18(2):127–133.
73. Filteau SM, Rollins NC, Coutsoudis A, et al. The effect of antenatal vitamin A and beta-carotene supplementation on gut integrity of infants of HIV-infected South African women. *J Pediatr Gastroenterol Nutr*. 2001;32(4):464–470.
74. Greenberg BL, Semba RD, Vink PE, et al. Vitamin A deficiency and maternal-infant transmissions of HIV in two metropolitan areas in the United States. *AIDS*. 1997;11(3):325–332.
75. Melikian G, Mmiro F, Ndugwa C, et al. Relation of vitamin A and carotenoid status to growth failure and mortality among Ugandan infants with human immunodeficiency virus. *Nutrition*. 2001;17(7–8):567–572.
76. Semba RD, Miotti PG, Chiphangwi JD, et al. Maternal vitamin A deficiency and mother-to-child transmission of HIV-1. *Lancet*. 1994;343(8913):1593–1597.
77. Villamor E, Mbise R, Spiegelman D, et al. Vitamin A supplements ameliorate the adverse effect of HIV-1, malaria, and diarrheal infections on child growth. *Pediatrics*. 2002;109(1):E6.
78. Baum MK, Mantero-Atienza E, Shor-Posner G, et al. Association of vitamin B6 status with parameters of immune function in early HIV-1 infection. *J Acquir Immune Defic Syndr*. 1991;4(11):1122–1132.
79. Semeere AS, Nakanjako D, Ddungu H, et al. Sub-optimal vitamin B-12 levels among ART-naive HIV-positive individuals in an urban cohort in Uganda. *PloS One*. 2012;7(7):e40072.
80. Alani A, Vincent O, Adewumi A, et al. Plasma folate studies in HIV-positive patients at the Lagos university teaching hospital, Nigeria. *Indian J Sex Transm Dis*. 2010;31(2):99–103.
81. Revell P, O'Doherty MJ, Tang A, Savidge GF. Folic acid absorption in patients infected with the human immunodeficiency virus. *J Intern Med*. 1991;230(3):227–231.
82. Fawzi WW, Msamanga GI, Wei R, et al. Effect of providing vitamin supplements to human immunodeficiency virus-infected, lactating mothers on the child's morbidity and CD4+ cell counts. *Clin Infect Dis*. 2003;36(8):1053–1062.
83. Fawzi WW, Msamanga GI, Spiegelman D, et al. A randomized trial of multivitamin supplements and HIV disease progression and mortality. *N Engl J Med*. 2004;351(1):23–32.
84. Sappey C, Legrand-Poels S, Best-Belpomme M, et al. Stimulation of glutathione peroxidase activity decreases HIV type 1 activation after oxidative stress. *AIDS Research and Human Retroviruses*. 1994;10(11):1451–1461.
85. Rodriguez JF, Cordero J, Chantry C, et al. Plasma glutathione concentrations in children infected with human immunodeficiency virus. *Pediatr Infect Dis J*. 1998;17(3):236–241.
86. Kupka R, Msamanga GI, Spiegelman D, et al. Selenium levels in relation to morbidity and mortality among children born to HIV-infected mothers. *Eur J Clin Nutr*. 2005;59(11):1250–1258.

87. Sazawal S, Black RE, Bhan MK, et al. Zinc supplementation in young children with acute diarrhea in India. *N Engl J Med*. 1995;333(13):839–844.
88. Graham NM, Sorensen D, Odaka N, et al. Relationship of serum copper and zinc levels to HIV-1 seropositivity and progression to AIDS. *J Acquir Immune Defic Syndr*. 1991;4(10):976–980.
89. Reich EN, Church JA. Oral zinc supplementation in the treatment of HIV-infected children. *Pediatr AIDS HIV Infect*. 1994;5(6):357–360.
90. Bobat R, Coovadia H, Stephen C, et al. Safety and efficacy of zinc supplementation for children with HIV-1 infection in South Africa: a randomised double-blind placebo-controlled trial. *Lancet*. 2005;366(9500):1862–1867.
91. Ndeezi G, Tylleskar T, Ndugwa CM, Tumwine JK. Multiple micronutrient supplementation does not reduce diarrhoea morbidity in Ugandan HIV-infected children: a randomised controlled trial. *Paediatr Int Child Health*. 2012;32(1):14–21.
92. Duggan C, Manji KP, Kupka R, et al. Multiple micronutrient supplementation in Tanzanian infants born to HIV-infected mothers: a randomized, double-blind, placebo-controlled clinical trial. *Am J Clin Nutr*. 2012;96(6):1437–1446. doi: 10.3945/ajcn.112.044263. Epub 2012 November 7.
93. Saint-Marc T, Partisani M, Poizot-Martin I, et al. Fat distribution evaluated by computed tomography and metabolic abnormalities in patients undergoing antiretroviral therapy: preliminary results of the LIPOCO study. *AIDS*. 2000;14(1):37–49.
94. Rollins NC, van den Broeck J, Kindra G, et al. The effect of nutritional support on weight gain of HIV-infected children with prolonged diarrhoea. *Acta Paediatr*. 2007;96(1):62–68.
95. Amadi B, Mwiya M, Chomba E, et al. Improved nutritional recovery on an elemental diet in Zambian children with persistent diarrhoea and malnutrition. *J Trop Pediatr*. 2005;51(1):5–10.
96. Ndekha MJ, Manary MJ, Ashorn P, Briend A. Home-based therapy with ready-to-use therapeutic food is of benefit to malnourished, HIV-infected Malawian children. *Acta Paediatr*. 2005;94(2):222–225.
97. Sunguya BF, Poudel KC, Mlunde LB, et al. Ready to Use Therapeutic Foods (RUTF) improves undernutrition among ART-treated, HIV-positive children in Dar es Salaam, Tanzania. *Nutr J*. 2012;11:60.
98. de Pee S, Manary M, Ashorn P. The devil is in the details. *Nutr Rev*. 2011;69(2):116–117; author reply 118.
99. WHO. Consolidated Guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: Recommendations for a Public Health Approach. Geneva, 2013; http://www.who.int/hiv/pub/guidelines/arv2013/en/. Accessed March 6, 2015.
100. Natchu U, Liu E, Duggan C, et al. Exclusive breastfeeding reduces risk of mortality in infants up to 6 mo of age born to HIV-positive Tanzanian women. *Am J Clin Nutr*. 2012;96:1071–1078.
101. Thior I, Lockman S, Smeaton LM, et al. Breastfeeding plus infant zidovudine prophylaxis for 6 months vs formula feeding plus infant zidovudine for 1 month to reduce mother-to-child HIV transmission in Botswana a randomized trial: the Mashi Study. *JAMA*. 2006;296:794–805.
102. Shingadia D, Viani RM, Yogev R, et al. Gastrostomy tube insertion for improvement of adherence to highly active antiretroviral therapy in pediatric patients with human immunodeficiency virus. *Pediatrics*. 2000;105(6):E80.
103. Temple ME, Koranyi KI, Nahata MC. Gastrostomy tube placement in

nonadherent HIV-infected children. *Ann Pharmacother*. 2001;35(4):414–418.
104. Miller TL, Awnetwant EL, Evans S, et al. Gastrostomy tube supplementation for HIV-infected children. *Pediatrics*. 1995; 96(4, pt 1):696–702.
105. Guarino A, Spagnuolo MI, Giacomet V, et al. Effects of nutritional rehabilitation on intestinal function and on CD4 cell number in children with HIV. *J Pediatr Gastroenterol Nutr*. 2002;34(4):366–371.
106. Ockenga J, Grimble R, Jonkers-Schuitema C, et al. ESPEN Guidelines on Enteral Nutrition: wasting in HIV and other chronic infectious diseases. *Clin Nutr*. 2006;25(2):319–329.
107. Clarick RH, Hanekom WA, Yogev R, Chadwick EG. Megestrol acetate treatment of growth failure in children infected with human immunodeficiency virus. *Pediatrics*. 1997;99(3):354–357.
108. Stockheim JA, Daaboul JJ, Yogev R, et al. Adrenal suppression in children with the human immunodeficiency virus treated with megestrol acetate. *J Pediatr*. 1999;134(3):368–370.
109. Beal JE, Olson R, Laubenstein L, et al. Dronabinol as a treatment for anorexia associated with weight loss in patients with AIDS. *J Pain Symptom Manage*. 1995;10(2):89–97.
110. Imdad A, Yakoob MY, Sudfeld C, et al. Impact of vitamin A supplementation on infant and childhood mortality. *BMC Public Health*. 2011;11 (suppl 3):S20.
111. WHO. *WHO Recommendations on the Management of Diarrhoea and Pneumonia in HIV-Infected Infants and Children: Integrated Management of Childhood Illness*. Geneva, Switzerland: WHO; 2010.
112. WHO. *Guidelines for an Integrated Approach to Nutritional Care of HIV-Infected Children (6 month-14 years)*. Geneva, Switerland: WHO; 2010.

Obesity in Developing Economies

45

Sumathi Swaminathan, PhD and Anura Kurpad, MD, PhD, FAMS

Now, populations in developing countries appear to be paradoxically at risk of high rates of obesity. One tends to associate developing countries with poverty, and the occurrence of overweight or obesity in these countries appears counter-intuitive. The picture of underweight children needs to be replaced with a picture in which poor countries have both underweight and overweight children. This is the so-called "dual burden of disease" in developing countries, since first, poverty is still difficult to eradicate, and second, economic development is never uniform and usually distorted, such that the benefits of economic growth are enjoyed by only a section of the population, with the attendant appearance of obesity.

Although childhood overweight and obesity rates are increasing at an alarming rate globally with an estimated 43 million, or approximately 7% of children affected under the age of 5 years,[1] the situation is perhaps worse in developing countries or economies with about 35 million children affected.[2] A large part of the world's population lives in developing countries that comprise a large part of Asia (except Japan), Africa, Central and South America. Childhood obesity is alarming in developing countries, since apart from developing risk factors for noncommunicable or chronic disease (NCD) or exhibiting frank signs of NCD such as diabetes or cardiovascular disease earlier in life, the likelihood of childhood overweight and obesity tracking into adulthood, concomitant with an increase in NCD, is a matter of great concern. In addition to the burden of disease, the economic burden of treatment is high as it is usually lifelong, and more often than not, met from personal finances in

developing economies.

The recent increase of childhood overweight and obesity in developing economies has largely been attributed to their economic and nutrition transition, and particularly with increasing urbanization, albeit at different rates in different countries. Therefore, changes in lifestyle and dietary habits, with an increase in food consumption and shifts in consumption patterns, particularly of calorie-dense foods, as well as inactivity, are largely implicated as drivers for this increase.[3,4]

TRENDS IN THE PREVALENCE OF CHILDHOOD OVERWEIGHT AND OBESITY

The prevalence of childhood overweight is highest in developed countries, with prevalences as high as 15% as measured in 2011. However, developing economies also show a trend of increasing overweight and obesity rates, as for example in Africa, where rates have increased from 4% in 1990, to 7% in 2011. Although Asia has a lower prevalence rate of 5%, in absolute numbers, about 17 million Asian children under 5 years of age are affected, versus about 12 million in Africa.[1] The worldwide trend of overweight and obesity in pre-school children suggests that the prevalence is expected to reach 9.1%, that is, approximately 60 million children in 2020.[5] Table 45-1 indicates secular trends of overweight in children below 5 years of age with prevalence estimates based on the WHO Child Growth Standards[6] median of weight-for-height above 2 standard deviations (SD). The dataset included 639 nationally representative surveys from 142 countries/territories conducted from 1985 to 2011.

On the basis of the World Bank's income classification of July 2012 (low income, middle income, upper middle income and high income, based on the gross national income), the rate of increase in overweight and obesity in children below 5 years of age from 1990, for low income and high income countries are similar, although at different levels (Figure 45-1A and B). Prevalence estimates among low-income countries are reaching the level of lower middle-income countries, but in absolute numbers, there is a threefold increase in obesity and overweight in low-income countries.[1]

Between the ages of 5 and 19 years, a global prevalence of about 10% has been reported with prevalences over 15% reported in Brazil, India, and Mexico (7). In Brazil, between 1974–1975 and 1996–1997, overweight and obesity in 6- to 18-year-old children increased more than threefold,

from 4.1% to 13.9%, in China from 0.93% to 14.9% in boys and from 1.6% to 8.9% in girls between 1985 and 2000 and in 14- to 17-year-old children in India from 9.8% to 11.7% between 2006 and 2009 (7). Although in developed economies, child overweight and obesity have increased particularly in lower income households and in ethnic minority groups; in contrast, in developing economies as in India and China, the rates have risen rapidly in the urban areas and in high-income households.[7,8]

The problem with childhood obesity is that it is likely to track into adulthood, and with the development of risk factors early in life leading to adverse consequences later, these trends are alarming. Understanding biological factors associated with obesity, as well as societal drivers are important.

TABLE 45-1 Estimated Prevalence and Number of Children Under 5 Years of Age Affected by Over-Weight (Including Obesity) by United Nations Region: 1990, 2010, 2011[a]

	Prevalence Estimate (%)			Number (Million)		
Region	**1990**	**2010**	**2011**	**1990**	**2010**	**2011**
AFRICA	4.2	7.1	7.3	4.6	11.0	11.5
Eastern	4.4	5.0	5.0	1.6	2.6	2.7
Middle	3.5	5.6	5.8	0.5	1.2	1.3
Northern	7.3	12.8	13.1	1.6	3.0	3.1
Southern	6.1	15.6	16.3	0.4	0.9	1.0
Western	1.9	6.2	6.6	0.6	3.2	3.4
ASIA[b]	3.7	4.6	4.7	14.4	16.5	16.9
Eastern	6.8	5.6	5.5	8.8	4.9	4.9
South-Central	2.0	3.0	3.1	3.6	5.6	5.8
South-Eastern	1.8	5.8	6.1	1.0	3.1	3.3
Western	4.4	10.8	11.3	1.0	2.8	3.0
LATIN AMERICA AND CARIBBEAN	6.5	7.1	7.1	3.6	3.8	3.8
Caribbean	4.0	7.3	7.5	0.2	0.3	0.3
Central America	5.1	6.4	6.4	0.8	1.0	1.0
South America	7.3	7.4	7.4	2.6	2.5	2.5
	2.6	3.6	3.7	0.0	0.0	0.0

OCEANIA[c]						
ALL DEVELOPING COUNTRIES	4.1	5.5	5.7	22.7	31.3	32.3
DEVELOPED COUNTRIES	7.4	14.1	14.5	5.7	9.9	10.3
GLOBAL	**4.5**	**6.5**	**6.6**	**28.4**	**41.2**	**42.6**

[a] Adapted from UNICEF-WHO-The World Bank, 2012.[1] Only prevalence estimates are provided without 95% confidence limits.

[b] Excluding Japan.

[c] Excluding Australia and New Zealand.

EARLY LIFE CYCLE FACTORS

Birth Weight and Breastfeeding

Of 18 million infants born of low birth weight (<2500 g) worldwide, 9.3 million are born in South Asia and 3.1 million in sub-Saharan Africa.[9] Both low birth weight, which reflects the pattern of intrauterine growth, and subsequent rapid catch-up growth during infancy, are associated with obesity, metabolic syndrome, dyslipidemia, hypertension, and insulin resistance later in life. The developmental origins of health and disease hypothesis propose that the early fetal environment programs metabolism of the individual, organ growth and functional development, thus affecting physiological functions at various stages of the life cycle.[10,11] This could explain why populations in developing countries like India and China, which are undergoing rapid nutrition transition, are also showing an increased prevalence of NCD including cardiovascular disease and diabetes.[10]

Infants born with intrauterine growth retardation (IUGR) are characterized by fetal hypoglycemia, possibly to maintain the maternal/fetal glucose concentration gradient and transport of glucose across the placenta to the fetus. In turn, this hypoglycemia limits insulin secretion, initiating fetal glucose production, but subsequently results in increased protein breakdown and decreased protein accretion, limiting the growth of lean body mass.[12] There is some evidence that Indian infants are born of low weight and low ponderal index but are of higher adiposity compared to Caucasian babies, the so-called “thin-fat” Indian infant, and that this continues further even later in life.[13] This phenomenon has not been shown consistently in other developing countries,[14] but if it is so, it indicates that the propensity to accrete fat and become overweight is high in such babies.

Although the early programming of metabolism and the propensity for weight gain are relevant to gaining excess weight gain after birth, it will also certainly depend on a number of factors that operate after birth. Immediately after birth, breastfeeding may protect a child from later weight gain and obesity, since it is possible that self-regulation of milk intake may be better in breast-fed than in bottle-fed infants. As much as this may seem intuitive, however, along with reports that exclusive breastfeeding causes a 20% reduction in obesity compared to exclusive formula feeding,[15] this effect has not been confirmed in randomized, controlled trials.[16]

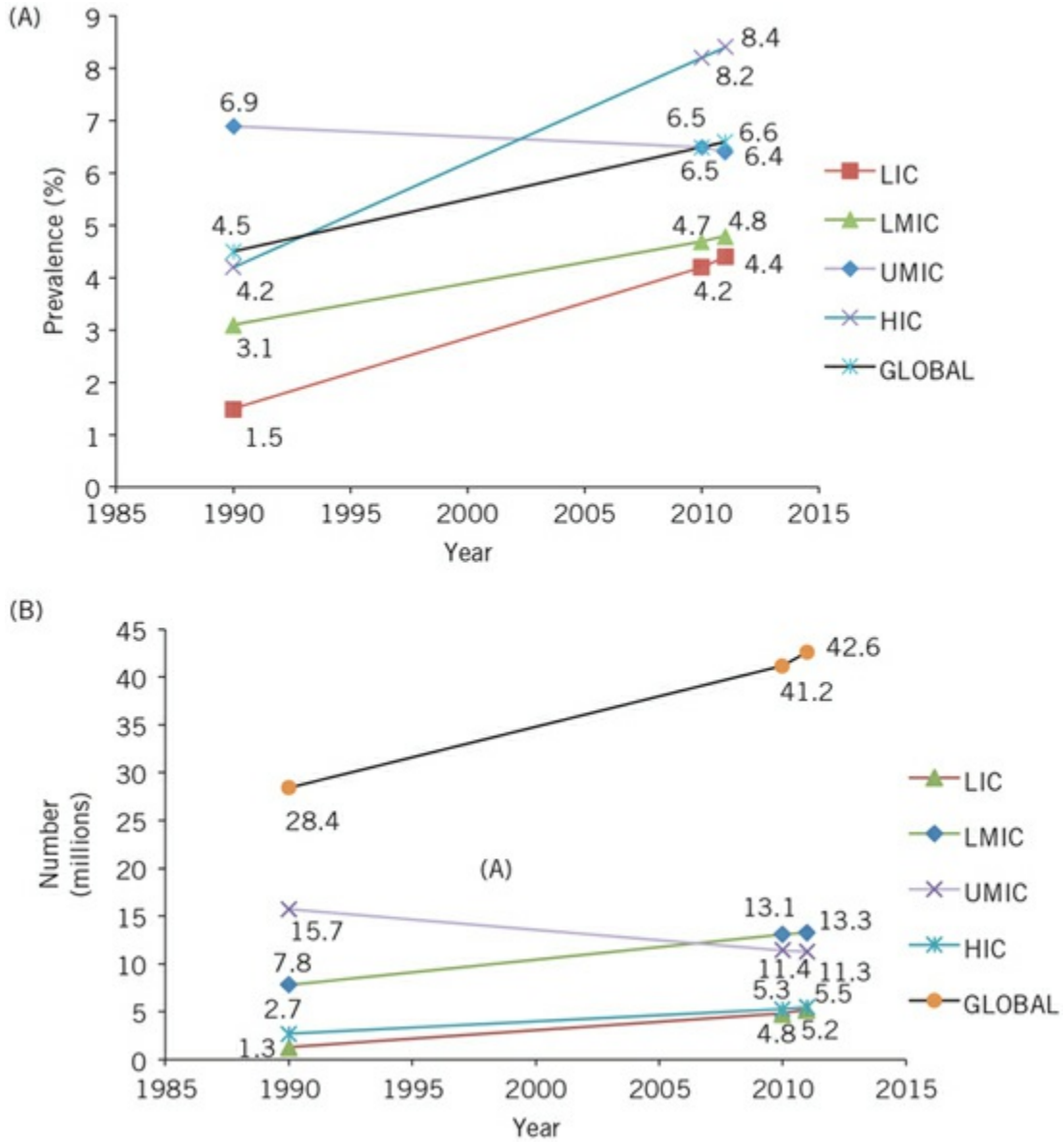


Figure 45-1 (A) Prevalence estimates of overweight and obesity in children below 5 years of age by countries classified based on world bank income group (years 1990, 2010, 2011) (B) Absolute numbers of children below 5 years of age with overweight and obesity by countries classified based on world bank income group (years 1990, 2010, 2011) LIC, Low-income countries; LMIC, Low middle-income countries; UMIC, Upper middle-income countries; HIC, High-income countries. (Data from UNICEF-WHO-The World Bank, 2012.[1])

Markets are also relevant, since mothers who want the best for their baby will be influenced by their peers, and may even get their education about a child's needs from the labels on complementary food packaging. The latest and more accurate Food and Agricultural Organization/World Health Organization/United Nations University consultation[17] on energy requirements has reduced the energy requirements of infants from 104 to 80 kcal/kg/d. On the basis of this definition, an analysis of the amount of complementary feed required (in commercial forms) would literally mean about one less feed if the new norm were followed. However, a recent study in India found that complementary feed packs were still being labeled to meet the older and higher energy requirements.[18] This problem would be trivial if women did not pass on feeding information from labels in their own peer networks, or if health education efforts were up to date and effective, or if women fed their children with foods prepared at home. Social conditions in the current world are such that that middle-class working women in developing countries look toward convenience and time-saving, and as a result, commercial complementary feeds are used more often than not in middle-class households, where the problem of childhood obesity is at its most relevant.

Rapid Catch-Up Growth and Adiposity Rebound

Growth in infancy up to 1 year and during childhood above 2 years of age have been found to be critical periods related to adult body composition, where rapid weight gain in infancy has been associated with higher lean mass in adulthood, while rapid weight gain above 2 years of age with higher fat mass.[11] A study of 1400 adults in New Delhi, India,[19] indicated that children with early adiposity rebound of 5 years or younger had the highest body mass index (BMI) in later childhood and this persisted into adulthood. Aggressive nutrition interventions, which are operational in many developing countries, that target undernourished children in early

infancy to ensure rapid catch up growth, could lead to adiposity and consequently to metabolic syndrome and other chronic diseases both in childhood and adulthood.[20] Relevant here are cultural norms and health education, given the concern of anxious mothers that their child's weight is an index of their health, and that the more their baby weighs, the healthier they are.

Stunting

In general, obesity is related to prosperity, but the relationship between stunting and obesity may be particularly operative in the context of developing economies, where children who were stunted during their earlier life are exposed to energy-dense foods and lower activity levels later on. An analysis of national surveys of Russia, Brazil, South Africa, and China showed that stunting was associated with being overweight among children[21]; similar findings were observed in young Mexican children.[22] However, this association is not uniform, for example, in Indonesia, stunting and lower socioeconomic status were more related to being underweight rather than overweight,[23] whereas in Guatemala, long-term improvements in height were not associated with equivalent increases in BMI.[24] In a Brazilian cohort of boys, stunting at 2 and 4 years of age was not associated with lean mass, or the ratio of fat to lean mass, during adolescence,[25] contrary to what has been reported in cross-sectional studies. Differences in economic development and access to energy-dense foods will confound the relation between stunting and overweight. In addition, it is important that this relationship is examined in longitudinal cohorts rather than a series of cross-sectional studies, as are available in many countries. Small errors in the measurement of height can also confound these analyses.[26] One potential mechanism that has been proposed for this putative relationship is that childhood nutritional stunting is associated with impaired fat oxidation, resulting in increased body fat in such children, and which is a factor that predicted obesity in other at-risk populations.[27,28] This phenomenon is still worth considering as economies develop, when the promotion of optimal growth of children is as important as preventing obesity.

DRIVERS OF CHILDHOOD OVERWEIGHT AND OBESITY

With rapid increases in the prevalence of overweight and obesity over the

past two to three decades, it is obvious that apart from genetics, the environment in which an individual lives must be contributing significantly to this rise, largely due to an imbalance between energy intake and expenditure. To maintain weight, a balance between energy intake and energy expenditure is necessary; a positive energy balance promotes weight gain. Energy intake and expenditure are very variable on a day to day basis. Although energy intake could vary in any of the proximal constituents of the diet, the most variable part of energy expenditure is physical activity. As a result, body fat and muscle mass are the two changeable or plastic elements of the body composition that are affected: excess energy is stored by the expansion of body fat, while muscle mass may be lost as physical activity is reduced.

An increase in energy intake and reduction in physical activity presumably increases overweight and obesity in children, although energy balance is determined by complex biological, behavioral, cultural, social, and environmental factors.[8] The problem is that measuring energy intake or expenditure with any degree of accuracy is difficult. This becomes relevant when one considers that weight gain is not acute, but occurs over a long period of time, in which a positive energy balance as small as 50–100 kcal/d may lead to significant gains in weight. Measuring energy intake or expenditure with our current tools to detect such a small imbalance is virtually impossible in the free world. Further, physiological adaptations in energy expenditure occur when energy intake is different from the actual required energy expenditure. For example, if energy balance is positive, energy expenditure may also rise over time through increments in muscle mass, with a new energy equilibrium achieved. Similarly, when energy balance is negative, adaptations to decrease energy expenditure are activated (e.g., a lowered basal metabolic rate), such that the energy balance is once again achieved.

Dietary Intake

At the individual level, overweight/obesity is due to a positive energy balance, with more calories consumed than what are expended daily. This is influenced by personal choices in everyday life, such as dietary habits and physical activity, and in turn, societal drivers such as urbanization, industrialization, and affluence shape these individual choices. For example, decreased occupational and environmental physical activity requirements have presumably led to a fall in daily energy expenditures. At the same time, more food is being produced than ever, and is available for human consumption.

With the growth of the food-processing industry, this abundance has translated into increased availability and access to energy-dense foods, even in developing nations. Rapid globalization has resulted in changes in patterns of food intake with the availability of refined, high-fat (particularly saturated fats) and high-sugar content foods being more easily available in most of the developing countries of the world.[4] In a study among 6- to 16-year-old children in India, a higher frequency of intake of fried foods was associated with a threefold greater risk of being overweight.[29] In urban India, and this is reflective of developing countries in general, the frequency of fast food, snacks, and bakery food consumption is rapidly increasing due to the attractiveness of fast food, large portion sizes, and easy access. Behaviors at home are also important, because negative behaviors such as eating while watching TV, snacking between meals, and regular dining out are related to overweight,[29] while some studies have shown a protective effect when family meals are eaten without simultaneously watching TV.[30] TV viewing is both a sedentary behavior and a source of exposure to food advertising for children.[31] Detailed data on diets of children in developing economies have not been reported yet. In general, however, the consumption of milk and meat products in the developing world has escalated.[3] This benefits infants and children in the first few years of life as the quality and quantity of protein as well as mineral and vitamin intakes improve, but higher intakes could lead to increase in saturated fatty acid and cholesterol intakes. This trend is now visible in developing countries.[3]

The problem with measuring food intake across populations is that surveys at scale are by necessity fairly broad and simple and are usually based on reported quantities of use and expenditure at the household level, which will not reflect actual consumption, but also will make it impossible to estimate intakes for different age groups in a population. Further, there is poor characterization of processed foods and food consumed outside the home. Even so, at least in India where obesity rates in children are showing an increase, surveys of food intake have actually documented relatively little change in the daily energy intake, but also a gradual decrease in the daily consumption of cereals and pulses, indicating substitution with other sources of energy such as fats.[32] With economic progress, such changes in dietary patterns have only been accentuated. The point is that energy intakes do not seem to mirror the gradual progression in overweight rates; while this may reflect the inability of surveys to accurately account for the increasing contribution of processed foods and

food consumed outside the home toward the daily energy intake, the problem may be due to positive energy balances emerging from falling energy requirements in an increasingly sedentary lifestyle.

Other important drivers are economics and agricultural policy. A balanced diet includes healthy proportions of fruits and vegetables, as well as low amounts of dietary fat. When one considers the intake of fruits and vegetables, however, available figures may be misleading. In India, for example, surveys indicate that dietary vegetable intake has increased by about 50% over the last two decades,[33] but in reality, this increase is mainly in the intake of potatoes and onions, whose nutritive value is relatively poor, especially with regard to micronutrients. In addition, vegetables and fruits are not stored effectively in developing economies, and therefore their prices are subject to large fluctuations depending on production and demand. As a consequence, the food consumption patterns of many poor homes are monotonous because they are simply reflective of these fluctuations, which limit the ability to have a varied dietary intake. On the other hand, there has been a sharp contrast in the pattern of intake of oils and fats: again, giving India as an example, there has been a 25% and 67% increase in consumption of oils and fats in rural and urban areas, respectively.[33] Other obesogenic trends include an increasing consumption of processed foods, snacks, fast food, and sweetened beverages, and a decrease in the number of meals eaten at home.[29]

What could be driving these dietary consumption patterns? Some factors can be deduced from agricultural policy: cash crops account for the greatest increases in area under cultivation. The production and yield among all food groups over the past 50 years in India has been for sugarcane and oil seeds.[34] Another important macroeconomic influence on consumption patterns is food price stability; this is variable across countries and depends on seasonal patterns, successful agriculture initiatives, and other factors.

Physical Activity

Regular physical activity is a metabolic need and influences the growth and development of children and adolescents. Over the past three decades, the use of technology in the activities of daily life has increased and consequently daily activity levels of children have also decreased. Spontaneous physical activity and the use of active transportation have declined with increased urbanization. Built environments in cities do not promote active living. With many parts of the developing world

undergoing rapid urbanization where over half of the world's urban population lives in Asia,[35] it is evident that the physical activity of children is affected.

The global school-based health survey (GSHS) conducted across 55 countries in the world indicates that in most of the developing countries approximately 20% of children 13–15 years of age are active while 30%–40% of children are inactive (that is, spending more than 3 h/d sitting, watching television, playing computer games, or doing other sitting activities).[36] Globally, about 80% of 13- to 15-year olds do not engage in the recommended level of ≥60 minutes of moderate to vigorous physical activity per day. This proportion was seen in over 56 of 105 countries in boys and 100 out of 105 countries for girls.[37]

Increased sedentary activity and increased sleep duration (within limits) have opposite effects on childhood weight status. The effect of sleep debt may probably operate through both direct metabolic effects and indirect pathways by disrupting hormone signaling pathways notably the activation of growth hormone-releasing hormone and inhibition of corticotrophin-releasing hormone which in turn mediates the release of pituitary-derived and dependent hormones such as human growth hormone and cortisol.[38] Short sleep duration is also thought to act through the modulation of leptin and ghrelin.[39] The passive viewing of screens, whether television or computer, can also have strong effects on the risk for overweight and obesity. In a study on urban and semi-urban children in India, increased television viewing of greater than or equal to 1.5 h/d was significantly associated with a risk for overweight compared to children viewing for ≤45 min/d.[40] This increased risk of obesity with a doubling of viewing time in effect means that something more than simply sitting in front of a screen is operational; in this context, behaviors such as eating and watching the screen at the same time can lead to passive overconsumption of energy. In addition to these behaviors, parental BMI, especially maternal BMI, is a known risk factor for childhood obesity,[41] and is likely to reflect the clustering of risk behaviors in families and genetic factors that affect obesity.

School-age children with a marginal to poor nutritional status have a lower total daily energy expenditure and lower energy expended through physical activity. This leads to poor physical performance mainly due to the small body size and lower muscle mass.[36] The lower daily physical activity is likely to happen in school as well as out of school. A recent longitudinal study among adolescent urban school children in India noted a

significant decline in moderate to vigorous physical activity within the course of a single year.[42] This may be because schools simply do not put much emphasis on physical activity and fitness, or do not have the means to do so in poor countries. Equally, there is also the need for excellence in academically competitive environments; this leads to children being made to choose extracurricular academic programs rather than sport in their extracurricular time.

Physical and social features of the urban environment are also known to influence physical activity levels. Actual access to open spaces or parks for leisure-time physical activity is poor in most developing countries, leading to poor walkability. Unplanned urban development, daily access to workplaces, schools and recreational facilities have become increasingly time consuming and dependent on motorized transport. As a case study, in India, while the population of six major metropolitan cities increased 1.9 times between 1981 and 2001, the number of vehicles increased by 7.8 times.[43]

Urbanization and Migration

In developing economies, the urban–rural divide in terms of overweight prevalence is often striking. Urbanization is associated with changes in dietary intake and energy expenditure patterns due to factors that have been explained above. However, increasing childhood obesity is also related to the relocation of populations for more prosperous livelihoods and the drivers of such migration. When there are no serious attempts by governments to increase agriculture and rural economies, concurrent with a push for urbanization, it is obvious that people will migrate from villages in search of better livelihoods offered by the services and industry sectors, which are concentrated in towns and cities. In many countries, this has led to a growing prosperity, and emergence of more middle- and high-income households, and an analysis of survey data in India[44] has shown how the BMI distribution of rural–urban migrants can shift toward the right as their duration of urban living increases (Figure 45-2). Migration is therefore a significant trend of population shift, but without planning, can lead to serious urban poverty as well.

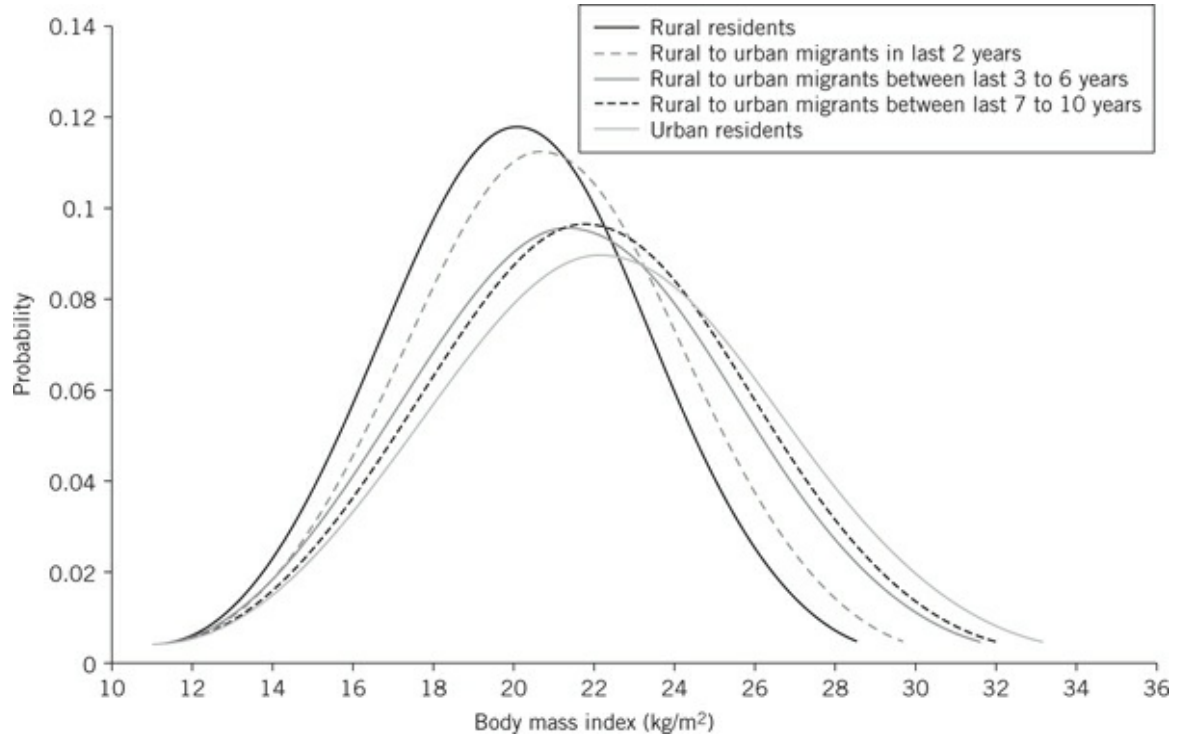


Figure 45-2 Probability distribution of BMI in rural–urban migrant and nonmigrant Indians. (From Kurpad Av, Varadharajan KS, Aeberli I.[45]) Distribution of BMI in populations migrating from rural to urban areas, stratified based on the number of years since migration. There is a continuous right-shift over time, with the BMI of migrants approaching the urban resident (nonmigrant) BMI.

Overweight/obesity rates are higher among those belonging to these higher socioeconomic groups. A recent analysis of nationally representative data from 38 low- and middle-income countries has indicated that higher risk of obesity in urban areas is driven by higher individual- and community level socioeconomic status in urban areas rather than urban living alone.[45] Even so, it is important to point out that these risk patterns are in concurrence with the trends that were observed in developed economies just two decades earlier. In this scenario, risks in the developed nations are believed to have reversed, with the greatest burden among rural residents and disadvantaged groups[46–49] and this shift is estimated to occur in developing countries when the per capita gross national income exceeds US$ 2500.[50] Brazil, an emerging economy, where obesity has spread rapidly among lower economic groups along with economic progress in the last decade, presents a case in point.[51]

POSSIBLE SOLUTIONS AND INTERVENTIONS

Earlier Country Experiences

Many countries have put public health interventions in place to address overweight and obesity; experiences from Chile and Singapore can yield lessons for developing economies. Chile is an example of a country with high levels of child undernutrition in the 1960s, which attempted to eradicate hunger but, inadvertently caused a rise in overweight/obesity. Singapore is an example of a prosperous country, which put in place strategies to combat overweight/obesity.

Chile underwent rapid modernization and economic growth in the early 1990s. Although great progress was made in public health and the prevalence of undernutrition, there was a negative impact with sedentary lifestyles and changes in dietary patterns. Growing incomes in the lower income groups were associated with corresponding increases in purchase of high-fat/high-carbohydrate, energy-dense foods.[52] Undernutrition among children below 6 years in Chile fell from 37% in 1960 to 2.9% in 2000, owing chiefly to the National Complementary feeding programme (PNAC), which focused on poor urban[53] (see Chapter 15 for details). This program delivered basic food supplements and rice to benefit all family members, to prevent redirection of food meant for infants. High coverage and well-established monitoring systems ensured that undernutrition was brought down to negligible levels by 1990, but distribution of food supplements to beneficiary households continued.[53,54] A small but nonsignificant gain in length for age of children was noted in a program evaluation in 1990, but weight for age and weight for length had increased significantly.[53] Studies based on accurate estimations of energy expenditure concluded that an excess of 200 kcal/d of energy was being supplied per child.[55] This could explain the twofold increase in overweight and obesity among children entering school between 1987 and 2000 (Figure 45-3).[55] Feeding mildly stunted children is thought to have contributed to overweight and obesity; however, the target-weight gain criterion, while being sensitive enough, was not specific for determining the need for food supplementation. The lesson to be learnt is that providing additional energy to all, through universal feeding programs, may promote obesity. Therefore, targeted approaches, though not easy, may be more important if obesity is to be prevented.

Singapore launched its National Healthy Lifestyle Program in 1992 to spearhead national strategies based on public education campaigns that focused on physical activity and healthy eating.[56] Official dietary and

physical activity guidelines were formulated, and health promotion grants and awards were instituted to sustain efforts in schools and workplaces. Media campaigns and events were used to raise awareness in the general public.[57,58] Programs also encouraged physical activity in all students, and targeted weight loss in overweight children through mandatory participation in intense exercises and food ration coupons that limited calories consumed during recess breaks. The consequent drop in the prevalence of overweight from 11.7% to 9.5% between 1993 and 2006 was partly attributed to this,[59] However, following reports of stigmatization, bullying and increase in eating disorders attributed to this program, the Holistic Health Framework focusing on general well-being of students was adopted in 2007. The Model School Tuck-shop Program in 2003 provided schools with healthier food service guidelines and placed restrictions on access to energy-dense foods, while similar initiatives were introduced at workplaces and for the general population.[57] The impact of such strategies is hard to ascertain, however, overweight/obesity rates seem to have stabilized or are falling in children and youth.[57,58]

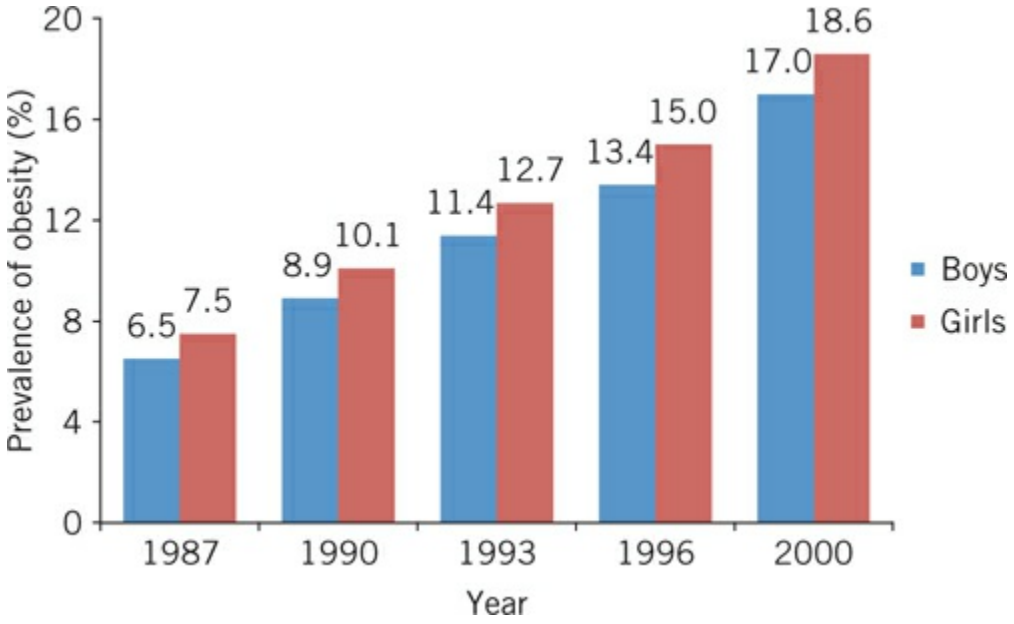


Figure 45-3 Prevalence of obesity in first-grade school children in Chile 1987–2000. (Adapted from O'Leary.[53])

Other countries intervening in the obesity epidemic also initially prioritized school-based strategies. Some of these strategies and their

limitations are outlined in Table 45-2. Coordinated national programs, such as “Agita São Paulo” in Brazil, “Vuka” in South Africa and “Let’s Move” in the United States, using a mix of strategies, are examples. Partnerships with the food and beverage industry are growing, in such areas as product reformulation (low salt, low saturated fats), portion size reduction, healthier menu alternatives, limiting targeted advertising, and nutritional labeling, along with initiatives in urban design and transport planning. Different countries may need different solutions; for adults, a modeling study in 2010 focused on interventions to promote healthy diets and physical activity in the BRIC nations found that the most cost-effective strategy in chronic diseases control was education and counseling by health workers.[59] Interventions targeting children were shown become cost-effective only in the long run as would mass-media campaigns. In the absence of locally relevant evidence, a broad-based strategy aimed at chronic diseases control, focusing on lifestyle changes should be the most sensible option.

In conclusion, with developing economies undergoing rapid increases in urbanization and changes in lifestyle, there is an increase in the prevalence of childhood overweight and obesity. This could track into adulthood and escalate the existing high rate of noncommunicable diseases even further in such populations. In turn, this would exert more strain on their economies, particularly because most of these countries already struggle with a high burden of undernutrition through the life cycle. Within countries, increasing weight is socially and culturally linked to prosperity, because the prevalence of overweight and obesity is higher among the higher socioeconomic groups rather than the lower socioeconomic groups, unlike in developed countries. Obesity prevention programs therefore need to be nuanced, based on socially and culturally accepted models of creating awareness, and targeted at educating individuals, communities, and populations.

TABLE 45-2 School-Based Strategies for Addressing Overweight and Obesity

	Strategy	Components of Strategy	Efficacy and Impact on Obesity/Overweight	Hindering Factors
1	Health education	Dietary and physical activity education	i. Consistent, but small positive effects on behavior	i. Lack of trained personnel in schools

			ii.	Evidence on change in BMI is limited, inconsistent	ii. Already crowded academic environment
2	Regulation of food environment in schools	i. Setting nutrition guidelines ii. Pricing strategies iii. Regulation of food and drink availability	i.	Consistent decreases in availability and consumption of high sugar/fat food in schools	i. Lunch meal scheme function poorly
			ii.	Increased availability of fruits	ii. Regulation of sale around schools will be difficult as trade is unorganized
			iii.	Evidence on change in BMI limited	
3	Regulation of physical activity environment in schools	i. Increase in frequency and duration of physical education classes ii. Development of physical activity infrastructure iii. Footpaths and bicycle ways in school vicinity	i.	Significant increases in time and intensity of physical activity, energy expenditure and aerobic capacity	i. Many schools lack playgrounds ii. Many urban schools exist in crowded environments and development of play grounds not possible iii. Lack of trained physical instructors
			ii.	Evidence on change in BMI limited, small and inconsistent	

REFERENCES

1. United Nations Children's Fund, the World Health Organization and the World Bank. Levels and trends in childhood malnutrition: UNICEF-WHO-The World Bank joint child malnutrition estimates; 2012.
2. de Onis M, Blossner M. Prevalence and trends of overweight among preschool children in developing countries. *Am J Clin Nutr*. 2000;72:1032–1039.
3. Schmidhuber J, Shetty P. The nutrition transition to 2030. Why developing countries are likely to bear the major burden. *Acta Agriculturae Scand Section C*. 2005;2:150–166.
4. Galal OM, Hulett J. Obesity among schoolchildren in developing countries. *Food Nutr Bull*. 2005;26:S261-S266.
5. de Onis M, Blössner M, Borghi E. Global prevalence and trends of overweight and obesity among preschool children. *Am J Clin Nutr*. 2010;92:1257–1264.
6. World Health Organization. WHO child growth standards: length/height-for-age, weight-for-age, weight-for-length, weight-for height and body mass index-for-age: methods and development. WHO; 2006. pp. 1–312.

7. Gupta N, Goel K, Shah P, Misra A. Childhood obesity in developing countries: Epidemiology, determinants, and prevention. *Endocr Rev.* 2012;33:48–70.
8. Wang Y, Lobstein T. Worldwide trends in childhood overweight and obesity. *Int J Ped Obes.* 2006;1:11–25.
9. UNICEF. Low birthweight. http://www.unicef.org/specialsession/about/sgreport-pdf/15_LowBirthweight_D7341Insert_English.pdf. Accessed March 4, 2015.
10. Gluckman PD, Hanson MA, Beedle AS. Early life events and their consequences for later disease: a life history and evolutionary perspective. *Am J Hum Biol.* 2007;19:1–19.
11. Uauy R, Kain J, Mericq V, et al. Nutrition, child growth, and chronic disease prevention. *Ann Med*. 2008;40:11–20.
12. Hediger ML, Overpeck MD, Kuczmarski RJ, et al. Muscularity and fatness of infants and young children born small- or large-for-gestational-age. *Pediatrics.* 1998;102:E60.
13. Yajnik CS. Obesity epidemic in India: intra-uterine origins? *Proc Nutr Soc.* 2004;63:387–396.
14. Andersen GS, Girma T, Wells JC, et al. Fat and fat-free mass at birth: air displacement plethysmography measurements on 350 Ethiopian newborns. *Pediatr Res*. 2011;70:501–506.
15. Singhal A, Lanigan J. Breastfeeding early growth and later obesity. *Obes Rev.* 2007;8:51–54.
16. Martin RM, Patel R, Kramer MS, et al. Effects of promoting longer-term and exclusive breastfeeding on adiposity and insulin-like growth factor-I at age 11.5 years: a randomized trial. *JAMA.* 2013;309:1005–1013.
17. FAO/WHO/UNU. Human energy requirements. Report of a Joint FAO/WHO/UNU Expert Consultation. Rome, Italy; 2004.
18. Kuriyan R, Kurpad AV. Complementary feeding patterns in India. *Nutr Metab Cardiovasc Dis*. 2012;22:799–805.
19. Bhargava SK, Sachdev HS, Fall CHD, et al. Relation of serial changes in childhood body mass index to impaired glucose tolerance in young adulthood. *N Engl J Med*. 2004;350:865–875.
20. Misra A, Khurana L. Obesity and the metabolic syndrome in developing countries. *J Clin Endocrinol Metab.* 2008;93:S9–30.
21. Popkin BM, Richards MK, Monteiro CA. Stunting is associated with overweight in children of four nations that are undergoing the nutrition transition. *J Nutr.* 1998;126:3009–3016.
22. Fernald LC, Neufeld LM. Overweight with concurrent stunting in very young children from rural Mexico: prevalence and associated factors. *Eur J Clin Nutr.* 2007;61:623–632.
23. Julia M, van Weissenbruch MM, de Waal HA, Surjono A. Influence of socioeconomic status on the prevalence of stunted growth and obesity in prepubertal Indonesian children. *Food Nutr Bull.* 2004;25:354–360.
24. Stein AD, Wang M, Digirolamo A, et al. Height for age increased while body mass index for age remained stable between 1968 and 2007 among Guatemalan children. *J Nutr*. 2009;139:365–369.
25. Gigante DP, Victora CG, Horta BL, Lima RC. Undernutrition in early life and body composition of adolescent males from a birth cohort study. *Br J Nutr.* 2007;97:949–954.
26. Timæus IM. Stunting and obesity in childhood: a reassessment using longitudinal data from South Africa. *Int J Epidemiol*. 2012;41:764–772.
27. Hoffman DJ, Sawaya AL, Verreschi I, et al. Why are nutritionally stunted children

at increased risk of obesity? Studies of metabolic rate and fat oxidation in shantytown children from São Paulo, Brazil. *Am J Clin Nutr.* 2000;72:702–707.
28. Clemente AP, Santos CD, Martins VJ, et al. Mild stunting is associated with higher body fat: study of a low-income population. *J Pediatr (Rio J).* 2011;87:138–144.
29. Kuriyan R, Thomas T, Sumithra S, et al. Potential factors related to waist circumference in urban South Indian children. *Indian Pediatr*. 2012;49:124–128.
30. Yuasa K, Sei M, Takeda E, et al. Effects of lifestyle habits and eating meals together with the family on the prevalence of obesity among school children in Tokushima, Japan: a cross-sectional questionnaire-based survey. *J Med Invest*. 2008;55:71–77.
31. Roblin L. Childhood obesity: food, nutrient, and eating-habit trends and influences. *Appl Physiol Nutr Metab.* 2007;32:635–645.
32. Shetty PS. Nutrition transition in India. *Pub Health Nutr*. 2002;5: 175–182.
33. National Sample Survey Office. Level and Pattern of Consumer Expenditure: NSS 66th Round, 2009–10. Ministry of Statistics and Programme Implementation, Government of India.
34. Agricultural Statistics At a Glance 2012. Directorate of Economics and Statistics, Ministry of Agriculture, Government of India.
35. United Nations Children's Fund (UNICEF). The State of the World's Children 2012: Children in an Urban World. New York, NY: UNICEF; 2012
36. Malina RM, Katzmarzyk PT. Physical activity and fitness in an international growth standard for preadolescent and adolescent children. *Food Nutr Bull.* 2006;27:S295-S313.
37. Hallal PC, Andersen LB, Bull FC, et al. Global physical activity levels: Surveillance progress, pitfalls and prospects. *Lancet.* 2012;380:247–257.
38. Must A, Parisi SM. Sedentary behaviour and sleep: paradoxical effects in association with childhood obesity. *Int J Obes.* 2009;33:S82-S86.
39. Zimberg IZ, Dâmaso A, Del Re M, et al. Short sleep duration and obesity: mechanisms and future perspectives. *Cell Biochem Funct.* 2012;30:524–529.
40. Kuriyan R, Bhat S, Thomas T, et al. Television viewing and sleep are associated with overweight among urban and semi-urban South Indian children. *Nutr J*. 2007;6:25.
41. Agras WS, Hammer LD, McNicholas F, Kraemer HC. Risk factors for childhood overweight: a prospective study from birth to 9.5 years. *J Pediatr*, 2004;145:20–25.
42. Swaminathan S, Selvam S, Thomas T, et al. Longitudinal trends in physical activity patterns in selected urban south Indian school children. *Indian J Med Res.* 2011;134:174–180.
43. National urban Transport Policy, 2006. Ministry of Urban Development, Government of India.
44. Kurpad AV, Varadharajan KS, Aeberli I. The thin-fat phenotype and global metabolic disease risk. *Curr Opin Clin Nutr Metab Care*. 2011; 14:542–547.
45. Neuman M, Kawachi I, Gortmaker S, Subramanian SV. Urban-rural differences in BMI in low- and middle-income countries: the role of socioeconomic status. *Am J Clin Nutr.* 2013;97:428–436.
46. Sobal J, Stunkard AJ. Socioeconomic status and obesity: a review of the literature. *Psychol Bull*. 1989;105:260–275.
47. McLaren L. Socioeconomic status and obesity. *Epidemiol Rev*. 2007; 29:29–48.
48. Befort CA, Nazir N, Perri MG. Prevalence of obesity among adults from rural and urban areas of the United States: findings from NHANES (2005–2008). *J Rural*

Health. 2012;28:392–397.
49. Ogden CL, Carroll MD, Curtin LR, et al. Prevalence of overweight and obesity in the United States, 1999–2004. *JAMA.* 2006;295:1549–1555.
50. Monteiro CA, Moura EC, Conde WL, Popkin BM. Socioeconomic status and obesity in adult populations of developing countries: a review. *Bull World Health Organ.* 2004;82:940–946.
51. Monteiro CA, Conde WL, Popkin BM. Income-specific trends in obesity in Brazil: 1975–2003. *Am J Public Health.* 2007;97:1808–1812.
52. Albala C, Vio F. Epidemiological transition in Latin America: the case of Chile. *Public Health*, 1995;109:431–432.
53. O'Leary R. The lessons of Chile's nutrition transition. http://www.worldfoodprize.org/documents/filelibrary/images/youth_programs/rese Accessed March 5, 2015.
54. Towards eradication of child undernutrition in Latin America and the Caribbean. Edition by the United Nations World Food Programme. Regional office for Latin America and the Caribbean; 2008.
55. Uauy R, Kain J. The epidemiological transition: need to incorporate obesity prevention into nutritional programmes. *Public Health Nutr*. 2002;5:223–229.
56. GL Gan, Pang J. Obesity in Singapore: Prevention and control. *Obes Prevent Manage*. 2012;38:8–13.
57. Soon G, Koh YH, Wong ML, Lam PW. Obesity prevention and control efforts in Singapore a 2008 case study. The National Bureau of Asian Research, Washington DC. 2008.
58. Toh CM, Cutter J, Chew SK. School based intervention has reduced obesity in Singapore. *BMJ*. 2002;16:324–427.
59. Cecchini M, Sassi F, Lauer JA, et al. Tackling of unhealthy diets, physical inactivity, and obesity: health effects and cost-effectiveness. *Lancet.* 2010;376:1775–1784.

Feeding Difficulties

46

Peter B. Sullivan, MA, MD, FRCP, FRCPCH

FEEDING, NUTRITION, AND GROWTH

Feeding is essential for survival. Children who cannot or do not eat properly may become unwell and fail to grow.[1] Thus, when feeding becomes problematic it is a source of great concern and anxiety for parents. Such concerns include the amount of food consumed, the rate of weight gain, possetting, restricted food preferences, delay in self-feeding, unacceptable mealtime behaviors, and peculiar food habits.[2]

In addition to its nutritional imperative, the process of feeding is important as a formative influence on the infant's emotional and social development. A major focus of the interaction between an infant and its parents surrounds feeding and thus early feeding experience is important to the psychological development of the child. Feeding is influenced by a wide range of factors including cultural background, parental experience and temperament, and the child's developmental status. This fundamental aspect of life is reflected in the interconnectedness of the words "nutrition," "nourish," "nurture," and "nurse." Feeding thus plays a central role in the life of the child not only in relation to their growth but also in its contribution to their social integration.

Problems with feeding can cause a major disruption to normal growth and development. In addition, they can pose additional complications for the child coping with a chronic disease. To protect the child from the adverse effects of these complications requires great patience and skill on the part of parents and clinicians alike. This chapter aims to describe the development of normal feeding skills and outline the etiology of common feeding problems together with an account of their assessment and management.

Epidemiology

Estimates suggest that up to 25% of infants and young children have some degree of feeding problem[3] although there is no hard epidemiological data on prevalence. Severe feeding problems associated with poor weight gain, such as refusal to eat or vomiting, have been reported to occur in 1%–2% of infants younger than 1 year of age,[4] and 70% of these infants continue to have feeding problems four and six years later.[5,6] Feeding disorders also have been linked to later deficits in cognitive development, behavioral problems, and eating disorders.

There has been remarkably little research on the development of infant feeding problems. Seminal studies by Dahl and colleagues in Sweden on 50 infants meeting strict diagnostic criteria, however, have helped to identify the characteristics and consequences of infant feeding problems.[4–7] Feeding problems in this group had been ongoing for a mean duration of five months. The commonest problems were refusal to eat (56%), colic (18%), and vomiting (16%). The majority (86%) of infants was underweight and 14% were malnourished (>2 SD below mean weight for age). Physical disorder (e.g., gastroesophageal reflux) was present in only 14% but in 6% this was a serious organic disease (e.g., congenital heart disease). Follow up studies at 2 years showed that over a third of the refusal to eat group had a persistent feeding problem.

The Physiology of Feeding

Understanding feeding problems in children rests on a detailed knowledge of oral-motor structure and function, the neurophysiology of feeding as well as child development and behavior. The process of taking in and swallowing food and coordinating this with breathing depends on three linked anatomical and physiological regions: the oral cavity (lips, tongue, and palate), the pharynx and the esophagus.

In a normal co-ordinated nutritive sucking cycle, breathing is intercalated between swallows. In a perfectly co-ordinated cycle suck, swallow and breathing movements are related in a 1 to 1 sequential pattern.[8] These form a rhythmically repetitive unit in which breathing appears to be continuous and uninterrupted. In less well co-ordinated feeding, breathing is always interrupted by swallowing. The co-ordination of the intrinsic muscles of the tongue to produce fully formed rhythmic, rolling movements seems to be in part a productive gestational maturity as premature babies have more poorly co-ordinated tongue movements. The linkage of sucking and swallowing with breathing appears to be achieved

consistently only by infants of more than 37 weeks postconceptional age.[8]

The development of oral-motor skills reflects general neurological maturation and requires the coordination of the movement of at least 31 pairs of striated muscles in the mouth, pharynx, and esophagus by 5 cranial nerves, the brain stem, and the cerebral cortex.[9]

The swallowing process occurs in four phases: oral preparatory, oral, pharyngeal, and esophageal. The first two phases are under voluntary control, with the pharyngeal phase partly voluntary and mostly involuntary, while the esophageal phase is entirely involuntary.

The *oral preparatory phase* involves the processing of food by the lips, teeth, cheeks, and tongue to form a bolus. Good lip seal, a coordinated range of tongue and jaw movements and normal oral sensation are all prerequisites for this phase to proceed without disruption.

In the *oral phase,* the food bolus is propelled towards the pharynx by elevation of the tongue with sequential contact with the hard and soft palates. When the bolus reaches the posterior oral cavity (e.g., soft palate and faucial pillars), the swallow reflex is activated. This sensory stimulus on the posterior oral structures ends the voluntary oral phase and triggers the involuntary pharyngeal phase of swallowing.

During the *pharyngeal phase*, the velum elevates, closing off the nasopharynx, and the pharyngeal constrictors propel the bolus through the faucial pillars to the upper esophageal sphincter (cricopharyngeus) in a peristaltic wave. Concomitantly, the larynx closes, elevates, and is pulled forward by the laryngeal strap muscles. The epiglottis and the aryepiglottic folds, the false vocal folds, and the true vocal folds form three separate levels of laryngeal closure, protecting the airway from penetration by food or liquid. Respiration stops briefly to allow the bolus to move to the relaxed upper esophageal sphincter. The cricopharyngeus muscle serves as the valve at the top of the esophagus. It remains in a fixed state of contraction and only relaxes to allow food or liquid to pass during a swallow.

The pharynx and larynx are richly endowed with chemoreceptors, baroreceptors, and temperature receptors whose purpose is to initiate and modify the swallow. The plethora of these sensitive receptors in the nasopharynx explains the aversive effects on normal swallowing of a noxious stimulus such as the presence of a nasogastric feeding tube.

The *esophageal phase* then begins and involves a continuation of peristaltic transport of the bolus to the stomach. Initiation of the swallowing reflex is often defective in neurologically impaired children and food can then accumulate in the vallecula or pyriform sinuses with

considerable risk of aspiration.

The Normal Development of Feeding Skills

Eating and swallowing are such a natural, subconscious act that it is easy to forget that eating is a learned skill. Unlike breathing, which is similar to feeding in that it is largely subconscious with some element of voluntary control, most of feeding is learned behavior. While the neonate feeding begins as a result of a range of reflexes (gag, phasic bite, tongue protrusion, rooting, suckling, and swallowing reflexes), it later becomes a voluntary act with only the pharyngeal and esophageal parts of the swallow remaining under reflex control. Development of sucking activity is both an inborn conditioned reflex dependent primarily on physiological maturation and it is capable of being strengthened or modified in accordance with learning experiences. Thus, the addition of non-nutritive sucking during gavage feeding, for instance accelerates the organization and efficiency of sucking pattern in premature infants. This has been shown to be associated with a more rapid weight gain [10] and change in gastrointestinal motility[11] during the postnatal period in these infants. Moreover, Morris and colleagues showed that the length of time premature infants needed to reach full enteral feeding was significantly associated with a poorer mental outcome.[12] Infants who reached full enteral feeding at an early age appeared to have a better development outcome despite their gestational age and the severity of respiratory complications.

During the first few months of life, patterns of feeding behavior become established. A typical normal infant will consume around 113 g of milk in 5 minutes and 230 g in 10–15 minutes. Transitional feeding from a predominantly sucking mode of feeding to mature solid food eating begins between 3 and 6 months of age. Growth in the upper digestive tract occurs as the mandible grows downwards and forwards and the hyoid bone and larynx move downwards. The sucking pads are gradually absorbed. All of these changes contribute to an enlargement of the buccal cavity, which allows food to be manipulated between the tongue and the buccal wall. The gradual eruption of teeth allows the infant to progress towards eating harder and lumpier foods. So growth and maturation play a role in feeding development, but learning from experience is also crucial. An important aspect of learning is sensation and sensory feedback. This involves proprioception, touch, pressure, temperature, and taste. Other important factors contributing to learning are gross and fine motor development, the methods of food presentation, and cognitive development. From three to

six months, the process of weaning starts and the early reflex mechanisms that subserve sucking are replaced by more volitional oral movements of the jaw and tongue. This is accompanied by the experience of an increasing range of food tastes and textures. By 6 months of age, an infant is capable of expressing hunger or satiation clearly and from this point is able to play an increasingly participatory role in mealtimes.

Infants and toddlers who take more than 30 minutes to feed are slow feeders and reports of behavioral feeding problems are common in toddlers and are related to slow feeding.[13] Reau demonstrated that toddlers with problematic feeding behaviors e.g., “not always hungry at mealtimes,” “does not always enjoy a feed,” or with strong food preferences at both 6 and 12 months eat more slowly than other toddlers. Thus, asking the mothers of infants and toddlers how long it takes the children to complete a meal should be an essential part of feeding evaluation. Further evaluation should be considered in those cases of reported meal length greater than 30 minutes, especially if lengthy feeding is persistent over time. Evaluation might include oral, motor, and psychosocial evaluation.

Weaning Failure

Weaning is the process whereby an infant becomes accustomed to an intake of solid food in preference to milk. This transitional phase may be associated with disorganization of feeding and infants are often seen to be agitated or unsettled at this stage. During this critical period, infants are vulnerable to develop feeding difficulties. Feeding is a reciprocal process that depends not only on the abilities and characteristics of the infant but also those of their parent. Avoidance by parents of encouraging the infant to make the transition from milk to solids at this time may make the process considerably more difficult at a later time. Similarly, prolonged nasogastric tube feeding or any other noxious stimulus in the oropharynx during this period may be associated with weaning failure.

Etiology of Feeding Disorders

Effective feeding depends on the normality of a wide range of anatomical, physiological, metabolic, and behavioral factors and thus the potential for disease processes to affect feeding is enormous. Table 46-1 itemizes some of the more common causes. It is important to recognize that chronic disease in any organ system in a child can be associated with poor feeding. Thus, anorexia is a consequence of chronic inflammatory diseases such as inflammatory bowel disease and of the metabolic derangement that

accompanies chronic renal failure. Dysphagia secondary to the esophagitis caused by gastroesophageal reflux is a common cause of poor feeding.[14] Suckle and swallow are complex co-ordinated motor actions, which are highly developed in the normal newborn. Because of the functional and anatomical interrelation of these actions with respiration, their impairment assumes great significance. The majority of symptoms and signs of severe dysphagia are respiratory in nature with nasal or laryngeal penetration. Bronchopulmonary dysplasia (BPD), a chronic lung disease of preterm babies with antecedent respiratory distress syndrome, ventilatory barotrauma, and oxygen toxicity is characterized by varying degrees of prolonged oxygen dependency. Infants with BPD can experience significant feeding difficulty, possibly secondary to tachypnea interfering with suck coordination.

TABLE 46-1 Etiology of Feeding Difficulties

Altered appetite
- Abnormal feeding patterns
- Supplemental feedings
- Metabolic disorders
- Inflammatory bowel disease

Diet
- Food allergy
- Inappropriate foods

Dysphagia
- Anatomic:
  - Macroglossia
  - Cleft lip and palate
  - Submucous cleft palate
  - Pierre Robin sequence
  - Laryngeal cleft
  - Tracheoesophageal fistula
  - Esophageal stricture
  - Retropharyngeal mass
  - Vascular ring
  - Foreign body
- Neuromuscular:
  - Prematurity
  - Cerebral palsy
  - Isolated neonatal swallowing dysfunction
  - Bulbar palsy
  - Rett syndrome
  - Infant botulism
  - Muscular dystrophy
  - Pseudo-obstruction
  - Connective tissue disease
  - Repaired tracheoesophageal fistula

Inflammatory:
Viral stomatitis
Candida stomatitis, pharyngitis
Peptic esophagitis (gastroesophageal reflux)
Crohn's Disease
Mucositis (graft-versus-host disease)
Systemic:
Cardiac disease
Pulmonary disease
Behavioral
Poor parent–infant interaction
Autonomy struggles
Picky eater
Delayed introduction of solids
Oral aversion
Conditioned dysphagia
Post-traumatic eating disorder
Vulnerable child
Parental responses

Source: Adapted from Rudolph & Link; Foy et al.; Illingworth.[44–46]

Rommel, characterized the etiology of feeding difficulties in 700 children with severe feeding difficulties and differentiated medical, oral, and behavioral categories.[15] Nearly half of the children had combined medical and oral conditions underlying their feeding difficulties. Oral sensory-based feeding problems appear to be related to past medical interventions. Children with feeding disorders had significant lower birth weight for gestational age and premature babies were over represented. In this study, 86% (603 of 700) of patients had a medical disorder, 61% (427 of 700) had oropharyngeal dysfunction, and 18.1% had a behavioral problem. Of medical diagnoses, a gastrointestinal problem was the most frequent and of these gastroesophageal reflux disease was the most frequent GI condition, diagnosed in 33% (228 of 700 of the total group). An oral feeding problem was diagnosed in 61% of patients and the majority of children with oral feeding problems were less than 2 years of age. The most frequent isolated oral feeding problem was related to sucking and was sensory based.

Oral sensory motor impairment may be present from birth and may often be unrecognized or mislabeled "nonorganic failure to thrive."[16] A common factor underlying such feeding difficulties may be neurophysiological abnormality manifesting as different degrees of oropharyngeal sensorimotor impairment.

Oral-Motor Impairment

Oral-motor impairment may occur as a result of structural lesions (e.g., cleft lip and palate, macroglossia, Pierre-Robin syndrome, esophageal atresia) or functional lesions (e.g., cerebral palsy, bulbar and pseudobulbar palsies, myopathies). In order to attain optimum oral-motor skill, a child must have the ability to move oral and facial structures independently of the rest of the body. Poor control of posture, uncoordinated movements of the upper limbs, lack of independent mobility, visual, hearing and communication problems all contribute to feeding difficulties in disabled children.

Cerebral Palsy

Severe disability is common in the graduates from neonatal intensive care units; the EPICure study group evaluated children who were born at 25 or fewer completed weeks of gestation at the time when they reached a median age of 30 months and 49% were disabled with 23% meeting the criteria for severe disability.[17] Epidemiological studies have shown that these are the children who will encounter feeding difficulties. The North American Growth in Cerebral Palsy Project is a population-based study that evaluated the growth and nutritional status in children with moderate-to-severe cerebral palsy and found the majority (58%) had reported feeding problems.[18] The Oxford Feeding Study examined 440 children with cerebral palsy and feeding problems and found that 89% required assistance with feedings and there were other concerns such as frequent choking (56%), stressful and prolonged feeding (43%) and vomiting (22%).[19] As has been outlined above, to feed effectively, an infant needs to co-ordinate sucking, swallowing, and breathing. Oral motor dysfunction is the primary cause of feeding difficulties in children with disabilities. Reilly et al. (1996) noted that greater than 90% of children with cerebral palsy had clinically significant oral motor dysfunction.[20] Prolonged feeding times, drooling, coughing, and choking with feeds and gagging are all signs of oral motor dysfunction. In one study of 271 children with cerebral palsy, the median length of time caregivers spent feeding per day was 2.5 hours with 28% (72/261) spending more than three hours a day on this activity alone.[19]

NEURODEVELOPMENTAL DISORDERS

Developmental disorders characterized by communication difficulties are

often accompanied by eating problems. Two conditions, autism and Rett syndrome will suffice as examples.

Autism is a developmental disorder characterized by severe deficits in social interaction and communication along with stereotypic behavior patterns. The unusual eating patterns and feeding difficulties of children with autism (which is more common in boys) has long been recognized. They often have an extremely limited food repertoire occasionally with apparent craving for certain foods. Research in this area is limited but in a parent survey of the eating habits of 340 autistic children,[21] two thirds of respondents considered their child to be a “picky” eater. The commonest behavioral problems reported were unwillingness to try new foods, mouthing objects, and rituals surrounding eating. Other problems were licking objects, smelling, and throwing food and pica. 60% of one series of 70 autistic patients exhibited pica.[22]

Rett syndrome may be confused with autism but arises from a mutation in the transcription regulating gene MECP2 on the X chromosome; it occurs almost exclusively in girls. Feeding problems are common in Rett syndrome in which there are characteristic oropharyngeal abnormalities.[23,24] Videofluoroscopic studies of feeding in girls with Rett syndrome have shown reduced movements of the mid and posterior tongue, with premature spillover of food and liquid from the mouth into the pharynx and laryngeal penetration of liquids and solid food during swallowing.[25,26] Air swallowing has also been noted as a problem in these patients.[27]

CONGENITAL HEART DISEASE

Difficulties with feeding are common in children with congenital heart disease (CHD). Both decreased energy intake and increased energy requirements in this group contribute to malnutrition.[28] For most parents feeding of infants and children with CHD poses significant difficulties, is time consuming and associated with considerable anxiety.[29] Maintenance of an adequate caloric intake, in order to achieve sustained growth, is often not possible without nutritional support, often requiring continuous enteral feeding.[30,31]

Cystic Fibrosis

Good nutritional care is an essential part of the management of the child with cystic fibrosis (CF) and is one of the major factors contributing to the

improved longevity of such children. Energy requirements vary but can be in excess of 150% of the daily recommended value for the normal child and this may pose a significant challenge or parents as they try to meet these requirements. A number of studies have highlighted that children with CF are at risk of developing behavior problems during mealtimes.[32,33] Behavioral therapy has been shown by meta-analysis of several studies to be as effective as oral supplementation and enteral and parenteral feeding in improving weight gain in young people with CF.[34]

Type 1 Diabetes

The adolescent years are often characterized by deterioration in the metabolic control of diabetes and it is not uncommon for adolescent girls particularly to be overweight. These changes coincide with the peak period of risk for the development of eating disorders. The commonest abnormality encountered in Engstrom's study of adolescent diabetic girls was binge eating and purging behavior (self-induced vomiting or insulin omission).[35] Diabetics with eating disorders had higher HbA_{1c} concentrations than those without eating disorders.[36] It is not surprising, therefore, that patients with type 1 diabetes and eating disorders have more long-term diabetic complications with an increased prevalence of retinopathy, microalbuminuria, and painful neuropathy when compared with those diabetics without an eating disorder.[37]

BEHAVIORAL DISORDERS

Feeding difficulties are some of the most common behavioral disturbances in young children. An estimated 24% of 2-year-old, 19% of 3–year-old, and 18% of 4-year-old children are reported by their parents as having problems with feeding.[38] These problems can range from nuisance behavior (messy, disruptive mealtime behavior) to total food refusal and life-threatening malnutrition. Any of the organic diseases referred to above can have an additional psychological and behavioral component, which should also be addressed for successful management.

Post-Traumatic Feeding Disorder

Post-traumatic feeding disorder (PTFD) also referred to as conditioned dysphagia, food aversion or food phobia is one type of feeding problem that occurs in infants who refuse to eat and have had distressing experiences involving the mouth, nose, throat, and esophagus.[39] The

prevalence of PTFD is unknown, although it may affect 4% of infants with gastroesophageal reflux who do not have a neurological or craniofacial problem, or a history of esophageal surgery and up to 40% of those who have had esophageal surgery.[40] The increase in PTFD mirrors the increasing number of infants whose survival depends upon advances in medical technology that require invasive procedures (e.g., suctioning, intubation, repeated insertion or removal of nasogastric feeding tubes) involving the oro- and nasopharynx. Such experiences may make the infant associate stimulation in and around the mouth with pain, discomfort or fear and lead to food phobia. With such a phobia, the child experiences intense anxiety if exposed to food and so refuses to eat to avoid the anticipated pain and discomfort. Another proposed mechanism to explain PTFD is visceral hyperalgesia, a neuropathic condition in which prior experience, for example, esophagitis changes sensory nerves in the esophagus so that previously innocuous stimuli, e.g., swallowing liquids or solids, are perceived as painful.

Parental Behavior

It is important to note that there is a clear association between parental behavior and child feeding behavior. Significant associations have been noted between coercive parental behaviors and children's food refusal and noncompliance.[41] In the study by Sanders and colleagues[41], feeding-disordered children engaged in higher levels of disruptive mealtime behavior (food refusal, noncompliance, complaining, oppositional behavior, and playing with food) and lower levels of chewing during mealtime. There were several significant age effects, with younger children (under age 3) engaging in more vomiting and less aversive demanding and verbalizations. Parents of feeding-disordered children were more negative and coercive in their feeding practices and engaged in higher levels of aversive instruction giving, aversive prompting, and negative eating related comments than parents of non-problem eaters.[41] Dahl et al. (1986) found that four factors were highly significant in predicting feeding problems in infants (1) feeding problems in the parents during their own infancy, (2) great anxiety experienced by the mother during pregnancy, (3) breastfeeding problems experienced by the mother, and (4) ill health in the mother. These studies provide support for the recommendation that evaluation of the feeding relationship should be an essential part of the diagnostic study of any child with feeding problems and for treatment methods that directly alter parents' feeding practices.

ASSESSMENT

Clinical History

A multidisciplinary team (MDT) approach offers the best way to diagnose the cause of the feeding problem and to assess the needs of the child. The members of this MDT typically comprise a pediatrician, a speech and language therapist, a dietitian, a psychologist, and an occupational therapist. Following assessment the MDT together with the parents can then formulate a management plan.

Southall and Martin[47] have produced clinical practice toolkits for assessment, problem formulation, and intervention with a child with feeding problems, which can be downloaded from http://www.radcliffehealth.com/shop/feeding-problems-children-practical-guide-0

A comprehensive clinical history is of particular value in the evaluation of infant dysphagia. As in other areas of clinical work, familiarity with the mechanisms and patterns of feeding disability results in a more perceptive enquiry and more meaningful description. The clinical history may identify the cause of the feeding difficulty and may also reveal associated conditions such as a history of pneumonia raising the suspicion of recurrent aspiration.

A detailed feeding history in indispensable and includes details of the child's feeding since birth, any changes of formula, introduction of solids, current diet, textures, feeding position, route and time of administration of feeds, and meal duration. Careful inquiry into the details of any food aversions and allergies is essential. In conjunction with the feeding history, a dietetic assessment is important to determine whether the caloric intake is adequate. Psychological assessment will help to identify behavioral and parental factors (e.g., family relationship issues, emotional problems) that contribute to the feeding disorder.

Examination

An important component of the detailed physical examination is observation of a feeding episode, as this will often provide significant insight into the underlying feeding problem. The level of interaction between the child and parent, verbal and nonverbal communications, the child's responses to offered foods, including disruptive behaviors such as spitting or turning the head away all give invaluable information. In conjunction with the dietitian who undertakes formal anthropometric analysis of nutritional status, the child's growth should be plotted on

appropriate reference charts. Feeding problems may be the harbinger of a serious underlying neurodevelopmental or metabolic problem and so such children need careful developmental assessment. Any suspicion of significant developmental delay indicates the need for more formal assessment by a neurodisability pediatrician.

Investigations

A careful developmental medical and feeding history, together with physical examination provides clues to the diagnosis and guides the selection of further investigations.

Perhaps the most relevant of these in the content of feeding problems is a videofluoroscopic swallowing study, which can assist in the identification of the conditions for safe swallowing and adequate protection of the airway. It is important to appreciate that not all aspiration or penetration identified by video fluoroscopy is, in fact, clinically significant in terms of the risk of chronic lung disease.[42]

Further details of the investigation and management of dysphagia (impaired swallowing) in children can be found in the excellent review by Anna Dusick.[43]

Management

Successful clinical management of infants with feeding difficulties depends upon the experience and skills of the multidisciplinary health care team and, crucially, the mother. It is impossible to generalize about best procedures or feeding routines apart from those few diagnoses (e.g., cleft lip and palate) where specific therapy is indicated.

Feeding problems that begin in infancy may persist and can result in reduced growth in height and weight. In some children, early oral motor dysfunction leads to slow and difficult feeding and the development of feeding aversion. Accordingly, it is observed in some older patients without current oral motor problems that early oral motor dysfunction which has resolved has led to acquired dysfunctional feeding behavior and poor nutritional intake. In such patients, early intervention to reduce oral motor dysfunction and consequent behavioral reactions might prevent growth compromise, long-standing behavioral problems and family discord.

The biomedical model of etiology proposes anatomical, oral-motor, or subtle sensory deficits as an explanation for feeding disorders. An appreciation of the additional insights provided by a biopsychosocial

model of causation[38] is necessary for a fuller understanding of this complex problem. Such an appreciation is essential to the development of effective management and intervention strategies for children with feeding disorders.

Reau and colleagues suggest that a combination of factors such as family dysfunction or maternal stress sometimes results in a preoccupied mother who is unaware of her infant's eating distress stemming from an organic problem.[13] In other cases, the mother is aware but is unsure what to do about it and the pediatrician may not think of oral motor dysfunction. As discussed above, PTFD is characterized by refusal of specific "feared" textures, for example, formula that has caused dyspepsia, lumpy foods that have made the infant gag. Other symptoms include acceptance of the "feared" food only when in a drowsy state (e.g., "dream feeding"), lack of awareness of hunger and thirst, and anxious anticipation of meal times as when events associated with being fed such as the site of food, feeding chair, or bottle trigger symptoms such as crying, gagging, coughing, retching, vomiting, and escape behavior such as squirming or crawling away. Four specific feeding behaviors (not chewing, sucking, or moving the food placed in the mouth for more than five seconds, passively refusing to swallow once the food was placed in the mouth, spitting out, and retching) exhibited by infants with PTFD will predict treatment outcome in most cases. These could be viewed as "bad habits" learnt by infants to avoid swallowing that need to be extinguished.

From a psychological perspective, feeding behaviors may result from either classical or operant conditioning and the distinction between these two has important clinical implications. Feeding behaviors resulting from classical conditioning (or stimulus response learning) are minimally influenced by consequences (e.g., punishment or reward) after the behaviors occur. For instance, withdrawing attention when an infant refuses food will not extinguish this behavior if it has been acquired by classic conditioning. Phobic responses, such as those seen in PTFD, are classically conditioned. Classical conditioning methods of treatment, therefore, are likely to be the most effective to help infants overcome their fear of swallowing or food phobia. Two commonly used treatment methods to extinguish behaviors resulting from classical conditioning are systematic desensitization and flooding.

Systematic desensitization provides slow, gradual exposure to the fear, for example, food with the infant largely controlling the rate and intensity of exposure. There is no evidence to support the efficacy of desensitization in the treatment of PTFD.

The behavioral technique that is believed to most helpful to treat PTFD is flooding.[3] Flooding provides rapid and intense exposure to the feared stimulus, for example, food with the therapist largely controlling the rate and intensity of exposure and providing active assistance with anxiety control. In the first few sessions, this technique is associated with much distress and fear and crying but with perseverance real improvement can be observed in days or weeks.

Feeding behaviors that result from operant conditioning are dependent on stimuli that precede the behaviors and on the responses that followed the behaviors. For example, if an infant is asked to swallow food placed in their mouth and the feeder responds with praise for swallowing then this behavior is reinforced and the infant is likely to comply the next time they are asked to swallow. Thus, the maintenance or extinction of specific mealtime behaviors learnt through operant conditioning will depend on the feeders' response to the behavior. Operant conditioning techniques range from contingency management, for example, extinction, timeout, positive reinforcement, to punishment, or aversive techniques such as bitter tastes. In PTFD, contingency management alone is particularly unhelpful, perhaps because infants with PTFD are more afraid of swallowing than eager to receive praise for swallowing.

CONCLUSION

Although common and affecting up to a quarter of normal small children, feeding disorders are often temporary. They are, however, very heterogeneous in manifestation including food refusal, dysphagia, hyperphagia, and pica. Feeding disorders arise from multiple etiologies but in general are based on three groups of children; those who are otherwise healthy, those with digestive disorders (especially gastroesophageal reflux) and those with neurological impairment. Special cases include children with congenital heart lesions, CF, type 1 diabetes, cancer, and developmental disorders (e.g., Rett syndrome and autism).

Multidisciplinary assessment and management is essential to a successful outcome. Both biomedical and biopsychosocial models of health care can contribute insights to help address underlying organic pathologies, support nutritional status and modify aberrant feeding behaviors.

REFERENCES

1. Lindberg L, Ostberg M, Isacson IM, Dannaeus M. Feeding disorders related to nutrition. *Acta Paediatr*. 2006;95(4):425–429.
2. Satter E. The feeding relationship: problems and interventions. *J Pediatr*. 1990;117(2, pt 2):S181–S189.
3. Chatoor I. Feeding disorders in infants and toddlers: diagnosis and treatment. *Child Adolesc Psychiatr Clin North Am*. 2002;11(2):163–183.
4. Dahl M, Sundelin C. Early feeding problems in an affluent society. I. Categories and clinical signs. *Acta Paediatr Scand*. 1986;75(3):370–379.
5. Dahl M, Eklund G, Sundelin C. Early feeding problems in an affluent society. II. Determinants. *Acta Paediatr Scand*. 1986;75(3):380–387.
6. Dahl M. Early feeding problems in an affluent society. III. Follow-up at two years: natural course, health, behaviour and development. *Acta Paediatr Scand*. 1987;76(6):872–880.
7. Dahl M, Kristiansson B. Early feeding problems in an affluent society. IV. Impact on growth up to two years of age. *Acta Paediatr Scand*. 1987;76(6):881–888.
8. Bu'Lock F, Woolridge MW, Baum JD. Development of co-ordination of sucking, swallowing and breathing: ultrasound study of term and preterm infants. *Dev Med Child Neurol*. 1990;32(8):669–678.
9. Bosma JF. Development of feeding. *Clin Nutr*. 1986;5:210–218.
10. Field T, Ignatoff E, Stringer S, et al. Nonnutritive sucking during tube feedings: effects on preterm neonates in an intensive care unit. *Pediatrics*. 1982;70(3):381–384.
11. Bernbaum JC, Pereira GR, Watkins JB, Peckham GJ. Nonnutritive sucking during gavage feeding enhances growth and maturation in premature infants. *Pediatrics*. 1983;71(1):41–45.
12. Morris BH, Miller-Loncar CL, Landry SH, et al. Feeding, medical factors, and developmental outcome in premature infants. *Clin Pediatr*. 1999;38(8):451–457.
13. Reau NR, Senturia YD, Lebailly SA, Christoffel KK. Infant and toddler feeding patterns and problems: normative data and a new direction. Pediatric Practice Research Group. *J Dev Behav Pediatr*. 1996;17(3):149–153.
14. Field D, Garland M, Williams K. Correlates of specific childhood feeding problems. *J Paediatr Child Health*. 2003;39(4):299–304.
15. Rommel N, De Meyer AM, Feenstra L, Veereman-Wauters G. The complexity of feeding problems in 700 infants and young children presenting to a tertiary care institution. *J Pediatr Gastroenterol Nutr*. 2003;37(1):75–84.
16. Reilly SM, Skuse DH, Wolke D, Stevenson J. Oral-motor dysfunction in children who fail to thrive: organic or non-organic? *Dev Med Child Neurol*. 1999;41(2):115–122.
17. Wood NS, Marlow N, Costeloe K, et al. Neurologic and developmental disability after extremely preterm birth. EPICure Study Group. *N Engl J Med*. 2000;343:378–384.
18. Fung EB, Samson-Fang L, Stallings VA, et al. Feeding dysfunction is associated with poor growth and health status in children with cerebral palsy. *J Am Diet Assoc*. 2002;102:3–73.
19. Sullivan PB, Lambert B, Rose M, et al. Prevalence and severity of feeding and nutritional problems in children with neurological impairment: Oxford Feeding Study. *Dev Med Child Neurol*. 2000;42:10–80.
20. Reilly JJ, Hassan TM, Braekken A, et al. Growth retardation and undernutrition in children with spastic cerebral palsy. *J Hum Nutr Diet*. 1996;9:429–435.
21. Williams PG, Dalrymple N, Neal J. Eating habits of children with autism. *Pediatr Nurs*. 2000;26(3):259–264.

22. Kinnell HG. Pica as a feature of autism. *Br J Psychiatry*. 1985;147:80–82.
23. Isaacs JS, Murdock M, Lane J, Percy AK. Eating difficulties in girls with Rett syndrome compared with other developmental disabilities. *J Am Diet Assoc*. 2003;103(2):224–230.
24. Morton RE, Bonas R, Minford J, et al. Feeding ability in Rett syndrome. *Dev Med Child Neurol*. 1997;39(5):331–335.
25. Morton RE, Bonas R, Minford J, et al. Respiration patterns during feeding in Rett syndrome. *Dev Med Child Neurol*. 1997;39(9):607–613.
26. Motil KJ, Schultz RJ, Browning K, et al. Oropharyngeal dysfunction and gastroesophageal dysmotility are present in girls and women with Rett syndrome. *J Pediatr Gastroenterol Nutr*. 1999;29(1):31–37.
27. Morton RE, Pinnington L, Ellis RE. Air swallowing in Rett syndrome. *Dev Med Child Neurol*. 2000;42(4):271–275.
28. Cameron JW, Rosenthal A, Olson AD. Malnutrition in hospitalized children with congenital heart disease. *Arch Pediatr Adolesc Med*. 1995;149(10):1098–1102.
29. Imms C. Impact on parents of feeding young children with congenital or acquired cardiac disease. *Cardiol Young*. 2000;10(6):574–581.
30. Vanderhoof JA, Hofschire PJ, Baluff MA, et al. Continuous enteral feedings. An important adjunct to the management of complex congenital heart disease. *Am J Dis Child*. 1982;136(9):825–827.
31. Ciotti G, Holzer R, Pozzi M, Dalzell M. Nutritional support via percutaneous endoscopic gastrostomy in children with cardiac disease experiencing difficulties with feeding. *Cardiol Young*. 2002;12(6):537–541.
32. Stark LJ, Jelalian E, Powers SW, et al. Parent and child mealtime behavior in families of children with cystic fibrosis. *J Pediatr*. 2000;136(2):195–200.
33. Duff AJ, Wolfe SP, Dickson C, et al. Feeding behavior problems in children with cystic fibrosis in the UK: prevalence and comparison with healthy controls. *J Pediatr Gastroenterol Nutr*. 2003;36(4):443–447.
34. Jelalian E, Stark LJ, Reynolds L, Seifer R. Nutrition intervention for weight gain in cystic fibrosis: a meta analysis. *J Pediatr*. 1998;132(3, pt 1):486–492.
35. Engstrom I, Kroon M, Arvidsson CG, et al. Eating disorders in adolescent girls with insulin-dependent diabetes mellitus: a population-based case-control study. *Acta Paediatr*. 1999;88(2):175–180.
36. Jones JM, Lawson ML, Daneman D, et al. Eating disorders in adolescent females with and without type 1 diabetes: cross sectional study. *BMJ*. 2000;320(7249):1563–1566.
37. Rydall AC, Rodin GM, Olmsted MP, et al. Disordered eating behavior and microvascular complications in young women with insulin-dependent diabetes mellitus. *N Engl J Med*. 1997;336(26):1849–1854.
38. Beautrais AL, Fergusson DM, Shannon FT. Family life events and behavioral problems in preschool-aged children. *Pediatrics*. 1982;70(5):774–779.
39. Benoit D, Coolbear J. Post-traumatic feeding disorders in infancy: Behaviors predicting treatment outcome. *Infant Mental Health J*. 1998;19(4):409–421.
40. Dellert SF, Hyams JS, Treem WR, Geertsma MA. Feeding resistance and gastroesophageal reflux in infancy. *J Pediatr Gastroenterol Nutr*. 1993;17(1):66–71.
41. Sanders MR, Patel RK, Le Grice B, Shepherd RW. Children with persistent feeding difficulties: an observational analysis of the feeding interactions of problem and non-problem eaters. *Health Psychol*. 1993;12:64–73.
42. Arvedson J, Brodsky L. *Pedaitric Swallowing and Feeding*. Albany, NY: Singular Publishing Group; 2002.

43. Dusick A. Investigation and management of dysphagia. *Semin Pediatr Neurol*. 2003;10(4):255–264.
44. Rudolph CD, Link DT. Feeding disorders in infants and children. *Pediatr Clin North Am*. 2002;49(1):97–112.
45. Foy T, Czyzewski D, Phillips S, et al. Treatment of severe feeding refusal in infants and toddlers. *Inf Young Chil*. 1997;9(3):26–35.
46. Illingworth RS. Sucking and swallowing difficulties in infancy: diagnostic problem of dysphagia. *Arch Dis Child*. 1969;44(238):655–665.
47. Southall A, Martin C. *Feeding Problems in Children: A Practical Guide*. Abingdon, UK: Radcliffe Publishing; 2010.

Failure-to-Thrive: Growth Failure in the Outpatient Setting

47

Jennifer A. Woo Baidal, MD, MPH Susanna Y. Huh, MD, MPH

BACKGROUND AND DEFINITIONS

Although the term "failure to thrive" (FTT) has been in use for over a century, a consensus definition does not exist.[1–3] FTT is not a diagnosis with a single etiology, but rather a description of an abnormal growth pattern reflecting failure to grow at an expected rate consistent with growth standards.[4] Terms such as growth failure, inadequate growth, and weight faltering have begun to replace the use of the term FTT, which may have negative connotations.

Specific definitions for growth failure are variable,[5] but there is general consensus that the definition should be based on anthropometric measurements.[3] A common static definition of growth failure based on attained growth is sex-specific weight-for-age or weight-for-length less than the 2 (3rd percentile) or more than 2 standard deviations below the mean for a reference population (z-score < −2). Other published cutoffs used to define failure-to-thrive include the 5th or 10th percentiles of weight-for-age or weight-for-height, or height-for-age less than the 3rd or 5th percentiles.[3] Optimally, measurements should be obtained on two or more occasions to support a definition of growth failure.

The World Health Organization (WHO) recommends expressing anthropometric measures in z-scores at both the individual and population level.[6] The weight-for-age z-score can be calculated using the following formula:

weight-for-age z-score = [actual weight - median weight] /standard deviation

where "z-score" refers to age- and sex-specific standard deviation of weight, and "median weight" refers to the median value for age and sex.[7] Online calculators and some electronic medical record software programs can assist in calculating z-scores, such as those found at www.statcoder.com. The use of z-scores allows for a more precise description of a child's growth compared to the use of percentiles. For example, a child whose weight-for-age plots "below the 3rd percentile" could have a weight-for-age z-score of either −2.1 or −3.5, encompassing a wide range of severity.[7] For a child with growth failure, the use of z-scores also allows for more precise quantitative longitudinal monitoring of a child's growth than centiles. A child whose weight-for-age improves from a z-score of −2.9 to −2.2 would have a weight-for-age below the 3rd percentile at each time point, masking the progress in growth that has occurred.

A dynamic definition of growth failure is based on decreased growth velocity, defined as weight-for-age or height-for-age curves that have crossed downward more than two major percentile lines (such as 5th, 10th, 25th, 50th, 75th, 90th, and 95th) after having previously achieved a stable growth pattern. Defining a stable pattern can be difficult, particularly in infancy when weight percentiles commonly fluctuate, as discussed in further detail below. The phenomenon of regression to the mean describes the tendency of large infants to cross percentiles downward and small infants to cross percentiles upward toward the mean of a reference population.[8] To account for regression to the mean, some research studies have used "conditional weight gain" <= 5th percentile to define growth failure, which compares an infant's weight gain with weight gain of other infants of the same sex and initial weight; this definition is difficult to apply to clinical practice.

The proximate cause of the majority of growth failure cases is malnutrition or undernutrition.[9] The terms malnutrition or undernutrition are often used interchangeably with growth failure, although the terms are not synonymous. Waterlow[10] proposed criteria for distinguishing between

acute and chronic undernutrition with the terms wasting and stunting. Wasting is defined as a weight-for-height <= 90% of the median and represents acute malnutrition. Stunting is defined as a length-for-age or height-for-age that is 90% or less of the median and represents chronic malnutrition. Table 47-1 shows the Waterlow classification for categorizing malnutrition.[10] To calculate the percentage of the median, the following formula can be used:

$$[\text{Actual weight}/\text{median weight}] \times 100 = \%\ \text{median}$$

where median weight is the median value for age and sex. Determining the severity and chronicity of malnutrition can help direct intervention as described below.

EPIDEMIOLOGY

The prevalence of pediatric growth failure is difficult to estimate for several reasons. The lack of a consistently used definition for growth failure impedes comparison of prevalence estimates across studies. Several other factors can interfere with nationally representative information on the prevalence of growth failure in infants, including sample and selection bias.[11] When interpreting national prevalence estimates, it is important to note that estimates of growth failure are equivalent to estimates of undernutrition; growth failure prevalence estimates frequently include low-birth-weight infants, who may account for 20%–40% of low-weight or short children.[4,12]

TABLE 47-1 Waterlow Criteria for Categorizing Type and Chronicity of Malnutrition

Type of Malnutrition	Acute (Weight for Height) (% of Median)	Chronic (Height for Age) (% of Median)
Mild	80–90	90–95
Moderate	70–80	85–90
Severe	<70	<85

Acute malnutrition (abnormal weight-for-height) is termed “wasting” and chronic malnutrition (abnormal height-for-age) is termed “stunting.”

Source: Adapted from Waterlow et al.[6]

Mei and colleagues studied nationally representative data from the National Health and Nutrition Surveys (NHANES) 1999–2004 to compare the prevalence of underweight using three cut-off points on the WHO and CDC weight-for-age growth curves.[13] Mei found that approximately 2% of infants were underweight when a cut-off of 2.3rd percentile, (−2 z-score) on WHO charts was used. When the cutoff was increased to the 5th percentile, the prevalence of underweight among infants under age 2 years was 3.8% on WHO charts and 6.8% on CDC charts. Similar trends were seen for weight-for-age estimates among older children.[13] Fewer US children are expected to be diagnosed with underweight when weight and length are plotted on WHO charts.[14] Figure 47-1 illustrates changes in the estimated prevalence of low weight-for-age according to different charts and cut-off values.

Recent epidemiologic evidence suggests that the number of underweight children in the United States has declined over the past three decades. Among children and adolescents in NHANES, the prevalence of underweight defined as BMI < 5th percentile declined from 5.1% in 1971–1974 to 3.5% in 2007–2010.[15] Among children aged 2–5 years, underweight decreased from 5.3% to 3.4% during the same time period.[15] These data excluded infants under the age of 24 months, the most vulnerable age group for low weight-for-age and low weight-for-length. The prevalence of underweight may be higher among specific subgroups. Frank and Zeisel reported in 1988 that an estimated 10% of low-income children in the United States were undernourished.[16]

Globally, an estimated 16% of children under 5 years of age were underweight (weight-for-age z-score < −2) in 2010 in the WHO Global Database on Child Growth and Malnutrition.[17] The prevalence of childhood underweight was 1.5% in developed nations and 17.5% in developing nations.[17]

RISK FACTORS

A newer understanding of the complex interactions between multiple factors that contribute to a child's nutritional status is replacing the previous perspective that growth failure results from either "organic" or "nonorganic" causes. Organic causes of growth failure refer to chronic medical disorders that interfere with growth, such as malabsorption, genetic syndromes, endocrine disorders, and neurologic dysfunction.[4] Nonorganic growth failure has traditionally been defined as a diagnosis of

exclusion, to label growth failure in a child when a medical condition is not identified.[4] However, this dichotomous approach does not recognize that children with growth failure commonly have more than one physiologic and psychosocial factor contributing to their undernutrition.[2,18] Further, this approach emphasizes the potential of a "missed" medical diagnosis and can lead to extensive invasive and unnecessary medical evaluation. In some series, as many as 90%–95% of children with growth failure had no underlying medical condition.[2,18] Growth failure can result from a "layering of risks," where multiple risk factors interact to cause growth failure in an infant who might have otherwise grown normally if only a single disease or a single psychosocial risk factor existed.[4]

Perinatal Risk Factors

Perinatal factors, including preterm birth and birth weight, genetic and developmental conditions, or environmental exposures can all predispose a child to growth failure. Infant preterm birth, including late preterm birth,[19] is an important risk factor for growth failure. Compared to term counterparts, infants who were both low birth weight (≤2500 g) and preterm (≤37 weeks) have been found to have smaller mean lengths, weights, and head circumferences in the first 3 years of life, even after correction for gestational age.[20,21] Preterm infants can have anthropometric measures plotted on curves developed for preterm infants until they achieve a postconceptual age of 40 weeks;[19,22,23] thereafter their measurements can be plotted on standard growth charts.

Both preterm and term infants who are born small for gestational age (SGA) are at risk for growth failure. Small for gestational age is usually defined as birth weight and/or length 2 standard deviations smaller than the mean for gestational age and sex in a reference population (z-score < −2).[24,25] Historically, the terms small for gestational age and intrauterine growth restriction (IUGR) have been used interchangeably, but the term IUGR should be reserved for infants with documented aberrant prenatal fetal dimensions[24]; the most common definition of IUGR is sonographically estimated fetal weight below the 10th percentile.[26] Term infants born SGA have growth failure that appears to be in part related to familial short stature. Data from the National Collaborative Perinatal Project, an 8-center US study of perinatal risk factors and growth and development in term infants (>=37 weeks gestation), reported that mean height of mothers of infants born SGA (defined as birth weight <=2500 g)

was 2 cm shorter than for mothers of infants without SGA.[27] Compared to preterm infants born appropriate for gestational age, preterm infants born SGA are at higher risk of growth failure in early childhood.[28]

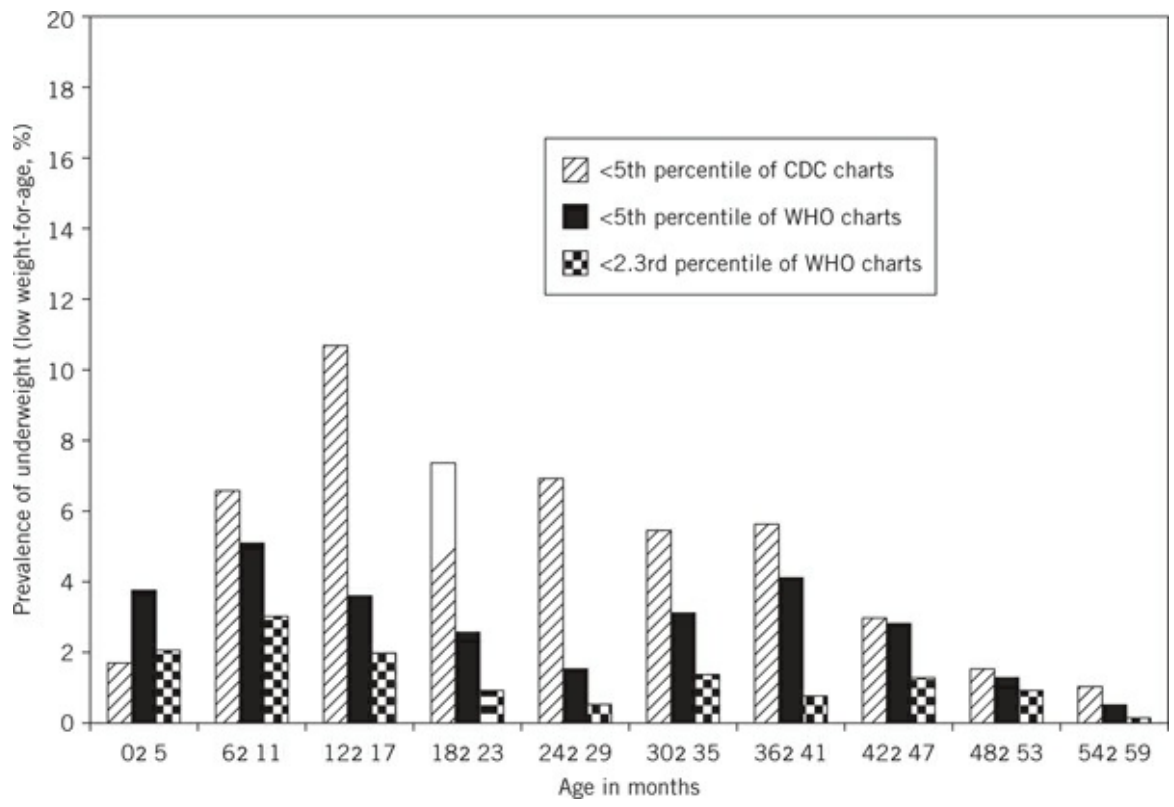


Figure 47-1 Comparison of prevalence of underweight (low weight-for-age) based on the CDC 2000 and WHO 2006 growth charts, NHANES, NHANES 1999–2004 for children aged 0–59 months. Reprinted with permission from the *Journal of Pediatrics*; Mei et al.[13]

Several reasons for in utero growth failure exist, including environmental exposures (maternal smoking, undernutrition, drug or toxin exposure), maternal medical complications, placental abnormalities, and fetal problems (chromosomal abnormalities, inborn errors of metabolism, or intrauterine infections).[10,24] Traditionally, growth symmetry has been thought to reflect the origins of in utero growth failure and prognosis for future somatic growth. Symmetric IUGR (proportional weight, height, and head circumference) was classically thought to result from undernutrition or other processes affecting the 1st or 2nd trimester, whereas asymmetric IUGR (disproportionately affected weight) was thought to result from

undernutrition or insults occurring during the 3rd trimester.[24] Recent data show that both symmetric and asymmetric IUGR can arise from the early second trimester.[29] Prenatal ultrasound measures of fetal growth parameters do not predict birth anthropometric measures well, perhaps due to in utero crossing of percentiles.[30] Villar et al.[31] suggested that infants with symmetric IUGR are less likely to respond to nutritional intervention, while those with disproportionately affected weight are more likely to have experienced in utero malnutrition and respond to nutritional intervention after birth. However, other data show that preterm infants born with either symmetric or asymmetric SGA have limited catch-up growth through 3 years of age.[32]

Medical Risk Factors

Congenital and chronic medical conditions can lead to poor weight gain through a variety of mechanisms.[33] Medical risk factors for the development of growth failure can be categorized as inadequate caloric intake, malabsorption, excessive caloric expenditure, and/or defective utilization of nutrients (Table 47-2). We briefly outline some common medical risk factors in this section; a detailed review of the many medical conditions associated with growth failure can be found elsewhere in this textbook.

Neurologic conditions, particularly cerebral palsy, are often associated with abnormal growth and nutrition in both preterm and term infants.[34–39] In addition to inadequate intake, other risk factors for growth failure in neurologically impaired children include oral motor dysfunction; increased nutrient losses due to emesis or spillage; and excessive energy expenditure related to hypertonia or movement disorders.[40] Thirty to forty percent of children with cerebral palsy are estimated to have feeding difficulties,[41,42] which can result in part from impaired central and enteric nervous system function.[43] Contractures or scoliosis can make measurement and interpretation of linear growth challenging in children with cerebral palsy; alternative measures such as upper arm and lower leg lengths can be plotted on available growth curves.[37,39] These measurements have been found to correlate with triceps skinfold thickness and mean arm circumference.[37] Triceps skinfold thickness can be helpful in the nutritional assessment in children with cerebral palsy, with values under the 10th percentile for age and sex indicating the need for additional assessment of nutritional and growth status.[36]

TABLE 47-2 Etiologies of Growth Failure

Mechanism	Examples of Associated Condition
Inadequate caloric intake	Reduced appetite Inappropriate dietary intake Improper formula mixing Excessive fruit juice consumption Mechanical feeding problems (Oral motor deficits) Congenital anomalies (Cleft lip or palate) Impaired swallowing (Dysphagia) Altered mental status Behavioral feeding disorders Disordered feeding techniques Infantile anorexia Sensory-based food aversions Inadequate food supply Insufficient breastmilk supply Neglect or abuse Poverty
Inadequate absorption	Gastrointestinal disease Vomiting Mucosal abnormality (Celiac disease, enzyme deficiency) Intestinal failure Pancreatic insufficiency (Shwachman-Diamond syndrome, cystic fibrosis) Chronic liver disease with cholestasis
Excessive energy expenditure	Hypertonia Chronic disease Congenital or acquired heart disease Chronic pulmonary disease Endocrine abnormalities Chronic or recurrent infections/fevers HIV or other immunodeficiency Malignancy (diencephalic syndrome)
Defective utilization	Genetic conditions Trisomies 21, 18, 13 Metabolic disorders

Source: Adapted from Gahagan; Markowitz et al.[4,7]

Estimation of energy needs in children with neurologic conditions can be a challenge. Estimation of energy needs using age-based EER/RDA or standard equations can result in excessive caloric intake, especially among nonambulatory children.[44–46] Table 47-3 lists suggested energy requirements based on calorie per centimeter of height for children with developmental delay.[47] Children with cerebral palsy may benefit from use

of indirect calorimetry to precisely estimate energy requirements.[46]

Children with cleft lip and palate have been considered to be at risk for undernutrition and growth failure, possibly related to an inadequate seal around the nipple, inadequate generation of negative sucking pressure, frequent respiratory and ear infections, and psychosocial factors.[48] In one cross-sectional study of 881 Brazilian children with cleft lip and/or palate, 20%–30% were found to have weight or height less than the 10th percentile for age during the first 2 years of life.[49] Other longitudinal studies have not shown significant FTT in children with nonsyndromic cleft lip and/or cleft palate through the first 2–5 years of life.[48,50] Children with cleft lip or palate as part of a syndrome may have a greater risk of growth failure than children with isolated cleft lip or palate.[48] Associated oral-motor dysfunction may lead to the need for special nipples, feeding instructions, or enteral tube feeding. A 2011 Cochrane review found limited data suggesting that breastfeeding may be associated with higher weight at six weeks postsurgery, as compared with spoon feeding, but no evidence that any particular bottle type was associated with improved infant growth.[51]

Difficulty with chewing, sucking, or swallowing adequate amounts of food may result in inadequate caloric intake leading to growth failure. Dysphagia can be defined broadly as a mechanical difficulty with swallowing. Oropharyngeal dysphagia can be defined as "a dysfunction of the pharyngeal phase of swallowing that allows laryngeal penetration and tracheal aspiration of small volumes of solid food or liquids."[52] Anatomic (e.g., Laryngeal cleft, tonsillar hypertrophy), physiologic (e.g., Eosinophilic esophagitis, food allergy, sinonasal secretions), and neuromuscular etiologies (e.g., Type I Chiari malformation) may lead to oropharyngeal dysphagia, in neurologically intact children[52] as well as children with genetic syndromes.[53] Chapter 48 discusses the causes and treatment of dysphagia and feeding difficulties in more depth.

Recent data link sleep problems in early childhood to feeding difficulties. In a case-control study, parents of children with feeding disorders reported shorter child sleep duration and delayed sleep time, compared with controls.[54] Sleep-disordered breathing in children, which is primarily related to adenotonsillar hypertrophy, has been associated with growth failure. A systematic review and meta-analysis found that child height and weight, and IGF-1 and IGFBP-3 significantly increased after adenotonsillectomy.[55]

Psychosocial Risk Factors

Poverty is the single greatest risk factor for undernutrition worldwide.[56,57] In low and middle-income countries, low weight, height, and BMI are clearly associated with short adult height and other adverse outcomes. In some developed nations such as the UK, prospective cohort studies have not found a clear association between socioeconomic status and poor conditional weight gain (<5th percentile) during the first 9 months of life, perhaps because of an adequate social safety net and study cohorts with a greater proportion of affluent participants than the general population.[8,58] In the United States, poverty remains a key social risk factor for childhood malnutrition. In 2011, almost 15% of US households reported food insecurity, defined as "uncertain of having, or unable to acquire, enough good to meet the needs of all their members because they had insufficient money or other resources for food."[59] Higher rates of food insecurity were reported in households with children under 6 years old (21%) and Black (25%), Hispanic (26%), and low-income households (35%).[59] Federal supplemental nutrition assistance programs appear to help alleviate food insecurity and improve nutrient intake; however, the low level of benefits —in 2006, SNAP was estimated to provide $1.40 per participant per meal —can mean that even concurrent participation in multiple programs may not adequately alleviate food insecurity.[9] Compared with infants receiving benefits, US infants < 12 months old who were eligible for but not receiving benefits from the Special Supplemental Program for Women, Infants, and Children (WIC) exhibited lower mean weight-for-age z-score (−0.23 vs. 0.009) and shorter stature (length-for-age z-score −0.23 vs. −0.02).[60] Housing insecurity has also been associated with lower weight-for-age z-scores.[61]

TABLE 47-3 Estimated Energy Needs for Developmentally Delayed Children

	Daily Caloric Recommendation
Ambulatory, ages 5–12 y	13.9 kcal/cm height
Nonambulatory, ages 5–12 y	11.1 kcal/cm height
Cerebral palsy with decreased levels of activity	10 kcal/cm height
Cerebral palsy with increased levels of activity	15 kcal/cm height
Athetoid cerebral palsy,	Up to 6000 kcal daily

adolescence	
Down syndrome, boys ages 5–12 y	16.1 kcal/cm height
Down syndrome, girls ages 5–12 year	14.3 kcal/cm height
Myelomeningocele	Approximately RDA for age may need as little as 7 kcal/cm height

Source: Adapted from Pemberton et al.[47]

Parental stress can impact a child's growth. Cross-sectional evidence has demonstrated a link between a mother's own experience of abuse as a child, and offspring growth failure. One case-control study of infants with growth failure found that 66% of mothers with infants experiencing growth failure had been abused in childhood, compared to 26% of mothers with children without growth failure.[62] A prospective study that recruited low-income mothers found that maternal report of experience of childhood abuse and unhappiness during pregnancy correlated with subsequent child growth failure through the age of 12 months.[63] Negative interactions between the child's mother and father, such as frequent separations and arguments, were also related to child growth failure.[63] Recent reviews report that maternal depression during an infant's first year appears to be a risk factor for poor growth through the first 2 years of life.[64,65] While the mechanisms underlying the association between maternal depression and child growth are not clear, maternal depression has been associated with suboptimal parenting behaviors, child behavioral difficulties and child developmental delays,[66,67] which themselves may be risk factors for underweight. Maternal anxiety and lack of maternal attachment have also been associated with growth failure.[68–70]

A systematic review of studies from developing countries found that maternal depression was associated with higher odds of underweight (usually defined as weight-for-age z-score <−2) and stunting (usually defined as height-for-age z-score <−2); the OR for underweight strengthened from 1.5 to 2.2 when the meta-analysis was restricted to longitudinal cohort studies.[65] Studies from developed countries also show that maternal depression is associated with lower child weight and height. In a U.K. case-control study, children ≤ 2 years of age with growth - faltering had mothers with higher rates of postnatal depression (OR 1.7, 95% CI 1.2–2.5) and anxiety (OR 2.1, 95% CI 1.3–3.3) than mothers of matched control children.[67] In a study of 181 U.S. Latina mothers, those with depressive symptoms both prenatally and at 4–6 weeks postpartum

were more likely to have children who were underweight (weight-for-age <5th percentile) in the first 2 years of life.[68] The nationally representative US Early Childhood Longitudinal study showed that children of mothers with moderate/severe depressive symptoms at 9 months postpartum had 1.5 higher odds (95% CI 1.03–2.13) of being ≤10 percentile in height-for-age at 5 years[71]; there was no increase in the odds of low weight-for-height. One cross-sectional U.S. study did not show an association between a positive 3-item maternal depression screen and odds of low child growth (weight-for-age <5th percentile or weight-for-height <10th percentile), among children aged 0–36 months old seen in urban hospital clinics or emergency departments.[66] Together these studies suggest that maternal depression prenatally or in the first postnatal year, but not later in childhood, may affect child growth during infancy and beyond, but more prospective data are needed.

Disturbances in feeding behaviors are associated with growth failure,[69,70,72–75] but in many studies it can be difficult to ascertain whether disruptive feeding behaviors are a cause or a consequence of growth failure. Severe food refusal resulting in markedly reduced intake of either solids or beverages can be an obvious cause of growth failure. Examples of behaviors exhibited by children with feeding disorders include food refusal, disruptive mealtime behavior, and food preferences limited to specific brands, types or textures.[76] Successful development of child feeding skills and behaviors depends on multiple factors, including: medical, physical, developmental and temperamental characteristics of the caregiver and child; psychosocial stressors; and positive caregiver-child interactions.[77]

There is no universally accepted classification system for feeding disorders. The central importance of the parent-child relationship to the development of feeding disorders led one group of experts to propose multiaxial diagnostic criteria for a “Feeding Disorder Between Parent and Child.”[77] Nicholls[78] described several common clinical presentations of childhood feeding disorders, including pica, rumination disorder, selective eating, food avoidance emotional disorder, food phobias, functional dysphagia, and food refusal. The Diagnostic Classification of Mental Health and Developmental Disorders of Infancy and Early Childhood (DC:0–3R), based on criteria developed by Chatoor,[79] defines “feeding behavior disorder” as difficulty in regulating feeding in response to physiological feelings of hunger or fullness.[80] The DC:0–3R also defines six subtypes of this disorder. The “Avoidant restrictive food intake

disorder" defined in DSM-5 replaces "feeding disorder of infancy and childhood" in DSM-4.[81] The three disorder subtypes under "Avoidant/restrictive food intake disorder" in DSM-5 mirror the DC:0-3R classification. Table 47-4[79–82] summarizes the DC:0–3R and DSM-5 classifications of feeding disorders.

Aberrant family perceptions of healthy eating can lead to parental restriction of a child's macro- or micronutrient intake, resulting in growth failure. Replacement of breast milk or formula with developmentally-inappropriate beverages, such as soy milk or rice milk, in order to align with family dietary practices, has been reported to lead to severe growth failure.[83] Several published cases of growth failure have been associated with prolonged duration of exclusive breastfeeding with delayed introduction of solid foods beyond 6–8 months of age.[84,85] Parental fear of obesity or cardiovascular disease can lead to suboptimal caloric intake or a diet with insufficient fat content that leads to poor growth.[86] Excessive fruit juice intake can lead to poor nutrition. Some fruit juices can lead to chronic diarrhea, as well as displacement of nutritionally-rich foods in the diet.[87,88] One case series of eight children with growth failure showed that reduction of juice intake was associated with improved weight gain.[89]

TABLE 47-4 Classification of Feeding Disorders Among Infant and Children

Diagnostic Classification of Developmental Disorders of Infancy and Early Childhood				DSM-5
Disorder Type	**Age of onset**	**Infant/Child Features**	**Treatment**	**Avoidant /Restrictive Food Intake Disorder**
Feeding Disorder of State Regulation	Age 0–6 mo	Disordered development of behavioral regulation Difficulty reaching and maintaining calm alertness for feedings or hard to awaken for feedings	Reduce stimulation at feedings if irritable Gentle stimulation prior to feedings if hard to awaken Treat parental depression, anxiety	Not included
Feeding Disorder of Caregiver-	Age 0–12 months	Limited emotional nurturing Reduced social	Intensive feeding intervention (multidisciplinary	Not included

Reciprocity		reciprocity at feedings. Caregiver unaware of or denies problems	clinic, home-based intervention, or hospitalization) Family therapy on nurturing emotional and physical development of child	
Infantile Anorexia	Ages 6 mo–3 y	Feeding refusal and poor hunger drive Refuses adequate amounts of food at least 1 month Behavioral difficulties (e.g., throw food, refuse to open mouth)	Parental education on separation of feeding roles, limit-setting Structured meals and snacks Foster self-feeding and eliminate distractions or coercive feeding	Individuals who do not eat enough or show little interest in feeding
Sensory Food Aversions	Age 0–3 y	Association of specific food characteristics with aversive response. Consistent refusal of specific foods or beverages based on taste, texture, temperature, or smell for at least 1 mo. Sensory difficulties (gag, vomit, or grimace with food)	Introduce variety of foods but avoid foods eliciting strong aversive reactions Model behavior (parents, peer feeding group) Supplemental macro- and micronutrients if needed Behavioral therapy	Individuals who only accept a limited diet in relation to sensory features
Post-Traumatic Feeding Disorder	Any age	Traumatic event involving oropharynx or gastrointestinal tract triggers intense and anticipatory fear of feeding Severe, consistent refusal of solids and/or beverages following traumatic event Child distress at sight of food or food reminders	Behavioral therapy (extinction or desensitization) Enteral feeding support	Individuals whose food refusal is related to aversive experience
Feeding disorder associated with a concurrent medical	Any age	Medical condition believed to cause distress Initiates feeding but becomes distressed or refuses to continue	Treat underlying medical condition Behavioral therapy	Exclusion criterion

condition	feeding

Source: Adapted from Chatoor et al.; zero to three; Boghani et al.[69,80,82]

Food allergy is estimated to affect 6%–8% of children at some point during childhood, with as many as 20% of children on an elimination diet due to perceived food allergy.[90] Unnecessary dietary restriction due to perceived food allergies – sometimes resulting from the use of alternative, non-validated allergy tests – can result in growth failure and micronutrient deficiencies.[91] In 2010, Tierney et al.[92] reviewed nineteen reports of severe dietary restriction, usually related to parental concern for allergic or other adverse reactions to specific foods, associated with kwashiorkor in US infants.

Over 12 million children are adopted by North American families from countries around the world each year, and these children commonly experience poor growth.[93] The degree of growth failure is proportional to the duration of time spent in an orphanage.[93] Growth failure among adopted children may be related to poor nutritional intake related to psychosocial deprivation, but stressors such as relocation, language barriers, extended periods in orphanages, or witnessing war and other acts of violence may also contribute to poor growth.[94–96]

Childhood neglect is a possible, but uncommon, contributor to growth failure.[62,97,98] Mediators of this link can include food withholding or intentional starvation.[99,100] Intentional starvation in infants and children has dire consequences. In one case series of 12 criminally prosecuted cases of intentional infant and child starvation, 10 of the children had severe wasting and stunting, and 2 had mild or moderate wasting.[100] Half of the infants and children died shortly before or after being assessed by medical or law enforcement personnel.[100] Although neglect or abuse are rarely the cause of growth failure, the severity of the consequences in cases of starvation underscore the need for providers to screen for serious social and psychological family problems.

Medical child abuse is defined as “unnecessary and harmful or potentially harmful medical care at the instigation of a caregiver,” and includes the commonly used term, Munchausen syndrome by proxy.[101] Although medical child abuse is rare, with incidence estimated at approximately 2 in 100,000 infants < 1 year old,[102] up to 25% of published reports of medical child abuse involved child anorexia or growth failure.[103] Mash et al.[103] compared characteristics of 17 cases of suspected or confirmed medical child abuse with 68 controls among children

evaluated in a pediatric gastroenterology clinic for failure-to-thrive over a 10-year period. They identified several risk indicators which can be used to screen for medical child abuse in cases of failure-to-thrive: involvement of five or more organs; absence of a serious congenital anomaly or -confirmed genetic disorder; five or more reported allergies; and parental refusal of services from a multidisciplinary feeding team.[103]

GROWTH CHARTS FOR SCREENING AND DIAGNOSIS

One goal of routine health supervision is to monitor the growth pattern of a child over time. Serial measurements of weight and length/height plotted on growth charts allow pediatricians to assess a child's growth trajectory and detect abnormal growth patterns. To appropriately screen for and diagnose growth abnormalities, infants and children should be weighed with minimal clothing on scales that measure to a precision of 100 grams.[7] Length for children under age 2 years should be obtained in the supine position using a length board. After 2 years of age, standing height should be obtained using a wall-mounted stadiometer.

Weight, height, and BMI for children age 2–19 years should be plotted on the 2000 CDC growth charts.[14] For infants and children under 2 years of age, the Centers for Disease Control and Prevention (CDC) and the American Academy of Pediatrics recommend that weight, length, weight-for-length, and head circumference be plotted on WHO growth charts.[14] The WHO charts were developed using longitudinal data from infants in six countries who were breastfed during the first year of life. In contrast with the cross-sectional data collected from predominantly formula-fed infants for the 2000 CDC growth charts, breastfed infants show a greater growth velocity than formula-fed infants during the first months of life, with subsequent slower rates of weight gain throughout infancy.[14] The use of WHO charts among US children will likely reduce the prevalence of growth failure diagnoses in early childhood,[13,14] but may increase the prevalence of growth failure diagnoses in the first six months of life.[104] Infants born preterm should have their chronologic age corrected for gestational age until they reach 24 months of age for weight, 40 months for length, and 18 months for head circumference.[105] Children with a genetic syndrome, including Down and Turner syndromes, should be plotted on syndrome-specific growth charts.

Shifting between growth percentiles is common in the first 2 years of

life, especially during the first six months. Mei[106] and colleagues studied over 10,000 children from birth to age 5 years participating in the California Child Health and Development Study from 1959 to 1967. The study included children from diverse socioeconomic backgrounds but did not include Latinos. 29% of infants between birth and 6 months of age, 6%–15% of children between 6 and 24 months of age, and 1%–5% of children between 24 and 60 months of age crossed 2 major weight-for-age percentiles. Shifts in weight-for-height occurred more commonly in each age group. The authors speculate that the frequent shifts in growth percentiles might be explained by regression to the mean, genetic predisposition, measurement error, or random variation.[106]

The common occurrence of shifting growth curves underscores the importance of a longitudinal approach to growth monitoring combined with a comprehensive history and physical examination to discern normal variations in growth from growth failure. Evaluation should include a careful family history with attention to the growth and pubertal history of parents and siblings. Genetic height potential should be calculated and compared to child centiles (Table 47-5).[107] Figure 47-2 illustrates characteristic linear growth curves for children with intrinsic short stature, constitutional or delayed growth, and growth failure related to undernutrition or disease (attenuated growth).[108]

TABLE 47-5 Calculating Genetic Height Potential

Boys	(Father's Height + Mother Height + 13 cm) divided by 2 Or (Father's Height + Mother's Height) divided by 2 + 6.5
Girls	(Father's Height + Mother's Height – 13 cm) divided by 2 Or (Father's Height + Mother's Height) divided by 2 – 6.5

Plot the end result on the appropriate growth chart at the age of 20 y to determine the predicted adult height percentile.

Source: Tanner et al.[107]

Normal variations in growth include genetic or familial short stature, and constitutional growth delay. Children with genetic or familial growth delay may cross percentiles downward but will maintain a normal growth velocity once they reach their genetically determined height percentile, and bone age will match chronological age.[109,110] Children with constitutional growth delay have a normal birth weight and length, and cross percentiles downward in the first 2–3 years of life, followed by a normal growth velocity.[110] In children with constitutional growth delay, bone age will be

delayed compared to the chronological age. They will also have puberty later than their peers, resulting in a normal adult height.[109]

Children with growth failure related to undernutrition will classically show a decline in weight percentile, followed by a decline in linear growth percentile. Weight-for-length reflects current nutritional status and helps to identify growth failure related to undernutrition or disease.[10] Children with malnutrition will usually display a drop in weight-for-length percentile over time,[10] although chronic malnutrition can result in stunting and a relatively normal weight-for-length, especially if a child has received nutritional intervention. Children with endocrine disorders will often have a normal or elevated weight-for-length in addition to short stature.[110] Head circumference is typically the last anthropometric measure to be affected by malnutrition.

APPROACH TO EVALUATION OF THE PATIENT WITH GROWTH FAILURE

A detailed history and physical examination are the most important components in the assessment of a child with growth failure, guide both diagnostic tests and treatment approach.[111,112] The process of history-taking can be the first step in establishing a therapeutic alliance between the medical team and the family or caregiver(s) that will allow successful treatment of medical, behavioral and psychosocial factors contributing to a child's growth failure.

Medical History

Past medical history, medication use, and immunization history should be detailed. Acute illness can increase metabolic demands through fever and stress responses, as well as decrease caloric intake through anorexia. A history of recurrent or unusual infections should increase suspicion for underlying immunodeficiency. Special attention should be paid to early life history. This includes maternal prenatal history, including use of drugs, possibility of prenatal infections, maternal nutrition, and health during pregnancy; birth history, including labor and delivery; birth size; and neonatal events. Difficulty with feeding and other diagnoses at nursery discharge have been associated with later growth problems.[63] Developmental history should be reviewed in detail.

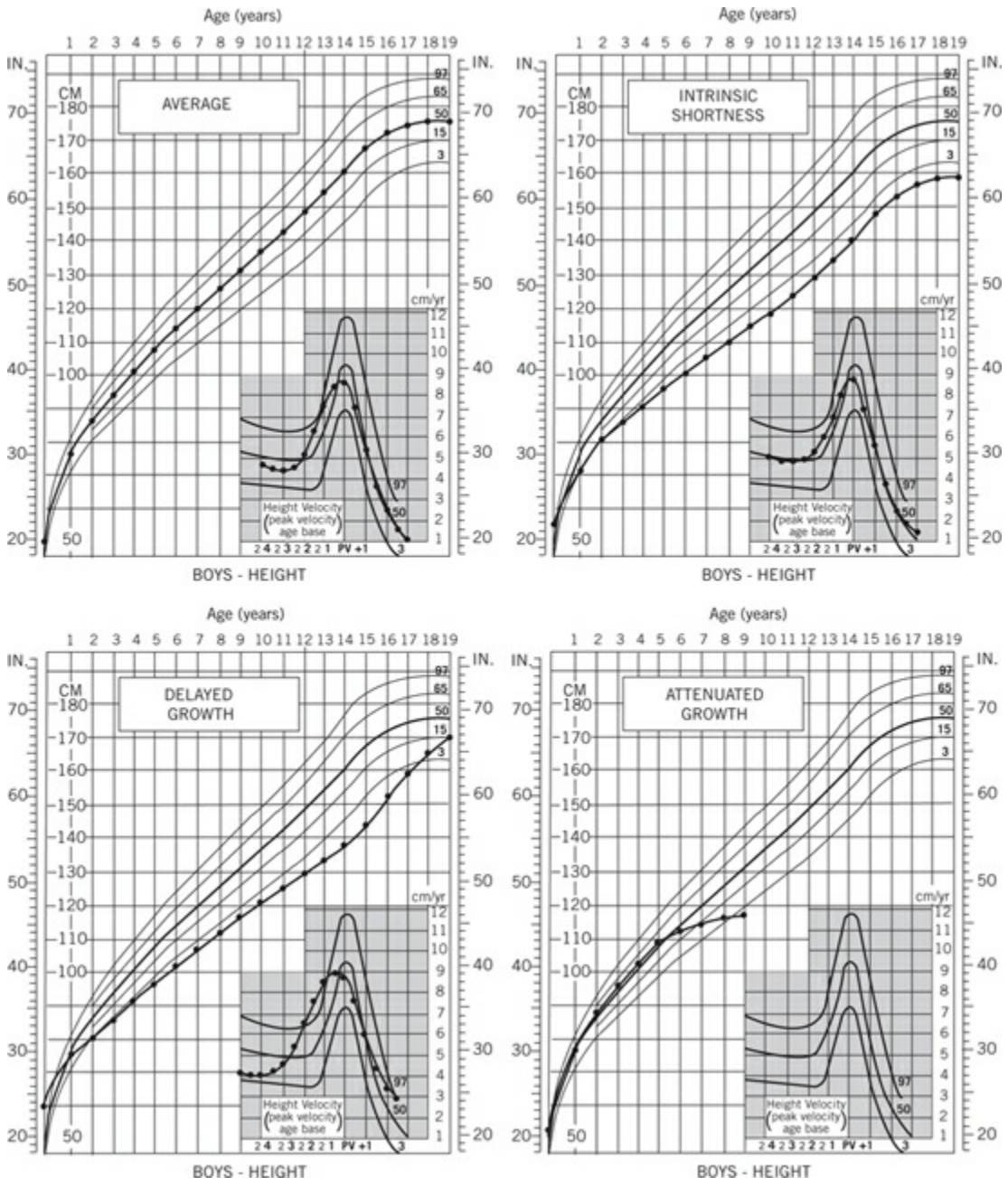


Figure 47-2 Linear growth curves (and height velocity curves, *insets*) of four different patterns of growth: average, intrinsic shortness or familial short stature; delayed or constitutional growth, and attenuated growth. The attenuated growth pattern is classically associated with malnutrition. Adapted from Schaff-Blass et al.[108]; reprinted with permission from the *Journal of Pediatrics*.

A thorough review of systems, with particular attention to the gastrointestinal, respiratory, allergy, and neurologic symptoms, will help

screen for underlying medical conditions. Gastrointestinal review should assess for dysphagia, vomiting, abdominal pain, and stool pattern (frequency, consistency). Symptoms suggestive of oromotor difficulties include choking, gagging, pouching, adverse reactions to foods, or aversion to specific textures or flavors. Symptoms and signs of aspiration are shown in Table 47-6.[43] An underlying inborn error of metabolism should be suspected when symptoms include: recurrent episodic vomiting, lethargy or dehydration; jaundice or liver dysfunction; developmental delay, seizures, hypotonia or other neurologic findings; impaired vision or hearing; organomegaly; and dysmorphic features.[113] The nutritional history should include a comprehensive, culturally appropriate assessment of food intake and feeding history from birth. The timing of introduction of solid foods should be obtained. The current types, quantities, and schedules of all foods and liquids should be obtained, and any adverse reactions to foods should be elicited. A 24-hour recall or 3-day food diary may be used, but a multiday food diary can be challenging in some families. Parents or caregivers should note the amount eaten rather than the amount served. For infants, breastfeeding latch, frequency, night feedings, and any difficulties with breastfeeding should be discussed. Formula type and preparation should be detailed; improper formula mixing can lead to insufficient caloric intake.[114] A feeding behavior history identifies feeding difficulties and may provide insight into their origins. Routines surrounding mealtime should be probed, and information including who provides meals, the setting in which meals are provided, the use of modeling at mealtime, the presence of distractions during mealtimes, and the duration of meals may provide information on specific behaviors that can be modified.[115] To identify potential goals for behavioral therapy, data regarding feeding patterns should be obtained, including: how children are introduced to new foods; whether infants or children are allowed to self-feed; whether cups or utensils are used; whether coercive or restrictive feeding patterns exist; and details regarding rewards or punishment used for feeding behaviors.

TABLE 47-6 Symptoms and Signs of Aspiration

Blinking, eye watering
Throat clearing, grimacing, head tilting
Difficulty handling oral secretions
Poor oral motor co-ordination
Limited endurance/increasing fatigue during feeding
Delayed swallow reflex

Multiple swallows to clear single bolus Wet voice quality during or after feeding Increased congestion during feeds Noisy/wet upper airway sounds associated with feeding Coughing or choking with feeds Change in breathing rhythm or apnea Recurrent wheeze Recurrent respiratory infection
Source: Reprinted with permission from Andrew et al.[43]

Family history should include parental and sibling weight and heights, and the age of onset of puberty. The child's genetic height potential (Table 47-5) should be calculated. A family history of illness that can cause growth failure, such as celiac disease, inflammatory bowel disease, thyroid dysfunction, cystic fibrosis, congenital heart disease, or other major organ disease, in addition to psychiatric or eating disorders, may be helpful in guiding the medical evaluation.

The social history should include the level of parental education, parent employment and job strain, household financial status and living conditions, and the mental health status of caregivers. The caregiver's perception of the child's nutritional status should be obtained as this can influence feeding patterns.[86] Family knowledge of appropriate child nutrition, as well as facilitators and barriers to purchasing, storing, and preparing food should be explored.

Physical Examination

Each child's height, weight and head circumference should be plotted on the appropriate WHO, CDC, or syndrome-specific growth chart (as described in the "Screening and Diagnosis" section above) and interpreted in the context of their family's growth history and pattern. The longitudinal trajectory of a child's growth curve can provide insight into an underlying medical condition or provide support of a diagnosis of health. In addition to the child's growth trajectory, the severity of malnutrition should also be evaluated. Gomez criteria, Waterlow criteria, or anthropometric z-scores can be used to categorize the severity of malnutrition. Severe, chronic malnutrition may indicate risk for refeeding syndrome and necessitate hospitalization for monitoring upon replenishment of calories.

Measurement of triceps skinfold values and mid-arm circumference is used to quantify adipose stores,[116] and can help the clinician distinguish whether slower than expected growth is related to genetic potential,

constitutional delays, or underlying illness. Depleted fat stores accompanying decreased growth velocity supports undernutrition as an underlying etiology. Use of skinfold measurements can be limited by the need for adequate training of personnel, special equipment and availability of reference standards. Published age- and sex-specific reference standards for pediatric skinfold and mid-arm circumference are available.[117]

A comprehensive physical examination to assess for underlying medical conditions, developmental status, and nutritional status should be performed. Table 47-7 summarizes possible findings on physical examination in children with growth failure that are suggestive of underlying medical problems or nutritional deficiencies.[118] Caregiver-child interaction is a critical component of the physical exam, including physical proximity, verbalization toward each other, affect, and eye contact. Attention to signs of abuse or neglect should be sought by examining general hygiene, oral health, and skin. Table 47-8[7,119] lists findings that should alert health care providers to explore neglect.[57]

TABLE 47-7 Physical Examination Findings that Suggest Underlying Cause of Growth Failure or Micronutrient Deficiency

	Clinical Sign(s)	Suggested Underlying Cause
Vital signs	--	Adrenal or thyroid insufficiency
	Hypertension	Renal disease
	Tachypnea/tachycardia	Increased metabolic demands
Skin	Pallor	Anemia
	Poor hygiene	Neglect
	Ecchymoses	Abuse
	Petechiae, ecchymoses	Vitamin C or K deficiency
	Candidiasis	Immunodeficiency
	Eczema	Allergic disease
	Erythema nodosum	Ulcerative colitis, vasculitis
	Diffuse pigmentation	Protein energy
	Xerosis, follicular keratosis	Vitamin A or essential fatty acid deficiency
	Perifolliculosis with blood or pigment	Vitamin C deficiency
	Pellagrous dermatosis	Niacin or tryptophan deficiency

	Scrotal or vulval dermatosis	Riboflavin
Hair	Hair loss	Stress
	Thin, sparse, easily pluckable	Protein energy malnutrition, zinc
Ears	Chronic otitis media	Immunodeficiency, structural orofacial defect
Eyes	Cataracts	Congenital infections, galactosemia
	Papilledema	Increased intracranial pressure
	Uveitis	Vasculitis
	Pale conjunctivae	Iron, folate, or vitamin B_{12} deficiency
	Bitot's spots, conjunctival or corneal xerosis, or keratomalacia	Vitamin A deficiency
	Angular palpebritis	Riboflavin or niacin deficiency
Nose	Nasolabial seborrhea	Riboflavin, niacin, or pyridoxine deficiency
Oropharynx	Aphthous stomatitis	Crohn's disease
	Tonsillar hypertrophy	
Lips	Angular stomatitis or cheilosis	Riboflavin, niacin, iron, or pyridoxine deficiency
Teeth and gums	Delayed tooth eruption	Delayed bone age
	Milk bottle caries	Neglect
	Swollen, bleeding gums	Vitamin C deficiency
Tongue	Ageusia, dysgeusia	Zinc
	Glossitis	Niacin, folate, riboflavin, iron, vitamin B_{12}, pyridoxine, tryptophan deficiency
	Atrophic filiform papillae	Folate, niacin, riboflavin, iron, or vitamin B_{12}
Neck	Thyroid enlargement	Thyroid disease, iodine deficiency
	Parotid enlargement	Protein energy
Chest	Wheeze	Cystic fibrosis, asthma
	Murmur	Congenital malformations
	Cardiomegaly, tachycardia	Thiamine deficiency
Abdomen	Distention, hyperactive bowel sounds	Malabsorption
	Hepatosplenomegaly	Liver disease, glycogen storage disease, malignancy; protein energy malnutrition

Genitourinary	Anomalies	Associated endocrinopathies
	Diaper rashes	Diarrhea, neglect
Rectum	Fistulae	Crohn's disease
	Empty ampulla	Hirschsprung's disease
Extremities	Edema	Hypoalbuminemia, protein energy malnutrition, thiamine deficiency
	Clubbing	Chronic lung disease
Nervous system	Abnormal deep tendon reflexes	Cerebral palsy; vitamin B_{12} or E deficiency
	Developmental delay	Altered caloric intake or requirements
	Cranial nerve palsy	Dysphagia
	Psychomotor changes, confusion irritability	Protein energy malnutrition
	Sensory loss, motor weakness, calf tenderness	Thiamine deficiency
	Loss of vibratory sense, decreased deep tendon reflexes	Vitamin B_{12} or E deficiency
Musculoskeletal system	Loss of muscle mass	Chronic or protein energy malnutrition
	Decreased subcutaneous fat	Protein energy malnutrition
	Craniotabes, frontal bossing, rachitic rosary, epiphyseal enlargement	Vitamin D deficiency
	Epiphyseal enlargement, subperiosteal hemorrhage	Vitamin C deficiency
Nails	Koilonychia	Iron

Source: Adapted from Markowitz et al.; Suskind & Varma.[7,119]

TABLE 47-8 Recognition of Growth Failure Secondary to Abuse or Neglect

Risk factors for neglect

- Parental depression, stress, marital strife, divorce
- Parental history of abuse as a child
- Mental retardation and psychological abnormalities in the parent(s)
- Young and single mothers without social supports
- Domestic violence
- Alcohol or other substance abuse
- Previous child abuse in the family
- Social isolation and/or poverty
- Parents with inadequate adaptive and social skills

Parents who are overly focused on career and/or activities away from home Failure to adhere to medical regimens Lack of knowledge of normal growth and development Infant with low birth or prolonged hospitalization
Signs of possible abuse or neglect that may arise during treatment
Intentional withholding of food Strong beliefs in health and/or nutrition regimens that jeopardize a child's well-being Family resistant to recommended interventions despite multidisciplinary team approach
Source: Adapted from Block, et al.[57]

Observation of a feeding in the clinic can provide insight into the medical and behavioral factors that may be contributing to the feeding difficulties.[120] Home visits by trained professionals, such as a behavioral psychologist or feeding specialist, or videotaped meals may provide additional insight in the infant or child's typical mealtime environment. In addition to identifying oromotor problems that impact chewing or swallowing, family dynamics and environmental interactions can be observed (Table 47-9).[120] Specific behavioral recommendations can be tailored to the individual patient and caregivers based on behaviors observed during the recorded feeding.

Ancillary Studies

Laboratory and radiographic studies should be guided by the history and physical examination, to look for specific underlying disease or complications related to undernutrition. More than three decades ago, Sills reported that laboratory testing in children with poor growth is unlikely to be diagnostically helpful in the absence of suggestive symptoms and/or signs.[112] Among 185 children under age 3 years hospitalized for FTT (weight < 3rd percentile or crossed 2 major percentiles downward), only 36 of the 2607 laboratory tests were of diagnostic utility.[112] All 36 of these test results were suspected based on information obtained during the medical history and physical examination. In a similar 1982 study of 122 hospitalized infants between 1 and 25 months of age, 0.8% of the approximately 40 tests per infant yielded diagnostic information toward the etiology of FTT.[111] These studies suggest that a careful history and physical examination acts as a less invasive and more efficient screen for medical disease in children with growth failure than a "shotgun" approach to laboratory screening. Still, some caution is needed in applying the results of these studies, both published more than 30 years ago, to modern

medical investigation of growth failure. For example, celiac disease, which can present with growth failure as the sole manifestation,[121,122] now has highly accurate screening blood tests and appears to be increasing in prevalence over time. Our understanding of the spectrum of early childhood clinical presentation of eosinophilic esophagitis, a recently identified condition, is evolving.[123,124] New studies to determine the utility of diagnostic testing in growth failure, particularly in the absence of other concerning symptoms or signs, would be helpful to guide clinicians in minimizing the number of tests while confidently identifying treatable causes of growth failure.

TABLE 47-9 Key Details of Feeding Observation

	Key Feeding Behaviors
Parent	Anxiety Inattention Force feeding Bothered by food messes Does not allow child participation in feeding Props bottles Ignores child's feeding signals
Child	Cries Spits up Gags Vomits or ruminates Holds food in mouth Back arching Plays with food or toys at mealtime Takes more than 30 min to eat Refuses to stay seated

Source: Reproduced with permission from Corrales et al.[181]

A complete blood count and iron studies can be considered to check for iron deficiency anemia. Iron deficiency anemia has been commonly reported in growth failure,[97] and children at particular risk include unsupplemented preterm infants, infants consuming excessive cow's milk or juice, or infants who have been exclusively breastfed past 6 months of age without iron supplementation. Urinalysis and urine culture are reasonable to check for urinary tract infection. Isolated renal tubular acidosis is a rare cause of growth failure; screening with venous blood gases rather than serum biochemistry may be more accurate in detecting acidosis.[125] Tissue transglutaminase IgA and total serum IgA should be

performed to screen for celiac disease. Stool studies are indicated for diarrhea, to assess for malabsorption and/or infection. A modified barium swallow may be needed if aspiration or oromotor dyscoordination is suspected. Head ultrasound or MRI should be considered for an increasing head circumference or other symptoms and signs suggestive of an intracranial mass.[126] In immigrant children and international adoptees, there is a higher risk for complications related to undernutrition and recommendations for health care and screening in this group should be followed.[93] For children from areas where HIV is endemic, HIV screening is indicated.

Lead concentration and erythrocyte protoporyphrin levels can be considered, especially in children at high risk for lead exposure. In a small case-control study, high lead levels were more common among children with FTT[127]; however, the current prevalence of high lead levels among US children with growth failure is likely to be lower, given that the prevalence of elevated blood lead levels >= 10 mcg/dL declined from 8.6% in 1988–1991 to 1.4% in 1999–2004.[128] Prevalence of high lead levels among US children has declined even in historically high-risk groups.[128]

For patients presenting with short stature, endocrine conditions such as hypothyroidism or growth hormone deficiency and genetic syndromes such as Russell-Silver, Turner, or Down syndrome must be considered as potential etiologies. Laboratory evaluation for short stature should include TSH, free T4, IGF-1, and IGFBP-3 levels in addition to the laboratory evaluation described above.[109] For female patients, karyotype can be ordered to check for Turner's Syndrome. A bone age radiograph can help determine a child's growth potential.[129] Referral to a pediatric endocrinologist should be made for suspected endocrine abnormalities. Children born small for gestational age with a length-for-age z-score < −2.5 at 2 years should be referred to endocrinology for consideration of growth hormone therapy[130]; in such children, growth hormone treatment started at age 2–4 years and continued over a three-year period has been associated with height gains ranging from 1.2 to 2.0 standard deviations.[25]

MANAGEMENT OF GROWTH FAILURE

The management of growth failure requires long-term treatment that may involve nutritional counseling, behavior modification, feeding therapy, and social work assistance based on the contributing factors identified during

the history and physical examination. Management of mild growth failure typically occurs in an outpatient setting. Successful treatment of any underlying medical conditions will often be associated with improved nutritional status and growth, but attention to nutrition is still needed to reduce complications and mortality.[131]

The medical, nutritional, feeding, and psychosocial aspects in growth failure are often best managed using a multidisciplinary team approach.[132–134] The team may include a physician, dietitian, feeding specialist (usually a speech and language pathologist or occupational therapist), social worker, and behavioral psychologist. In addition to a team approach in the clinic setting, home visits by a home health nurse or community health worker may have some benefit on growth outcomes in childhood.[135,136]

Hospitalization may be warranted in cases of severe malnutrition to allow for monitoring of hemodynamics, electrolytes (refeeding syndrome), and feeding tolerance, or when there are concerns regarding caregiver competence, child neglect or abuse.[137] In cases of severe feeding refusal, admission may be required for initiation of nasogastric tube feeds. Children recovering from severe malnutrition may gain weight safely on caloric intakes as high as 170 kcal/kg/d and protein intakes of 4–5 g/kg/d.[138] However, in the United States, this extreme level of macronutrient needs is unusual.

Caloric needs for catch-up growth can be estimated using prediction equations that take estimated stress and activity into account. In a study of children with growth failure that compared predictive equations to measured resting energy expenditure (REE), the Schofield equation was least likely to underestimate REE.[139] Additional calories for catch-up growth can be calculated in the following manner[82]:

1. Determine the child's Dietary Reference Intake (DRI) for age
2. Determine the child's ideal weight-for-height or ideal weight-for-age (50th percentile on appropriate sex-specific growth chart)
3. Multiply DRI for age by ideal weight
4. Divide by child's actual weight

Protein needs can be calculated in a similar manner. The caloric requirements used as a basis for nutritional counseling should be intermittently adjusted based on weight and height gain over time. Nutritional and feeding interventions should be tailored to the appropriate developmental stage.

For a child with growth failure, the goal for catch-up growth is to cross percentiles upward to attain a normal weight-for-height and ensure that they reach their genetic potential for linear growth. One published criterion for adequate catch-up growth is to exceed, by 2–3 fold,[9] the average age-specific growth rates published by Guo et al.[116] These widely cited growth rates were calculated using data from two small, regional US cohorts that were not nationally representative and were comprised of predominantly formula-fed infants; calculated age-specific mean growth rates based on WHO growth data would differ slightly.

The ideal target amount of catch-up growth, and the speed at which it should be achieved, continues to be debated, with several factors to consider.[140,141] In resource-poor environments, among children born low birth weight or with a history of growth failure, catch-up growth is associated with reduced morbidity and mortality from infections,[141] and improved cognitive function. In a study pooling data from longitudinal cohorts in five developing nations, higher weight gain between 0 and 2 years was associated with more schooling and decreased risk of grade failure.[142] SGA infants who show catch-up growth display improved cognitive outcomes.[143] However, in both developing and developed nations, excessive catch-up growth has also been associated with greater adiposity, insulin resistance, and cardiovascular disease in childhood and adulthood[140]; rapid infant weight gain during the first few months of life may confer the greatest cardiometabolic risk in later life.[140] Further research is required to determine the optimal timing, magnitude and tempo of catch-up growth that best balances associated benefits and potential adverse effects.[140,144]

There is a paucity of high quality studies (longitudinal data and/or randomized controlled trials) examining which nutritional and feeding interventions in toddlers and children are most effective in treating growth failure related to undernutrition in the outpatient setting, especially among children without cognitive impairment. Further, there is no clear consensus as to how best to describe feeding behavior interventions and outcomes that may affect growth. A 2011 systematic review of 31 studies from high-income countries reported that a parental restrictive feeding style was associated with higher BMI, and parental pressure during feeding was associated with lower BMI and weight gain in childhood[145]; the authors pointed out that 25 of the 31 reviewed studies were cross-sectional, raising the possibility of reverse-causality, and 16 different feeding measures were used across studies. Childhood growth is a commonly studied outcome in

intervention trials in developing nations, but the myriad of factors affecting growth failure specific to these settings may limit the generalizability of these studies to developed nations.[146] Consequently, recommendations discussed below are based on limited controlled studies, published guidelines by experts in the field, and our own clinical experience.

Nutritional and Feeding Interventions for Infants

Growth failure in breastfeeding infants may be related to maternal factors, such as milk supply, or infant factors, such as poor latch or suck. These factors should be thoroughly explored as a potential cause for inadequate energy intake, to allow infants continued access to the nutritional, immunologic, and psychosocial benefits of breastfeeding. Maternal milk supply may be increased by stimulating milk supply with breast pumping or increased feeding frequency, advising maternal rest and hydration, targeting stress reduction, and modifying home and work environments to allow for breastfeeding.[4] Consultation with a lactation specialist may assist with infant latch and suck, and may also be helpful for infants who are irritable during feeds as a response to strong milk let down. Infants with neurodevelopmental or anatomical conditions that preclude safe and sufficient breastfeeding may need to drink expressed human milk from a bottle or feeding tube; thickened feeds may be indicated for infants with aspiration.

Infants who are irritable with feeds may have cow's milk protein intolerance, gastroesophageal reflux, or difficulty with swallowing. Treatment of these conditions is discussed in detail elsewhere in this text. Optimal postdischarge nutrition for preterm infants is also discussed elsewhere in this text.

When a breastfeeding infant with inadequate caloric intake is unable to gain weight despite interventions to increase milk supply and improve breastfeeding technique, supplementation with formula should be considered.[147] Growth failure should be verified on WHO growth charts and referral to a lactation consultant should be considered before starting formula supplementation. If formula supplementation is started, powdered formula can be added to expressed breast milk, or bottle feeds with formula can be introduced after breastfeeds based on information obtained during evaluation.

In formula-fed infants, communication with caregivers is a key to understanding the underlying reasons for poor growth so that specific

behaviors that may be contributing to undernutrition can be targeted for intervention. Infrequent, short duration, or small volume of feeds may be identified as underlying reasons for insufficient caloric intake. If an increase in the volume of daily feeds is not tolerated or is ineffective, the caloric density and protein content of formula can be increased.[148] Powdered formula can be increased from the standard 20 kcal/oz up to a maximum of 26 kcal/oz through concentration. If further increases in caloric density are needed, commercially available modulars may be added to a typical maximum of 30 kcal/oz. The daily caloric needs and fluid requirements of infants should be kept in mind and hydration status monitored.

In older infants, age-appropriate solids may be targeted for increasing caloric intake. Adding powdered formula or cooking oil can increase the caloric density of cereals or purees. A dietitian should be consulted when increasing the caloric density of breastmilk, formula, or solids.

Nutritional and Feeding Interventions for Toddlers and Children

The main goal of nutrition therapy is to increase oral intake of calories and nutrients. Healthy, nutrient-rich and calorically dense foods should be offered. The density of calories can be increased in healthy foods through the addition of oils, avocado, sour cream, heavy cream, butter, peanut butter, and cheese. High calorie foods with low-nutritional value such as candies, desserts, and fried foods should be avoided as they can lead to a preference for unhealthful foods associated with adverse cardiometabolic health risks in later life. Fruit juice or sugar-sweetened beverages should be avoided.

Age-appropriate nutritional supplement drinks typically provide 30 kcal/oz, and if used, should be offered after solid foods rather than in lieu of them. Feeding recommendations should be given within the context of cultural preferences, particularly in the case of immigrant families and adoptees. A multivitamin containing iron and zinc may be helpful to ensure that common micronutrient deficiencies are avoided.

Cyproheptadine, an antihistamine appetite stimulant, can be a useful adjunct to stimulate appetite in toddlers and children with inadequate intake. In a systematic review of 12 placebo-controlled randomized controlled trials in children and adults, we found that cyproheptadine is efficacious in achieving weight gain.[149] The dose used in the five pediatric studies of children age 3 months to 14 years ranged from 2 to 12 mg/d

divided two to three times daily, and the medication was generally well-tolerated. Many physicians will alternate a few weeks' use of cyproheptadine with a one to two weeks drug holiday, due to possible tachyphylaxis, but none of the reviewed studies used a cycling regimen.

If feeding difficulties are present, information regarding normal developmental progression of feeding and guidelines regarding behavioral feeding strategies should be provided. It is common for caregivers of a child with growth failure to experience stress around mealtimes or to feel the need to control the amount and type of foods that their child eats.[150] Concrete guidelines regarding the recommended meal schedule, setting, and specific foods to be offered are helpful in both reducing caregiver stress and in treating or preventing problematic feeding behaviors.

As part of normal development, older infants and young children want to touch their food and self-feed as they develop independence and motor ability. Physicians can help parents understand their own concerns about mess, control, and loss associated with their baby growing up.[4] Allowing self-feeding with spoons or fingers, even in children with growth failure, is important to promote healthy eating and development.[151] However, a young child may not be able to feed themselves sufficient calories for growth without assistance. Parents should not force a child to drink or eat predetermined amounts of liquids or foods because forceful feeding can worsen feeding disorders or lead to post-traumatic stress disorder.[79] Feeding and behavioral therapists may be able to assist parents in understanding hunger and satiety cues and avoid restrictive or forceful feeding habits that can perpetuate growth failure or lead to oral aversion.[79]

The concept of the division of feeding responsibilities as discussed by Satter may be helpful for parents to develop strategies to reduce stress around mealtime[151]: parents are responsible for providing safe and appropriate foods in a supportive manner while children are responsible for deciding how much and what they will eat. Parents should be advised that they are shaping the child's food preferences in early life.[152] Most children will refuse new tastes and textures the first time they are exposed, but as foods become more familiar they are more likely to be accepted.[153] If families can set realistic goals about the variety and amount of foods that children eat, mealtime battles, and aversive behaviors may be avoided.

The meal setting and schedule should be structured to promote healthy eating behaviors and improve caloric intake.[79] One case control study found that disorganized mealtime structure and family dysfunction was more prevalent in 4-year-old children whose growth fell below the 10th

percentile for height and weight, compared with controls with normal growth.[154] Offering meals or snacks every three hours and with the child sitting in a high chair or booster seat can promote child recognition of hunger and satiety cues and ability to self-regulate feeding. Limiting meal and snack duration to 30 minutes and avoiding grazing between meals and snacks may facilitate greater intake. Providing foods first and then liquids afterwards may prevent children from feeling full from liquids before eating their meals. Parental modeling of eating nutritious foods and the mechanisms of chewing may encourage child intake of nutritious foods. Elimination of distractions such as TV or toys can also be helpful. Making mealtime social with caregivers providing positive comments about food and eating may help encourage children to take in sufficient amounts of food.

For infants or toddlers with severe eating disorders, such as severe feeding refusal or selectivity among cognitively impaired children, long-term treatment in intensive day programs or inpatient treatment may be needed.[76] The behavioral modification interventions used in programs for feeding difficulties vary, but usually combine more than one technique, including escape prevention, extinction, contingent or positive reinforcement, texture fading, planned ignoring, among other techniques.[76] More research is needed regarding which techniques and what combinations of techniques might be effective in nondevelopmentally delayed children.

CONSEQUENCES AND PROGNOSIS

Limited information exists regarding the long-term growth outcomes of full-term infants with growth failure in developed countries. A few small case-control studies suggest that children with severe growth failure in early life experience improvement in their somatic growth, but continue to have lower weights and heights than age-matched controls later in childhood.[155–158] Boddy et al.[158] reported that among 42 children from a 1986 UK birth cohort with a WFA < 3rd percentile for at least three months duration identified at 12 months of age, mean weight-for-age z-score improved from −2.09 at 12 months of age to −0.97 at 6 years of age. Among controls from the same birth cohort matched for age, birth weight, gender, social position, ethnic group, and area of residence, mean weight for age z-score was 0.094 at 12 months and 0.68 at 6 years. Mean height for age z-score was −1.26 in cases and −1.76 in controls at 12 months, and −0.91 in cases and 0.27 in controls at 6 years. A 2005 systematic review

identified eight controlled studies and five uncontrolled studies assessing the growth and cognitive outcomes of children with FTT[159]; the authors concluded that children with FTT were shorter and weighed less than control children at follow-up ranging from three to nine years later, but that few children were below the 3rd percentile at follow-up. Subsequent to publication of the systematic review, Drewett et al.[156] compared children from a 1987 birth cohort within the slowest 5th percentile of weight gain, to 91 controls, and found that the adjusted mean difference at age 12 was 4.3 cm for height, and 7.2 kg for weight. Interpretation of these studies is challenging because of the potential for selection bias in case-control studies, and the inability to clearly delineate the role of nutrition, genetics, or other factors in the growth of study subjects. For example, only 2 of the 5 case-control studies in the systematic review controlled for parental height.[159]

Among term infants born small for gestational age, the majority will show improvement in anthropometric parameters within the first 2 years of life,[160,161] but mean weight and height in childhood and adulthood remain lower than for infants of normal birth weight.[162] In the National Perinatal Collaborative Cohort, compared with children with a birth weight > 2500 g, children born at ≥ 37 weeks with a birth weight ≤ 2500 g had lower mean weight-for-age z-score (−0.60 SD) and height-for-age z-score (−0.50 SD) at age 7 years.[27] These differences correspond to an average height deficit of 2 cm and an average weight deficit of 1.6 kg at 7 years of age. In the same study, 22% of siblings born SGA had age 7 weight < 10th percentile, compared with 12% of siblings not born SGA, suggesting that genetic rather than environmental factors account for the persistent effects of SGA on growth.[27] In a French cohort, term infants born with a birth weight or length below the 3rd percentile were a mean of 3.9–4.5 cm shorter at age 20 years, than infants born with a normal birth weight (25th–75th percentile).[163]

Several retrospective and prospective case-control studies show that the majority of preterm infants with growth failure do attain normal weights and heights in later life, but compared to term, normal birth weight peers, infants born with extremely or very low birth weight exhibit lower mean weight-for-age and/or height-for-age in the first few years of life,[20,164–166] in adolescence,[167] and in adulthood.[166,168] One small study of 195 US VLBW preterm infants (birth weight < 1500 g) suggested sex-specific differences in adult body composition.[168] Compared with normal birth weight, term, sex-matched controls, VLBW males were a mean of

3.1 cm shorter ($P < .01$) and weighed 10.5 kg less at age 20 years, whereas females were a mean of 1.2 cm shorter and weighed 2 kg less and these differences were not statistically significant. Nevertheless, the majority of VLBW infants achieved adult weight and stature within normal limits. Among male VLBW infants, 11% had adult height-for-age z-score < −2 and 7% had adult weight-for-age z-score < −2. Among females, 2% had adult height-for-age z-score < −2 and 5% had adult weight-for-age < −2; the mean differences in these outcomes were not statistically significant, although there was probably insufficient power to detect a subtle effect. Moderate (32–35 weeks)[169] and late preterm birth (34–36 weeks)[19] has also been associated with lower weight and height in childhood.

Separating the individual effects of preterm birth and small for gestational age on growth outcomes was difficult in early cohort studies which tended to use weight-based inclusion criteria, but recent studies have attempted to examine the individual effects. Preterm infants with short maternal height and those born SGA appear to be at the highest risk of short stature.[166] In one Dutch study of 1338 children born at < 32 weeks gestation and/or birth weight < 1500 g, the height of 10-year-old children born SGA was −0.29 SD lower than for children born AGA.[170] In a French study of 1,597 infants born at < 33 weeks gestation, 24% of infants born SGA had short stature at age 5 year (height z-score for age and sex < −2).[171] International adoptees from orphanages often have significant catch-up growth in weight and height after adoption.[93,172] A meta-analysis of 33 studies showed substantial gains in height and weight after adoptive placement.[172] Among 148, 7 to 59-month-old children adopted from Eastern Europe, 62% showed a gain of >+0.5 units in height z-score at six months after adoption.[173] Only 7% of children had a height z-score < −2 at six months after adoption, and two of these children had growth hormone deficiency.[173] Physicians should advise adoptive parents to allow three to six months for evidence of improved growth velocity before intervening, unless a medical condition is suspected. This period of monitoring may help avoid the negative consequences of rapid weight gain, including obesity, cardiovascular, and adverse metabolic outcomes.[93]

Early life feeding difficulties have been associated with long-term feeding habits. Children with picky eating or feeding refusal may have feeding problems later in childhood.[79,174] Less commonly, an undiagnosed anxiety or autistic spectrum disorder may be uncovered later in childhood.[4,79] Children with severe eating disorders may require long-term intensive multimodal therapy that includes behavior modification, family

counseling, education, and feeding therapy. Some children with underlying medical or neurologic disorders may need ongoing enteral tube feeding supplementation.

In developing nations, severe childhood malnutrition has been clearly associated with adverse cognitive, health, and social outcomes.[175] In the United States and other developed nations, the effect of slow weight gain on cognitive outcomes is less clear.[159,176,177] Two systematic reviews published several years ago examining the cognitive sequelae of failure-to-thrive came to differing conclusions. Definitions for FTT were variable across studies. A 2004 systematic review of 31 studies (11 uncontrolled) of "nonorganic failure-to-thrive" concluded that "FTT in infancy is associated with adverse intellectual outcomes sufficiently large to be of importance at a population level."[177] The overall weighted standard mean difference for 10 controlled studies was estimated at −0.28, corresponding to a mean IQ difference of 4.2 points. The magnitude of the associations with cognitive outcomes was larger among the three studies where cases were identified from hospitals or specialist clinics vs. population screening. The effect size decreased substantially between infancy and later in childhood; in a few longitudinal studies with repeat measures, the mean summary effect estimates (D-statistics) were −0.51 to −0.61 at 1 year, and −0.22 to −0.24 in the same children at 6 years of age. A 2005 systematic review included 13 studies of failure-to-thrive for any reason and had growth or psychomotor development outcomes at 3 years or older, of which eight had a comparison group and two were randomized controlled trials.[159] A meta-analysis of the four studies that reported IQ scores at follow-up yielded a pooled standardized mean difference of -0.22 (95% CI −0.41 to −0.03), roughly equivalent to −3 IQ points.[159] Although the effect size was fairly consistent with the prior meta-analysis, the authors felt that this effect size was "of questionable clinical significance." They also concluded that it was unclear to what extent observed differences reflected confounding by variables such as birth weight, maternal height, and socioeconomic status, which were not consistently controlled for across studies; in most of the studies, the control groups displayed mean cognitive testing scores that were lower than population norms, bringing into question whether the results are generalizable to populations with higher cognitive function.

Nutritional, social, environmental, and genetic factors have the potential to modify the associations between growth failure in early childhood and later cognitive and other health outcomes. The association between chronic iron deficiency anemia and long-term irreversible

developmental delays best illustrates the link between micronutrient deficiency and developmental outcomes.[178,179] In addition to iron, it is possible that other micronutrient deficiencies could modify the associations between early growth failure and health outcomes in later life. In one small intervention study of weekly structured play home visits delivered over two years, malnourished children who received the intervention had better cognitive test scores at age 6, than malnourished controls who had not received the intervention.[180] These findings emphasize the importance of comprehensively addressing the psychosocial, medical, nutritional, and environmental factors to optimize anthropometric, cognitive, and other health outcomes for each child who presents with growth failure.

SUMMARY

Growth failure usually arises from the interaction of medical, social, and environmental factors that result in a final common pathway of undernutrition. Commonly associated risk factors include short parental height, preterm and/or SGA size at birth, maternal depression during infancy, and behavioral feeding disorders. Each potential contributing domain is best explored by a multidisciplinary team to tailor the assessment and management. The history and physical examination should guide the judicious use of laboratory investigations. Treatment in the outpatient setting focuses on optimizing the feeding environment, schedule and caloric density of offered foods and beverages. Key research priorities include development of consensus regarding the optimal criteria for the diagnosis of growth failure and feeding disorders; additional randomized studies of feeding interventions; and long-term prospective studies of health outcomes among children with growth failure in developed nations.

REFERENCES

1. Holt L. *The Diseases of Infancy and Childhood: For the Use of Students and Practitioners of Medicine*. New York, NY: Appleton and Company; 1879.
2. Jaffe AC. Failure to thrive: current clinical concepts. *Pediatr Rev*. 2011;32:100–107.
3. Olsen EM. Failure to thrive: still a problem of definition. *Clin Pediatr (Phila)*. 2006;45:1–6.
4. Gahagan S. Failure to thrive: a consequence of undernutrition. *Pediatr Rev*. 2006;27:e1–e11.
5. Wilcox WD, Nieburg P, Miller DS. Failure to thrive. A continuing problem of definition. *Clin Pediatr (Phila)*. 1989;28:391–394.

6. Waterlow JC, Buzina R, Keller W, et al. The presentation and use of height and weight data for comparing the nutritional status of groups of children under the age of 10 years. *Bull World Health Organ*. 1977;55:489–498.
7. Markowitz R, Duggan C, Watkins JB. Failure to thrive: malnutrition in the pediatric outpatient setting. In: Duggan C, Watkins JB, Walker WA, eds. *Nutrition in Pediatrics: Basic Science, Clinical Application*. 4th ed. Hamilton, Canada: BC Decker; 2008:xvii, 923.
8. Blair PS, Drewett RF, Emmett PM, et al. Family, socioeconomic and prenatal factors associated with failure to thrive in the Avon Longitudinal Study of Parents and Children (ALSPAC). *Int J Epidemiol*. 2004;33:839–847.
9. American Academy of Pediatrics. *Committee on Nutrition., Barness LA. Pediatric Nutrition Handbook*. 6th ed. Elk Grove Village, IL: American Academy of Pediatrics; 2009.
10. Waterlow JC. Classification and definition of protein-calorie malnutrition. *Br Med J*. 1972;3:566–569.
11. Drotar D. Sampling issues in research with nonorganic failure-to-thrive children. *J Pediari Psychol*. 1990;15:255–272.
12. Gayle HD, Dibley MJ, Marks JS, Trowbridge FL. Malnutrition in the first two years of life. The contribution of low birth weight to population estimates in the United States. *Am J Dis Child*. 1987;141:531–534.
13. Mei Z, Ogden CL, Flegal KM, Grummer-Strawn LM. Comparison of the prevalence of shortness, underweight, and overweight among US children aged 0 to 59 months by using the CDC 2000 and the WHO 2006 growth charts. *J Pediatr*. 2008;153:622–628.
14. Grummer-Strawn LM, Reinold C, Krebs NF. Use of World Health Organization and CDC growth charts for children aged 0-59 months in the United States. *MMWR Recomm Rep*. 2010;59:1–15.
15. Fryar CD, Ogden CL. *Prevalence of Underweight Among Children and Adolescents Aged 2-19 Years: United States, 1963–1965 Through 2007-2010*. Atlanta, GA: CDC\NCHS Health E-Stat; 2012.
16. Frank DA, Zeisel SH. Failure to thrive. *Pediatr Clin North Am*. 1988;35:1187–1206.
17. Lutter CK, Daelmans BM, de Onis M, et al. Undernutrition, poor feeding practices, and low coverage of key nutrition interventions. *Pediatrics*. 2011;128:e1418–e1427.
18. Shields B, Wacogne I, Wright CM. Weight faltering and failure to thrive in infancy and early childhood. *BMJ*. 2012;345:e5931.
19. Goyal NK, Fiks AG, Lorch SA. Persistence of underweight status among late preterm infants. *Arch Pediatr Adolesc Med*. 2012;166:424–430.
20. Casey PH, Kraemer HC, Bernbaum J, et al. Growth patterns of low birth weight preterm infants: a longitudinal analysis of a large, varied sample. *J Pediatr*. 1990;117:298–307.
21. Guo SS, Roche AF, Chumlea WC, et al. Growth in weight, recumbent length, and head circumference for preterm low-birthweight infants during the first three years of life using gestation-adjusted ages. *Early Hum Dev*. 1997;47:305–325.
22. Santos IS, Matijasevich A, Domingues MR, et al. Late preterm birth is a risk factor for growth faltering in early childhood: a cohort study. *BMC Pediatr*. 2009;9:71.
23. Ehrenkranz RA, Younes N, Lemons JA, et al. Longitudinal growth of hospitalized very low birth weight infants. *Pediatrics*. 1999;104:280–289.
24. Saenger P, Czernichow P, Hughes I, Reiter EO. Small for gestational age: short

stature and beyond. *Endocr Rev*. 2007;28:219–251.
25. Clayton PE, Cianfarani S, Czernichow P, et al. Management of the child born small for gestational age through to adulthood: a consensus statement of the International Societies of Pediatric Endocrinology and the Growth Hormone Research Society. *J Clin Endocrinol Metab*. 2007;92:804–810.
26. Maulik D. Fetal growth compromise: definitions, standards, and classification. *Clin Obstet Gynecol*. 2006;49:214–218.
27. Strauss RS, Dietz WH. Growth and development of term children born with low birth weight: effects of genetic and environmental factors. *J Pediatr*. 1998;133:67–72.
28. De Jesus LC, Pappas A, Shankaran S, et al. Outcomes of small for gestational age infants born at <27 weeks' gestation. *J Pediatr*. 2013;163:55–60.e1–e3.
29. Hindmarsh PC, Geary MP, Rodeck CH, et al. Intrauterine growth and its relationship to size and shape at birth. *Pediatr Res*. 2002;52:263–268.
30. Bryan SM, Hindmarsh PC. Normal and abnormal fetal growth. *Horm Res*. 2006;65 (suppl 3):19–27.
31. Villar J, Smeriglio V, Martorell R, et al. Heterogeneous growth and mental development of intrauterine growth-retarded infants during the first 3 years of life. *Pediatrics*. 1984;74:783–791.
32. Strauss RS, Dietz WH. Effects of intrauterine growth retardation in premature infants on early childhood growth. *J Pediatr*. 1997;130:95–102.
33. Blecker U, Mehta DI, Davis R, et al. Nutritional problems in patients who have chronic disease. *Pediatr Rev*. 2000;21:29–32.
34. Kelleher KJ, Casey PH, Bradley RH, et al. Risk factors and outcomes for failure to thrive in low birth weight preterm infants. *Pediatrics*. 1993;91:941–948.
35. Dusick AM, Poindexter BB, Ehrenkranz RA, Lemons JA. Growth failure in the preterm infant: can we catch up? *Semin Perinatol*. 2003;27:302–310.
36. Samson-Fang LJ, Stevenson RD. Identification of malnutrition in children with cerebral palsy: poor performance of weight-for-height centiles. *Dev Med Child Neuro*. 2000;42:162–168.
37. Stallings VA, Charney EB, Davies JC, Cronk CE. Nutrition-related growth failure of children with quadriplegic cerebral palsy. *Dev Med Child Neuro*. 1993;35:126–138.
38. Stallings VA, Charney EB, Davies JC, Cronk CE. Nutritional status and growth of children with diplegic or hemiplegic cerebral palsy. *Dev Med Child Neuro*. 1993;35:997–1006.
39. Stallings VA, Cronk CE, Zemel BS, Charney EB. Body composition in children with spastic quadriplegic cerebral palsy. *J Pediatr*. 1995;126:833–839.
40. Marchand V, Motil KJ. Nutrition support for neurologically impaired children: a clinical report of the North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition. *J Pediatr Gastroenterol Nutr*. 2006;43:123–135.
41. Fung EB, Samson-Fang L, Stallings VA, et al. Feeding dysfunction is associated with poor growth and health status in children with cerebral palsy. *J Am Diet Assoc*. 2002;102:361–373.
42. Sullivan PB, Lambert B, Rose M, et al. Prevalence and severity of feeding and nutritional problems in children with neurological impairment: Oxford Feeding Study. *Dev Med Child Neuro*. 2000;42:674–680.
43. Andrew MJ, Parr JR, Sullivan PB. Feeding difficulties in children with cerebral palsy. *Arch Dis Child Educ Pract Ed*. 2012;97:222–229.
44. Hogan SE. Energy requirements of children with cerebral palsy. *Can J Diet Pract Res*. 2004;65:124–130.

45. Walker JL, Bell KL, Boyd RN, Davies PS. Energy requirements in preschool-age children with cerebral palsy. *Am J Clin Nutr*. 2012;96:1309–1315.
46. Magnuson B, Peppard A, Auer Flomenhoft D. Hypocaloric considerations in patients with potentially hypometabolic disease States. *Nutr Clin Pract*. 2011;26:253–260.
47. Pemberton C, Moxness K, German M, et al., eds. *Mayo Clinic Diet Manual*. Philadelphia, PA: BC Decker, Inc; 1988.
48. Zarate YA, Martin LJ, Hopkin RJ, et al. Evaluation of growth in patients with isolated cleft lip and/or cleft palate. *Pediatrics*. 2010;125:e543–e549.
49. Montagnoli LC, Barbieri MA, Bettiol H, et al. Growth impairment of children with different types of lip and palate clefts in the first 2 years of life: a cross-sectional study. *J Pediatr (Rio J)*. 2005;81:461–465.
50. McHeik JN, Levard G. Growth in infants in the first two years of life after neonatal repair for unilateral cleft lip and palate. *Int J Pediatr Otorhinolaryngol*. 2010;74:465–468.
51. Bessell A, Hooper L, Shaw WC, et al. Feeding interventions for growth and development in infants with cleft lip, cleft palate or cleft lip and palate. *Cochrane Database Syst Rev*. 2011;2:CD003315.
52. Richter GT. Management of oropharyngeal dysphagia in the neurologically intact and developmentally normal child. *Curr Opin Otolaryngol Head Neck Surg*. 2010;18:554–563.
53. Cooper-Brown L, Copeland S, Dailey S, et al. Feeding and swallowing dysfunction in genetic syndromes. *Dev Disabil Res Rev*. 2008;14:147–157.
54. Tauman R, Levine A, Avni H, et al. Coexistence of sleep and feeding disturbances in young children. *Pediatrics*. 2011;127:e615–e621.
55. Bonuck KA, Freeman K, Henderson J. Growth and growth biomarker changes after adenotonsillectomy: systematic review and meta-analysis. *Arch Dis Child*. 2009;94:83–91.
56. Listernick R, Christoffel K, Pace J, Chiaramonte J. Severe primary malnutrition in US children. *Am J Dis Child*. 1985;139:1157–1160.
57. Block RW, Krebs NF; American Academy of Pediatrics Committee on Child A, Neglect, American Academy of Pediatrics Committee on N. Failure to thrive as a manifestation of child neglect. *Pediatrics*. 2005;116:1234–1237.
58. Wright CM, Parkinson KN, Drewett RF. The influence of maternal socioeconomic and emotional factors on infant weight gain and weight faltering (failure to thrive): data from a prospective birth cohort. *Arch Dis Child*. 2006;91:312–317.
59. Coleman-Jensen A, Nord M, Andrews M, Carlson S. *Household Food Security in the United States in 2011*. Washington, DC: U.S. Department of Agriculture, Economic Research Service; 2012.
60. Black MM, Cutts DB, Frank DA, et al. Special Supplemental Nutrition Program for Women, Infants, and Children participation and infants' growth and health: a multisite surveillance study. *Pediatrics*. 2004;114:169–176.
61. Cutts DB, Meyers AF, Black MM, et al. US Housing insecurity and the health of very young children. *Am J Public Health*. 2011;101:1508–1514.
62. Weston JA, Colloton M, Halsey S, et al. A legacy of violence in nonorganic failure to thrive. *Child Abuse Negl*. 1993;17:709–714.
63. Altemeier WA 3rd, O'Connor SM, Sherrod KB, Vietze PM. Prospective study of antecedents for nonorganic failure to thrive. *J Pediatr*. 1985;106:360–365.
64. Stewart RC. Maternal depression and infant growth: a review of recent evidence. *Matern Child Nutr*. 2007;3:94–107.
65. Surkan PJ, Kennedy CE, Hurley KM, Black MM. Maternal depression and early

childhood growth in developing countries: systematic review and meta-analysis. *Bull World Health Organ*. 2011;89:608–615.
66. Casey P, Goolsby S, Berkowitz C, et al. Maternal depression, changing public assistance, food security, and child health status. *Pediatrics*. 2004;113:298–304.
67. O'Brien LM, Heycock EG, Hanna M, et al. Postnatal depression and faltering growth: a community study. *Pediatrics*. 2004;113:1242–1247.
68. Wojcicki JM, Holbrook K, Lustig RH, et al. Chronic maternal depression is associated with reduced weight gain in latino infants from birth to 2 years of age. *PloS One*. 2011;6:e16737.
69. Chatoor I, Ganiban J, Hirsch R, et al. Maternal characteristics and toddler temperament in infantile anorexia. *J Am Acad Child Adolesc Psychiatry*. 2000;39:743–751.
70. Black MM, Papas MA, Hussey JM, et al. Behavior problems among preschool children born to adolescent mothers: effects of maternal depression and perceptions of partner relationships. *J Clin Child Adolesc Psychol*. 2002;31:16–26.
71. Surkan PJ, Ettinger AK, Ahmed S, et al. Impact of maternal depressive symptoms on growth of preschool- and school-aged children. *Pediatrics*. 2012;130:e847–e855.
72. Robb AS. Eating disorders in children. Diagnosis and age-specific treatment. *Psychiatr Clin North Am*. 2001;24:259–270.
73. Chatoor I, Ganiban J, Surles J, Doussard-Roosevelt J. Physiological regulation and infantile anorexia: a pilot study. *J Am Acad Child Adolesc Psychiatry*. 2004;43:1019–1025.
74. Stein MT, Robinson J. Feeding problems, sleep disturbances, and negative behaviors in a toddler. *J Dev Behav Pediatr*. 2001;22:S81–S85.
75. McDougall P, Drewett RF, Hungin AP, Wright CM. The detection of early weight faltering at the 6-8-week check and its association with family factors, feeding and behavioural development. *Arch Dis Child*. 2009;94:549–552.
76. Williams KE, Field DG, Seiverling L. Food refusal in children: a review of the literature. *Res Dev Disabil*. 2010;31:625–633.
77. Davies WH, Satter E, Berlin KS, et al. Reconceptualizing feeding and feeding disorders in interpersonal context: the case for a relational disorder. *J Fam Psychol*. 2006;20:409–417.
78. Nicholls D, Bryant-Waugh R. Eating disorders of infancy and childhood: definition, symptomatology, epidemiology, and comorbidity. *Child Adolesc Psychiatr Clin N Am*. 2009;18:17–30.
79. Chatoor I. *Diagnosis and Treatment of Feeding Disorders in Infants, Toddlers, and Young Children*. Washington, DC: Zero To Three; 2009.
80. Zero to Three (Organization). *DC:0-3R Revision Task Force. DC:0-3R : Diagnostic Classification of Mental Health and Developmental Disorders of Infancy and Early Childhood. Rev. ed*. Washington, DC: Zero To Three Press; 2005.
81. Bryant-Waugh R, Markham L, Kreipe RE, Walsh BT. Feeding and eating disorders in childhood. *Int J Eat Disord*. 2010;43:98–111.
82. Boghani M, Huh SY. Growth failure. In: Duggan C, Sonneville K, eds. *Manual of Pediatric Nutrition*, 5th ed. Shelton, CT: People's Medical Publishing House; 2014: 401–416.
83. Carvalho NF, Kenney RD, Carrington PH, Hall DE. Severe nutritional deficiencies in toddlers resulting from health food milk alternatives. *Pediatrics*. 2001;107:E46.

84. Weston JA, Stage AF, Hathaway P, et al. Prolonged breast-feeding and nonorganic failure to thrive. *Am J Dis Child*. 1987;141:242–243.
85. O'Connor ME, Szekely LJ. Frequent breastfeeding and food refusal associated with failure to thrive. A manifestation of the vulnerable child syndrome. *Clin Pediatr (Phila)*. 2001;40:27–33.
86. Pugliese MT, Weyman-Daum M, Moses N, Lifshitz F. Parental health beliefs as a cause of nonorganic failure to thrive. *Pediatrics*. 1987;80:175–182.
87. Hyams JS, Leichtner AM. Apple juice. An unappreciated cause of chronic diarrhea. *Am J Dis Child*. 1985;139:503–505.
88. Lifshitz F, Ament ME, Kleinman RE, et al. Role of juice carbohydrate malabsorption in chronic nonspecific diarrhea in children. *J Pediatr*. 1992;120:825–829.
89. Smith MM, Lifshitz F. Excess fruit juice consumption as a contributing factor in nonorganic failure to thrive. *Pediatrics*. 1994;93:438–443.
90. Sinagra JL, Bordignon V, Ferraro C, et al. Unnecessary milk elimination diets in children with atopic dermatitis. *Pediatr Dermatol*. 2007;24:1–6.
91. Noimark L, Cox HE. Nutritional problems related to food allergy in childhood. *Pediatr Allergy Immunol*. 2008;19:188–195.
92. Tierney EP, Sage RJ, Shwayder T. Kwashiorkor from a severe dietary restriction in an 8-month infant in suburban Detroit, Michigan: case report and review of the literature. *Int J Dermatol*. 2010;49:500–506.
93. Mason P, Narad C. Long-term growth and puberty concerns in international adoptees. *Pediatr Clin North Am*. 2005;52:1351–1368, vii.
94. Jenista JA. The immigrant, refugee, or internationally adopted child. *Pediatr Rev*. 2001;22:419–429.
95. Health care for children of immigrant families. American Academy of Pediatrics. Committee on Community Health Services. *Pediatrics*. 1997;100:153–156.
96. Miller LC, Hendrie NW. Health of children adopted from China. *Pediatrics*. 2000;105:E76.
97. Black MM, Dubowitz H, Casey PH, et al. Failure to thrive as distinct from child neglect. *Pediatrics*. 2006;117:1456–1458.
98. Skuse DH, Gill D, Reilly S, et al. Failure to thrive and the risk of child abuse: a prospective population survey. *J Med Screen*. 1995;2:145–149.
99. Krieger I. Food restriction as a form of child abuse in ten cases of psychosocial deprivation dwarfism. *Clin Pediatr (Phila)*. 1974;13:127–133.
100. Kellogg ND, Lukefahr JL. Criminally prosecuted cases of child starvation. *Pediatrics*. 2005;116:1309–1316.
101. Roesler TA, Jenny C, American Academy of Pediatrics. *Medical Child Abuse: Beyond Munchausen Syndrome by Proxy*. Elk Grove Village, IL: American Academy of Pediatrics; 2009.
102. Brown P, Tierney C. Munchausen syndrome by proxy. *Pediatr Rev*. 2009;30:414–415.
103. Mash C, Frazier T, Nowacki A, et al. Development of a risk-stratification tool for medical child abuse in failure to thrive. *Pediatrics* 2011;128:e1467–e1473.
104. Mei Z, Grummer-Strawn LM. Comparison of changes in growth percentiles of US children on CDC 2000 growth charts with corresponding changes on WHO 2006 growth charts. *Clin Pediatr (Phila)*. 2011;50:402–407.
105. Bithoney WG, Dubowitz H, Egan H. Failure to thrive/growth deficiency. *Pediatr Rev*. 1992;13:453–460.
106. Mei Z, Grummer-Strawn LM, Thompson D, Dietz WH. Shifts in percentiles of growth during early childhood: analysis of longitudinal data from the California

Child Health and Development Study. *Pediatrics*. 2004;113:e617–e627.
107. Tanner JM, Goldstein H, Whitehouse RH. Standards for children's height at ages 2-9 years allowing for heights of parents. *Arch Dis Child*. 1970;45:755–762.
108. Schaff-Blass E, Burstein S, Rosenfield RL. Advances in diagnosis and treatment of short stature, with special reference to the role of growth hormone. *J Pediatr*. 1984;104:801–813.
109. Halac I, Zimmerman D. Evaluating short stature in children. *Pediatr Ann*. 2004;33:170-176.
110. Weintraub B. Growth. *Pediatr Rev*. 2011;32:404–406.
111. Berwick DM, Levy JC, Kleinerman R. Failure to thrive: diagnostic yield of hospitalisation. *Arch Dis Child*. 1982;57:347–351.
112. Sills RH. Failure to thrive. The role of clinical and laboratory evaluation. *Am J Dis Child*. 1978;132:967–969.
113. Ficicioglu C, An Haack K. Failure to thrive: when to suspect inborn errors of metabolism. *Pediatrics*. 2009;124:972–979.
114. McJunkin JE, Bithoney WG, McCormick MC. Errors in formula concentration in an outpatient population. *J Pediatr*. 1987;111:848–850.
115. Reau NR, Senturia YD, Lebailly SA, Christoffel KK. Infant and toddler feeding patterns and problems: normative data and a new direction. Pediatric Practice Research Group. *J Dev Behav Pediatr*. 1996;17:149–153.
116. Guo SM, Roche AF, Fomon SJ, et al. Reference data on gains in weight and length during the first two years of life. *J Pediatr*. 1991;119:355–362.
117. Addo OY, Himes JH. Reference curves for triceps and subscapular skinfold thicknesses in US children and adolescents. *Am J Clin Nutr*. 2010;91:635–642.
118. Collins J, Mezey AP. Failure to thrive. In: Shelov SP MA, Edelman CM, Barnett HL, eds. *Primary Care Pediatrics*. Norwalk, CT: Appleton-Century-Crofts; 1984:327–329.
119. Suskind R, Varma R. Assessment of nutritional status of children. *Pediatr Rev*. 1984;5:195–202.
120. Rudolph CD, Link DT. Feeding disorders in infants and children. *Pediatr Clin North Am*. 2002;49:97–112, vi.
121. Steens RF, Csizmadia CG, George EK, et al. A national prospective study on childhood celiac disease in the Netherlands 1993-2000: an increasing recognition and a changing clinical picture. *J Pediatr*. 2005;147:239–243.
122. Hoffenberg EJ, Emery LM, Barriga KJ, et al. Clinical features of children with screening-identified evidence of celiac disease. *Pediatrics*. 2004;113:1254–1259.
123. Mukkada VA, Haas A, Maune NC, et al. Feeding dysfunction in children with eosinophilic gastrointestinal diseases. *Pediatrics*. 2010; 126(3):e672–e677.
124. Haas AM, Maune NC. Clinical presentation of feeding dysfunction in children with eosinophilic gastrointestinal disease. *Immunol Allergy Clin North Am*. 2009;29:65–75, ix.
125. Adedoyin O, Gottlieb B, Frank R, et al. Evaluation of failure to thrive: diagnostic yield of testing for renal tubular acidosis. *Pediatrics*. 2003;112:e463.
126. Wilne S, Koller K, Collier J, et al. The diagnosis of brain tumours in children: a guideline to assist healthcare professionals in the assessment of children who may have a brain tumour. *Arch Dis Child*. 2010;95:534–539.
127. Bithoney WG. Elevated lead levels in children with nonorganic failure to thrive. *Pediatrics*. 1986;78:891–895.
128. Jones RL, Homa DM, Meyer PA, et al. Trends in blood lead levels and blood lead testing among US children aged 1 to 5 years, 1988–2004. *Pediatrics*. 2009;123:e376–e385.

129. Cohen P, Rogol AD, Deal CL, et al. Consensus statement on the diagnosis and treatment of children with idiopathic short stature: a summary of the Growth Hormone Research Society, the Lawson Wilkins Pediatric Endocrine Society, and the European Society for Paediatric Endocrinology Workshop. *J Clin Endocrinol Metab*. 2008;93:4210–4217.
130. Houk CP, Lee PA. Early diagnosis and treatment referral of children born small for gestational age without catch-up growth are critical for optimal growth outcomes. *Int J Pediatr Endocrinol*. 2012;2012:11.
131. Chen LC, Chowdhury A, Huffman SL. Anthropometric assessment of energy-protein malnutrition and subsequent risk of mortality among preschool aged children. *Am J Clin Nutr*. 1980;33:1836–1845.
132. Peterson KE, Washington J, Rathbun JM. Team management of failure to thrive. *J Am Diet Assoc*. 1984;84:810–815.
133. Bithoney WG, McJunkin J, Michalek J, et al. The effect of a multidisciplinary team approach on weight gain in nonorganic failure-to-thrive children. *J Dev Behav Pediatr*. 1991;12:254–258.
134. Sampson P. *Interdiscipinary Teamwork*. Baltimore, MD: Paul H. Brookes; 1999.
135. Black MM, Dubowitz H, Hutcheson J, et al. A randomized clinical trial of home intervention for children with failure to thrive. *Pediatrics*. 1995;95:807–814.
136. Black MM, Dubowitz H, Krishnakumar A, Starr RH Jr. Early intervention and recovery among children with failure to thrive: follow-up at age 8. *Pediatrics*. 2007;120:59–69.
137. Schmitt BD, Mauro RD. Nonorganic failure to thrive: an outpatient approach. *Child Abuse Negl*. 1989;13:235–248.
138. Fjeld CR, Schoeller DA, Brown KH. Body composition of children recovering from severe protein-energy malnutrition at two rates of catch-up growth. *Am J Clin Nutr*. 1989;50:1266–1275.
139. Sentongo TA, Tershakovec AM, Mascarenhas MR, et al. Resting energy expenditure and prediction equations in young children with failure to thrive. *J Pediatr*. 2000;136:345–350.
140. Jain V, Singhal A. Catch up growth in low birth weight infants: striking a healthy balance. *Rev Endocr Metab Disord*. 2012;13:141–147.
141. Adair LS. Developing world perspective: the importance of growth for short-term health. *Nestle Nutr Workshop Ser Pediatr Program*. 2010;65:71–79.
142. Martorell R, Horta BL, Adair LS, et al. Weight gain in the first two years of life is an important predictor of schooling outcomes in pooled analyses from five birth cohorts from low- and middle-income countries. *J Nutr*. 2010;140:348–354.
143. Lee PA, Houk CP. Cognitive and psychosocial development concerns in children born small for gestational age. *Pediatr Endocrinol Rev*. 2012;10:209–216.
144. Belfort MB, Gillman MW. Healthy infant growth: what are the trade-offs in the developed world? *Nestle Nutr Inst Workshop Ser*. 2013;71:171–184.
145. Hurley KM, Cross MB, Hughes SO. A systematic review of responsive feeding and child obesity in high-income countries. *J Nutr*. 2011;141:495–501.
146. Deboer MD, Lima AA, Oria RB, et al. Early childhood growth failure and the developmental origins of adult disease: do enteric infections and malnutrition increase risk for the metabolic syndrome? *Nutr Rev*. 2012;70:642–653.
147. Hren I, Mis NF, Brecelj J, et al. Effects of formula supplementation in breast-fed infants with failure to thrive. *Pediatr Int*. 2009;51:346–351.
148. Clarke SE, Evans S, Macdonald A, et al. Randomized comparison of a nutrient-dense formula with an energy-supplemented formula for infants with faltering growth. *J Hum Nutr Diet*. 2007;20:329–339.

149. Huh SY, Pappa H. Efficacy of cyproheptadine in promoting weight gain: a meta-analysis. *J Pediatr Gastroenterol Nutr*. 2004;2004:S507.
150. Piazza CC. Feeding disorders and behavior: what have we learned? *Dev Disabil Res Rev*. 2008;14:174–181.
151. Satter E. *Child of Mine: Feeding with Love and Good Sense*. Berkeley, CA: Bull Pub.; Distributed in the USA by Publishers Group West; 2000.
152. Birch LL. Development of food preferences. *Annu Rev Nutr*. 1999;19:41–62.
153. Birch LL, Marlin DW. I don't like it; I never tried it: effects of exposure on two-year-old children's food preferences. *Appetite*. 1982;3:353–360.
154. Heptinstall E, Puckering C, Skuse D, et al. Nutrition and mealtime behaviour in families of growth-retarded children. *Hum Nutr Appl Nutr*. 1987;41:390–402.
155. Mitchell WG, Gorrell RW, Greenberg RA. Failure-to-thrive: a study in a primary care setting. Epidemiology and follow-up. *Pediatrics*. 1980;65:971–977.
156. Drewett RF, Corbett SS, Wright CM. Physical and emotional development, appetite and body image in adolescents who failed to thrive as infants. *J Child Psychol Psychiatry*. 2006;47:524–531.
157. Drewett RF, Corbett SS, Wright CM. Cognitive and educational attainments at school age of children who failed to thrive in infancy: a population-based study. *J Child Psychol Psychiatry*. 1999;40:551–561.
158. Boddy J, Skuse D, Andrews B. The developmental sequelae of nonorganic failure to thrive. *J Child Psychol Psychiatry*. 2000;41:1003–1014.
159. Rudolf MC, Logan S. What is the long term outcome for children who fail to thrive? A systematic review. *Arch Dis Child*. 2005;90:925–931.
160. Hokken-Koelega AC, De Ridder MA, Lemmen RJ, et al. Children born small for gestational age: do they catch up? *Pediatr Res*. 1995;38:267–271.
161. Karlberg JP, Albertsson-Wikland K, Kwan EY, et al. The timing of early postnatal catch-up growth in normal, full-term infants born short for gestational age. *Horm Res*. 1997;48 (suppl 1):17–24.
162. Hediger ML, Overpeck MD, McGlynn A, et al. Growth and fatness at three to six years of age of children born small- or large-for-gestational age. *Pediatrics*. 1999;104:e33.
163. Leger J, Levy-Marchal C, Bloch J, et al. Reduced final height and indications for insulin resistance in 20 year olds born small for gestational age: regional cohort study. *BMJ*. 1997;315:341–347.
164. Kitchen WH, Doyle LW, Ford GW, et al. Very low birth weight and growth to age 8 years. II: Head dimensions and intelligence. *Am J Dis Child*. 1992;146:46–50.
165. Casey PH, Kraemer HC, Bernbaum J, et al. Growth status and growth rates of a varied sample of low birth weight, preterm infants: a longitudinal cohort from birth to three years of age. *J Pediatr*. 1991;119:599–605.
166. Bocca-Tjeertes IF, Reijneveld SA, Kerstjens JM, et al. Growth in small-for-gestational-age preterm-born children from 0 to 4 years: the role of both prematurity and SGA status. *Neonatology*. 2013;103:293–299.
167. Peralta-Carcelen M, Jackson DS, Goran MI, et al. Growth of adolescents who were born at extremely low birth weight without major disability. *J Pediatr*. 2000;136:633–640.
168. Hack M, Schluchter M, Cartar L, et al. Growth of very low birth weight infants to age 20 years. *Pediatrics*. 2003;112:e30–e38.
169. Bocca-Tjeertes IF, Kerstjens JM, Reijneveld SA, et al. Growth and predictors of growth restraint in moderately preterm children aged 0 to 4 years. *Pediatrics*. 2011;128:e1187–e1194.
170. Knops NB, Sneeuw KC, Brand R, et al. Catch-up growth up to ten years of age in

children born very preterm or with very low birth weight. *BMC Pediatr*. 2005;5:26.
171. Pierrat V, Marchand-Martin L, Guemas I, et al. Height at 2 and 5 years of age in children born very preterm: the EPIPAGE study. *Arch Dis Child Fetal Neonatal Ed*. 2011;96:F348–F354.
172. Van Ijzendoorn MH, Bakermans-Kranenburg MJ, Juffer F. Plasticity of growth in height, weight, and head circumference: meta-analytic evidence of massive catch-up after international adoption. *J Dev Behav Pediatr*. 2007;28:334–343.
173. Miller BS, Kroupina MG, Mason P, et al. Determinants of catch-up growth in international adoptees from eastern europe. *Int J Pediatr Endocrinol*. 2010;2010:107252.
174. Ammaniti M, Lucarelli L, Cimino S, et al. Feeding disorders of infancy: a longitudinal study to middle childhood. *Int J Eat Disord*. 2012;45:272–280.
175. Victora CG, Adair L, Fall C, et al. Maternal and child undernutrition: consequences for adult health and human capital. *Lancet*. 2008;371:340–357.
176. Belfort MB, Rifas-Shiman SL, Rich-Edwards JW, et al. Infant growth and child cognition at 3 years of age. *Pediatrics*. 2008;122:e689–e695.
177. Corbett SS, Drewett RF. To what extent is failure to thrive in infancy associated with poorer cognitive development? A review and meta-analysis. *J Child Psychol Psychiatry*. 2004;45:641–654.
178. Lozoff B, Jimenez E, Hagen J, et al. Poorer behavioral and developmental outcome more than 10 years after treatment for iron deficiency in infancy. *Pediatrics*. 2000;105:E51.
179. Lozoff B, Beard J, Connor J, et al. Long-lasting neural and behavioral effects of iron deficiency in infancy. *Nutr Rev*. 2006;64:S34-S43; discussion S72–S91.
180. Grantham-McGregor S, Schofield W, Harris L. Effect of psychosocial stimulation on mental development of severely malnourished children: an interim report. *Pediatrics*. 1983;72:239–243.
181. Corrales KM. Growth failure. In: Hendricks KM, Duggan C, eds. *Manual of Pediatric Nutrition*, 4th ed. Hamilton, Ontario, Canada: BC Decker; 2005: 569–85.

Developmental Disabilities

48

Kelly A. Dougherty, PhD
Babette S. Zemel, PhD

Children with moderate-to-severe disabilities have a markedly increased risk for malnutrition. Neurologic abnormalities—resulting in altered physical activity (voluntary and involuntary), fine motor patterns, and oral-motor patterns—will often significantly affect food intake and nutrient requirements. Chronic undernutrition, characterized by growth failure and reductions in fat stores and lean body mass, is the most common form of malnutrition, but overweight and obesity also occur. Despite these clinical observations, only limited research-based information on the nutrient needs and nutritional status of children with severe disabilities is available. Consequently, recommended nutrient intakes and growth pattern expectations for this group of patients are poorly defined and must be approached on an individual basis.

Nutritional status assessment is essential for identifying the undernourished or overnourished child, estimating the optimum energy intake to promote growth and well-being, and monitoring treatment. In children with moderate to severe disabilities, nutritional assessment is complicated by the interaction of the primary disease process (e.g., muscle atrophy, contractures, central nervous system pathology, or movement disorders) and both chronic and acute malnutrition. Cerebral palsy (CP), a common disability with significant risk for malnutrition, will be used as a model to describe the nutritional needs and nutritional status of children with severe disabilities.

Close to 90% of children with CP require at least some assistance with feeding based upon oral-motor dysfunction[1,2] and acquisition of eating skills is associated with gross motor functional ability.[3] Established in 1996, the North American Growth in Cerebral Palsy Project (NAGCPP)

was a large, population-based study that recruited subjects from six centers in the United States and Canada to investigate growth, nutrition, functional outcomes and overall quality of life in children with CP. Results showed that in children ages 2 to 18 years with moderate to severe CP, 30%–40% had severe feeding difficulties.[4] Parent-reported feeding dysfunction was strongly associated with poor health and nutritional status as well as severity of motor impairment. Those orally fed (nontube fed) showed a dose response relationship between severity of feeding dysfunction and growth and body fat (energy) stores, suggesting a pattern of inadequate energy intake. The authors recommended using a structured feeding dysfunction questionnaire to screen for nutritional risk in children with CP. Similar findings from a large population-based cohort study conducted in England showed that feeding difficulties reported by parents in the first four weeks of life in children with CP were associated with more severe neurodevelopmental impairments and an increased likelihood of being underweight by school age, suggesting that early, persistent, and severe feeding difficulties are a marker for future growth failure.[5] The association of feeding difficulties and nutritional status in CP has been demonstrated in every population studied.[1,4–8] Feeding difficulties and nutritional status also are associated with increased health care utilization and decreased societal participation by the child and parents.[9,10]

The timing of introduction of feeding assistance for those with oral-motor dysfunction may be important. In a recent registry-based study in Norway,[11] encompassing approximately 80% of children born with CP born between January 1, 1996 and December 31, 2003 (n = 661), 21% of the sample was completely dependent on feeding assistance, including tube feeding (in 14%). Speech function was highly associated with the need for feeding assistance. Duration of gastrostomy tube feeding was positively associated with weight and BMI z-scores, but was not associated with improved height status. However, age at placement of the gastrostomy tube was inversely associated with both height and weight status, independent of gross motor impairment, underscoring the importance of early intervention to promote growth and nutritional status.

Several important nutritional issues were recognized in these studies. Assistance with feeding, prolonged feeding times, and choking significantly affect food intake.[1,2] In addition, gastrointestinal problems, such as reflux, vomiting, and constipation, and some medications, such as those used to treat seizures, contribute to alterations in both nutritional intake and requirements.[2,12] Food intake low in energy density is of

particular concern because of the marked linear growth failure,[6,7,9] delayed skeletal[13–16] and sexual maturation,[17] poor weight gain, and reduced energy reserves seen in patients with CP.[18] Despite lower total energy expenditure in children with CP,[12] the energetic cost of physical activity in ambulatory children is greater than in typical children.[19] Increased muscle tone is also associated with greater resting energy expenditure.[20] Although protein intake usually meets recommended levels, lower concentrations of markers of protein metabolism in children with CP, particularly those fed enterally, suggest that needs are not met.[21] Abnormal body composition, with reductions in muscle and fat stores, and a high prevalence of undernutrition (failure to thrive, marasmus) and less frequently overnutrition (overweight, obesity) have also been noted.[18,22,23] Some specific nutrients are at risk, particularly vitamins C, A, and D; iron; and calcium, but micronutrient needs of children with neurologic impairment requires further research.[24] Low bone density,[22,25–27] associated with degree of immobilization, hypovitaminosis D, feeding difficulties, and lower triceps skinfold measurements in children with CP, also has been reported. Risk of fracture in children with moderate to severe CP is high, and is associated with low bone density,[28] greater body fat, feeding by gastrostomy tube, and prior history of fracture.[29]

For children with CP, the drug–nutrient interactions of anticonvulsant medications and vitamin D were demonstrated by several investigators in the 1970s. Vitamin D deficiency and insufficiency continues to be a concern in the nutritional care of these children.[30–32] Some classes of anticonvulsants, through the action of the hepatic microsomal enzymes, cause increased vitamin D catabolism. In a survey of 125 noninstitutionalized children with heterogeneous types of CP in the United States, nearly 20% had low serum 25-hydroxyvitamin D levels. However, low vitamin D status was not associated with severity of CP, growth failure, or use of anticonvulsants.[32] Another study of institutionalized children in South Africa found that anticonvulsant therapy was associated with long-bone fracture, and that 3 months of vitamin D and calcium supplementation resulted in marked improvement in serum calcium and phosphorus, and reduced alkaline phosphatase and fracture rate.[33] Another vitamin D and calcium supplementation study demonstrated improved bone mineral density (BMD) in children with CP.[32] Weight-bearing physical activity focused on normal movement patterns[34] vibration platforms,[35] or loaded sit to stand exercises combined with virtual training

cycling[36] also has a beneficial effect on bone. Thus, in addition to anticonvulsants, low dietary intake of vitamin D and calcium, limited physical activity and low sunlight exposure also contribute to rickets, osteopenia, and pathologic fractures in children with CP.[37–40]

Nutritional assessment of children with severe disabilities has several components, including evaluation of dietary intake, growth status, body composition, energy expenditure, bone density and laboratory data in the context of the nutritional risk factors related to the medical history, physical examination, diagnosis, and current therapy. The need for interdisciplinary evaluation and treatment of nutrition and feeding abnormalities for children and adults with CP has been addressed by numerous investigators and position papers.[10,41–44] Inadequate energy intake is a primary concern. Children with CP have abnormal growth and body composition. Therefore, conventional methods for estimating energy requirements based upon prediction equations that use body weight or surface area, may be inappropriate for nonambulatory children with CP.[45,46] The Dietary Reference Intake (DRI) recommendations for energy from the US Food and Nutrition Board[47] base the estimated energy requirement (EER) on age group, gender, physical activity level, height, weight and weight status (healthy weight vs. overweight), so these recommendations may prove to be more appropriate for children less severely affected with CP. The importance of determining energy requirements and meeting those requirements in children with CP is underscored by several nutrition intervention trials in which energy supplementation by gastrostomy or nasogastric feeding tube resulted in improvements in various components of nutritional status.[48–52]

Another approach to determining energy needs in children with severe CP has been to determine the extent of growth failure and poor nutritional status, and their relationship to nutritional and disease-related factors and energy expenditure. A multicenter study of growth and health outcomes in North America documented the association of growth failure with indicators of overall health and social participation across the spectrum of CP, showing that children with the best growth status had lower health care utilization and fewer days of social participation missed.[9] In a large single center study (*N* = 142), children ages 2–18 years with spastic quadriplegic CP (SQCP) participated in a survey of growth and nutritional status.[6,7] Linear growth, assessed by upper-arm and lower-leg lengths, was significantly reduced (z-scores, –1.57 to –2.38) relative to typically developing children.[53] Weight and triceps skinfold thickness were, on

average, about 65% of age- and sex-specific medians for children in the United States; subscapular skinfold thickness was 81% of the median. Poor nutritional status was evident in children as young as 2–4 years of age. Linear growth retardation in upper-arm and lower-leg length was significantly associated with poor nutritional status (arm muscle area, percent body fat), after adjusting for disease severity (diagnosis, cognitive ability, oral-motor function, ambulatory status, gastrostomy feeding, and dyskinetic component) and nondisease variables (age, gender, race, pubertal status, and midparental height). This analysis demonstrated that the large effect of poor nutritional status on linear growth was independent of, and additive to, the effects of the severity of the CP and nondisease factors.

Energy expenditure and dietary intake were measured in a subsample of these children with SQCP. Resting energy expenditure (REE), expressed as the percentage of the predicted value from World Health Organization (WHO)[54] prediction equations, was considerably lower than in typically developing children (91% predicted in CP vs. 105% in controls). REE was lowest (88% predicted) among the majority group of poorly nourished children with CP.[12] Total energy expenditure, measured by the doubly labeled water method, was also lower than in control children, in keeping with reduced physical activity in children with severe SQCP. The physical activity factor, or ratio of total to REE, was 1.23 for children with CP as opposed to 1.57 for controls. This is similar to the patterns of resting and total energy expenditure observed in adults[55] with CP. Thus, in children with SQCP, energy requirements are lower than in normal children. However, determination of energy intake proved to be far more difficult. The ratio of total energy intake, measured by a three-day weighed food intake diary, to total energy expenditure indicated a large—about 50%—overestimation of food intake by the caregivers of these children with poor oral-motor function.

Other studies have confirmed the lower total energy expenditure and physical activity factors for children with CP,[19,56,57] except for children with good ambulation. These findings illustrate the complexity of assessing energy requirements in disabled children. Although total energy requirements are lower in children with severe CP, dietary intake is likely to be grossly overestimated in the clinical setting. The prevalence of chronic undernutrition and growth failure in many children with SQCP indicates that energy needs are not being met. The next sections outline an approach to determining energy requirements in children with CP or other severe disabilities. The first step is the assessment of growth and

nutritional status to identify those children whose energy needs are not being met and the degree of past or current undernutrition. Methods for estimating energy requirements based on nutritional status and related factors are reviewed.

NUTRITION STATUS ASSESSMENT

Inadequate energy intake appears to be a major cause of nutrition-related growth failure in children with CP. Thus, two clinical management needs arise: (1) methods to assess nutritional and growth status to identify patients who are undernourished or overnourished and (2) methods to determine the energy requirement of the individual patient. The elements of complete nutritional assessment are based on the standard pediatric medical evaluation and are presented in more detail in Chapters 2 and 3.

The medical history includes assessment of acute and chronic medical problems and medications. In addition, a nutritional history, including review of a "typical" day's food intake; dietary history; use of energy, vitamin, mineral, herbal, or other nutritional supplements; and unusual food practices by the child or family are obtained. The past growth patterns, including pubertal history and growth charts, are reviewed. The history includes a review of systems, with emphasis on dental and oral-motor function and on the gastrointestinal tract (emesis, gastroesophageal reflux, diarrhea, constipation).

The physical examination includes current pubertal status[58]; weight; height, length, or alternative linear growth measures; head circumference; mid-arm circumference; and skinfold measurements. Interpretation of anthropometric measures requires comparison with children of similar age and sex in order to determine growth and nutritional "status." In addition to the reference ranges established for typically developing children, a number of publications offer values based on children with CP. Krick et al.[59] provided the first CP-specific growth charts for height, weight, and weight-for-height for children with quadriplegic CP based on 1,630 observations on 360 children at a single center. Malnourished children were not excluded from the sample, so these curves do not represent the optimal growth pattern of adequately nourished, healthy children with quadriplegic CP. The NAGCPP multicenter study also produced growth curves for weight, knee height, upper arm length, mid upper arm muscle area, triceps skinfold and subscapular skinfold for children with moderate to severe CP,[9] but the curves were not published as a reference; they were created to demonstrate the effect of nutritional status and degree of

neurologic impairment (i.e., gross motor function classification) on growth status. Height, weight and BMI growth charts stratified by motor and feeding skills in children with CP have since been published based on close to 142,000 observations on 24,920 children with CP in the California Department of Developmental Services.[60] Using a similar data source, Brooks et al.[61] published growth charts for weight stratified by age and gross motor function classification system constructed from over 100,000 weight measures in children with CP. These authors showed increased morbidity and mortality of children with low weight-for-age depending on gross motor function classification. These charts were designed to mimic the CDC charts and there were no exclusion criteria for poor nutritional status. Studies are needed to establish their utility in clinical care and validate their effectiveness in recognizing unhealthy weights to support treatment changes. Details of the recommended methods, equipment, and reference standards for the anthropometric measurements are in the following section. (For an overview of some of the physical findings associated with nutritional deficiencies, see Appendix, Table I-8.)

The laboratory evaluation is focused by recognizing the risk factors identified from the history and physical examination for each individual patient. The primary nutritional problem is usually inadequate energy intake, and laboratory assessment of this pattern of malnutrition is of limited use. Both serum albumin and prealbumin can be indicators of the adequacy of calorie and protein intake, with albumin reflecting the past month (half-life, approximately 18–20 days) and prealbumin reflecting the past week (half-life, approximately 2–3 days) of nutrient intake. However, these are often in the normal range with the chronic under-nutrition associated with CP. Iron-deficiency anemia is seen in groups of patients who have limited energy intake or who have monotonous food intake with few iron-rich foods. Iron status evaluation includes a complete blood count (hemoglobin, hematocrit, and red cell indices) as a screen, followed by other iron studies (serum iron, total iron-binding capacity, transferrin, ferritin) as indicated. Response to iron therapy is monitored by changes in the components of the complete blood count and a reticulocyte count.

Laboratory tests are selected depending on the patient's history and physical examination and could include serum values for vitamin D, calcium, phosphorus, and alkaline phosphatase, and radiologic examination of long bones to diagnose rickets. Clinically significant osteopenia often occurs without rachitic changes and can be diagnosed by determining bone mineral content or bone density [dual-energy x-ray absorptiometry (DXA)]. Bone density of the distal femur has been

proposed as a measure for children with CP and contractures who are difficult to position for standard measurement sites.[62–65] Skeletal age determination[66,67] is useful for diagnosing nutritional dwarfism in children with a delay of 2 or more years in bone age and for determining the potential for catch-up growth. Blood values for selected vitamins and minerals are available when indicated.

ANTHROPOMETRY

Anthropometric evaluation of growth and nutritional status is an essential component of nutritional care. It is a rapid, inexpensive, noninvasive means of determining both short- and long-term nutritional status. A well-trained anthropometrist is required in order to ensure accurate, reproducible measurements. Numerous anthropometric measures are used in the assessment of nutritional status. Each measure offers different information, and no single measure is sufficient for characterization of nutritional status. Proper equipment should be used, and it should be calibrated regularly to ensure accurate measurements. When accurate measurements are obtained and compared with appropriate reference standards, the clinician obtains a current classification of nutritional status and an opportunity to see changes in status over time. Guidelines for anthropometric measurement techniques are available[68] and are summarized below with consideration of the child with developmental disabilities.

Measurement of Growth

Weight, stature or length, and head circumference should be measured at routine office visits and every hospital admission. Current status and increments in growth between visits are very informative, especially for tracking progress in the individual patient. For children with severe disabilities, segmental measures of the arm or leg offer alternative measures of linear growth (upper-arm length, lower-arm length, lower-leg length, and knee height). These are described below for assessment when a length or stature measurement is not possible.

Weight

Because growth retardation is common in children with severe disabilities, it is of extra importance that both weight and linear growth are measured at the same time. Excess weight can be attributable to excess fat, excess

water caused by edema, or organomegaly, so other anthropometric measures, such as arm circumference and skinfold thicknesses, are important complements in interpreting a weight measurement.

Equipment, Measurement Technique, and Reference Standards

Weight should be measured on a digital electronic or beam balance scale. The scale should be checked weekly with known calibration weights. The measurement should be taken to the nearest 0.1 kg in older children and to the nearest 0.01 kg in infants.

The scale should be reset to zero prior to placing a child on it. Children should be weighed wearing little or no outer clothing and without shoes. Infants should be measured without diapers. Wheelchair-accessible scales are available for older children who cannot stand unattended on a scale. The measured weight of the wheelchair alone is deducted from the weight of the child (with little or no clothing) in the wheelchair.

For reference standards for weight, see WHO growth charts[69] for infants and children ages 0–2 years of age and Centers for Disease Control and Prevention (CDC) 2000 growth charts[70] for children age 2 years and older. Additionally, CP-specific growth charts are available[60,61] (see also http://www.lifeexpectancy.org/articles/GrowthCharts/All.pdf).

Length or Stature

Linear growth is a measure of a child's nutritional history and genetic potential; it helps to distinguish between short- and long-term nutritional problems. Length or stature measurements are appropriate for children who can be properly positioned, as described below. For children less than 2 years of age or children older than 2 who are not able to stand erect unsupported, a supine length measurement is taken. Standing height, or stature, is measured at age 2 and older. In older children with developmental disabilities, a length measurement is often required past 2 years of age. When comparing a supine length measurement to a growth chart for stature, the length should be decreased by 0.7 cm[71] to adjust for the known difference between length and stature measurements.

Equipment, Measurement Technique, and Reference Standards

To measure supine length, an infantometer or an inflexible length board with a fixed headboard and a movable footboard should be used. Length is measured to the nearest 0.1 cm. Infantometers and stadiometers with digital counters should be calibrated daily with a calibration bar of fixed

length, or following manufacturer's guidelines.

Measurement of supine length requires two people to position and hold the child. The head should be placed at the top of the board, with the Frankfurt plane perpendicular to the floor.[72] The Frankfurt plane extends from the lower margin of the orbit to the upper margin of the auditory meatus (Figure 48-1). An assistant is needed to hold the head firmly in this position. The knees should be flattened to fully extend the legs. The feet should be together and flexed to a 90° angle by the footboard with the infant fully stretched.

To measure stature, the child should stand with the heels, buttocks, and back of the head against the stadiometer. The arms should be down and relaxed and the heels together. The position of the head should be adjusted so that the Frankfurt plane is parallel to the floor. Stature should be measured using a stadiometer with a head paddle that glides smoothly but is firmly perpendicular to the back of the stadiometer. Alternatively, a tape measure permanently fixed to a wall or doorframe can be used provided that a head paddle is available that will fit at a 90° angle to the wall. Both length and stature measurements should be accurate to 0.1 cm.

Length and stature measures are inappropriate for children with spinal curvature, contractures, or other conditions that prevent proper positioning for the measurement. Segmental measures of arm or leg length are alternatives under these conditions (see below).

For both length and stature, measurements should be taken in triplicate, with the measurements in agreement within 0.5 cm, and the mean recorded. Whenever possible, the statures of both biological parents should be obtained and midparental stature computed (the average stature of the mother and father). Adjustment of stature or length measurements using midparental stature charts is a technique that recognizes and corrects for differences in the genetic potential for linear growth.[73]

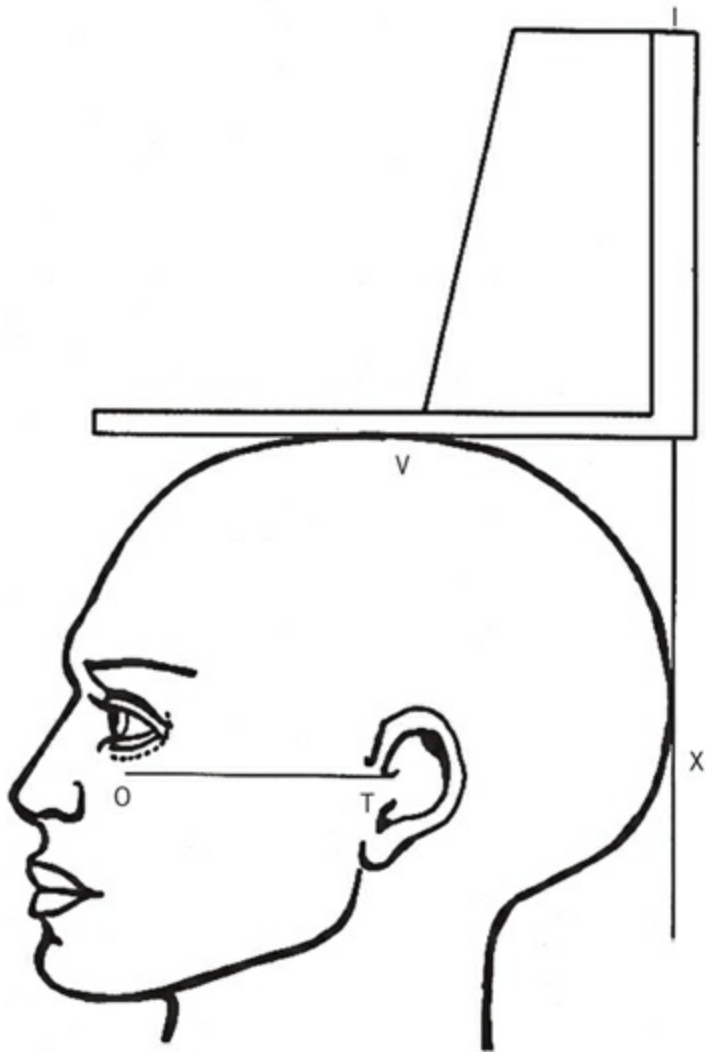


Figure 48-1 The Frankfurt plane extends from the auditory meatus to the lower border of the orbit. This plane should be parallel to the paddle or headboard when measuring stature or length. Reproduced from Ross et al.[72]

For reference standards for length and stature, see WHO growth charts[69] for infants and children ages 0–2 years of age and CDC 2000 growth charts[70] for children age 2 years and older. Additionally, CP-specific growth charts are available.[59,60] (see also (http://www.lifeexpectancy.org/articles/GrowthCharts/All.pdf).

Weight-for-Height Indices

Weight-for-length and weight-for-height indices are important in the

assessment of nutritional status to ensure energy reserves adequate for maintaining growth and over-all health. For infants and children ages 0–2 years of age, weight-for-length and weight-for-height charts are available[69] to assess nutritional status. Additionally, CP-specific weight-for-height[59] are available. For children more than 2 years of age, the body mass index (BMI) can be used. BMI is calculated as weight in kilograms divided by height in meters squared (wt/ht^2). Reference standards for BMI for typically developing children from birth to 20 years are available.[69,70] Use of these charts is essential for interpretation of BMI in the pediatric range. Additionally, CP-specific charts for BMI are also available.[60] (see also http://www.lifeexpectancy.org/articles/GrowthCharts/All.pdf).

Head Circumference

Brain growth is most rapid in the first 3 years of life. Head circumference is a good indicator of brain growth and therefore should be included in the assessment of growth and nutritional status. Because of the high risk of growth failure in children with developmental disabilities, it is important to measure head circumference until age 3, or longer in some children, to determine whether growth of the brain and head is also compromised. However, head circumference is of limited use in nutritional status assessment of patients with central nervous system damage and micro- or megalocephaly of non-nutritional etiology.

Equipment, Measurement Technique, and Reference Standards

A flexible metal or non-stretch plastic-coated measuring tape scaled to 0.1 cm should be used. The tape is placed over the supraorbital ridge and around the occiput so that a maximum circumference is obtained. Care should be taken that the tape is evenly placed on all sides and is flat against the skull. Three measurements should be obtained and the mean recorded.

For reference standards for head circumference, WHO growth charts[74] for infants and children ages 0–2 years of age, and CDC 2000 growth charts for children ages 2–3 years of age[70] are recommended. For older children see Roche and colleagues.[75]

Arm and Leg Length Measures

Segmental measures of the arm and leg are alternative measures of linear growth when length or stature measurements are not possible. This commonly occurs in children with developmental disabilities. Both upper-

arm and lower-leg length should be measured because they provide measures of upper- and lower-body long-bone growth. Frequently, lower-leg length is more compromised than upper-arm length in children who are nonambulatory. Measurement of knee height[76] and ulnar length[77] are additional alternative measure of linear growth and can be used in prediction equations to estimate height.

Equipment, Measurement Technique, and Reference Standards

For young infants, sliding calipers (0–200 mm) are used. For older infants and children, an anthropometer (50–570 mm) or segmometer (Rosscraft Innovations, Inc.) is used. All measurements should be taken in triplicate to the nearest 0.1 cm and the average used. For infants and young children up to 24 months of age, upper-arm length is measured as shoulder-to-elbow length. With the arm flexed at a 90° angle, the upper arm is measured from the superior lateral surface of the acromion to the inferior surface of the elbow. In children 2–18 years of age, upper-arm length is measured from the superior lateral surface of the acromion to the radiale (head of the radius) with the arm relaxed, as shown in Figure 48-2.

Ulna length[77] is measured with the patient seated and left arm resting on a table with the elbow bent at 90°–110°. The hand should be positioned with the palm faced down and fingers extended and together. The measurement is taken from the proximal end of the ulna (olecranon) to the tip of the styloid process.



Figure 48-2 Upper arm length measurement technique.

For infants and young children, lower-leg length is measured as a knee-to-heel length. With the infant lying supine—with the leg flexed to 90° at the hip, the knee, and the ankle—the lower-leg length is measured from the heel to the superior surface of the knee. For older children, a comparable measure is knee height, taken in a seated position with similar 90° flexion at the hip, knee, and ankle. For children 2–18 years of age, the tibia length, also known as the lower-leg length measure is taken from the distal margin of the medial malleolus to the proximal border of the medial tibial condyle while the patient is sitting in a relaxed position, as shown in Figure 48-3. The right side should be measured unless there is unilateral involvement affecting the right side. The least affected side should be measured and thus noted on the growth chart so that repeat measures are always taken on the same limb.

Reference percentiles for upper-arm length and lower-leg length for

typically developing infants and toddlers are given in Figures 48-4 and 48-5, and for older children (3–18 y) in Figures 48-6 through 48-9. These are derived from work by Spender and colleagues[53] and Snyder and colleagues.[78] Recently, tibia length growth curves for ambulatory children with CP (gross motor function classification system levels I, II, and III) have been published based on 750 participants and nearly 1200 observations.[79] Reference percentiles for ulna length in typically developing children ages 5–19 years, and corresponding height prediction equations based on ulna length are presented by Gauld et al.[77] and for knee height by Chumlea et al.[80] Choice of which segment to measure may depend on characteristics of the individual patient, such as ambulation status, contractures and deformities, and difficulty in positioning. Selection of reference percentiles based on children with CP can be used to assess the status of an individual child relative to other children with CP. However, because of the pervasive nutritional issues in children with CP, these reference percentiles are unlikely to represent “optimal” growth.

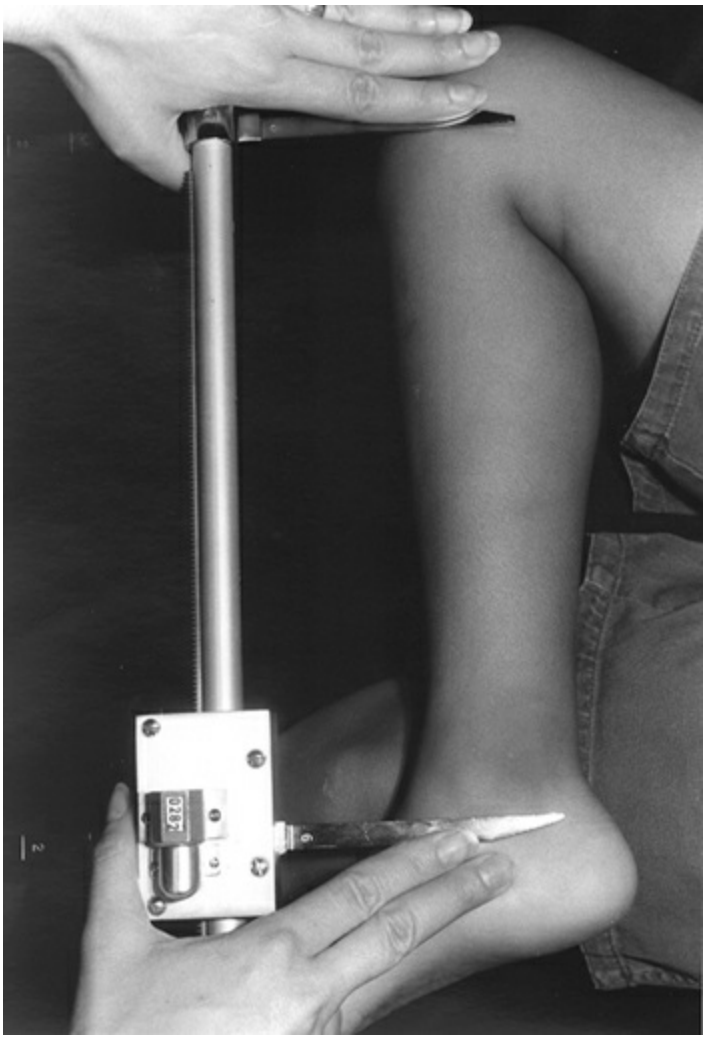


Figure 48-3 Lower leg length measurement technique.

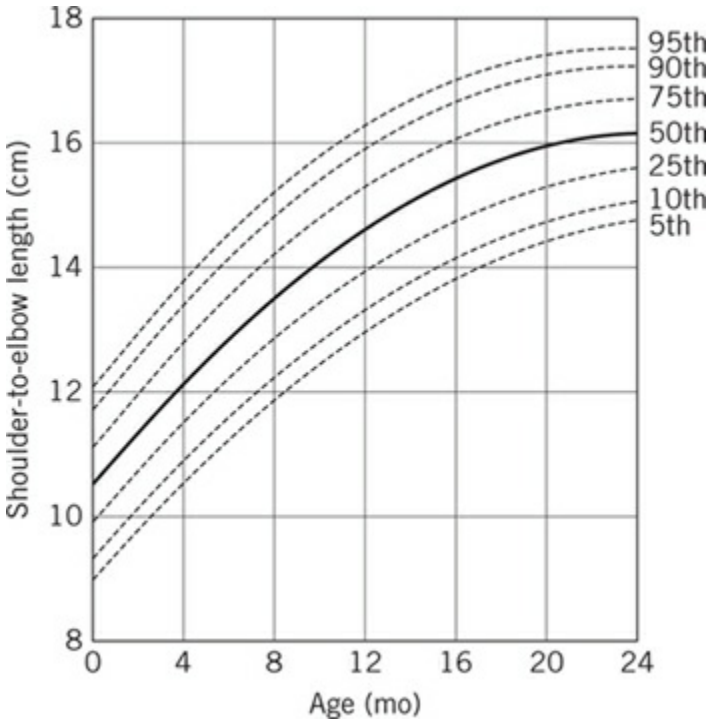


Figure 48-4 Shoulder-to-elbow lengths for boys and girls from birth to age 2 years. Reproduced from Stallings & Zemel.[94]

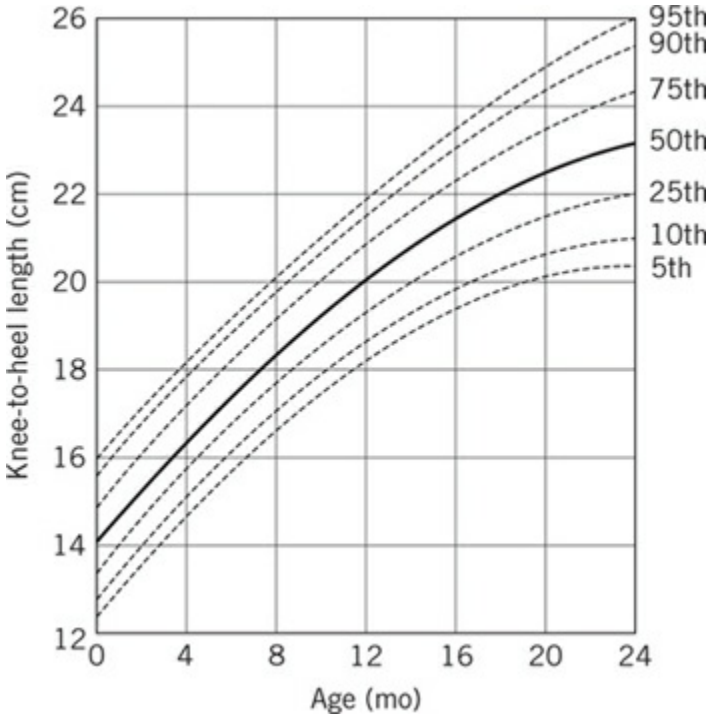


Figure 48-5 Knee-to-heel lengths for boys and girls from birth to age 2 years. Reproduced from Stallings & Zemel.[94]

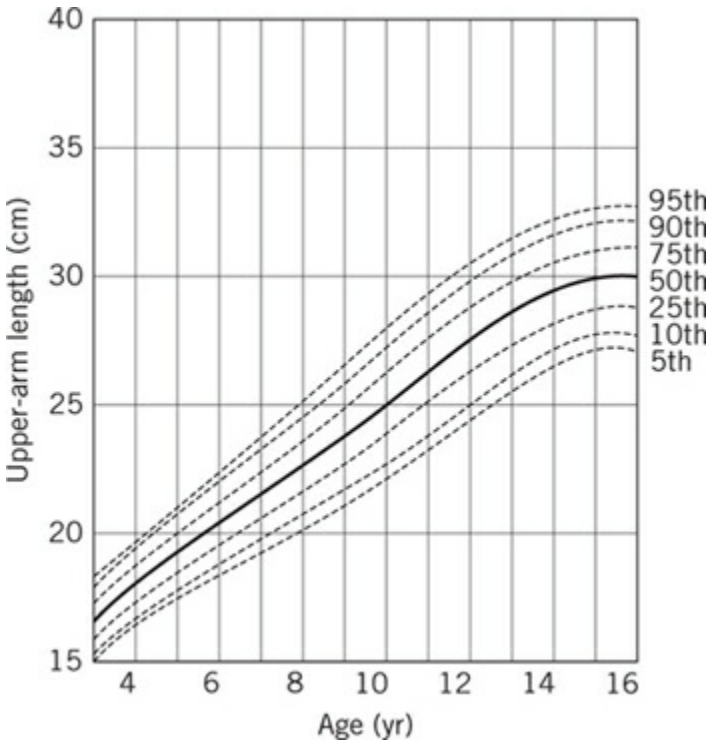


Figure 48-6 Upper-arm lengths for girls 3–16 years of age. Reproduced from Stallings & Zemel.[94]

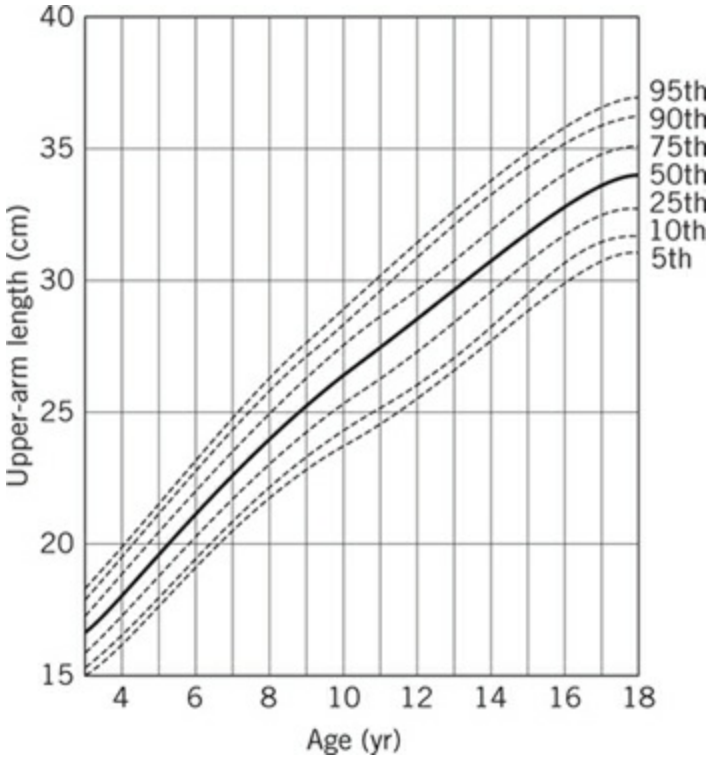


Figure 48-7 Upper arm lengths for boys 3–18 years of age. Reproduced from Stallings & Zemel.[94]

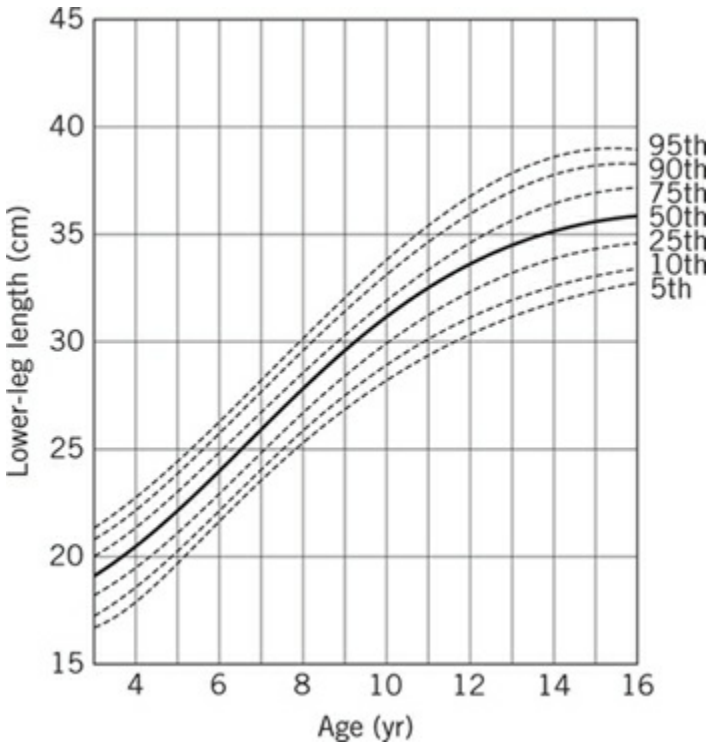


Figure 48-8 Lower leg length for girls 3–16 years of age. Reproduced from Stallings & Zemel.[94]

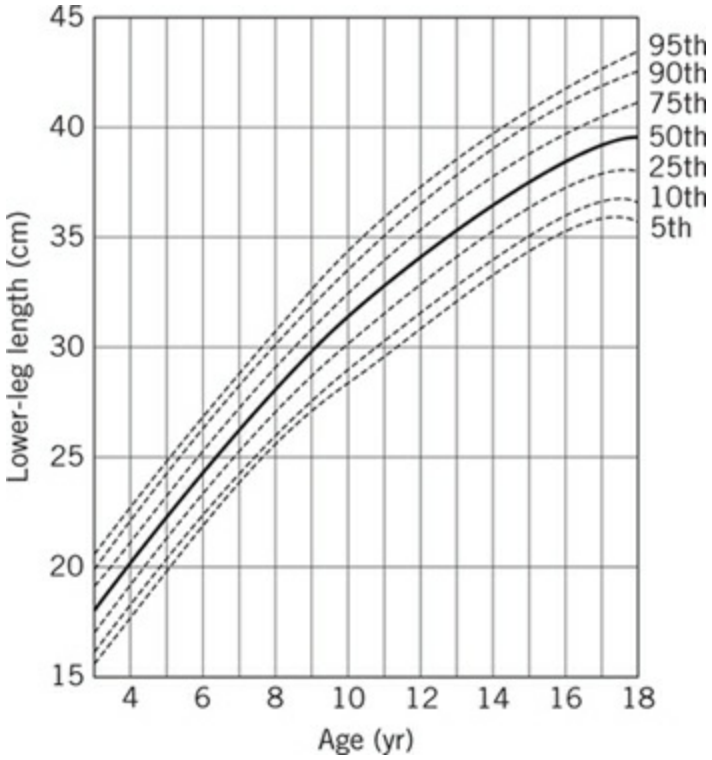


Figure 48-9 Lower leg length for boys 3–18 years of age. Reproduced from Stallings & Zemel.[94]

Nutritional Status

Anthropometric indicators of nutritional status consist of measurements of lean and fat tissues at sites known to be sensitive indicators of nutrition and health. Upper-arm anthropometry, the measurement of arm circumference and skinfold thickness, is particularly useful because excellent reference data are available for clinical interpretation. An additional measure of subcutaneous fat on the trunk, the subscapular skinfold thickness measure, is helpful for further assessing total body fat stores, especially when fat distribution is abnormal.

Mid-Upper-Arm Circumference

Mid-upper-arm circumference is a composite measure of muscle, fat, and

bone in the arm. It is sensitive to current nutritional status and can be used in combination with the triceps skinfold thickness measure to estimate upper-arm muscle area and upper-arm fat area.

Equipment, Measurement Technique, and Reference Standards

A nonstretch flexible tape measure is used. All measurements should be taken in triplicate to the nearest 0.1 cm. The midpoint of the upper arm is located by measuring the length of the upper arm (from the tip of the acromion to the olecranon with the arm flexed at 90° and the palm facing upward) and marking the midpoint with a washable ink pen. The mid-arm circumference is taken with the arm down and in a fully relaxed position. Gently shaking the arm usually ensures that it is relaxed. The tape is placed over the midpoint perpendicular to the long axis of the arm. Care should be taken so that there is no pinching or gaping of the tape as it encircles the arm. This process improves the accuracy and reproducibility of the measure.

For reference standards for mid-upper arm circumference, see Frisancho[81] and the WHO Multicentre Growth Study.[74]

Skinfold Thickness

Skinfold thickness measures assess subcutaneous fat at specified sites. The triceps and subscapular skinfold measures are indicators of total body fat stores and are sensitive to changes in nutrient intake and nutritional status. Depletion of fat stores at the triceps site is common in children with CP and in other chronically undernourished groups, whereas fatness at the subscapular site is not affected as severely.[6,7,82] Reference data for the sum of these two skinfold measures are available[83] and are used in the research setting as a characterization of total body fat.

Equipment, Measurement Technique, and Reference Standards

Holtain skinfold calipers are preferred because they are scaled to 0.2 mm. Lange calipers can be used, but with some loss of accuracy as they are scaled to the nearest 0.5 mm. Calipers that are not spring-loaded and therefore do not provide a constant tension are not recommended.

The triceps skinfold thickness is measured over the triceps muscle, along the back of the arm. The measurement is taken at the same level at which the mid-upper-arm circumference is measured. The patient should be upright with the arm down and fully relaxed for the measurement. The fold of fat and skin is lifted away from the underlying muscle and held

throughout the measurement. The calipers are placed next to the fingers lifting up the skinfold; the skinfold is held in position while the thickness is measured with calipers, and the reading should be taken three to four seconds after releasing the caliper's handles.

For the subscapular skinfold, the patient should be upright with the arm and shoulder down and fully relaxed. The skinfold is lifted 1 cm below the tip of the scapula at a 45°angle following the natural contour of the body. The fold is held in place, and the calipers are placed next to the fingers holding the fold. The reading should be taken three to four seconds after releasing the caliper's handles.

A number of different sources for skinfold thickness reference standards based on typically developing children are available.[74,83–85]

Body Composition

Body composition assessment, the measurement of fat and lean body mass stores, is an essential component of nutritional assessment of the disabled child. Whereas linear growth reflects the longer-term nutritional history and genetic potential and is often compromised in children with disabilities, fat and lean body mass stores reflect short-term nutritional adequacy. Body composition can be estimated using the prediction equations of Slaughter and colleagues[86] based on typically developing children ages 8–18 years. These equations use the triceps and subscapular skinfold thickness measures shown in Table 48-1. Specific equations are given based on race, gender, and pubertal status groups. Adjustment factors (for sex, race, puberty status, level of obesity, and gross motor function) to the Slaughter equations for children with CP (ages 8–18) have also been published,[87] but have not been independently validated.

Body composition values estimated from the Slaughter et al.[86] anthropometric prediction equations have been compared with measures determined from total body water[18] in children with severe SQCP. Fat mass measured by these two methods is significantly correlated (r = .69), indicating that the anthropometric prediction equations is a reasonable estimate to be used as a clinical, noninvasive method to estimate total body fat in children with CP. Other methods, such as bioelectrical impedance analysis (BIA)[57] and DXA,[87] are possible. However, motion artifact and inability to properly position the patient could increase the measurement error and give inconsistent results in children with moderate to severe disabilities. In addition, most BIA devices require a height measurement, although a recently published prediction equation by Reiken et al.[57] uses

tibia length. For children whose height can be estimated only from long-bone measures, the measurement error is greatly increased so that BIA methods may become less effective measures of body composition. In a small sample (n = 8), Liu et al.[88] compared estimates of body composition by BIA and anthropometry with DXA in eight children with CP and severe motor impairment. Results showed correlation coefficients for DXA versus other methods for fat-free mass determination were excellent (r = .9) but only moderate for fat mass and percent body fat (range 0.4–0.8). The best predictor of percent body fat was skinfolds and of fat mass was bioelectrical impedance.

Bone Density Assessment

Children with moderate to severe CP are at increased risk of osteopenia and fracture,[27–29] so assessment of bone density is often warranted. Bone density is measured by DXA, a rapid, noninvasive technique that involves very low levels of ionizing radiation. The current recommendation of the International Society for Clinical Densitometry for assessment of bone density in children is to measure the BMD of the lumbar spine and whole body, and compare the results to age and gender-specific reference data to determine the z-score (standard deviation). A z-score less than or equal to –2 is considered "low" BMD.[89]

For many children with developmental disabilities, particularly for those with CP, a whole body and spine measurement may not be feasible due to altered posture, spasticity, voluntary and involuntary movements, and metal spine implants.[64] In addition, the BMD measure is influenced by height and weight, so for children with developmental disabilities and growth failure, a low BMD may be related to poor growth rather than bone strength.[90] An alternative scan technique with accompanying reference data for the distal femur has been developed.[62–65] This has proven to be a more feasible approach for assessing BMD in children with CP and is associated with fracture risk.[28] An example of a distal femur image is given in Figure 48-10. Peripheral quantitative computed tomography (pQCT) has also been used in the research setting to assess bone strength in children with CP.[26] This is not a clinically available assessment technique at this time.

TABLE 48-1 Anthropometric Equations for Prediction of Percent Fat (Ages 8–18

Years)	
White males	
Prepubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 1.7
Pubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 3.4
Postpubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 5.5
Black males	
Prepubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 3.2
Pubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 5.2
Postpubescent:	1.21 (triceps + subscapular) – 0.008 (triceps + subscapular)^2 – 6.8
All females	1.33 (triceps + subscapular) – 0.013 (triceps + subscapular)^2 – 2.5
If (triceps + subscapular) greater than 35 mm	
All Males:	0.783 (triceps + subscapular) + 1.6
All Females:	0.546 (triceps + subscapular) + 9.7
CP-specific adjustments	
Overall adjustment	+12.2
Additional adjustment for:	
Males	–5.0
More severe GMFCS	+5.1
Black race	–3.1
Pubescent	+2.0
Postpubescent	–4.6
If (triceps + subscapular) greater than 35 mm	–3.2

Example: for a black postpubescent male at GMFCS level 1 with a triceps + subscapular less than 35 mm, the estimated percent fat = Slaughter percent fat + 12.2–5.0–3.1–4.6.

CP, Cerebral Palsy; GMFCS, Gross Motor Function Classification System.

Adapted from Slaughter et al.[86] and Gurka et al.[87]

ENERGY INTAKE RECOMMENDATIONS

Assessment of energy intake compared to requirement is an essential component of the nutritional care of children with significant disabilities.

Undernutrition and overnutrition are the common nutritional pathologies and are the result of energy imbalance. True energy intake is difficult to determine accurately in children with poor oral-motor function and other gastrointestinal problems. Food loss during feeding and regurgitation contribute to the inaccurate estimation of energy intake, even when weighed food diaries recorded over several days are obtained.[12] Daily energy expenditure and its components can be measured in individuals in a research setting (doubly labeled water, 24-h indirect room calorimetry, REE, thermic effect of food, physical activity). Only REE by indirect calorimetry, a measurement of basal metabolic needs, is available as a clinical evaluation and can be used with estimates of physical activity to predict total energy needs.

There is little information regarding the accuracy of the revised DRI recommendations for energy intake,[47] now designated the EER, for estimating the energy needs of individual patients with developmental disabilities with atypical body size, body composition, and physical activity. The EER is based on gender, age, height, weight and level of physical activity. The formulas for determining the EER are given in Table 48-2. A study of 52 children with CP, ages 2–19 years, generated correction factors (a general correction factor for CP, and additional corrections for high degree of movement and gross motor function classification) to existing prediction equations for total energy expenditure.[57] These equations have not been independently validated.

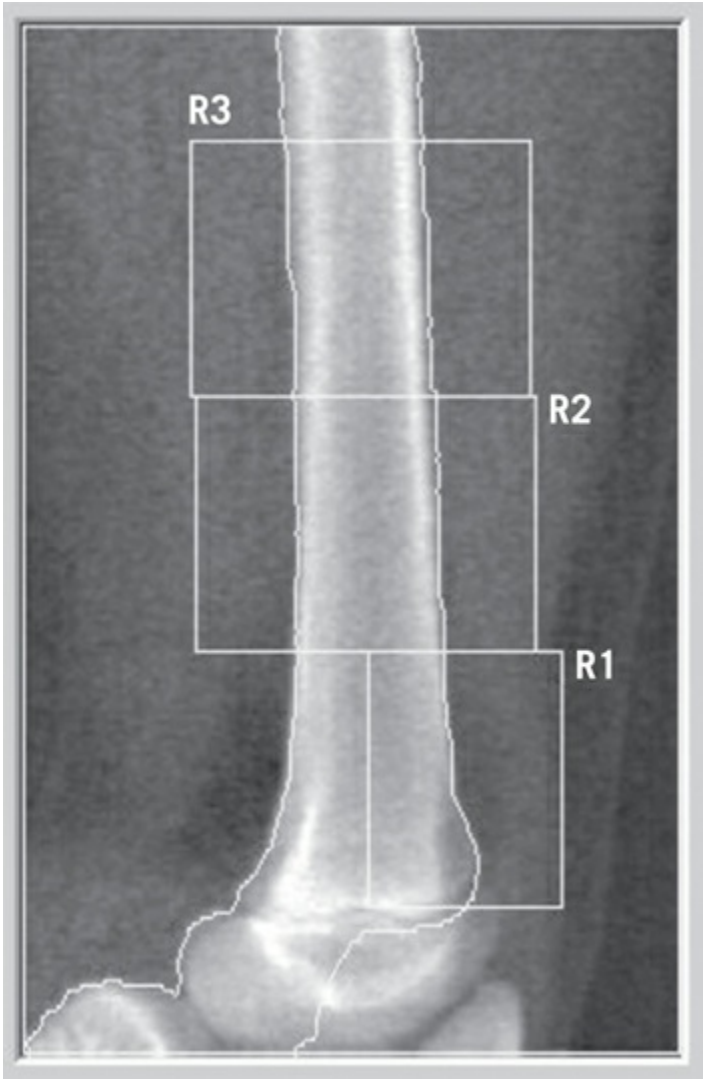


Figure 48-10 DXA scan of a lateral distal femur for assessment of bone mineral density in children. The bone mineral density of regions R1, R2, and R3 are determined separately.

The formulas for energy requirement from WHO[54] allow for individualized prediction of REE based on gender, age, and body weight, as shown in Table 48-3. Whenever possible, REE should be measured by indirect calorimetry in the early morning with the child fully at rest and in a fasting state. Under these conditions, measurements obtained over 30–60 minutes of inactivity can provide an excellent estimate of resting energy needs. Recently, in children with moderate to severe CP, changes in REE over four hours in response to intrathecal baclofen injection was investigated to determine the contribution of high muscle tone to energy

needs in this population.[91] Results showed a significant drop in REE from 101 ± 17% to 84 ± 24% of predicted after injection, suggesting high muscle tone greatly contributes to the energy needs of children with CP. The value for REE, whether from indirect calorimetry or a prediction equation, is adjusted by an activity factor to determine total energy expenditure and daily energy requirements. In normally active typically developing children and in adults with light or moderate activity, the activity factors range between 1.50 and 1.89 times the REE. Overall physical activity is lower in children with CP, but they have a higher energy expenditure during walking compared to typically developing children.[92] Studies are needed to determine how the energetic cost of usual activities affects activity level and total energy expenditure. In children with mild[19] and severe[12] CP, the physical activity factor is much lower ranging from 1.3 to 1.5 times the REE.

TABLE 48-2 Prediction Equations for Estimated Energy Requirements (Kcal per day) for Healthy Children[47]

Infants	Prediction Equations				
0–3 mo	89*weight (kg) – 100 + 175				
3– 6 mo	89*weight (kg) – 100 + 56				
6–12 mo	89*weight (kg) – 100 + 22				
12–24 mo	89*weight (kg) – 100 + 20				
Males	**General Prediction Equation***	**Sedentary PA Coefficient**	**Low Active PA Coefficient**	**Active PA Coefficient**	**Very Active Coeffi**
3–8 y	88.5 – 61.9*age + PAL*((26.7*weight) + 903*(height)) + 20	1	1.13	1.26	1.42
9–18 y	88.5 – 61.9*age + PAL*((26.7*weight) + 903*(height)) + 25	1	1.13	1.26	1.42
> 18 y	662 – 9.53*age + PAL*((15.91*weight)+	1	1.11	1.25	1.48

	539.6*(height))				
Overweight 3–18 y	114 – 50.9*age + PAL*((19.5*weight)+ 1161.4*(height))	1	1.12	1.24	1.45
Females	**General Prediction Equation***	**Sedentary PA Coefficient**	**Low Active PA Coefficient**	**Active PA Coefficient**	**Very Active Coeffi**
3–8 y	135.3 – 30.8*age + PAL*((10*weight) + 934*(height)) + 20	1	1.16	1.31	1.56
9–18 y	135.3 – 30.8*age + PAL*((10*weight) + 934*(height)) + 25	1	1.16	1.31	1.56
> 18 y	354 – 6.91*age + PAL*((9.36*weight) + 726*(height))	1	1.12	1.27	1.45
Overweight 3–18 y	389 – 41.2*age + PAL*((15*weight) + 701.6*(height))	1	1.18	1.35	1.6

*Each prediction equation uses weight (kg) and height (kg), and requires that a physical activity coefficie (PA) be included in the calculation of the EER. The PA categories, based on the physical activity level (calculated as the ratio of total energy expenditure to resting energy expenditure), are as follows:

Sedentary: PAL is estimated to be ≥1.0 and <1.4.

Low Active: PAL is estimated to be ≥1.4 and <1.6.

Active: PAL is estimated to be ≥1.6 and <1.9.

Very Active: PAL is estimated to be ≥1.9 and <2.5.

TABLE 48-3 Equations for Predicting Resting Energy Expenditure (Kcal/d) from Body Weight (w)

Age,y	Male	Female
0–3	60.9 w – 54	61.0 w – 51
3–10	22.7 w + 495	22.5 w + 499
10–18	17.5 w + 651	12.2 w + 746
18–30	15.3 w + 679	14.7 w + 496
30–60	11.6 w + 879	8.7 w + 829
> 60	13.5 w + 487	10.5 w + 596

Adapted from World Health Organization.[54]

Two older methods for estimating energy requirements have been published. Culley and Middleton developed a method for disabled children that takes level of motor function into account.[93] The energy consumption based on motor function status was 14.7 kcal/cm of height and 77 kcal/kg for children without motor dysfunction, 13.9 kcal/cm and 75 kcal/kg for ambulatory children with some motor dysfunction, and 11.1 kcal/cm and 64 kcal/kg for nonambulatory children with motor dysfunction. Krick and colleagues reported a factorial method approach in determining energy needs in children with CP.[46] This method accounts for energy intake based on estimated REE needs, muscle-tone alterations, normal growth needs, and catch-up growth or nutritional repletion in malnourished children. The formula is summarized as

SUMMARY

Monitoring nutrition and growth status in children and adults with CP and other severe types of developmental disabilities is an essential component of care. This chapter summarizes the previous work in the area and outlines clinical approaches to determining nutritional status and estimating energy needs. Although data are not available to provide precise definitions of the levels of severity of malnutrition and growth failure and their effect on long-term outcome, it is clear that many patients with moderate to severe CP or other disabilities have malnutrition and growth failure as the result of inadequate energy intake. Nutritional status assessment every three to six months will enable the clinical care team to identify malnutrition and growth failure and will provide an objective means of following the response to nutritional intervention.

REFERENCES

1. Reilly S, Skuse D, Poblete X. Prevalence of feeding problems and oral motor dysfunction in children with cerebral palsy: a community survey. *J Pediatr*. 1996;129:877–882.
2. Sullivan PB, Juszczak E, Lambert BR, et al. Impact of feeding problems on nutritional intake and growth: Oxford Feeding Study II. *Dev Med Child Neurol*. 2002;44:461–467.
3. Weir KA, Bell KL, Caristo F, et al. Reported eating ability of young children with cerebral palsy: is there an association with gross motor function? *Arch Phys Med Rehabil*. 2013;94:495–502.
4. Fung EB, Samson-Fang L, Stallings VA, et al. Feeding dysfunction is associated with poor growth and health status in children with cerebral palsy. *J Am Diet Assoc*. 2002;102:361–373.
5. Motion S, Northstone K, Emond A, et al. Early feeding problems in children with

cerebral palsy: weight and neurodevelopmental outcomes. *Dev Med Child Neurol*. 2002;44:40–43.
6. Stallings VA, Charney EB, Davies JC, Cronk CE. Nutritional status and growth of children with diplegic or hemiplegic cerebral palsy. *Dev Med Child Neurol*. 1993;35:997–1006.
7. Stallings VA, Charney EB, Davies JC, Cronk CE. Nutrition-related growth failure of children with quadriplegic cerebral palsy. *Dev Med Child Neurol*. 1993;35:126–138.
8. Troughton KE, Hill AE. Relation between objectively measured feeding competence and nutrition in children with cerebral palsy. *Dev Med Child Neurol*. 2001;43:187–190.
9. Stevenson RD, Conaway M, Chumlea WC, et al. Growth and health in children with moderate-to-severe cerebral palsy. *Pediatrics*. 2006;118:1010–1018.
10. Position of the American Dietetic Association: nutrition in comprehensive program planning for persons with developmental disabilities. *J Am Diet Assoc*. 1992;92:613–615.
11. Dahlseng MO, Finbraten AK, Juliusson PB, et al. Feeding problems, growth and nutritional status in children with cerebral palsy. *Acta Paediatr*. 2012;101:92–98.
12. Stallings VA, Zemel BS, Davies JC, et al. Energy expenditure of children and adolescents with severe disabilities: a cerebral palsy model. *Am J Clin Nutr*. 1996;64:627–634.
13. Gilbert SR, Gilbert AC, Henderson RC. Skeletal maturation in children with quadriplegic cerebral palsy. *J Pediatr Orthop*. 2004;24:292–297.
14. Henderson RC, Gilbert SR, Clement ME, et al. Altered skeletal maturation in moderate to severe cerebral palsy. *Dev Med Child Neurol*. 2005;47:229–236.
15. Kong CK, Tse PW, Lee WY. Bone age and linear skeletal growth of children with cerebral palsy. *Dev Med Child Neurol*. 1999;41:758–765.
16. Roberts CD, Vogtle L, Stevenson RD. Effect of hemiplegia on skeletal maturation. *J Pediatr*. 1994;125:824–828.
17. Worley G, Houlihan CM, Herman-Giddens ME, et al. Secondary sexual characteristics in children with cerebral palsy and moderate to severe motor impairment: a cross-sectional survey. *Pediatrics*. 2002;110:897–902.
18. Stallings VA, Cronk CE, Zemel BS, Charney EB. Body composition in children with spastic quadriplegic cerebral palsy. *J Pediatr*. 1995;126:833–839.
19. Bell KL, Davies PS. Energy expenditure and physical activity of ambulatory children with cerebral palsy and of typically developing children. *Am J Clin Nutr*. 2010;92:313–319.
20. Liu LF, Moyer-Mileur L, Gooch J, Samson-Fang L. The contribution of tone to resting energy expenditure in children with moderate to severe cerebral palsy: evaluation utilizing intrathecal baclofen injection. *J Pediatr Rehabil Med*. 2008;1:163–167.
21. Schoendorfer N, Tinggi U, Sharp N, et al. Protein levels in enteral feeds: do these meet requirements in children with severe cerebral palsy? *Br J Nutr*. 2012;107:1476–1481.
22. Chad KE, McKay HA, Zello GA, et al. Body composition in nutritionally adequate ambulatory and non-ambulatory children with cerebral palsy and a healthy reference group. *Dev Med Child Neurol*. 2000;42:334–339.
23. Tomoum HY, Badawy NB, Hassan NE, Alian KM. Anthropometry and body composition analysis in children with cerebral palsy. *Clin Nutr*. 2010;29:477–481.
24. Schoendorfer N, Boyd R, Davies PS. Micronutrient adequacy and morbidity: paucity of information in children with cerebral palsy. *Nutr Rev*. 2010;68:739–

748.
25. Henderson RC, Lark RK, Gurka MJ, et al. Bone density and metabolism in children and adolescents with moderate to severe cerebral palsy. *Pediatrics*. 2002;110:e5.
26. Binkley T, Johnson J, Vogel L, et al. Bone measurements by peripheral quantitative computed tomography (pQCT) in children with cerebral palsy. *J Pediatr*. 2005;147:791–796.
27. Al Wren T, Lee DC, Kay RM, et al. Bone density and size in ambulatory children with cerebral palsy. *Dev Med Child Neurol*. 2011;53:137–141.
28. Henderson RC, Berglund LM, May R, et al. The relationship between fractures and DXA measures of BMD in the distal femur of children and adolescents with cerebral palsy or muscular dystrophy. *J Bone Miner Res*. 2010;25:520–526.
29. Stevenson RD, Conaway M, Barrington JW, et al. Fracture rate in children with cerebral palsy. *Pediatr Rehabil*. 2006;9:396–403.
30. Bergqvist AG, Schall JI, Stallings VA. Vitamin D status in children with intractable epilepsy, and impact of the ketogenic diet. *Epilepsia*. 2007;48:66–71.
31. Henderson RC. Vitamin D levels in noninstitutionalized children with cerebral palsy. *J Child Neurol*. 1997;12:443–447.
32. Jekovec-Vrhovsek M, Kocijancic A, Prezelj J. Effect of vitamin D and calcium on bone mineral density in children with CP and epilepsy in full-time care. *Dev Med Child Neurol*. 2000;42:403–405.
33. Bischof F, Basu D, Pettifor JM. Pathological long-bone fractures in residents with cerebral palsy in a long-term care facility in South Africa. *Dev Med Child Neurol*. 2002;44:119–122.
34. Chad KE, Bailey DA, McKay HA, et al. The effect of a weight-bearing physical activity program on bone mineral content and estimated volumetric density in children with spastic cerebral palsy. *J Pediatr*. 1999;135:115–117.
35. Wren TA, Lee DC, Hara R, et al. Effect of high-frequency, low-magnitude vibration on bone and muscle in children with cerebral palsy. *J Pediatr Orthop*. 2010;30:732–738.
36. Chen CL, Chen CY, Liaw MY, et al. Efficacy of home-based virtual cycling training on bone mineral density in ambulatory children with cerebral palsy. *Osteoporos Int*. 2013;24:1399–1406.
37. Crosley CJ, Chee C, Berman PH. Rickets associated with long-term anticonvulsant therapy in a pediatric outpatient population. *Pediatrics*. 1975;56:52–57.
38. Lifshitz F, Maclaren NK. Vitamin D-dependent rickets in institutionalized, mentally retarded children receiving long–term anticonvulsant therapy. I. A survey of 288 patients. *J Pediatr*. 1973;83:612–620.
39. Mimaki T, Walson PD, Haussler MR. Anticonvulsant therapy and vitamin D metabolism: evidence for different mechanisms for phenytoin and phenobarbital. *Pediatr Pharmacol (New York)*. 1980;1:105–112.
40. Tolman KG, Jubiz W, Sannella JJ, et al. Osteomalacia associated with anticonvulsant drug therapy in mentally retarded children. *Pediatrics*. 1975;56:45–50.
41. Position of the American Dietetic Association: nutrition in comprehensive program planning for persons with developmental disabilities. *J Am Diet Assoc*. 1997;97:189–193.
42. Boyle J. Nutritional management of the developmentally disabled child. *Pediatr Surg Int*. 1991;6:76–81.
43. Johnson RK, Maeda M. Establishing outpatient nutrition services for children with

cerebral palsy. *J Am Diet Assoc*. 1989;89:1504–1506.
44. Wodarski LA. An interdisciplinary nutrition assessment and intervention protocol for children with disabilities. *J Am Diet Assoc*. 1990;90:1563–1568.
45. Eddy TP, Nicholson AL, Wheeler EF. Energy expenditures and dietary intakes in cerebral palsy. *Dev Med Child Neurol*. 1965;7:377–386.
46. Krick J, Murphy PE, Markham JF, Shapiro BK. A proposed formula for calculating energy needs of children with cerebral palsy. *Dev Med Child Neurol*. 1992;34:481–487.
47. Food and Nutrition Board The Institute of Medicine. *Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids (Macronutrients)*. Washington, DC: National Academies Press; 2002.
48. Fried MD, Pencharz PB. Energy and nutrient intakes of children with spastic quadriplegia. *J Pediatr*. 1991;119:947–949.
49. Patrick J, Boland M, Stoski D, Murray GE. Rapid correction of wasting in children with cerebral palsy. *Dev Med Child Neurol*. 1986;28:734–739.
50. Rempel GR, Colwell SO, Nelson RP. Growth in children with cerebral palsy fed via gastrostomy. *Pediatrics*. 1988;82:857–862.
51. Sanders KD, Cox K, Cannon R, et al. Growth response to enteral feeding by children with cerebral palsy. *JPEN J Parenter Enteral Nutr*. 1990;14:23–26.
52. Shapiro BK, Green P, Krick J, et al. Growth of severely impaired children: neurological versus nutritional factors. *Dev Med Child Neurol*. 1986;28:729–733.
53. Spender QW, Cronk CE, Charney EB, Stallings VA. Assessment of linear growth of children with cerebral palsy: use of alternative measures to height or length. *Dev Med Child Neurol*. 1989;31:206–214.
54. World Health Organization. *Energy and Protein Requirements*. Geneva, Switzerland: World Health Organization Technical Report Series No. 724; 1985.
55. Johnson RK, Hildreth HG, Contompasis SH, Goran MI. Total energy expenditure in adults with cerebral palsy as assessed by doubly labeled water. *J Am Diet Assoc*. 1997;97:966–970.
56. Walker JL, Bell KL, Boyd RN, Davies PS. Energy requirements in preschool-age children with cerebral palsy. *Am J Clin Nutr*. 2012;96:1309–1315.
57. Rieken R, van Goudoever JB, Schierbeek H, et al. Measuring body composition and energy expenditure in children with severe neurologic impairment and intellectual disability. *Am J Clin Nutr*. 2011;94:759–766.
58. Tanner JM. *Growth at Adolescence*. 2nd ed. Oxford, UK: Blackwell Scientific Publication; 1962.
59. Krick J, Murphy-Miller P, Zeger S, Wright E. Pattern of growth in children with cerebral palsy. *J Am Diet Assoc*. 1996;96:680–685.
60. Day SM, Strauss DJ, Vachon PJ, et al. Growth patterns in a population of children and adolescents with cerebral palsy. *Dev Med Child Neurol*. 2007;49:167–171.
61. Brooks J, Day S, Shavelle R, Strauss D. Low weight, morbidity, and mortality in children with cerebral palsy: new clinical growth charts. *Pediatrics*. 2011;128:e299–e307.
62. Harcke HT, Taylor A, Bachrach S, et al. Lateral femoral scan: an alternative method for assessing bone mineral density in children with cerebral palsy. *Pediatr Radiol*. 1998;28:241–246.
63. Henderson RC, Lark RK, Newman JE, et al. Pediatric reference data for dual X-ray absorptiometric measures of normal bone density in the distal femur. *AJR Am J Roentgenol*. 2002;178:439–443.
64. Moyer-Mileur LJ, Mughal Z, Fung EB. Children with special considerations. In:

Sawyer AJ, Bachrach LK, Fung EB, eds. *Bone Densitometry in Growing Patients*. Totowa, NJ: Humana Press; 2006:137–158.
65. Zemel BS, Stallings VA, Leonard MB, et al. Revised pediatric reference data for the lateral distal femur measured by Hologic Discovery/Delphi dual-energy X-ray absorptiometry. *J Clin Densitom*. 2009;12:207–218.
66. Greulich W, Pyle S. *Radiographic Atlas of Skeletal Development of the Hand and Wrist*. CA: Stanford University Press; 1950.
67. Tanner J, Healy M, Goldstein H, Cameron N. *Assessment of Skeletal Maturity and Prediction of Adult Height (TW3) Method*. London, UK: W.B. Saunders; 2001.
68. Lohman TG, Roche AR, Martorell R. *Anthropometric Standardization Reference Manual*. Champaign, IL: Human Kinetics; 1988.
69. World Health Organization. *WHO child growth standards: length/height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age*. Geneva, Switzerland: WHO Press; 2006.
70. Ogden CL, Kuczmarski RJ, Flegal KM, et al. Centers for Disease Control and Prevention 2000 growth charts for the United States: improvements to the 1977 National Center for Health Statistics version. *Pediatrics*. 2002;109:45–60.
71. WHO Multicentre Growth Reference Study Group. *WHO Child Growth Standards: Growth Velocity Based on Weight, Length and Head Circumference: Methods and Development*. Geneva, Switzerland: WHO; 2009.
72. Ross W, Carr RV, Carter JL. *Anthropometry Illustrated. The Human Animal Series*. Vol. 1. Surrey, BC: Turnpike Electronic Publications; 1999.
73. Himes JH, Roche AF, Thissen D, Moore WM. Parent-specific adjustments for evaluation of recumbent length and stature of children. *Pediatrics*. 1985;75:304–313.
74. World Health Organization. *WHO child growth standards: head circumference-for-age, arm-circumference-or-age, triceps skinfold-for-age and subscapular skinfold-for-age*. Geneva, Switzerland: WHO Press; 2006.
75. Roche AF, Mukherjee D, Guo SM, Moore WM. Head circumference reference data: birth to 18 years. *Pediatrics*. 1987;79:706–712.
76. Stevenson RD. Use of segmental measures to estimate stature in children with cerebral palsy. *Arch Pediatr Adolesc Med*. 1995;149:658–662.
77. Gauld LM, Kappers J, Carlin JB, Robertson CF. Height prediction from ulna length. *Dev Med Child Neurol*. 2004;46:475–480.
78. Snyder RG, Schneider L, Owings S. *Anthropometry of Infants, Children and Youths of Age 18 for Project Safety Design*. Bethesda, MD: Consumer Product Safety Commission Report No. UM-HSR1-77-17; 1977.
79. Oeffinger D, Conaway M, Stevenson R, et al. Tibial length growth curves for ambulatory children and adolescents with cerebral palsy. *Dev Med Child Neurol*. 2010;52:e195–e201.
80. Chumlea WC, Guo SS, Steinbaugh ML. Prediction of stature from knee height for black and white adults and children with application to mobility-impaired or handicapped persons. *J Am Diet Assoc*. 1994;94:1385–1388, 1391; quiz 1389–1390.
81. Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr*. 1981;34:2540–2545.
82. Spender QW, Cronk CE, Stallings VA, Hediger ML. Fat distribution in children with cerebral palsy. *Ann Hum Biol*. 1988;15:191–196.
83. Frisancho AR. *Anthropometric Standards for the Assessment of Growth and Nutritional Status*. Ann Arbor, MI: University of Michigan Press; 1990.
84. Cronk CE, Roche AF. Race- and sex-specific reference data for triceps and

subscapular skinfolds and weight/stature. *Am J Clin Nutr*. 1982;35:347–354.
85. Johnson CL, Fulwood R, Abraham S, Bryner JD. Basic data on anthropometric measurements and angular measurements of the hip and knee joints for selected age groups 1-74 years of age. *Vital Health Stat 11*. 1981;219:1–68.
86. Slaughter MH, Lohman TG, Boileau RA, et al. Skinfold equations for estimation of body fatness in children and youth. *Hum Biol*. 1988;60:709–723.
87. Gurka MJ, Kuperminc MN, Busby MG, et al. Assessment and correction of skinfold thickness equations in estimating body fat in children with cerebral palsy. *Dev Med Child Neurol*. 2010;52:e35–e41.
88. Liu LF, Roberts R, Moyer-Mileur L, Samson-Fang L. Determination of body composition in children with cerebral palsy: bioelectrical impedance analysis and anthropometry vs dual-energy x-ray absorptiometry. *J Am Diet Assoc*. 2005;105:794–797.
89. Rauch F, Plotkin H, DiMeglio L, et al. Fracture prediction and the definition of osteoporosis in children and adolescents: the ISCD 2007 Pediatric Official Positions. *J Clin Densitom*. 2008;11:22–28.
90. Zemel BS, Leonard MB, Kelly A, et al. Height adjustment in assessing dual energy x-ray absorptiometry measurements of bone mass and density in children. *J Clin Endocrinol Metab*. 2010;95:1265–1273.
91. Liu LF, Moyer-Mileur L, Gooch J, Samson-Fang L. The contribution of tone to resting energy expenditure in children with moderate to severe cerebral palsy: evaluation utilizing intrathecal baclofen injection. *J Pediatr Rehabil Med*. 2008;1:163–167.
92. van den Hecke A, Malghem C, Renders A, et al. Mechanical work, energetic cost, and gait efficiency in children with cerebral palsy. *J Pediatr Orthop*. 2007;27:643–647.
93. Culley WJ, Middleton TO. Caloric requirements of mentally retarded children with and without motor dysfunction. *J Pediatr*. 1969;75:380–384.
94. Stallings VA, Zemel BS. Nutritional assessment of the disabled child. Chapter 7. In: Sullivan PR, Rosenbloom L, eds. *Feeding the Disabled Child (Clinics in Developmental Medicine, No. 140)*. London, UK: Mac Keith Press; 1996:62–76.

Inborn Errors of Metabolism and the Liver

49

Charles P. Venditti, MD, PhD and Gerard T. Berry, MD

INTRODUCTION

The liver is a nutritional clearing house, metabolizing, partitioning, and distributing dietary nutrients and their metabolites both within the liver and among the different organs in the body. Inborn errors of metabolism involve small molecules, as well as large complex molecules and organelles.[1] It is not surprising that the liver with its varied array of anabolic and catabolic pathways features prominently in hereditary enzyme deficiencies, solute transport defects, and perturbations in organelle biogenesis.

Furthermore, the liver plays a central role in whole body metabolism. Hepatocytes, functionally heterogeneous in their spatial distribution along the tracts from the periportal to the centrilobular zones, are the prime movers in a complex process that involves (1) transport of water and solutes across membrane bilayers; (2) biosynthesis of small and complex molecules, the latter of which are largely destined for export; (3) interconversion of metabolites assimilated from ingested food such as amino acids; (4) catabolism of decaying macromolecules; (5) oxidation of fuels such as the free fatty acids, oleic, stearic, and palmitic acids, the most important energy source for hepatocytes; and (6) synthesis and export of glucose, as well as the hormonal regulation of its storage form, glycogen, the first line of defense in fasting adaptation.

The experiments of nature that result in the heritable defects in glycogenolysis, gluconeogenesis, mitochondrial fatty acid oxidation,

carnitine metabolism, ketogenesis, and glucose transport underscore the essential role of the liver in fasting adaptation. The endocrine system and especially circulating insulin govern the interorgan flux of metabolites that are converted to glucose in the liver or to the ketone bodies, acetoacetate and 3-hydroxybutyrate, that spare glucose. The liver is the site of ketogenesis but oxidation of ketone bodies only occurs in extrahepatic tissues as the hepatocytes are devoid of succinyl-CoA:3-oxyacid-CoA transferase activity (SCOT), the enzyme needed to utilize ketone bodies. Hepatic ketogenesis hinges on the carnitine-dependent translocation of long-chain fatty acids across the inner mitochondrial membrane. The temporal regulation of glycogen phosphorolysis, glucose-6-phosphate hydrolysis, gluconeogenic enzyme activities, carnitine palmitoyltransferase activity and, hence, fatty acid oxidation and ketogenesis depends on changing concentrations of hormones such as insulin and glucagon. Fasting produces a decrease in the ratio of insulin to glucagon, and the magnitude of this alteration depends on the duration of fasting. Inborn errors of hepatic glucose release and export, glycogenolysis, gluconeogenesis, and fatty acid oxidation/ketogenesis characteristically surface at different postprandial times following the last meal. Specifically, hypoglycemia may occur within just a few postprandial hours in a patient with glucose-6-phosphatase deficiency, whereas, of all the inborn errors, a defect in fatty acid oxidation requires the longest period of fasting to result in hypoglycemia.

INBORN ERRORS OF METABOLISM

Many inborn errors of metabolism affect the liver. In some of these biochemical genetic disorders, such as phenylketonuria (PKU), the enzyme function in the hepatocyte is defective but the complications related to accumulation of phenylalanine metabolites are extrahepatic. Other disease types globally affect the hepatocyte or damage one or more organelles. In addition to hepatocytes, the liver contains other cell populations, such as the Kupffer cells, which can also be affected; this may lead to alterations in hepatic function or size. Mechanisms of hepatic injury, including discrete involvement of organelles (as evidenced by biochemical or pathologic findings) are discussed for selected disorders of amino acid, ammonia, organic acid, carbohydrate, glycogen, fatty acid, complex lipid, cholesterol, bile salt, glycosaminoglycan, glycoprotein, and metal metabolism.

Timing of the Presentation of Inborn Errors of Metabolism

Inborn errors of metabolism, especially as envisioned by Garrod in 1909, are most commonly thought of as being the so-called small molecule diseases, in which the pathophysiology is usually a result of the excess of a potentially toxic intermediate. The absence of a necessary substrate more often concerns the symptoms in small molecular diseases. Nutritional treatment approaches typically focus on minimizing pathometabolite production, usually by dietary precursor restriction, while trying to balance demands for substrate utilization and interconversion with obligatory metabolism. Anaplerotic strategies, aimed at relieving product deficiency past the site of a block in a pathway, also have a place in the treatment of inborn errors of metabolism. Yet, the large molecule or complex macromolecule disorders, and, in particular, the storage diseases are now the subject of intense scrutiny as enzyme replacement therapy has transformed molecular history. This division is artificial in that all of the biochemical genetic diseases arise as a result of abnormalities in protein structure and function. In turn, these abnormalities are caused by alterations in nuclear or mitochondrial genes and their expression.

Most disorders of intermediary metabolism may not show signs in the fetus. The placenta and maternal circulation provide a constant supply of calories, which reduces the metabolic stress on the affected pathway of the fetus. Maternal hepatic function is also a means of elimination of many of the toxic metabolites that would otherwise cause symptoms. The free exchange of metabolites from the affected fetus to the maternal circulation has been demonstrated in a number of disorders. However, a metabolic toxicity syndrome featuring maternal liver disease can be seen when the fetus is affected with one of several disorders. The best-described syndrome is when the fetus is affected with long-chain hydroxylacyl-CoA dehydrogenase (LCHAD) deficiency.[2] Other inborn errors in which this can be seen include carnitine palmitoyl transferase type I (CPT I) deficiency[3] and short-chain acyl-CoA dehydrogenase (SCAD) deficiency.[4] A history of maternal illness during pregnancy should always be sought when evaluating patients for metabolic disease.

Prenatal effects can be seen in several different classes of inborn errors. Lesions of the electron transport chain can feature multiple antenatal effects such as intrauterine growth retardation.[5] Other disorders of energy metabolism—such as deficiencies of pyruvate carboxylase, pyruvate dehydrogenase, and fumarase can feature central nervous system

(CNS) dysgenesis.[6] Renal cystic changes are seen in multiple acyl-CoA dehydrogenase deficiency,[7] the neonatal variant of carnitine palmitoyl transferase II (CPT II) deficiency,[8] and in the congenital disorders of glycosylation (CDG).[9] Peroxisomal disorders may have a combination of dysmorphic features and hepatocellular involvement by birth.[10] A number of sterol synthetic defects—including Smith–Lemli–Opitz syndrome, lathosterolosis, desmosterolosis, and Conradi-Hunerman syndrome—can feature multiple malformations.[11] These patients can develop cholestatic liver disease and may secrete inadequate quantities of normal bile acids. More specifically, hepatobiliary malformations and developmental defects have been noted to also occur in the setting of hereditary metabolic disease such as ductal plate malformations, which is seen in phosphomannose isomerase deficiency[12]; a CDG; and biliary atresia, which has been documented in one set of siblings with Niemann-Pick type C disease[13] and one patient with deficiency of the mitochondrial trifunctional protein.[14]

A few storage disorders may appear with prenatal or peripartum pathology caused by the accumulation of normal cellular materials as the result of a block at one or more steps of their degradation. Examples in which the disease may be evident at birth include several lysosomal storage disorders, glycogen storage disease (GSD) type IV, CDG type I, and Niemann-Pick type C.[15] The recognition of accumulated material in the liver and in other tissues, or the secondary alteration of the somatic structure as a result of the perturbations in metabolism, may take months to occur. Early nutritional manipulations have not been studied in the settings described above.

Each inborn error of metabolism can arise as the result of more than one DNA mutation within the gene. Each mutation can have different effects on the phenotype because of varying levels of residual function of the affected protein. In the case of multimeric proteins, the net activity of the complex may be different based on the structure of the altered subunits. The residual activity, the net metabolic stress on the body (in the case of catabolic errors in intermediary metabolism), and the amount of substrate combine to determine the timing of the recognition of the altered phenotype. Epigenetic or non-genic factors also can influence the phenotype seen in recessive enzymopathies in a less predictable fashion, best evidenced in the intrafamilial variability seen in cystic fibrosis.

Categories of Inborn Errors of Metabolism Involving the Liver and Gastrointestinal Tract

Table 49-1 outlines many of the disorders that have a known disturbance in the structure or function of the liver and GI tract as a result of inborn errors of metabolism in the pediatric population. Selected examples of these disorders are discussed briefly in the subsection on mechanisms of pathogenesis and comments on the nutritional approaches to management are highlighted in the text and listed in Table 49-1.

A few examples of metabolic diseases in which the liver serves the primary role in maintaining normal extrahepatic concentrations of metabolites and in which the signs are exclusively extrahepatic should be mentioned. A prime example of the importance of hepatic metabolism as it relates to CNS effects is PKU due to defects in phenylalanine hydroxylase. The enzymatic blockage prevents the conversion of phenylalanine to tyrosine. Consequences of the increased phenylalanine and lowered tyrosine concentrations include alterations in the transport of neutral aromatic amino acids into the brain and the production of neurotransmitters. As a result, mental retardation and seizures can occur. By lowering the blood phenylalanine concentration, many of the effects on cognition can be prevented or reversed. Hepatic size, synthetic functions, and laboratory tests are normal. A low phenylalanine diet provides the mainstay of therapy for these patients.

Nonketotic hyperglycinemia (NKH) is due to defects in the glycine cleavage system. Although the system is found in the brain and kidney, the majority of the activity is expressed in the liver. Except in the atypical form, the failure of the cleavage system results in the accumulation of glycine in all tissues; however, the pathophysiology is due to elevated glycine levels in the CNS. Unlike PKU, in which no symptoms are seen in the early neonatal period, infants with NKH may present with intractable seizures within hours of birth. Glycine levels are massively elevated in the cerebrospinal fluid in the neonatal period, implying that intrauterine elevations are a feature of this disorder. The liver in these patients is unremarkable.

Another example of an inborn error of metabolism where the liver is responsible for whole body metabolism is hyperoxalosis type I, a disorder of glyoxyalate metabolism. It is caused by a deficiency of alanine-glyoxylate aminotransferase, which is due to either a mutant protein or a mistargeting event whereby the enzyme localizes to the mitochondria in lieu of the peroxisomes, where it normally participates in glyoxyalate metabolism. Patients with this disorder progressively deposit calcium oxalate crystals throughout the body because of increased production of oxalate by the liver. The liver is spared, but many other organs—including

the kidneys, heart, skin, retina, peripheral nerves, arteries and skeleton—are not. Nephrocalcinosis eventually leads to end-stage renal failure and ultimately death. Liver or combined liver–kidney transplantation is needed to treat this disorder, depending on the duration and magnitude of the blood oxalate elevation. A liver is transplanted to stop the production of oxalate and provide a reserve for the defective enzyme, and in patients with coexisting renal failure, kidney transplantation is also required to provide essential functions and ensure adequate oxalate clearance.

TABLE 49-1 Metabolic Disorders that Feature Liver Disease/Gastrointestinal Dysfunction

Class of Disorder	Enzyme Defect	Nutritional/Medical/S Treatment
Carbohydrate Metabolism		
Galactosemia	Galactose-1-phosphate uridyltransferase	Remove lactose from diet
Hereditary fructose intolerance	Fructose 1-phosphate aldolase	Remove sucrose, fructose sorbitol from diet
Fructose 1,6-bisphosphatase deficiency	Fructose 1,6-bisphosphatase	Avoidance of fasting; Rem sucrose, fructose and sor diet
Glycogen synthase defect (GSD0)	Glycogen synthase	Avoidance of fasting
Glycogen storage disease (GSD)	Glucose-6-phosphatase	Continuous calories, noct cornstarch feedingstype I Liver transplantation?
Glycogen storage disease (GSD type Ib)	Glucose-6-phosphate translocase (G6PT)	Continuous calories, noct cornstarch feedings Liver transplantation?
GSD, type III	Amylo-1, 6-glucosidase (debrancher enzyme)	High carbohydrate diet, fr feedings may be needed
GSD, type IV	Amylo 1,4 ->1,6 transglucosidase (brancher enzyme)	Pts may need liver transp
GSD, type VI	Liver phosphorylase	Therapy depends on sym
GSD-IXa		
GSD-IXb		
GSD-IXc (PHKG2)		
Phosphorylase kinase deficiency	Phosphorylase kinase	Therapy depends on sym
Fanconi Bickel Syndrome	GLUT2	Pts always have a renal F syndrome

Transaldolase deficiency[34]	Transaldolase	Cirrhosis in early infancy ; children with hypomorphic mutations
Glycerol-3-phosphate dehydrogenase I deficiency		Neutral lipid accumulation and transient hyperlipiden infancy
Citrullinemia type 2	Citrin (SLC25A13 gene)	Transient infantile cholest syndrome
Amino- and Organic Acid Disorders		
Tyrosinemia, type I	Fumarylacetoacetate hydrolase	NTBC, dietary tyrosine re Eventual need for liver transplantation
Maple syrup urine disease	Branch chain oxoacid dehydrogenase	Leucine restriction; isoleu valine supplementation us needed thiamine in respo Liver transplantation?
Propionic acidemia	Propionyl-coenzyme A carboxylase A, B	Branched chain amino ac restriction, carnitine supplementation Liver transplantation?
Methylmalonic Acidemia	L-Methylmalonyl coenzyme A mutase (*mut* class) MMAA (*cblA* class) MMAB (*cblB* class)	Branched chain amino ac restriction, carnitine supplementation hydroxylcobalamin IM (no patients respond) Liver transplantation?
Urea Cycle and Related Disorders		
Carbamyl phosphate synthase deficiency	Carbamyl phosphate synthetase	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-citrulline Liver transplantation?
Ornithine transcarbamylase (OTC) deficiency	Ornithine transcarbamylase	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-citrulline Liver transplantation?
Citrullinemia	Argininosuccinate synthase	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-arginine Liver transplantation?
Argininosuccinic acidemia	Argininosuccinate lyase	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-arginine Liver transplantation?

N-Acetyl glutamate synthase deficiency	N-acetyl glutamate synthetase	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-citrulline N-carbamyl glutamate Liver transplantation?
Hyperornithinemia-hyperammonemia-homocitrullinuria (HHH) syndrome	ORNT1 mitochondrial ornithine transporter	Stringent dietary protein r Sodium benzoate and/or phenylbutyrate L-citrulline
Lysinuric protein intolerance	SLC7A7	Stringent dietary protein r L-citrulline, HSM
Fatty acid oxidation defects		
Carnitine palmitoyltransferase-I (CPT-I) deficiency	Carnitine palmitoyltransferase-I	Avoidance of fasting Carnitine may be useful Can be associated with m HELLP syndrome
Carnitine palmitoyltransferase-II (CPT-II) deficiency	Carnitine palmitoyltransferase-II	Avoidance of fasting Carnitine may be useful Can be associated with d kidneys and brain malforn
Primary carnitine deficiency	Na^+/carnitine cotransporter	Treatment with L-carnitine eliminate fasting hypoglyc liver disease, cardiomyop myopathy
Carnitine-acylcarnitine translocase deficiency	Carnitine-acylcarnitine translocase	Neonatal lethality in most
Very long chain acyl-CoA dehydrogenase (VLCAD) deficiency	Very long chain acyl-CoA dehydrogenase	Avoidance of fasting Carnitine may be useful Probably not a Mendelian
ACAD9 deficiency	Very long chain acyl-CoA dehydrogenase	RC complex I assembly fa
Long chain 3-hydroxyl acyl-CoA dehydrogenase (LCHAD) deficiency	Long chain 3-hydroxy acyl-CoA dehydrogenase	Avoidance of fasting Carnitine therapy may pro dysrythmias Can be associated with m HELLP syndrome
Medium chain acyl-CoA dehydrogenase deficiency	Medium chain acyl-CoA dehydrogenase	Avoidance of fasting Carnitine may be useful
Short chain acyl-CoA dehydrogenase (SCAD) deficiency	Short chain acyl-CoA dehydrogenase	Can be associated with m HELLP syndrome
HMG-CoA lyase deficiency	3-hydroxy-3-methylglutaryl-CoA lyase	Avoidance of fasting
Multiple Acyl-CoA dehydrogenase deficiency	ETFA, ETFB, ETFDH	Can be associated with c anomalies

		Riboflavin therapy is bene certain mutations
Mitochondrial energy production/Primary lactic acidosis syndromes		
LARS disease	Leucyl-tRNA synthetase deficiency	Translational defect
Electron transport chain (ETC) deficiencies	Complexes I, II, III, IV (Cytochrome c oxidase), V (ATPase) of the ETC	Clinically heterogeneous, prenatal manifestations
POLG1 MPV17 TWINKLE Deoxyguanosine kinase (DGOUK)	DNA Polymerase-gamma 1	Mitochondrial DNA deplet disorder Mitochondrial DNA deplet disorder Mitochondrial DNA deplet disorder Mitochondrial DNA deplet disorder
GRACILE syndrome	BCS1L gene defect	Neonatal lethality
Succinyl-CoA synthetase deficiency	SUCLA2 SUCLG1	Liver disease/mitoDNA de Liver disease/mitoDNA de
Thymidine phosphorylase deficiency		MNGIE/mitoDNA depletio Bone marrow transplanta
Pyruvate carboxylase deficiency	Pyruvate carboxylase	Features neurodegenerat
PEPCK deficiency	Phosphoenolpyruvate carboxykinase	Extremely rare, may be a with cirrhosis
Storage Disorders		
Gaucher, type I, II and type III	β-glucosidase	Can feature nonimmune h fetalis
Niemann Pick, type A	Acid sphingomyelinase	
Niemann Pick, type B		
Niemann Pick, type C (NPC type 1)	NPC1 gene	
Niemann Pick, type C (NPC type 2)	NPC2 gene, NPC2	Can feature nonimmune h fetalis
Sandhoff disease	HEXB gene	
Sialidosis, type II	Neuraminidase or alpha-N-acetyl neuroaminidase	Can feature nonimmune h fetalis
Galactosialidosis	Protective protein deficiency	Can feature nonimmune h fetalis
Sialic acid transport disorder (infantile free sialic acid storage disease)	SLC17A5	Can feature nonimmune h fetalis
I-cell disease	*N*-acetylglucosaminyl-1-	Can feature nonimmune h

	phosphotransferase	fetalis
GM1 gangliosidosis	β-galactosidase	Can feature nonimmune h fetalis
Multiple sulfatase deficiency	SUMF1 (sulfatase-modifying factor-1 gene)	
Wolman disease	Lysosomal acid lipase	Can feature nonimmune h fetalis
Farber Disease type I	Ceramidase	Can feature nonimmune h fetalis
Mucopolysaccharidosis, type VII	β-glucuronidase	Can feature nonimmune h fetalis
Congenital disorders of glycosylation		
Multiple enzymopathies; phosphomannomutase (type I) N-acetylglucosaminyl-transferase II (type II),	GDP-Man:GlcNAc2-PP-dolicholmannosyltransferase	Can feature hyperinsulinis protein-losing enteropathy nonimmune hydrops fetal Oral mannose for phosphomannomutase de
Phosphoglucomutase 1 deficiency		Hepatomegaly, hypoglyce uvula/cleft palate, Certain features respond galactose therapy
Sterol Synthesis Defects		
Smith-Lemli-Opitz syndrome	7-dehydrocholesterol reductase	Malformation syndrome
Lathosterolosis	3B-hydroxy steroid-delta-5 desaturase	Malformation syndrome
Desmosterolosis	24-dehydrocholesterol reductase	Malformation syndrome
Conradi-Hünermann syndrome	3B-hydroxysteroid-8,7-isomerase, others	Malformation syndrome
Mevalonic acidemia	Mevalonate kinase	Dysmorphic features, HSI
Peroxisomal Biosynthesis and Function		
Zellweger disease	Peroxisomal biogenesis defects	Dysmorphic features, malformations
Infantile Refsum disease	Peroxisomal biogenesis defects	Dysmorphic features
Bile salt metabolic defects	Delta(4)-3-oxosteroid 5β reductase, 3 β-hydroxy-Δ5 steroid dehydrogenase-isomerase, Oxysterol 7α-hydroxylase	Cholestatic liver disease treat first two with chenodeoxycholic acid +/– cholic acid
Metal Metabolism		
Neonatal iron storage disease/perinatal	Defect(s) uncertain	Prenatal and neonatal letl Liver transplantation?

hemochromatosis		
Juvenile hemochromatosis	hemojuvelin; hepcidin antimicrobial peptide	
Hemochromatosis, other	HFE 3 (transferrin receptor-2) HFE4 (ferroportin) Atransferrinemia Aceruloplasminemia	Autosomal dominant inhe Neurological syndrome
Wilson disease	ATP7B	Penicillamine
Indian childhood cirrhosis	Defect(s) uncertain	
Idiopathic copper toxicosis	Defect(s) uncertain	
Other disorders		
Glycine-*N*-methyltransferase	Glycine-*N*-methyltransferase	A cause of hypermethione
Progressive Familial Intrahepatic Cholestasis Syndromes (PFIC1-4), including Byler disease (PFIC1)	ATP8B1, ABCB11, ABCB4; Progressive PFIC3-beta-hydroxy-delta-5-C27-steroid oxidoreductase	PFIC1-3 may encode tran proteins
Vitamin Malabsorption Disorders and Chronic Diarrhea		
Congenital folate malabsorption	SLC	IM folinic acid
Imerslund-Grasbeck disease	CUBN (cubilin)	IM OH cobalamin
Intestinal biotin transporter defect	Unknown etiology	One patient reported to da
Chronic diarrhea	Congenital chloride diarrhea	High chloride diet, electro replacement
	Congenital secretory diarrhea due to defective sodium/hydrogen exchange	Electrolyte replacement, a balance
	glucose-galactose malabsorption	Remove glucose and gala from diet, provide fructose
	lactase deficiency	Avoid lactose
	sucrase-isomaltase deficiency	Avoid sucrose, symptoma with oral yeast and yeast-extracts
A/hypobetalipoproteinemia (A)	MTTP gene (recesive)	
Hypo A/hypobetalipoproteinemia	APOB gene (dominant)	

The last group of metabolic disorders includes those in which the signs and symptoms can be hepatic and extrahepatic. Included are a large

number of conditions, such as the urea cycle, fatty acid oxidation, and organic acid disorders, as well as others that affect storage and transport. Liver transplantation has been employed as a treatment modality in some of these diseases with variable success.[16]

Hepatic Injury

The metabolic processes within the hepatocyte, as in any cell, are closely regulated and interactive; they are responsive to external as well as internal stimuli. Central to the well-being of any cell are adequate stores of energy or the means to produce energy from readily available substrates as needed. With this energy, the cell maintains its homeostasis, grows, synthesizes material for export, exports, transports, and recycles unneeded or decaying materials. All of these processes are affected by one or more biochemical genetic disorders.

Compartmentalization of cellular functions is a necessary part of the synthetic and degradative pathways, as well as being essential for the production and maintenance of electrochemical gradients in energy production. Hepatic metabolism is quantitatively and even qualitatively different in the periportal inflow zone compared to the perivenous outflow area. Periportal cells are responsible for the majority of gluconeogenesis, glycogen synthesis by the indirect pathway, urea synthesis, and fatty acid oxidation. This regionalization may allow for discontinuous histopathologic findings during acute and less than massive metabolic insult. As the degree of hepatic injury becomes more severe, or as the duration of repeat injury increases and fibrosis occurs, the distinctions become less obvious.

Pathophysiology

For many of the biochemical genetic disorders, common histologic appearance and laboratory perturbations exist, precluding exact diagnosis without the use of extraordinary testing. The common pathways of hepatic pathophysiology include inflammation, necrosis, and cholestasis as the result of alterations in homeostasis; steatosis as the result of the accumulation of intermediates and the response to metabolic stress; and the accumulation of relatively nontoxic macromolecules, which usually alter cellular function on the basis of mass effects rather than chemical composition. It is also clear that the pathophysiology in some instances is different with many metabolic diseases if the patient is a newborn infant rather than a child or an adult. A factor that can confound diagnosis of a

metabolic disorder is the nonspecific malhandling of metabolites by a diseased liver, particularly methionine and tyrosine.

Cholestasis and Hepatobiliary Disease

Cholestasis can result from multiple groups of metabolic disorders. Defects in bile acid metabolism, absence of peroxisomes, and disorders of amino acid, lipid, carbohydrate, or complex molecule degradation, metal metabolism, and transport can lead to the recognition of cholestasis.

Cystic fibrosis, an extremely common autosomal recessive disease, is due to defects in the cystic fibrosis transmembrane conductance regulator (CFTR), whose gene is located on chromosome 7q31.2. Although respiratory disease, pancreatic exocrine insufficiency, and failure to thrive are easily recognized as components of the clinical spectrum, hepatobiliary disease may be seen in very early infancy with impaired intrahepatic and extrahepatic biliary drainage. CFTR is localized to the intrahepatic biliary epithelium and is a regulator of cholangiocellular bile production. Cholestasis is felt to be secondary to defective chloride transport across this epithelium. Focal biliary cirrhosis can occur early with later evidence of steatosis and fibrosis. In a minority of patients, prolonged neonatal jaundice with cirrhotic change is seen. In rare instances, liver transplantation may be required (see Chapter 55).

Other transport system defects can also cause a cholestatic change in the liver. Citrullinemia Type 2, a urea cycle disorder, can feature cholestasis as the neonatal hepatic presentation. A series of disorders—the progressive familial intrahepatic cholestasis syndromes—feature mutations in a variety of proteins likely involved in cannilicular transport. Four distinct genetic loci exist, and mutations have been demonstrated in patients from diverse backgrounds, including the Old Order Amish[17] and an Eskimo isolate from Greenland.[18] Three of the genes appear to encode transport-related proteins while the fourth encodes a steroid oxidoreductase.[19–21]

The genetic basis of other cholestatic syndromes, such as North American Indian childhood cirrhosis, appears to involve novel proteins and illustrate that the current understanding of the mechanisms of cholestasis are far from complete[22] (see Chapter 56).

Bile acid synthetic defects can present with cholestatic liver disease and progress to cirrhosis.[23] A number of these rare enzymopathies are known to exist and it is likely that others remain to be discovered. Cheondeoxycholic acid and cholic acid can be used to treat patients with

these disorders, in some instances, with great efficacy.

Hepatocellular Necrosis

Hepatocellular necrosis manifesting with jaundice, edema, ascites, hepatic synthetic failure, or hepatic encephalopathy is a major common pathology due to catabolic errors. The laboratory studies show variable degrees of elevation of serum transaminases, hypoglycemia, hyperammonemia, hypofibrinogenemia, and hypoprothrombinemia. The clinical picture is similar to that of sepsis, and the laboratory findings can be indistinguishable from those of a severe viral hepatitis (see Chapter 62).

Galactosemia is a typical example of a disorder in this group. Classic galactosemia is due to a severe deficiency of hepatic galactose-1-phosphate uridyl transferase (GALT) activity. As a result of significant impairment of the function of the enzyme, a syndrome of neonatal-onset toxicity occurs when the at-risk infant is exposed to significant amounts of dietary lactose or galactose. The toxic metabolites are galactose-1-phosphate (a substrate of the enzyme) and galactitol (the result of aldose reductase activity on the sugar). Because there is trapping of cellular phosphate as galactose-1-phosphate, hepatic intracellular ATP/ADP ratios may be altered. Interference with other enzymatic systems has been postulated. Both mechanisms can lead to severe acute hepatocellular damage, which is reversible if treated immediately. The acute manifestations of the disease in the extended neonatal period often are noted within a few days of the onset of milk feedings. Vomiting and diarrhea are common. Most patients present with jaundice caused by an unconjugated hyperbilirubinemia. Severe hemolysis occurs in some patients so that the clinical presentation resembles erythroblastosis fetalis. Ascites has been a prominent early finding. With the progression of hepatic disease, bile stasis, pseudoacinar formation, and portal fibrosis are seen. An end-stage cirrhotic liver can result whose histology is indistinguishable from that of other metabolic etiologies. Laboratory findings include derangements of hepatic function, albuminuria, hyperaminoaciduria, hyperchloremic metabolic acidosis, hypergalactosuria, and elevated blood galactose concentrations. There is increased production and elimination of galactitol and the presence of increased concentrations of tissue galactose-1-phosphate. In the untreated state, alterations in carbohydrate composition of glycoproteins, such as transferrin, can be seen paralleling the CDG.

The other affected organ systems in the neonate include the lens of the eye, the CNS, the immune system, and the renal tubules. Increased

synthesis of galactitol in the lens is associated with cataract formation. CNS findings in the infant include alterations in the level of consciousness and hypotonia. Cerebral edema and increased intracranial pressure can occur. A high frequency of *Escherichia coli* sepsis has been noted with fulminant courses. With prompt supportive therapy and the elimination of lactose and galactose from the diet, the acute toxicity syndrome resolves. Chronic, or long-term, effects of GALT deficiency occur in the CNS and ovary. Dietary therapy attempts to minimize offensive carbohydrates precursors from the diet but may never eliminate aberrant metabolites derived from endogenous production.

Hereditary fructose intolerance (HFI) usually presents with vomiting, diarrhea, and sometimes hypoglycemia. Symptoms are related to the ingestion of fructose or sucrose. The pathophysiology includes phosphate trapping, with resultant similar intrahepatic pathologic progression in the face of additional carbohydrate, and, as in galactosemia, there is renal tubular dysfunction. Beneficial effects from the withdrawal of the offending carbohydrates are seen in 2–3 days. Older patients characteristically avoid sucrose and have an unusual history of not having any dental caries. Two common mutations are known in the Caucasian population and molecular diagnosis is possible in some cases. Dietary therapy to eliminate fructose is completely effective at preventing the complications of this disorder.

Neonatal iron storage disease, or perinatal hemochromatosis, is a disorder resulting from severe prenatal hepatic disease. There is no single etiology for the disease; nor is there a clear genetic link to any single enzymatic defect, although a recessive pattern of inheritance seems likely. The liver shows diffuse fibrosis with hepatocellular nodular regeneration and ductal transformation. Siderosis in the apical cytoplasm of the tubular hepatocytes is typical. Some patients have had Down syndrome. A newer entity, GRACILE, reported in Finish infants, presents with growth retardation, lactic acidemia, anemia, and hemochromatosis.[24] The defect is in the BCS1L gene, involved in the assembly of complex III of the respiratory chain, and has been mapped to a locus on 2q33.37 and cloned. Other conditions may feature neonatal hemochromatosis, such as the tricho-hepato-enteric syndrome.[25] The neonatal onset cases, regardless of distinct etiology, all seem to be severe entities with a difficult clinical course. Although liver transplantation can be curative, some patients may benefit from antioxidant treatment without surgical intervention.

Juvenile hemochromatosis, another form of primary hemochromatosis, can be caused by mutations in hemojuvelin[26] or the gene encoding

hepcidin antimicrobial peptide.[27] This condition, like the more commonly seen adult variant caused by mutations in the HFE gene, can feature severe iron overload with hepatocytic iron deposition, liver fibrosis, hypogonadotrophic hypogonadism, and cardiac involvement. Two other genetic forms of primary hemochromatosis are also known. HFE3 is caused by mutations in the gene encoding transferrin receptor-2 and is intermediate in severity.[28] HFE4, the only documented autosomal dominant form of the disease, is caused by a mutant ferroportin gene.[29] Dietary restriction of iron is usually not effective in patients with hemochromatosis and many adults require prophylactic phlebotomy to reduce body iron stores.

Other inborn errors of metabolisms can feature hepatocytic iron deposition by unclear mechanisms. Atransferrinemia, caused by mutation in the structural gene for transferrin, produces a syndrome of microcytic anemia and iron overload and is treated by plasma infusions to provide functional transferrin. Aceruloplasminemia produces a syndrome of hepatic and CNS iron deposition. The basal ganglia seem to be most affected and the syndrome is dominated by neurological signs. Ceruloplasmin is low, as is serum iron, but excessive iron deposition is seen in the liver, brain, and pancreas (see Chapter 9).

Disorders of copper metabolism are another group of important metal metabolic diseases. The prototype is Wilson disease, an autosomal recessive disorder characterized by intracellular hepatic copper accumulation, cirrhosis, and, in a sizable number of patients, neurologic abnormalities. The primary defect resides in the ATP7B gene, which encodes a polypeptide that acts as a membrane copper-transport protein. The protein likely plays a role in intracellular copper delivery as well as release of copper into the bile. Like galactosemia and hereditary fructose intolerance, patients with Wilson disease can have a renal tubulopathy featuring aminaciduria and phosphaturia. Neurological and psychiatric presentations are well described. Chelation therapy can be used to enhance copper excretion. Some patients with hepatic failure will need liver transplantation. Another group of less well-characterized disorders of copper overload exist; these include Indian childhood cirrhosis and idiopathic copper toxicosis. The exact basis of these disorders is not known at present.

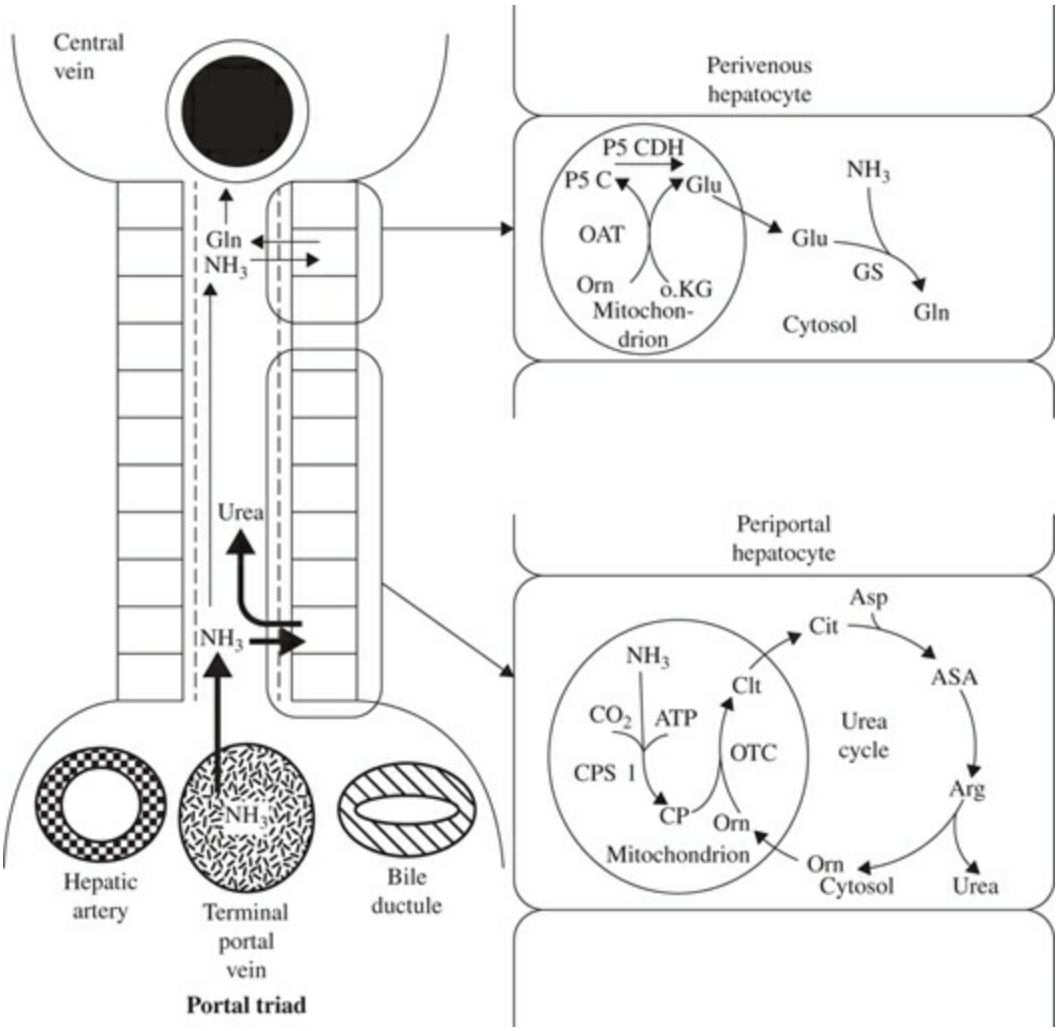


Figure 49-1 Depicted is the nonhomogeneous distribution of enzymes involved in ammonia metabolism in the hepatocytes of an acinar sinusoid as they are linearly distributed from the portal triad to the region of the central vein or terminal hepatic venule. The specific enzymatic reactions are shown for an individual periportal and perivenous hepatocyte. The glutamine synthetase (GS) and ornithine aminotransferase (OAT) enzyme activities are expressed exclusively in the one to three cell layers surrounding the central vein, that is, the region of zone 3 of the liver lobule, whereas the urea cycle enzymes are concentrated within the periportal hepatocytes. However, the urea cycle enzyme activities are higher in zone 1 immediately surrounding the portal triad than in the middle zone 2. The hepatocytes are shown as *squares*, the hepatic mitochondria as *shaded circles*, and the lining of the space of Disse as the *interrupted lines* on either side

of the linear array of hepatocytes. Arg, arginine; ASA, argininosuccinate; Asp, aspartate; ATP, adenosine triphosphate; Cit, citrulline; CP, carbamylphosphate; CPS-I, carbamylphosphate synthetase type 1; Gln, glutamine; Glu, glutamate; α-KG, α-ketoglutarate; NH_3, ammonia; CO_2, CO_2 or bicarbonate; Orn, ornithine; P5C, pyrroline 5-carboxylate; P5CDH, pyrroline-5-carboxylate dehydrogenase. B. The medications sodium phenylacetate and phenylbutyrate and sodium benzoate promote alternative waste nitrogen disposal by participating in these two mitochondrial reactions. (Adapted from Tuchman M, Lichenstein GR, Rajagopal BS, et al. Hepatic glutamine synthetase deficiency in fatal hyperammonemia after lung transplantation. *Ann Intern Med*. 1997;127:447.)

Urea cycle enzyme disorders may appear as early as 1–3 days of age. Figure 49-1 shows a functional overview of the urea cycle and highlights the compartmentalization of metabolism within the cellular zones of the liver. Infants show combinations of vomiting, lethargy, increased respiratory drive, and CNS dysfunction that may progress to seizure activity. Their glucose concentrations are normal, with normal or slightly alkalotic blood pHs. Markedly elevated plasma ammonium concentrations are a common feature. Blood urea nitrogen concentrations may be low. The common pathogenesis is an inability to detoxify ammonia, which is produced as the result of the catabolism of amino acids. Thus, either protein ingestion or catabolic stress can trigger hyperammonemia. The presentation of the individual disorders is nonspecific, and specialized laboratory testing such as quantitative plasma amino acid analysis and measurement of urinary orotic acid concentration is used to separate the diseases. The enzymes of ureagenesis are present in intestinal, renal, and hepatic compartments, but only in the liver are the full complement of activities of the urea cycle present to enable the conversion of NH_3 to urea. In the liver, only the proximal enzymes *N*-acetyl glutamate synthase, carbamoylphosphate synthetase, and ornithine transcarbamylase are localized within the mitochondrial matrix. In biopsied material examined by electron microscopy, pleomorphic mitochondria with swollen cristae and electron-dense matrices are seen. Other hepatocellular changes include alterations in the appearance of the endoplasmic reticulum. Chronic injury, as the result of either prolonged hyperammonemia or repeated episodes of metabolic crisis, may cause portal fibrosis. Synthetic function of the liver

is normal, although elevations in serum transaminases can occur. Such elevations may be present between episodes of hyperammonemia and despite the use of ammonia-trapping agents and enhancers of alternative nitrogen metabolic pathways. In the acute hyperammonemic crisis, detoxification by hemodialysis or hemofiltration may be essential.

In alpha-1 antitrypsin deficiency due to the PI ZZ phenotype, the protein has a lysine residue instead of a glutamic acid at position 342. The altered protein is not secreted, because the reactive loop center of one molecule is inserted into the beta-pleated sheet structure of a second molecule. This protein is retained within the endoplasmic reticulum, as is reflected by the accumulation of periodic acid–Schiff (PAS)–positive diastase-resistant material. In some susceptible individuals, the accumulation of the abnormal protein damages the endoplasmic reticulum or Golgi membranes, leading to hepatocellular damage and cirrhosis. Liver transplantation is required in some instances.

Tyrosinemia type I can present in early infancy with varying degrees of hepatomegaly and a mild elevation of serum transaminases. The biochemical lesion is a defect in fumarylacetoacetate hydrolase. Common features also include abnormalities of coagulation, with clotting factors often markedly reduced. Vitamin K does not correct the disorder. Renal dysfunction is usually evident. A porphyuric presentation with neuropathy due to increased succinylacetone can be seen in the acute setting. Specialized testing (i.e., quantitative analysis of plasma amino acids by anion exchange chromatography) usually reveals hypertyrosinemia, an increased urinary excretion of succinylacetone, and an elevated serum alpha-fetoprotein concentration. The hepatic disease is progressive and advances from microscopic cirrhosis, which can be seen in the first weeks of life, to macronodular cirrhosis with regenerating nodules. Hepatocellular carcinomas are frequent. In this disorder, biochemical abnormalities may be seen prenatally. The molecular mechanism of the hepatic and renal damage is not known, although the intracellular compartmentalization of toxic metabolites has been proposed to induce local damage. The inhibition of 4-hydroxyphenylpyruvate dioxygenase by fumarylacetoacetate is a major factor in the recognized biochemical abnormalities, and treatment with 2-(nitro-4-trifluoromethylbenzoyl)-1,3-cyclohexanedione (NTBC) corrects these but does not always prevent the progression to hepatocellular carcinoma. Dietary therapy consists of limiting tyrosine intake in conjunction with NTBC administration. Serum alpha-fetoprotein, a marker of hepatocellular carcinoma, and routine liver imaging studies are longitudinally followed to help decide which patients

need to undergo liver transplantation.

The ACAD9 deficiency is a defect in very long chain fatty acid oxidation but secondarily results in a mitochondrial respiratory chain defect involving Complex I. Severe defects may be associated with hepatic failure (reference Vockley).

Steatosis

Steatosis can be associated at some point in the natural history of a number of the organic acidopathies that result from enzymatic defects in the metabolism of branched-chain amino acids or odd-chain fatty acids. Propionic acidemia and methylmalonic acidemia, due to defects in either subunit of the heterodimeric propionyl coenzyme A carboxylase or the activity of the enzyme methylmalonyl-CoA mutase, are the prototypes for these disorders. Figure 49-2 displays the location of the blocks in the pathway of propionyl-CoA metabolism. Affected infants present with severe metabolic acidosis, refusal to feed, vomiting, and alterations in CNS function. Hepatomegaly is found in a fraction of the patients. The livers at autopsy have shown fatty infiltration and necrosis, and the reticuloendothelial cells may have increased iron deposits. The exact mechanism of the fat deposition is unknown but may relate to an imbalance between the rate of fat synthesis and the rate of export of lipoproteins. Laboratory findings of a metabolic ketoacidosis with hyperammonemia and hyperglycinemia are characteristic during the initial presentation and subsequent metabolic crises.

Almost all the disorders of beta-oxidation of fatty acids from defects in carnitine palmitoyltransferase I, carnitine/acylcarnitine translocase, and carnitine palmitoyltransferase II through to the utilization of hydroxy-, medium-chain, or short-chain fatty acids have been associated with some degree of hepatic dysfunction and fat accumulation. An overview of these disorders is depicted in Figure 49-3. In the majority of cases when histology has been performed, the accumulation occurs as macrovesicular lipid storage. Hypothetical mechanisms of fat accumulation in these disorders include effects of a constant supply of fatty acids with a block in their utilization, increased availability of glycerol, and net intracellular accumulation. Dietary management of this group of disorders varies with the nature of the block but generally includes avoidance of fasting, restriction of dietary amino acid (for organic acidurias) and fat intake, and careful monitoring for nutritional status. Anaplerotic therapy remains investigational. The newly described glycerol-3-phosphate dehydrogenase 1 deficiency is associated with hepatic steatosis and transient

hyperlipidemia.[30]

Hepatic Pathology Resulting From Peroxisomal Disease

The absence of hepatocyte organelles and their functions can result in recognized hepatic disease in late fetal or early infant life. The best characterized group of these disorders includes those due to the peroxisomal diseases, which comprise a group of disorders with effects on multiple viscera, skeletal development, and the CNS. Zellweger disease, the paradigm for peroxisomal disorders, is caused by biosynthetic defects in the formation of the peroxisome. As a result, more than 50 enzymes involved in multiple pathways can be perturbed. The peroxisomes are responsible for the oxidation of amino acids, alcohols, dicarboxylic acids, pipecolic acid, and phytanic acid, as well as very long-chain fatty acids. They are also essential for the synthesis of bile acids, ether-phospholipids such as the plasmalogens, and cholesterol, and they are a major reservoir of the hydrogen peroxide–degrading enzyme, catalase. Zellweger disease was also called cerebrohepatorenal disease because of the organ systems involved in the neonate. The infants have distinct dysmorphic features, an enlarged fontanelle, profound hypotonia, hepatomegaly, and neurodegeneration. Neuronal migratory defects are often seen. The liver has micronodular cirrhosis and fibrosis on examination. The laboratory abnormalities are explained by the missing multiple enzyme functions. Diagnosis can be strongly suspected by the constellation of physical findings and is confirmed by the abnormalities in very long-chain fatty acid metabolism. The dysmorphia varies with the severity of the defect. For example, infantile Refsum disease, considered to be a milder phenotype of the peroxisomal biogenesis disorders, generally presents with failure to thrive and milder degrees of hepatocellular dysfunction in older infants or toddlers with dysmorphic facial features.

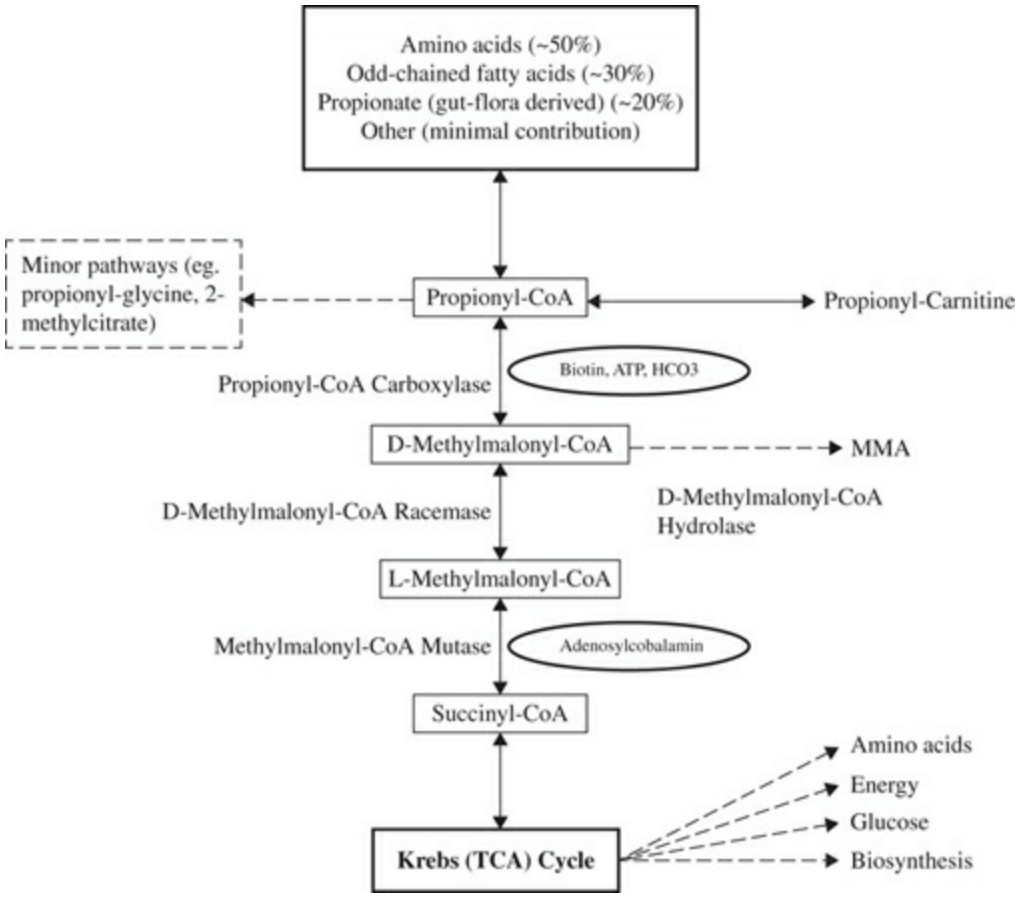


Figure 49-2 Major pathway of the conversion of propionyl-CoA, a major 3-carbon metabolite, into succinyl-CoA. The precursors are indicated with their approximated contribution to whole body propionate metabolism. The biotin-dependent enzyme, propionyl-CoA carboxylase converts propionyl-CoA into D-methylmalonyl-CoA that is then racemized into L-methylmalonyl-CoA and isomerized into succinyl-CoA, a Krebs cycle intermediate. The L-methylmalonyl-CoA mutase reaction requires adenosylcobalamin, an activated form of vitamin B_{12}. The formation of adenosylcobalamin requires intracellular metabolism of cobalamin. (Adapted from Chandler RJ, Venditti CP. Genetic and genomic systems to study methylmalonic academia. *Mol Genet Metab.* 2005;86(1–2):34–43.)

Hepatic Disease due to Glycosylation Defect

Disorders of glycosylation result from various defects in the formation or processing of the carbohydrate residues attached by N- or O-linkages of

peptides. These disorders can be considered “prelysosomal” storage disorders, as improperly glycosylated proteins initially accumulate in the endoplasmic reticulum. This may, in turn, lead to cellular dysfunction via the unfolded peptide response. The most common hepatic pathology is steatosis, but hydrops and severe hepatocellular dysfunction have been reported. Common clinical manifestations include failure to thrive, GI dysfunction or dysmotility, hypotonia, cognitive delays, and alterations in fat distribution. Mannose supplementation has been used as a treatment for mannose phosphate isomerase deficiency (CDG 1b). The newly described PGM1 enzyme deficiency is associated with fasting hypoglycemia, hepatic glycogenosis, abnormal circulating glycan-containing proteins and may respond to galactose therapy.[31]

Hepatic Disease due to Mitochondrial Dysfunction

Multiple reports have described infants with severe or fatal hepatic disease of early onset due to a mitochondrial depletion syndrome. In some cases, increased fat has been seen histologically; however, ultrastructural examination of other patients’ tissues has shown increased numbers of mitochondria with morphologic abnormalities, and even glycogen storage within the mitochondria. Focal biliary necrosis has been reported, but fibrosis is a later finding. Lactic acidosis, hypoglycemia, and synthetic defects have also been reported. In all cases, activities of the electron transport chain complexes with subunits encoded by mitochondrial DNA have been reduced. The amount of mitochondrial DNA has been reduced to less than 10% of normal. Mutations in the nuclear genes encoding deoxyguanosine kinase (DGOUK), thymidine phosphorylase (MNGIE), MPV17 (causing Navajo hepatoencephalopathy), polymerase–gamma (POLG), and succinyl-CoA synthase (SUCLG1) genes have been identified in some patients. The gastrointestinal phenotypes are variable and can feature hepatic failure, seen in patients with mutations in DGUOK, POLG, and MP17; hepatomegaly with encephalopathy, seen in the one patient with succinyl-CoA synthase deficiency and intestinal pseudo-obstruction, a prominent feature of thymidine phosphorylase (MNGIE) deficiency.

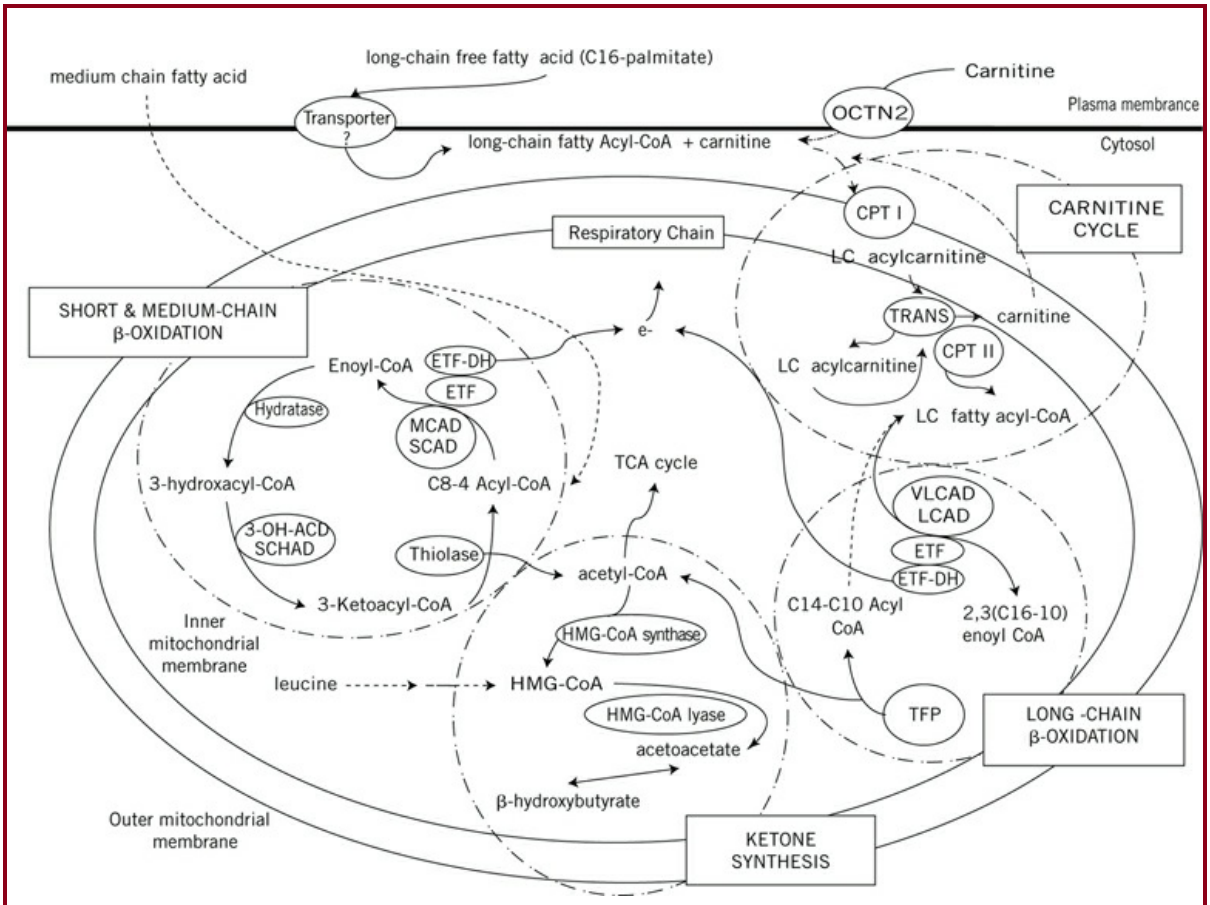


Figure 49-3 An overview of mitochondrial fatty acid oxidation. Carnitine enters the cell through the action of the organic cation/carnitine transporter (OCTN2). Palmitate, a typical 16-carbon long-chain fatty acid, is transported across the plasma membrane and can be activated to form a long-chain (LC) fatty acyl-CoA. It then enters into the carnitine cycle, where it is transesterified by carnitine palmitoyltransferase-I (CPT-I), translocated across the inner mitochondrial membrane by carnitine/acylcarnitine translocase (TRANS), and then reconverted into a long-chain fatty acyl CoA by carnitine palmitoyltransferase-II (CPT-II) to undergo β-oxidation. Very long chain acyl-CoA dehydrogenase (VLCAD/LCAD) leads to the production of (C16-10) 2,3 enoyl-CoA. Trifunctional protein (TFP) contains the activities of enoyl-CoA hydratase (hydratase), 3-OH-hydroxyacyl CoA dehydrogenase (3-OH-ACD), and β-ketothiolase (thiolase). Acetyl CoA, FADH, and NADH are produced. Medium- and short-chain fatty acids (C8-4) can enter the mitochondrial matrix independent of the carnitine cycle. Medium-chain acyl-CoA dehydrogenase (MCAD), short-chain

acyl-CoA dehydrogenase (SCAD), and short-chain hydroxy acyl-CoA dehydrogenase (SCHAD) are required. Acetyl-CoA can then enter the Krebs (TCA) cycle. Electrons are transported from FADH to the respiratory chain via the electron-transfer flavoprotein (ETF) and the electron-transfer flavoprotein dehydrogenase (ETF-DH). NADH enters the electron transport chain through complex I. Acetyl-CoA can be converted into hydroxymethylglutaryl-CoA (HMG-CoA) by β-hydroxy-β-methylglutaryl-CoA synthase (HMG-CoA synthase) and then the ketone body acetoacetate by the action of β-hydroxy-β-methylglutaryl-CoA lyase (HMG-CoA lyase). (Adapted from Venditti CP, Stanley CA. Disorders of mitochondrial fatty acid oxidation. In: Behrman RE, Kliegman RM, Jenson HB, eds. *Nelson Textbook of Pediatrics*, 17th edition. Philadelphia, PA: W.B. Saunders; 2003.)

Hepatic Disease due to Storage Disorders

Material either synthesized within the cell or accumulated through endocytic processes as part of the body's recycling of macromolecules is the sine qua non of the heterogeneous storage disorders. In the GSDs type I, III, or IV presenting in early infancy, there is accumulation of glycogen as well as fat. GSD type I should also be thought of as a steatosis disorder. In Niemann-Pick type A, sphingomyelin accumulates in hepatocytes, as well as in the reticuloendothelial elements, but in Niemann-Pick type C, the accumulated material includes both sphingomyelin and cholesterol. In the latter disorder, hepatocellular necrosis and a progression to fibrosis can also occur, blurring the distinctions made here about disorders primarily associated with hepatocytic dysfunction or storage with minimally altered hepatocytes. The accumulation and storage of glucocerebroside in Kupffer cells in Gaucher disease and of glycolipids and complex lipids in other disorders lead to liver enlargement but relatively little hepatocellular functional abnormality. In many cases, the histopathology and histochemical staining patterns can identify the disorder.

Hepatic Disease due to Porphyrias

The heme biosynthetic pathway begins in the mitochondria with a condensation reaction of glycine and succinyl-CoA and ends with the production of heme, which is then used for hemoglobin formation in the bone marrow and, of equal importance, for cytochrome formation. Defects

in any of the steps in this pathway represent a group of eight disorders that can feature cutaneous, psychiatric, neurologic, and hepatic symptoms. One disorder of this group, protoporphyria, can be associated with severe liver disease that can require liver transplantation.

Other Disorders With Primary Gastrointestinal Disturbances

Chronic diarrhea can be a manifestation of a number of inborn errors of metabolism, including lactase and sucrose–isomaltase deficiencies, glucose–galactose malabsorption, congenital chloride diarrhea, and congenital secretory diarrhea due to defective sodium/hydrogen exchange. Diarrhea, malabsorption, and protein-losing enteropathy can also be features of other disorders discussed in this chapter, such as cystic fibrosis, lysinergic protein intolerance, and phosphomannose isomerase deficiency. Selective vitamin malabsorption disorders, such as congenital folate malabsorption, the intestinal biotin transporter defect,[32] and Imerslund-Grasbeck disease[33] might also be included as metabolic disorders where the primary defect lies in the GI tract.

CONCLUSION

A wide variety of catabolic, anabolic, and transport defects affecting the GI tract can be recognized. The age of presentation ranges from mid-gestation to adulthood and encompasses a wide range of symptoms and signs, some of which are nonspecific. For some disorders, the pathophysiology has been delineated and molecular determinants characterized, but in the majority only the broader features are currently comprehended. The vast array of biochemical genetic diseases, these unfortunate experiments of nature, continues to provide great insight into the secrets of normal hepatic metabolism in man.

REFERENCES

1. Scriver CR, Beaudet AL, Sly WS, et al., eds. *The Metabolic and Molecular Bases of Inherited Disease*, 8th ed. New York: McGraw-Hill; 2001: 2165–2192.
2. Ibdah JA, Bennett MJ, Rinaldo P, et al. A fetal fatty-acid oxidation disorder as a cause of liver disease in pregnant women. *N Engl J Med*. 1999;340(22):1723–1731.
3. Innes AM, Seargeant LE, Balachandra K, et al. Hepatic carnitine palmitoyltransferase I deficiency presenting as maternal illness in pregnancy. *Pediatr Res*. 2000;47(1):43–45.

4. Matern D, Hart P, Murtha AP, et al. Acute fatty liver of pregnancy associated with short-chain acyl-coenzyme A dehydrogenase deficiency. *J Pediatr*. 2001;138(4):585–588.
5. von Kleist-Retzow JC, Cormier-Daire V, Viot G, et al. Antenatal manifestations of mitochondrial respiratory chain deficiency. *J Pediatr*. 2003;143(2):208–212.
6. Kerrigan JF, Aleck KA, Tarby TJ, et al. Fumaric aciduria: clinical and imaging features. *Ann Neurol*. 2000;47(5):583–588.
7. Chisholm CA, Vavelidis F, Lovell MA, et al. Prenatal diagnosis of multiple acyl-CoA dehydrogenase deficiency: association with elevated alpha-fetoprotein and cystic renal changes. *Prenat Diagn*. 2001;21(10):856–859.
8. Strom EH, Stromme P, Westvik J, Pedersen SJ. Renal cysts in the carbohydrate-deficient glycoprotein syndrome. *Pediatr Nephrol*. 1993;7(3):253–255.
9. Wanders RJ. Metabolic and molecular basis of peroxisomal disorders: a review. *Am J Med Genet A*. 2004;126(4):355–375.
10. Elpeleg ON, Hammerman C, Saada A, et al. Antenatal presentation of carnitine palmitoyltransferase II deficiency. *Am J Med Genet*. 2001;102(2):183–187.
11. Kelley RI, Herman GE. Inborn errors of sterol biosynthesis. *Annu Rev Genomics Hum Genet*. 2001;2:299–341.
12. de Koning TJ, Nikkels PG, Dorland L, et al. Congenital hepatic fibrosis in 3 siblings with phosphomannose isomerase deficiency. *Virchows Arch*. 2000;437(1):101–105.
13. Adam G, Brereton RJ, Agrawal M, Lake BD. Biliary atresia and meconium ileus associated with Niemann-Pick disease. *J Pediatr Gastroenterol Nutr*. 1988;7(1):128–131.
14. Matthews RP, Russo P, Berry GT, et al. Biliary atresia associated with a fatty acid oxidation defect. *J Pediatr Gastroenterol Nutr*. 2002;35(5):624–628.
15. Stone DL, Sidransky E. Hydrops fetalis: lysosomal storage disorders in extremis. *Adv Pediatr*. 1999;46:409–440.
16. Leonard JV, Walter JH, McKiernan PJ. The management of organic acidaemias: the role of transplantation. *J Inherit Metab Dis*. 2001;24(2):309–311.
17. Bull LN, van Eijk MJ, Pawlikowska L, et al. A gene encoding a P-type ATPase mutated in two forms of hereditary cholestasis. *Nat Genet*. 1998;18:219–224.
18. Klomp LW, Bull LN, Knisely AS, et al. A missense mutation in FIC1 is associated with Greenland familial cholestasis. *Hepatology*. 2000;32(6):1337–1341.
19. Schwarz M, Wright AC, Davis DL, et al. The bile acid synthetic gene 3-beta-hydroxy-delta-5-C27-steroid oxidoreductase is mutated in progressive intrahepatic cholestasis. *J Clin Invest*. 2000;106:1175–1184.
20. Strautnieks SS, Bull LN, Knisely AS, et al. A gene encoding a liver-specific ABC transporter is mutated in progressive familial intrahepatic cholestasis. *Nat Genet*. 1998;20(3):233–238.
21. de Vree JM, Jacquemin E, Sturm E, et al. Mutations in the MDR3 gene cause progressive familial intrahepatic cholestasis. *Proc Natl Acad Sci U S A*. 1998;95(1):282–287.
22. Chagnon P, Michaud J, Mitchell G, et al. A missense mutation (R565W) in cirhin (FLJ14728) in North American Indian childhood cirrhosis. *Am J Hum Genet*. 2002;71(6):1443–1449.
23. Bove KE, Heubi JE, Balistreri WF, Setchell KD. Bile acid synthetic defects and liver disease: a comprehensive review. *Pediatr Dev Pathol*. 2004;7(4):315–334.
24. Visapaa I, Fellman V, Vesa J, et al. GRACILE syndrome, a lethal metabolic disorder with iron overload, is caused by a point mutation in BCS1L. *Am J Hum*

Genet. 2002;71(4):863–876.
25. Verloes A, Lombet J, Lambert Y, et al. Tricho-hepato-enteric syndrome: further delineation of a distinct syndrome with neonatal hemochromatosis phenotype, intractable diarrhea, and hair anomalies. *Am J Med Genet*. 1997;68(4):391–395.
26. Papanikolaou G, Samuels ME, Ludwig EH, et al. Mutations in HFE2 cause iron overload in chromosome 1q-linked juvenile hemochromatosis. *Nat Genet*. 2004;36(1):77–82.
27. Roetto A, Papanikolaou G, Politou M, et al. Mutant antimicrobial peptide hepcidin is associated with severe juvenile hemochromatosis. *Nat Genet*. 2003;33(1):21–22.
28. Camaschella C, Roetto A, Cali A, et al. The gene TFR2 is mutated in a new type of haemochromatosis mapping to 7q22. *Nature Genet*. 2000;25:14–15.
29. Njajou OT, Vaessen N, Joosse M, et al. A mutation in SLC11A3 is associated with autosomal dominant hemochromatosis. *Nat Genet*. 2001;28(3):213–214.
30. Joshi M, Eagan J, Desai NK, et al. A compound heterozygous mutation in GPD1 causes hepatomegaly, steatohepatitis, and hypertriglyceridemia. *Eur J Hum Genet*. 2014; Oct;22(10):1229–32. doi: 10.1038/ejhg.2014.8.
31. Tegtmeyer LC, Rust S, van Scherpenzeel M, Ng BG, et al. Multiple phenotypes in phosphoglucomutase 1 deficiency. *N Engl J Med*. 2014; Feb 6;370(6):533–42. doi: 10.1056/NEJMoa1206605. PubMed PMID: 24499211.
32. Mardach R, Zempleni J, Wolf B, et al. Biotin dependency due to a defect in biotin transport. *J Clin Invest*. 2002;109(12):1617–1623.
33. Aminoff M, Carter JE, Chadwick RB, et al. Mutations in CUBN, encoding the intrinsic factor-vitamin B12 receptor, cubilin, cause hereditary megaloblastic anaemia 1. *Nat Genet*. 1999;21(3): 309–313.
34. Verhoeven NM, Huck JH, Roos B, et al. Transaldolase deficiency: liver cirrhosis associated with a new inborn error in the pentose phosphate pathway. *Am J Hum Genet*. 2001;68(5):1086–1092.

Inborn Errors of Fasting Adaptation—Glycogen Storage Disease

50

David A. Weinstein, MD, MMSc and Katalin M. Ross, RD, LDN

Glycogen storage disease (GSD) is caused by a defect in any of multiple enzymes involved in the synthesis or utilization of glycogen.[1,2] Glycogen is stored predominantly in the liver and muscles, and the symptoms of the disorder depend on the location of the defect. Hypoglycemia is the cardinal manifestation of the hepatic glycogenoses due to the inability to utilize glycogen as a source of glucose in a fasting state. Weakness, muscle cramps, and myopathy are the main features of the muscle forms of GSD. Since the disease processes are very different, this chapter is focused on the hepatic glycogenoses that are more dependent on nutritional therapies.

NORMAL PHYSIOLOGY OF GLUCOSE METABOLISM

Following consumption of a meal, carbohydrates absorbed into the blood stream provide fuel for the brain, muscle, red blood cells, and kidneys. The liver is freely permeable to glucose. After being taken up by the liver, glucose is phosphorylated by glucokinase to form glucose-6-phosphate (G6P). G6P is subsequently converted to glucose-1-phosphate, and

glycogen synthase creates chains of glucose molecules by catalyzing the formation of α-1,4-linkages. A branching enzyme forms α-1,6-linkages approximately every 10 glucose units along the chain. During periods of fasting, debranching enzyme hydrolyzes the α-1,6-linkages at branch points and leads to the formation of free glucose. In addition, a cascade of enzymatic reactions activates hepatic glycogen phosphorylase, the rate-limiting enzyme in glycogenolysis, which hydrolyzes the α-1,4-linkages from the outer branches of glycogen to form G6P. Glucose-6-phosphatase (G6Pase) catalyzes the terminal reaction of glycogenolysis and gluconeogenesis, the hydrolysis of G6P to glucose and inorganic phosphate in hepatocytes and renal epithelial cells, allowing glucose to be released into the systemic circulation.[3]

Glucose is stored predominantly in muscle, liver, and kidneys as glycogen for use during fasting. Although muscle stores at least twice as much glycogen as the liver, muscle lacks G6Pase. Thus, muscle is unable to release glucose for systemic use, and the muscle forms of GSD are not associated with hypoglycemia. There are five major types of hepatic GSD that affect glucose homeostasis. The location of each of the enzymatic defects is shown in Figure 50-1.

The clinical severity of the glycogen storage diseases varies widely depending on multiple factors including the degree of impairment of glycogenolysis, whether endogenous glucose can be produced by gluconeogenesis, and the availability of alternative fuels (lactate, ketones) during periods of hypoglycemia. Patients with severely limited endogenous glucose production (GSD I) present with seizures, failure to thrive, or developmental delay. In contrast, patients with normal gluconeogenesis and abundant alternative fuels have a milder phenotype (GSD 0 or GSD IX), and they can be asymptomatic until an illness impairs normal dietary carbohydrate intake.

HEPATIC GLYCOGEN SYNTHASE DEFICIENCY (GLYCOGEN STORAGE DISEASE TYPE 0)

Type 0 glycogen storage disease (GSD 0) is caused by a deficiency in hepatic glycogen synthase, which leads to a marked decrease in liver glycogen content.[4] GSD 0 is due to mutations in the GYS2 gene located on chromosome 12p12.2 and is inherited in an autosomal recessive manner. To date, approximately 20 mutations have been reported with no identified dominant mutation. Although no high-risk population has been identified,

a common mutation, R246X, has been found in people of Italian ancestry.[5]

Pathophysiology

After consumption of carbohydrate, the inability to store glucose as glycogen in the liver results in postprandial hyperglycemia. Glucose taken up by the liver gets shunted down the glycolytic pathway leading to postprandial hyperlactatemia. During fasting, fatty acid oxidation is robust leading to formation of high concentrations of ketones that can be used as an alternative fuel during period of hypoglycemia. Amino acids and lactate can also undergo gluconeogenesis, and these alternative fuels can serve as substrate for endogenous glucose production.

Presentation

The typical presentation of GSD 0 is recurrent ketotic hypoglycemia associated with intercurrent illness.[6] The physical examination is usually normal, and the liver is not enlarged unlike the other hepatic glycogenoses. During infancy, fasting ketotic hypoglycemia develops on cessation of night-time feeding. Weaning from overnight feeds proves to be difficult with hypoglycemia and symptoms of agitation and fussiness are often misconstrued as colic. Significant ketonemia develops with fasting and serves as an alternative energy source for the brain.[7] The protection from the prominent ketonemia may explain why developmental delay has only been described in 22% of children with GSD 0.[8] Other manifestations are often subtle including short stature, failure to thrive, hyperlipidemia, or elevation of hepatic transaminase concentrations.[5]

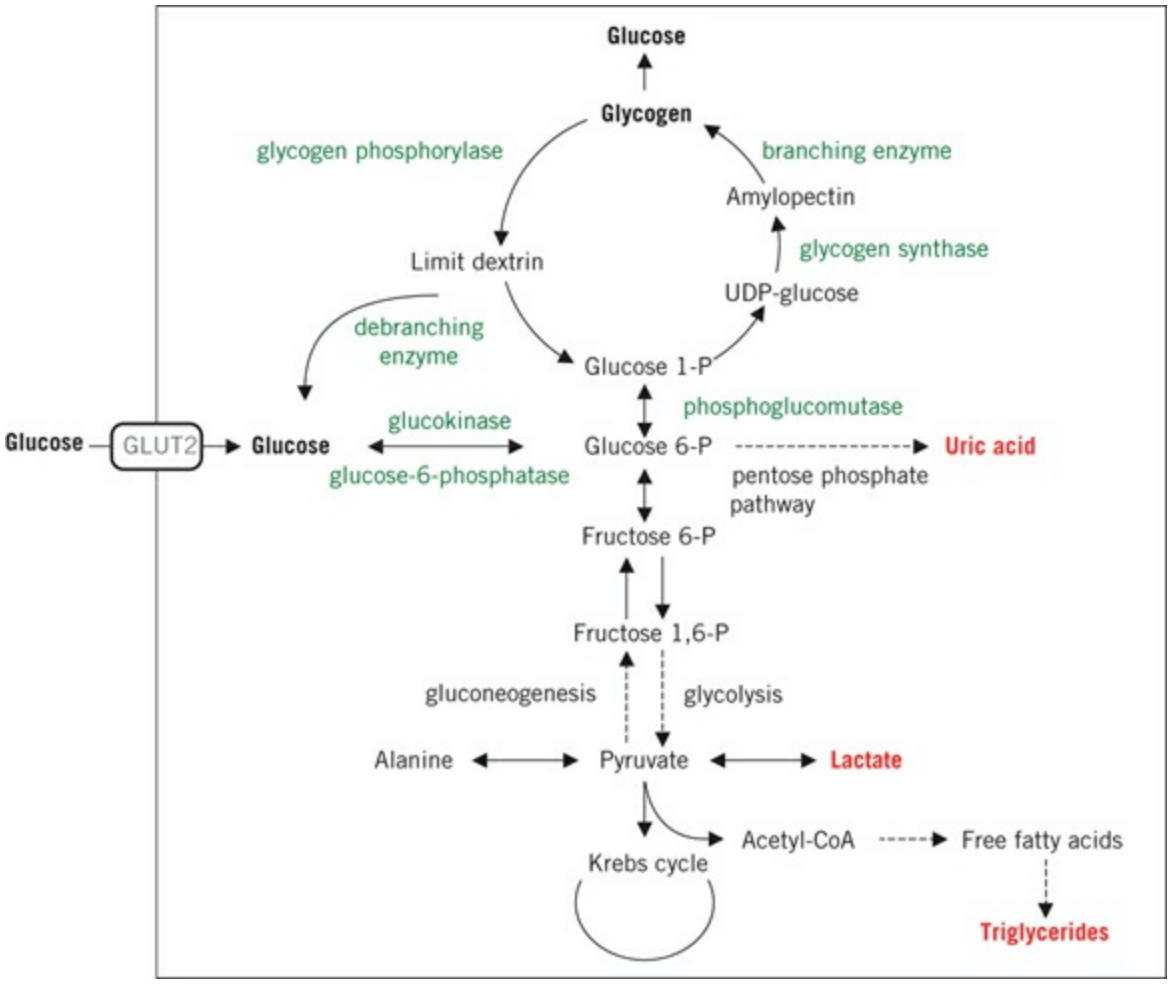


Figure 50-1 Schematic representation of the glycogen storage diseases.

Most children with GSD 0 are identified incidentally during a gastrointestinal illness or period of poor nutrition when hypoglycemia is discovered in a lethargic child. GSD 0 can also be misdiagnosed as diabetes because of postprandial hyperglycemia and fasting ketonuria; however, this presentation would lack symptoms of polyuria and polydipsia.[8]

Diagnosis

The diagnosis of GSD 0 is usually made when fasting ketotic hypoglycemia is found to alternate with postprandial hyperglycemia and hyperlactatemia.[8] Patients usually present after an overnight fast for baseline assessment, and a glucose or galactose tolerance test can be used to assess for the unique biochemical pattern. The gold standard for making

the diagnosis of GSD 0 is by mutation analysis using DNA extracted from blood.[4] Testing in commercial laboratories is now available. Although liver biopsies are no longer recommended to make the diagnosis, a few cases with no identifiable mutation in the GYS2 gene have been diagnosed by liver biopsy with demonstration of decreased hepatic glycogen content or abnormal enzyme activity.

Nutritional Management

The goal of treatment is to prevent hypoglycemia and minimize the associated lactic acidosis. Amino acids can serve as the precursors for gluconeogenesis, and thus a high protein diet is preferred (2–3 g/kg). Since hyperlactatemia and hyperlipidemia occur after ingestion of carbohydrates, a low carbohydrate diet is preferred, and low glycemic index, complex carbohydrates are preferred over simple sugars.

Infants with GSD 0 are usually asymptomatic due to frequent feeds. As children grow; however, it may be difficult to wean the infant from overnight frequent feeds without additional support. In children, uncooked cornstarch (UCS) with 1–1.5 g/kg administered at bedtime is used to prevent hypoglycemia in conjunction with a high protein, low carbohydrate diet as previously discussed. In addition to the bedtime cornstarch dose, some children require frequent snacks every 2–4 hours during the day to prevent hypoglycemia during the daytime.[5] Daytime cornstarch is occasionally used (0.5 g/kg) for children and adults who have higher energy needs secondary to an active lifestyle.

In addition to assessing the patient's intake to monitor compliance with the above guidelines, it is critical to ensure that patients receive adequate calories and that no vitamin or mineral deficiencies are present. Unlike other forms of GSD that have a more restricted diet, vitamin and mineral deficiencies are rare in GSD 0. Osteoporosis is common in GSD 0 due to abnormal bone mineralization in the setting of acidosis, and it is important to ensure that appropriate calcium and vitamin D intake is occurring.

Monitoring of blood glucose and blood ketone concentrations is recommended on a routine basis. Cornstarch therapy overnight is titrated to keep blood ketone concentrations under 0.5 mmol/L. During illness and periods of suboptimal food intake, blood ketone monitoring is the first clue that a child or adult with GSD 0 is becoming catabolic. Supplementation with glucose and extra cornstarch is recommended if ketones are increasing as high concentrations of ketones may lead to vomiting and worsening hypoglycemia.

Natural History and Prognosis

Although overnight hypoglycemia is common in infants and young children, fasting is usually better tolerated with age. Most children over 7 years of age tolerate a typical overnight fast without development of hypoglycemia, and fasting up to 18 hours has been reported in teenagers.[9] This improvement, however, is not universal, and some children continue to develop hypoglycemia with overnight fasts. Although severe hypoglycemia is unusual in older children and adults, ketosis remains common in untreated individuals, and most patients report feeling better with continued glucose supplementation overnight. In addition, fatigue with exertion is common in untreated individuals, and glucose supplementation improves stamina during sports and other active periods.

There is limited experience with adults confirmed to have GSD 0, but there is no reason to believe the prognosis is not excellent for these patients. Short stature and osteopenia are common in untreated children, but improve with prevention of hypoglycemia, lactic acidosis, and ketosis. The long-term complications commonly seen in the other forms of GSD, such as hepatic adenomas, cirrhosis, kidney dysfunction, and muscular abnormalities, have not been reported in teenagers and adults with GSD 0. There are no formal guidelines for screening in this condition.

GLUCOSE-6-PHOSPHATASE DEFICIENCY (TYPE I GSD; VON GIERKE DISEASE)

G6Pase converts G6P to glucose in the final step of both glycogenolysis and gluconeogenesis.[10] The enzyme is present in the liver, kidneys, and intestines, but it is not present in muscle. At the cellular level, G6Pase is a complex enzyme system located in the endoplasmic reticulum (ER) membrane and consists of a catalytic unit that faces into the ER lumen, a calcium-binding protein, and three transport proteins that facilitate transport of G6P (T1), phosphate (T2), and glucose (T3).[11] Abnormalities of any of these components can cause abnormal enzyme function and the clinical phenotype of GSD I. There are three major subtypes of GSD I. More than 85% of patients have mutations that impair the catalytic function of the enzyme (GSD type Ia). Defects in the T1 glucose-6-phosphate transporter account for most other cases (GSD type Ib). Abnormality of the phosphate transporter (GSD type Ic) is rare.

GLYCOGEN STORAGE DISEASE TYPE IA (VON GIERKE DISEASE)

GSD Ia is caused by mutations in the G6PC gene on chromosome 17q21, which encodes G6Pase.[12] The disease is inherited in an autosomal recessive manner, and the incidence is estimated to be 1 in 100,000 live births. GSD type Ia is particularly common in people of Ashkenazi Jewish heritage, and 1 in 68 individuals in this population have been reported to be carriers of the disease.[13] Although GSD type Ia occurs in all ethnic groups, common mutations have also been reported in people of Chinese, Japanese, and Mexican heritage.

Pathophysiology

The inability to convert G6P to glucose results in hypoglycemia whenever exogenous glucose administration is not occurring. Increased concentrations of G6P develop in hepatocytes and shunting into alternative pathways results in the following:

- Lactic acidosis (as a result of increased glycolysis)[14]
- Hyperuricemia (as a consequence of shunting into the pentose phosphate shunt leading to increased synthesis of uric acid; decreased renal excretion also occurs as lactate and uric acid share the same transporter in the kidney)
- Hypertriglyceridemia (as a result of increased synthesis of acetyl CoA)[15]

With prevention of hypoglycemia and glycogenolysis, all metabolic abnormalities abate.

Presentation

Most children do not present with symptoms until 3–6 months of age although hypoglycemia may occur soon after birth. During infancy, as the period between feeds increases or as an acute illness disrupts normal feeding, symptomatic hypoglycemia develops. The symptoms of agitation and fussiness associated with hypoglycemia may go unrecognized until the child presents with failure to thrive, tachypnea (from lactic acidosis), seizures, lethargy, and developmental delay. If hypoglycemia in infancy is missed, GSD I occasionally is diagnosed when hepatomegaly and a protuberant abdomen are discovered during a routine physical exam.

Diagnosis

The simplest and most rapid means of determining the defect in a child with hypoglycemia suspected to have GSD is to obtain serial blood measurements of glucose, lactate, and ketones during a fasting study. Hypoglycemia and lactic acidosis will manifest in a child with GSD type Ia after a brief fast of 3–4 hours. During the study, children should have secure intravenous access to allow for rapid correction as hypoglycemia can develop precipitously. Glucagon must be administered with extreme caution as it will not rescue the child from hypoglycemia, and it can exacerbate hyperlactatemia. Typically, glucose concentrations are monitored every 15–30 minutes after the initial two hours. After the plasma glucose has fallen below 50 mg/dL, a sample for blood glucose and lactate determination should be obtained. As many of the portable meters are inaccurate in the setting of lactic acidosis, close clinical observation is needed during the test. As soon as the test is completed, intravenous glucose is administered to bring down lactate concentrations and maintain euglycemia.

Mutation analysis on DNA extracted from leukocytes is the recommended test for confirming the diagnosis in a child suspected of having GSD Ia. A total of 88 different mutations have been reported. Since many of the genetic tests for GSD type Ia are designed to pick up only the common mutations, complete gene sequencing is recommended for anyone not in a high-risk population.[16] Mutation analysis allows for noninvasive diagnosis, and the technique can be utilized for preimplantation screening and prenatal diagnosis. Liver biopsy is no longer recommended for diagnosing a patient with suspected GSD I, and it should be reserved for those patients in whom GSD Ia is suspected but the mutation analysis is nondiagnostic.

Nutritional Management of the Infant with Type I

Treatment consists of providing a continuous dietary source of glucose to prevent blood glucose from falling below the threshold for glucose counter-regulation, approximately 70 mg/dL (4 mmol/L).[17] A continuous source of glucose can be provided by nocturnal intragastric infusion (via nasogastric tube or gastrostomy) or by using uncooked (raw) cornstarch. An estimate of the minimum amount of glucose required can be obtained by using the following formula to calculate the basal glucose production rate:

$$y = 0.0014x^3 - 0.214x^2 + 10.411x - 9.084,$$

where y = milligrams of glucose per minute and x = body weight in kilograms.[18]

To avoid excessive caloric intake, the optimum feeding schedule and amounts of cornstarch is determined by metabolic monitoring to ensure that the biochemical goals of therapy are achieved. Cornstarch doses should be weighed on a gram scale to avoid over treatment that commonly occurs when doses are estimated.

Since nonglucose sugars cannot be converted back to glucose, patients with GSD, type I must restrict intake of fructose, sucrose, and galactose. Mothers of infants with type I GSD are often encouraged not to breast-feed as the main source of carbohydrate in breast milk is lactose. The formulas most commonly used to treat GSD type I are outlined in Table 50-1.

TABLE 50-1 Commonly Used Formulas in GSD Type I

Formula	Carbohydrate Source	Grams Carbohydrate per 100 mL	Calories/Ounce	General Commen
Enfamil Prosobee Lipil Mead Johnson	Corn syrup solids	7.2	20	Most commonly used formu
Nutramigen Lipil Mead Johnson	Corn syrup solids and modified cornstarch	7	20	Used with protein allergies an malabsorpti issues
Pregestimil Mead Johnson	Corn syrup solids, dextrose, modified cornstarch	6.9	20	High conter of MCTs as fat (55% of calories)
Neocate Nutricia	Corn syrup solids	7.9	20	Commonly used with food allergi
Elecare Abbott	Corn syrup solids	7.2	20	Commonly used with food allergi malabsorpti
Tolerex Novartis	Maltodextrin, modified corn	23	30	Commonly used in sick

	starch			day manageme
Vivonex RTF Novartis	Maltodextrin, modified corn starch	17.5	30	Can also be used in sick day manageme
Osmolite Abbott	Maltodextrin	14.4	32	Isotonic Kosher
Enfagrow Soy Mead Johnson	Corn syrup	7.9	20	Iron-fortified formula for toddlers

MCT = medium-chain triglyceride.

Source: Information obtained from "Pediatric Nutrition Reference Guide" by Texas Children's Hospital, 9th edition, 2010.

The formula chosen must not contain the nonutilizable sugars. Prosobee is the most commonly used formula as it is a nonspecialized, standard infant formula that is easy to obtain and less expensive than the other specialized formulas. The specialized formulas may be indicated, however, if there is a medical necessity, such as malabsorption or intolerance. Tolerex is commonly used for sick day management as a smaller volume is required to meet the carbohydrate needs of the child. It is commonly well tolerated in infants even though it is an adult formula, but it should not be used as the primary source of nutrition due to the relatively low concentration of protein.

Hypoglycemia is prevented in the infant with GSD I through frequent formula feeds provided every 1.5–3 hours. As a result of the need to frequently eat, feeding aversions commonly develop. When solids are introduced, complex carbohydrates such as pastas, legumes, vegetables, and oatmeal are encouraged. UCS is typically not tolerated in infants younger than 6 months due to lack of pancreatic and intestinal amylase that is required to digest cornstarch. The ability to digest UCS therapy, however, differs for each infant, and many infants cannot absorb cornstarch adequately until around 12 months of age, and many children with GSD Ib will not tolerate cornstarch well until 2 years of age.[17] Pancreatic enzyme replacement therapy has been attempted in some infants who develop severe diarrhea with cornstarch, but this intervention is not recommended. Variable response to this intervention has occurred, and some patients have worsened due to possible overdigestion of cornstarch. Use of exogenous enzymes may also delay endogenous enzyme production.

Although it is commonly theorized that UCS therapy can lead to obesity in GSD I patients, it must be noted that this is a misconception. In comparing an infant receiving overnight cornstarch supplementation with overnight continuous formula feeds, the infant receiving the cornstarch actually receives fewer calories and a more optimal feeding schedule is promoted. For example, a typical 12-month-old patient with GSD I requires approximately 4 g/h of glucose. To meet the glucose needs, 60 mL/h of Prosobee is required providing ~480 kcal over the 12-hour period. In contrast, when treated with 15 g cornstarch in 2 ounces Prosobee every four hours, only 284 cal are administered during the same 12-hour period. Not only is there the 41% reduction in calories, but the cornstarch therapy is associated with lower insulin concentrations and more stability in the morning.

In addition to saving calories, cornstarch therapy is associated with fewer neurologic complications when compared with continuous feeds. If a continuous feed is interrupted due to a mechanical problem or leakage of formula, severe hypoglycemia rapidly develops due to the high insulin state. In contrast, the low insulin state associated with cornstarch administration minimizes plasma glucose fluctuations and, blood lactate concentrations increase sufficiently to provide the brain with an alternative fuel. This decreases the risk of hypoglycemia-induced seizures.

Nutritional Management of Children and Adolescents With Type I

Children and adolescents with GSD type I are usually treated with UCS therapy given in boluses around the clock. If regular UCS is used, feeds should be administered every 4–5 hours around the clock. With aging, many children and adults clinically are stable with feeds administered every six hours, but it comes at the expense of elevated triglycerides and lactate concentrations. Spacing to six hours even at night should not occur unless all markers of metabolic control are optimal.[19]

When dosing cornstarch, it is critical to note that glucose requirements do not continue to rise with aging and weight.[18] Basal glucose needs are primarily dependent on brain glucose utilization, and glucose requirements stabilize after 7 years of age. Although additional support is needed during puberty, older children and adults usually have similar glucose requirements (10–12.5 g/h). Weight-based dosing will lead to overtreatment.

Recently, an extended release cornstarch preparation (Glycosade) was

introduced. The new therapy typically extends the duration of fasting by 2–3 hours overnight, but some adults can maintain optimal metabolic control for up to 10 hours.[20] The extended release cornstarch is only recommended for night use at this time as it may be too slow to adequately meet the metabolic needs during the day. Standard dosing is 120 g of Glycosade at bedtime for a prepubertal child, 135–150 g for a pubertal adolescent, and 135 g for an adult. Initiation of the therapy should be performed with very close monitoring as the response can be variable, and most children have initiation of the therapy in the hospital. Although the therapy has been used in Europe since 2009, long-term studies on Glycosade are lacking, and close follow-up is strongly recommended for anyone on the therapy.

Dietary fat should be restricted to about 20% of the total energy intake and cholesterol is restricted to <300 mg/d. Carbohydrates, mostly in the form of starches, typically provide about 60%–65% of the daily calories, of which cornstarch accounts for 30%–45%. Dietary galactose and fructose continue to be restricted along with restriction of sucrose.

Due to dependence on starch and the restrictions on fruit and dairy products, patients often have dietary intakes that are deficient in vitamins and essential minerals. Calcium and vitamin D supplementation are extremely important in combination with a daily multivitamin supplementation. Adult multivitamins can be used in children as long as the patient is monitored to avoid vitamin A toxicity. It is also important for iron concentrations to be monitored as iron absorption is typically abnormal in GSD I due to effects of cornstarch and abnormal hepcidin regulation.

Natural History and Prognosis

Prior to the 1970s, many children with GSD I died in infancy or early childhood. Recurrent severe hypoglycemia can cause brain damage, but the prognosis has improved dramatically with early diagnosis and long-term maintenance of optimal metabolic control.[19] Although most people with GSD Ia do well into adulthood, both acute and long-term complications remain common (Figure 50-2).

Hyperlactatemia is the most common acute complication of GSD Ia. Lactate is generated as a consequence of counter-regulation associated with hypoglycemia or secretion of stress hormones (cortisol, epinephrine, glucagon, growth hormone). Lactate can also result from increased counter-regulation when glucocorticoids, stimulants, or sympathomimetics are utilized. Elevated lactate concentrations are an under-recognized cause

of gastroesophageal reflux in young children. During illness, lactic acidosis alters gastric motility and causes vomiting. Lactate elevation acutely also presents as tachypnea, and it frequently is misdiagnosed as bronchiolitis in many infants and young children. Anesthesia and the stress of surgery cause increased secretion of counter-regulatory hormones, and severe lactic acidosis can develop despite maintenance of normal blood glucose concentrations.

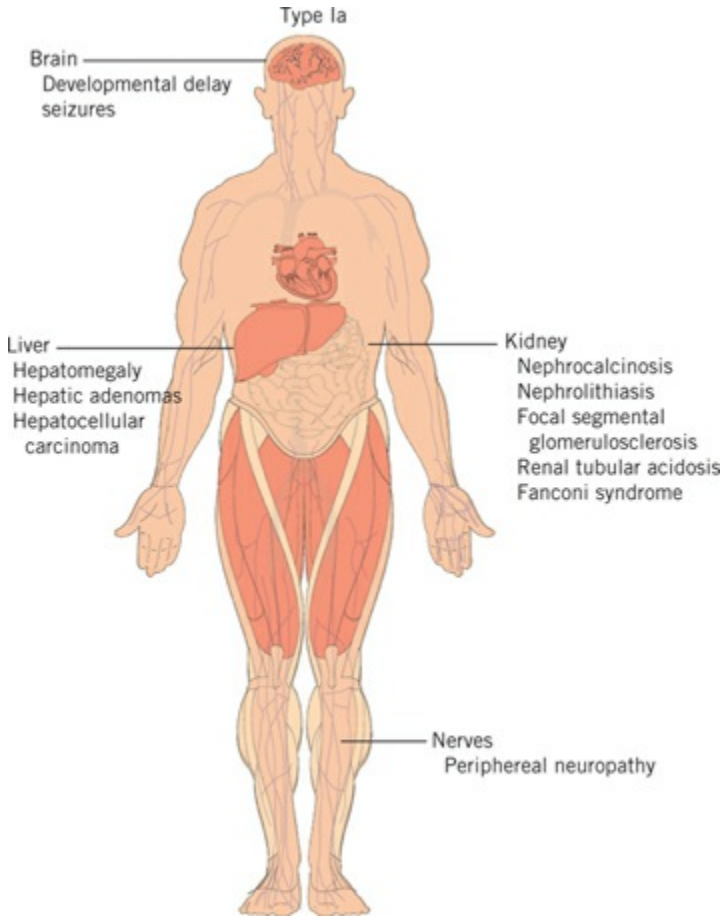


Figure 50-2 Complications associated with GSD type Ia.

There is increasing evidence that optimal metabolic control can prevent most long-term complications.[21] A strong link between suboptimal control (triglycerides over 500 mg/dL or lactates over 3 mmol/L) and complications including hepatic adenomas, renal disease, and osteoporosis has been demonstrated. Hepatic adenomas previously were detected in approximately 70% of patients by the end of puberty, but the prevalence of adenoma formation has been falling with improved metabolic control.[21,22]

The mean age at diagnosis is approximately 15 years, but adenomas occasionally develop during childhood.[23] There is no gender predisposition, and no difference has been found in the rate of adenoma formation in patients treated with cornstarch compared with those treated with continuous overnight intragastric drip feeding. Most adenomas remain stable over time, but they may be associated with hemorrhage or degenerate into hepatocellular carcinoma.[24] High alkaline phosphatase concentrations and elevation in the erythrocyte sedimentation rate (ESR) may be seen in patients with adenomas. Ultrasonography is the preferred screening method. When malignant transformation is a concern, magnetic resonance imaging using ferumoxide contrast provides greater definition and may be the best at differentiating benign adenomas from hepatocellular carcinoma.[25] Imaging is recommended at least annually after 5 years of age, and every six months if adenomas appear to be enlarging. Both α-fetoprotein and carcinoembryonic antigen have proven unreliable markers for determining malignant transformation.[24] As there are no good markers for determining development of hepatocellular carcinoma, resection of any lesion over 5 cm that is growing is recommended.

Treated children usually show no significant impairment of renal function except glomerular hyperfiltration. Some patients have a distal renal tubular acidification defect associated with hypercalciuria.[26] Urinary citrate excretion normally increases with age, whereas in GSD Ia, there is an inverse relationship between age and citrate excretion.[27] The combination of low citrate excretion and hypercalciuria appears to be important in the pathogenesis of nephrocalcinosis and nephrolithiasis. Citrate supplementation may prevent or ameliorate nephrocalcinosis and the development of urinary calculi. Increased albuminuria may be observed in adolescents. More severe renal injury with proteinuria, hypertension, and decreased creatinine clearance due to focal segmental glomerulosclerosis and interstitial fibrosis, which ultimately progresses to renal failure, may be seen in young adults.[28] Patients with persistently elevated concentrations of blood lactate, lipids, and uric acid appear to be at increased risk of nephropathy.[29] Normalization of metabolic parameters decreases proteinuria, and optimal therapy from an early age may delay or prevent renal disease.[19,29]

Anemia is common in patients with GSD Ia and is associated with suboptimal metabolic control.[30] Whereas anemia is usually mild, an unusual, unremitting iron resistant anemia occurs in a subset of patients.[31]

The anemia can be profound with hemoglobin concentrations as low as 4 g/dL and is characterized by microcytosis (mean corpuscular volume 50–70 fL), widening of the red cell distribution width (RDW), and iron studies consistent with iron deficiency. Neither erythropoietin nor treatment with oral and intravenous iron significantly improves the anemia. This form of iron resistant anemia has been associated with large hepatic adenomas (>7 cm in diameter) which inappropriately express hepcidin, a peptide that inhibits intestinal absorption of iron and macrophage recycling of iron. Resection of the hepatic adenomas has been associated with rapid correction of the anemia.[31]

With patients surviving into adulthood, osteoporosis is likely to be a common cause of morbidity.[32] Osteoporosis develops without abnormalities in calcium, phosphate, parathyroid hormone, or vitamin D metabolism.[33] Poor metabolic control has been associated with decreased bone mineral content. The etiology is likely multifactorial, including systemic acidosis, elevated cortisol concentrations, delayed pubertal development, inadequate dietary calcium, and lack of physical activity.[34]

Obesity is another common complication of GSD due to the need for constant carbohydrate intake. The caloric intake of patients should be monitored closely by a registered dietician. Patients benefit from carbohydrate counting as overconsumption of carbohydrate is common. Other complications, seen rarely in type Ia GSD, include pancreatitis, polycystic ovaries, and a bleeding diathesis.[35] Pancreatitis is related to hyperlipidemia, and treatment with Gemfibrozil or Niacin is recommended when hypertriglyceridemia persists despite attempts to improve metabolic control. Pancreatitis has been associated with use of growth hormone and glucocorticoids due to stimulation of glycogenolysis and worsening hypertriglyceridemia. Polycystic ovaries are common in poorly controlled GSD, but it does not appear to be inherent to the disease.[36] Fertility appears to be unaffected and more than 100 children have been born to mothers with GSD Ia. Platelet dysfunction associated with hyperlipidemia and acidosis is also a consequence of poorly controlled GSD. Bleeding is usually not a problem in patients in good metabolic control.

GLYCOGEN STORAGE DISEASE TYPE 1B

Type Ib GSD is caused by defects in the G6P transporter T1 that is responsible for transport of G6P into the lumen of the ER. The transporter is encoded by G6PT1 gene on chromosome 11q23 and is inherited in an

autosomal recessive manner.[37] The incidence is estimated to be 1 in 1,000,000 live births although the incidence of GSD Ib appears higher in Italy.

Pathophysiology

The inability of G6P to interact with the G6Pase enzyme prevents endogenous glucose production. Similar to GSD Ia, hepatomegaly and hypoglycemia develop shortly after birth. In contrast, however, patients with GSD Ib also manifest neutropenia and inflammatory bowel disease (IBD).[38] Immune dysfunction develops as a consequence of both disturbed myeloid maturation and functional defects of circulating neutrophils and monocytes. IBD is also attributed to neutropenia and impaired neutrophil function but persists despite granulocyte colony stimulating factor (G-CSF) therapy.[39]

Presentation

Symptomatic hypoglycemia may occur soon after birth, but most children present around 3–6 months of age. Neutropenia is occasionally the first abnormality noted during an evaluation for a serious bacterial infection usually from *Staphylococcus aureus*, *Streptococcus pneumoniae*, and *Escherichia coli*. IBD typically develops in the second decade, but it can present within the first few years of life.

Diagnosis

Full gene sequencing is commercially available. To date, approximately 70 mutations in the G6PT gene have been described.[40] Diagnosis by liver biopsy is not recommended; if performed, differentiation between the types requires an analysis of G6Pase activity in intact and fully disrupted microsomes as enzyme activity normalizes in type Ib when freezing disrupts the integrity of the ER. Typically, the diagnosis is made clinically when a patient with biochemical features of GSD I develops neutropenia and/or IBD.

Nutritional Management

The nutritional management of GSD Ib is the same as GSD Ia. Although GSD Ib is easier to manage metabolically, restriction of nonutilizable sugars is also critical in this condition.

Natural History and Prognosis

Long-term complications appear to be less frequent than in GSD Ia (Figure 50-3). IBD is the major cause of morbidity, and a Crohn's-like IBD almost universally develops by the end of puberty.[41] Periodic screening of inflammatory markers is recommended, and serologic studies may be of benefit if there is laboratory evidence of systemic inflammation.[42] A colonoscopy often is performed when IBD is suspected based on clinical features and laboratory studies, but a normal study does not rule out the condition. The IBD in GSD Ib frequently is isolated to the small intestine; consequently, a capsule endoscopy may reveal disease in patients in whom a colonoscopy reveals no evidence of bowel inflammation. Although the IBD responds to therapy with G-CSF, this comorbidity continues to occur even when neutropenia is treated.[43] Salicylate therapy (i.e., Pentasa) is the first line treatment of GSD enterocolitis. Entocort helps when terminal ileum disease is present, and Humira has been rarely needed when severe disease is present.[44] Patients with GSD Ib may benefit from probiotic therapy, but formal studies of this intervention are still lacking. Probiotics appear to prevent overgrowth of pathologic bacterial species, and treatment may decrease the frequency of IBD exacerbations and *Clostridium difficile* infections.

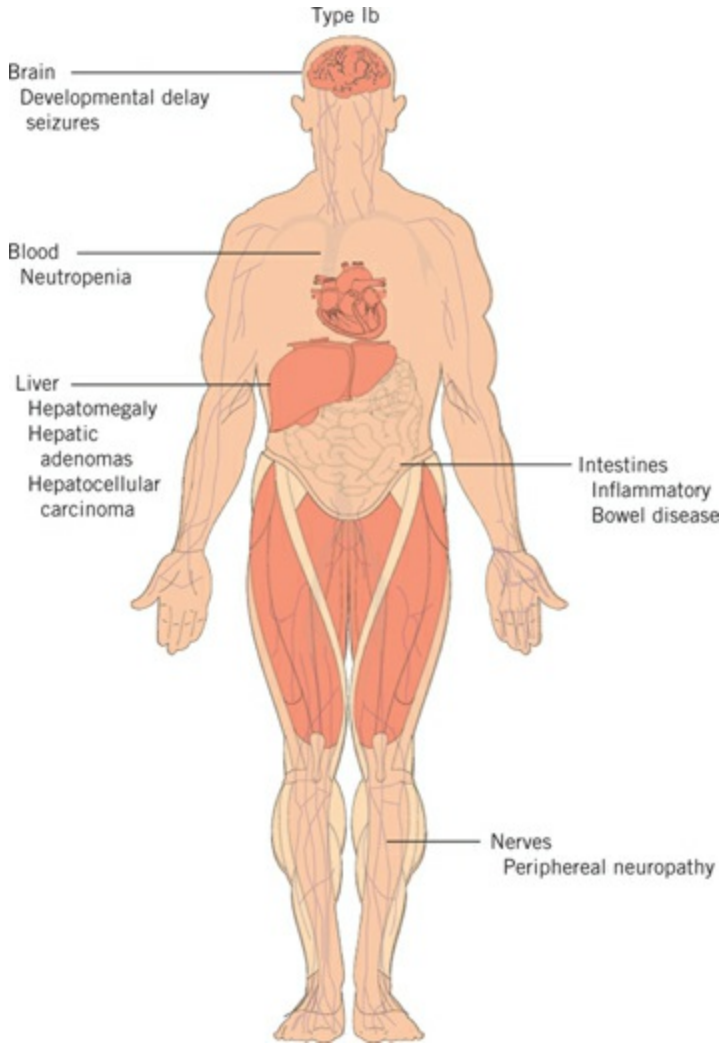


Figure 50-3 Complications associated with GSD type Ib.

Neutropenia occurs almost universally in GSD Ib, but the onset of this complication is variable. Low neutrophils are documented before the age of one year in two-thirds of patients, but neutropenia is first noted between the ages of six and nine years in 18% of patients.[38] Most patients had intermittent neutropenia without any clear cyclical course. Children with GSD Ib are prone to oral complications, including recurrent mucosal ulceration, gingivitis, and rapidly progressive periodontal disease. Therapy with recombinant human G-CSF improves infection-related morbidity by increasing numbers of circulating neutrophils and improving in vitro neutrophil function.[39] There is increasing evidence that overtreatment with G-CSF can be significant cause of morbidity, and recommended dosing in GSD Ib is lower than in other conditions. The recommended starting dose is 2.5 mcg/kg/d, and the lowest dose should be used that prevents serious

infections. Most children respond well to doses of 1–2 mcg/kg/d. Higher doses of G-CSF have been associated with hypersplenism, splenic ruptures, and esophageal varicies. Of concern is recent development of leukemia in several patients. Vitamin E supplementation has recently been reported to improve immune function, but studies on this intervention are limited.[45]

DEBRANCHING ENZYME DEFICIENCY (TYPE III GSD; CORI DISEASE; FORBES DISEASE)

Type III GSD is caused by a deficiency of the glycogen debranching enzyme. The incidence of GSD III is estimated to be 1 in 100,000 live births. Populations with a high incidence of GSD III include people from the Faroe Islands, people from the Indian subcontinent, and Jews of North African descent.[46,47] GSD III is inherited in an autosomal recessive manner with the gene for the debranching enzyme located on chromosome 1p21.[48] There are four distinct isoforms with differentially expressed clinical phenotypes associated with specific mutations.[49] Type IIIa affects both liver and muscle and accounts for 80%–85% of patients with GSD III in the United States; type IIIb affects only the liver and accounts for approximately 15% of patients.[2]

Pathophysiology

Release of glucose from glycogen requires sequential action of both glycogen phosphorylase and glycogen debranching enzyme. After glycogen phosphorylase has acted on the outer branches of glycogen leaving four glucose residues, the debranching enzyme utilizes two distinct catalytic activities to remove them. The first is a transferase activity that transfers three of the four glucosyl residues distal to the glycogen branch point to the outer end of an adjacent chain. Secondly, the amylo-1,6-glucosidase activity of the same enzyme also hydrolyses the branch point glucose residue. Without the function of the debranching enzyme, the breakdown of glycogen is interrupted with an abnormal form (limit dextrin) left to accumulate in affected tissues.[3]

Presentation

Type III GSD exhibits wide clinical and genetic variability. In infancy and early childhood, fasting hypoglycemia and hepatomegaly may be indistinguishable from GSD I; however, unlike GSD I, blood levels of

lactate and uric acid are normal. Biochemical abnormalities include elevated aspartate transaminase (AST), alanine transaminase (ALT), creatine kinase concentrations, and hyperlipidemia.[50] Serum transaminase concentrations are typically considerably more elevated than in GSD I, and often the AST and ALT concentrations are over 1000 U/L at presentation. Creatine kinase concentrations may be normal at presentation, and normal concentrations do not exclude muscle involvement.

Diagnosis

Mutation analysis from full gene sequencing is commercially available for the diagnosis of both type IIIa and IIIb GSD. Measurements of enzyme activity from biopsy of liver and muscle as well as demonstration of abnormal glycogen (limit dextrin) were the standard in diagnosis prior to mutation analysis. Skin fibroblast or lymphocyte enzyme activity can be used to screen for GSD III, but these studies may not be definitive and cannot be used for subtyping. A fasting study demonstrates ketotic hypoglycemia without the hyperlactatemia characteristic of GSD I. Glucagon fails to elicit a glycemia response after a fast, but does elicit a glycemic response when given two hours after a carbohydrate-rich meal.

Nutritional Management

In this type of GSD, gluconeogenesis can occur and protein can therefore be utilized as a substrate for endogenous glucose production. In infancy, hypoglycemia can be problematic, and patients are treated with a high protein diet (3 g/kg) along with intermittent cornstarch therapy. Without cornstarch, hypoglycemia and ketosis invariably occur, but treatment with 1–1.75 g/kg at four- to six-hour intervals during both day and night maintains normoglycemia, increases growth velocity, and decreases serum transaminase concentrations.[51] As patients get older, hypoglycemia is less common, and the minimum amount of cornstarch is used to maintain normoglycemia. Continuous nocturnal feeding of a nutrient mixture consisting of glucose or glucose oligosaccharides, and protein or amino acids, combined with intermittent high protein feedings during the day may be especially beneficial for patients who have significant growth retardation and myopathy.[52] In GSD III, galactose and fructose can be converted into glucose, and strict restriction of these sugars is not necessary. Overstorage of carbohydrates can be problematic, and concentrated sweets should be avoided to avoid excessive glycogen storage.

Natural History and Prognosis

Patients with type III GSD typically do well throughout childhood and as young adults, but complications can occur as patients get older (Figure 50-4). Hepatic adenomas can develop in adults with GSD III, but the frequency is much less than in the GSD I population. Cases of hepatocellular carcinoma have been reported.[22] Hepatic fibrosis is common in children with both GSD IIIa and IIIb, and cirrhosis and portal hypertension can develop.

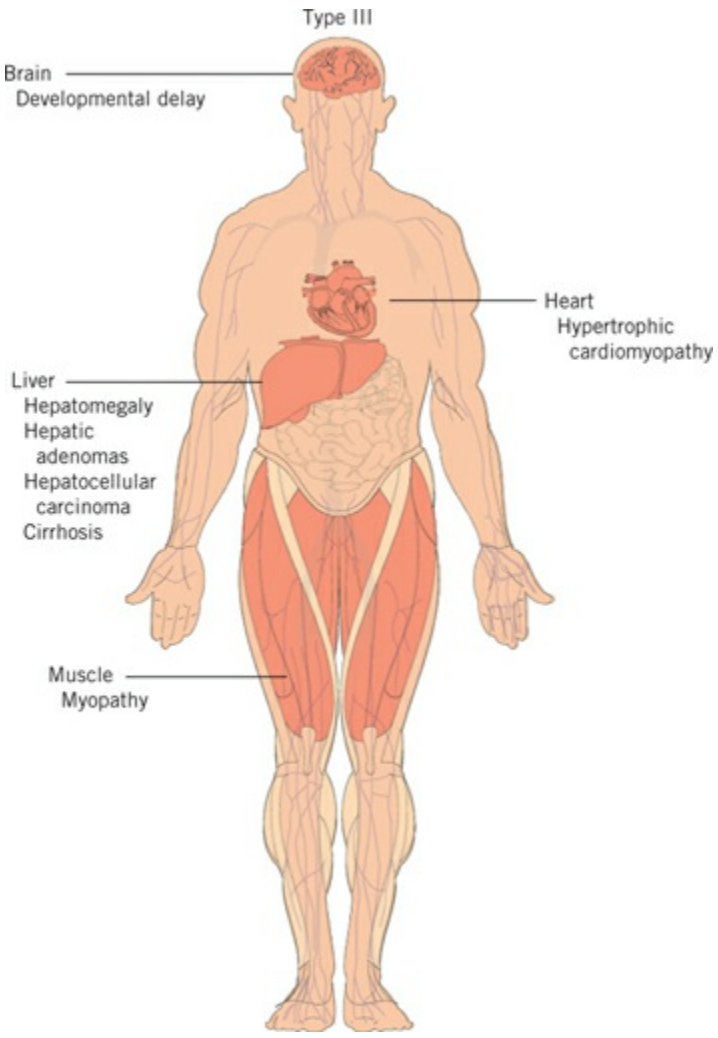


Figure 50-4 Complications associated with GSD type III.

With GSD IIIa, complications related to the muscle disease become increasingly common with aging. In the third to fourth decades of life, myopathy becomes prominent with slowly progressive muscle weakness.[53] Exercise is restricted because it causes elevations in serum creatine kinase

and aldolase concentrations. Unlike in McArdle's disease (GSD V), treatment with carbohydrate prior to exercise does not prevent muscle damage or symptomatology.[54]

Cardiac complications may also develop in patients with GSD IIIa. Usually by the time a child has finished puberty, concentric left ventricular hypertrophy is detectable by echocardiography. Ventricular function is usually normal, but severe cardiac dysfunction and arrhythmias can occur.[55] Rare cases of severe cardiomyopathy in infants and children have been reported, so annual echocardiograms are recommended for all children with GSD IIIa. The hypertrophic cardiomyopathy in GSD III appears largely due to overtreatment with carbohydrate. Restriction of carbohydrate intake and a high protein diet has successfully resulted in decreased ventricular hypertrophy and improved function.[56,57]

Liver transplantation in GSD III is not recommended. Normalization of glucose concentrations does not prevent cardiac or muscular disease, and transplantation may hasten the development of complications.[58] Several of the immunomodulators, including cyclosporine, may exacerbate the myopathy leading to earlier clinical deterioration. Since liver pathology is not the major source of morbidity in GSD type IIIa, liver transplantation should only be performed when there is high risk for hepatocellular carcinoma or evidence of substantial cirrhosis or liver dysfunction.

GLYCOGEN PHOSPHORYLASE DEFICIENCY (TYPE VI GSD; HERS DISEASE) AND GLYCOGEN PHOSPHORYLASE KINASE DEFICIENCY (TYPE IX GSD)

Both types VI and IX GSD result from impaired glycogenolysis. Type VI GSD is due to hepatic phosphorylase deficiency, whereas type IX GSD results from abnormal phosphorylase kinase (PHK) activity, the enzyme that activates hepatic phosphorylase. Phenotypically, these diseases are similar, and they are therefore discussed as one entity in this section. Their estimated prevalence is approximately 1 in 100,000, but this is almost certainly an underestimation. Together, they account for approximately 25%–30% of all cases of GSD.[59] Type IX GSD is more common in people of Scandinavian heritage, and 1 in 1000 people of Mennonite heritage have type VI GSD.[60]

The hepatic glycogen phosphorylase gene (PYGL) is located on

chromosome 14q21-22 and is inherited in an autosomal recessive manner.[2] PHK is present in liver and muscle, and it is a complex enzyme consisting of four subunits: α, β, δ, and γ. The α-subunit is encoded by the PHKA2 gene located on Xp22.2-p22.1 and is inherited as an X-linked disease. The β-subunit is encoded by PHKB2 gene on chromosome 16q12-q13, and mutations are inherited as an autosomal recessive variant of GSD IX. The γ-subunit, encoded by the PHKG2 gene on chromosome 16p11-p12, contains the catalytic site of the PHK enzyme and is inherited autosomal recessively.[2]

Pathophysiology

Glycogen phosphorylase is the rate-limiting step in glycogenolysis stimulating the hydrolysis of the outer branches of glycogen. Glycogen PHK deficiency results in failure to activate glycogen phosphorylase. Fortunately, these disorders are typically mild and can be confused with "ketotic hypoglycemia." Mild hyperlipidemia and elevation in liver enzyme concentration are common in untreated children, whereas lactic acid and uric acid concentrations are typically normal.

Presentation

Fasting ketotic hypoglycemia is the cardinal manifestation although typically milder than GSD I and III. Children are usually discovered when hepatomegaly is detected on a routine physical examination or when hypoglycemia occurs as a consequence of prolonged fasting during an intercurrent illness.[61]

Diagnosis

It is possible to diagnose glycogen PHK deficiency by assaying the activity of the enzyme in leukocytes and erythrocytes. The blood assay, however, lacks sensitivity as there are liver, muscle, heart, and brain isoforms of this enzyme. Mutation analysis is now the preferred method for diagnosing these conditions. Sequencing of the PHKA2 gene (GSD IX alpha) is usually performed first in males; GSD VI is more common in females suspected of having one of these conditions. Mutations in the PHKB or PHKG2 genes are less common causes of GSD IX.[62]

Nutritional Management

The goal of treatment is to prevent hypoglycemia, normalize plasma ketone concentrations, and minimize systemic acidosis. Frequent small

meals are supplemented with UCS (1.5–2 g/kg given one to two times per day). Occasionally, children with GSD IX may have severe hypoglycemia and a phenotype similar to that of GSD I. Even when more prolonged fasting can be tolerated without development of hypoglycemia, most children and adults report improved energy and well-being with bedtime cornstarch administration because ketosis is prevented.[63] A high protein (2–3 g/kg/d) and a complex carbohydrate diet is recommended. Restriction of simple sugar intake results in decreased hepatomegaly.

Natural History and Prognosis

The prognosis of patients with GSD VI and IX is excellent. Treatment, however, improves growth and stamina in patients with these milder forms of GSD. Although most patients do very well, a more severe variant associated with significant hepatic transaminase elevation, hepatic fibrosis, and cirrhosis has been described in patients with mutations in PHKA2 or PHKG2.[64] There is evidence that aggressive treatment may delay or prevent onset of these complications.[65] Muscle symptoms occur occasionally, and these disorders are rarely associated with a proximal renal tubular acidosis and neurologic abnormalities.[66] A severe cardiac-specific PHK variant that can lead to cardiac failure has been reported.[67] Hyperlipidemia is common in untreated children and adults, and untreated adults may be at higher risk for cardiovascular disease.

REFERENCES

1. Wolfsdorf JI, Weinstein DA. Glycogen storage disease. *Rev Endocr Metab Disord*. 2003;4(1):95–102.
2. Elpeleg ON. The molecular background of glycogen metabolism disorders. *J Ped Endocr Metab*. 1999;12(3):363–379.
3. Roach PJ. Glycogen and its metabolism. *Curr Mol Med*. 2002; 2(2):101–120.
4. Orho M, Bosshard N, Buist NR, et al. Mutations in the liver glycogen synthase gene in children with hypoglycemia due to glycogen storage disease type 0. *J Clin Invest*. 1998;102(3):507–515.
5. Weinstein DA, Correia CE, Saunders AC, et al. Hepatic glycogen synthase deficiency: an in frequently recognized cause of ketotic hypoglycemia. *Mol Genet Metab*. 2005;87(4):284–288.
6. Aynsley-Green A, Williamson DH, Gitzelmann R. Hepatic glycogen synthetase deficiency. Definition of syndrome from metabolic and enzyme studies on a 9-year-old girl. *Arch Dis Child*. 1977;52(7):573–579.
7. Hasselbalch SG, Knudsen GM, Jakobsen J, et al. Blood-brain barrier permeability of glucose and ketone bodies during short-term starvation in humans. *Am J Physiol*. 1995;268(6 Pt1):E1161–E1166.
8. Bachrach BE, Weinstein DA, Orho-Melander M, et al. Glycogen synthase

deficiency (glycogen storage disease type 0) presenting with hyperglycemia and glucosuria: report of three new mutations. *J Pediatr*. 2002;140(6):781–783.
9. Dykes JR, Spencer-Peet J. Hepatic glycogen synthetase deficiency. Further studies on a family. *Arch Dis Child*. 1972;47(253):558–563.
10. Nordlie RC, Foster JD, Lange AJ. Regulation of glucose production by the liver. *Annu Rev Nutr*. 1999;19:379–406.
11. Burchell A. The molecular basis of the type 1 glycogen storage diseases. *Bioessays*. 1992;14(6):395–400.
12. Lei KJ, Shelly LL, Pan CJ, et al. Mutations in the glucose-6-phosphatase gene that cause glycogen storage disease type 1a. *Science*. 1993;262(5133):580–583.
13. Ekstein J, Rubin BY, Anderson SL, et al. Mutation frequencies for glycogen storage disease Ia in the Ashkenazi Jewish population. *Am J Med Genet A*. 2004;129(2):162–164.
14. Fernandes J, Berger R, Smit GP. Lactate as a cerebral metabolic fuel for glucose-6-phosphatase deficiency. *Pediatr Res*. 1984; 18(4):335–339.
15. Alaupovic P, Fernandes J. The serum apolipoprotein profile of patients with glucose-6-phosphatase deficiency. *Pediatr Res*. 1985;19:380–384.
16. Rake JP, ten Berge AM, Visser G, et al. Glycogen storage disease type Ia: recent experience with mutation analysis, a summary of mutations reported in the literature and a newly developed diagnostic flow chart. *Eur J Pediatr*. 2000;159(5):322–330.
17. Wolfsdorf JI, Keller RJ, Landy H, Crigler JR. Glucose therapy for glycogenosis type 1 in infants: comparison of intermittent uncooked cornstarch and continuous overnight glucose feedings. *J Pediatr*. 1990;117(3):384–391.
18. Bier DM, Leake RD, Haymond MW, et al. Measurement of "true" glucose production rates in infancy and childhood with 6,6-dideuteroglucose. *Diabetes*. 1977;26(11):1016–1023.
19. Weinstein DA, Wolfsdorf JI. Effect of continuous glucose therapy with uncooked cornstarch on the long-term clinical course of type 1a glycogen storage disease. *Eur J Pediatr*. 2002;161 Suppl 1:S35–S39.
20. Correia CE, Bhattacharya K, Lee PJ, et al. Use of modified cornstarch therapy to extend fasting in glycogen storage disease types Ia and Ib. *Am J Clin Nutri*. 2008;88(5):1272–1276.
21. Wang DQ, Fiske LM, Correras CT, Weinstein DA. Natural history of hepatocellualar adenoma formation in glycogen storage disease type I. *J Pediatr*. 2011; 159(3):442–446.
22. Bianchi L. Glycogen storage disease I and hepatocellular tumours. *Eur J Pediatr*. 1993;152 Suppl 1:S63–S70.
23. Labrune P, Trioche P, Duvaltier I, et al. Hepatocellular adenomas in glycogen storage disease type I and III: a series of 43 patients and review of the literature. *J Pediatr Gastroenterol Nutr*. 1997;24(3):276–279.
24. Franco LM, Krishnamurthy V, Bali D, et al. Hepatocellular carcinoma in glycogen storage disease type Ia: a case series. *J Inherit Metab Dis*. 2005;28(2):153–162.
25. Pauleit D, Textor J, Bachmann R, et al. Hepatocellular carcinoma: detection with gadolinium and ferumoxides-enhanced MR imaging of the liver. *Radiology*. 2002;222(1):73–80.
26. Restaino I, Kaplan BS, Stanley C, Baker L. Nephrolithiasis, hypocitraturia, and a distal renal tubular acidification defect in type 1 glycogen storage disease. *J Pediatr*. 1993;122(3):392–396.
27. Weinstein DA, Somers MJ, Wolfdorf JI. Decreased urinary citrate excretion in type 1a glycogen storage disease. *J Pediatr*. 2001;138(3):378–382.

28. Chen Y.-T, Coleman RA, Scheinman JI, et al. Renal disease in type I glycogen storage disease. *N Engl J Med*. 1988;318(1):7–11.
29. Wolfsdorf JI, Laffel LMB, Crigler JF. Metabolic control and renal dysfunction in type I glycogen storage disease. *J Inher Metab Dis*. 1997;20(4):559–568.
30. Wang DQ, Carreras CT, Fiske LM, et al. Characterization and pathogenesis of anemia in glycogen storage disease type Ia and Ib. *Genet Med*. 2012;14(9):795–799.
31. Weinstein DA, Roy CN, Fleming MD, et al. Inappropriate expression of hepcidin is associated with iron refractory anemia: implications for the anemia of chronic disease. *Blood*. 2002;100(10):3776–3781.
32. Minarich LA, Kirpich A, Fiske LM, Weinstein DA. Bone mineral density in glycogen storage disease type Ia and Ib. *Genet Med*. 2012. doi: 10.1038/gim.2012.36. [epub ahead of print].
33. Lee PJ, Patel JS, Fewtrell M, et al. Bone mineralization in type I glycogen storage disease. *Eur J Pediatr*. 1995;154(6):483–487.
34. Wolfsdorf JI. Bones benefit from better biochemical control in type I glycogen storage disease. *J Pediatr*. 2002;141(3):308–310.
35. Lee PJ, Leonard JV. The hepatic glycogen storage diseases—problems beyond childhood. *J Inher Metab Dis*. 1995;18(4):462–472.
36. Sechi A, Deroma L, Lapolla A, et al. Fertility and pregnancy in women affected by glycogen storage disease type I, results of a multicenter Italian study. *J Inherit Metab Dis*. 2013;36(1):83–89.
37. Annabi B, Hiraiwa H, Mansfield BC, et al. The gene for glycogen-storage disease type 1b maps to chromosome 11q23. *Am J Hum Genet*. 1998;62(2):400–405.
38. Visser G, Rake JP, Fernandes J, et al. Neutropenia, neutrophil dysfunction, and inflammatory bowel disease in glycogen storage disease type Ib: results of the European Study on Glycogen Storage Disease Type I. *J Pediatr*. 2000;137(2):187–191.
39. Visser G, Rake JP, Labrune P, et al. Granulocyte colony-stimulating factor in glycogen storage disease type Ib. Results of the European Study on Glycogen Storage Disease Type I. *Eur J Pediatr*. 2002;161 Suppl 1:S83–S87.
40. Chen LY, Pan CJ, Shieh JJ, Chou JY. Structure-function analysis of the glucose-6-phosphate transporter deficient in glycogen storage disease type Ib. *Hum Mol Genet*. 2002;11(25):3199–3207.
41. Roe TF, Thomas DW, Gilsanz V, et al. Inflammatory bowel disease in glycogen storage disease type Ib. *J Pediatr*. 1986;109(1):55–59.
42. Davis MK, Valentine JK, Weinstein DA, Polyak S. Antibodies to CBir1 are associated with glycogen storage disease type Ib. *J Pediatr Gastroenterol Nutr*. 2010;51(1):14–18.
43. Roe TF, Coates TD, Thomas DW, et al. Treatment of chronic inflammatory bowel disease in glycogen storage disease type Ib with colony-stimulating factors. *N Engl J Med*. 1992;326(25):1666–1669.
44. Davis MK, Rufo PA, Polyak SF, Weinstein DA. Adalimumab for the treatment of Crohn-like colitis and enteritis in glycogen storage disease type Ib. *J Inherit Metab Dis*. 2008 [epub ahead of print Jan 5, 2008; PMID 18172743].
45. Melis D, Della Casa R, Parini R, et al. Vitamin E supplementation improves neutropenia and reduces the frequency of infections in patients with glycogen storage disease type 1b. *Eur J Pediatr*. 2009; 168(9):1069–1074.
46. Parvari R, Moses S, Shen J, et al. A single-base deletion in the 3'-coding region of glycogen debranching enzyme is prevalent in glycogen storage disease type IIIA in a population of North African Jewish patients. *Eur J Hum Genet*.

1997;5(5):266–270.
47. Santer R, Kinner M, Steuerwald U, et al. Molecular genetic basis and prevalence of glycogen storage disease type IIIa in the Faroe Islands. *Eur J Hum Genet*. 2001;9(5):388–391.
48. Bao Y, Dawson TL, Chen YT. Human glycogen debranching enzyme gene (AGL): complete structural organization and characterization of the 5' flanking region. *Genomics*. 1996;38(2):155–165.
49. Shen J, Bao Y, Liu HM, et al. Mutations in exon 3 of the glycogen debranching enzyme gene are associated with glycogen storage disease type III that is differentially expressed in liver and muscle. *J Clin Invest*. 1996;98(2):352–357.
50. Coleman RA, Winter HS, Wolf B, Chen YT. Glycogen debranching enzyme deficiency: long-term study of serum enzyme activities and clinical features. *J Inherit Metab Dis*. 1992;15(6):869–881.
51. Borowitz SM, Greene HL. Cornstarch therapy in a patient with type III glycogen storage disease. *J Ped Gastroeneterol Nutr*. 1987;6(4):631–634.
52. Slonim A, Weisberg C, Benke P, et al. Reversal of debrancher deficiency myopathy by the use of high-protein nutrition. *Ann Neurol*. 1982;11(4):420–422.
53. Moses S, Gadoth N, Bashan N, et al. Neuromuscular involvement in glycogen storage disease type III. *Acta Paediatr Scand*. 1986;75(2):289–296.
54. Preisler N, Pradel A, Husu E, et al. Exercise intolerance in glycogen storage disease type III: weakness or energy deficiency? *Mol Genet Metab*. 2013;109(1):14–20.
55. Lee PJ, Deanfield JE, Burch M, et al. Comparison of the functional significance of left ventricular hypertrophy in hypertrophic cardiomyopathy and glycogenosis type III. *Am J Card*. 1997;79(6):834–838.
56. Dagli AI, Zori RT, McCune H, et al. Reversal of glycogen storage disease type IIIa-related cardiomyopathy with modification of diet. *J Inherit Metab Dis*. 2009;32 Suppl 1:S103–S106.
57. Sentner CP, Caliskan K, Vletter WB, Smit GP. Heart failure due to severe hypertrophic cardiomyopathy reversed by low calorie, high protein dietary adjustments in a glycogen storage disease type IIIa patient. *JIMD Rep*. 2012;5:13–16.
58. Davis MK, Weinstein DA. Liver transplantation in children with glycogen storage disease: controversies and evaluation of the risk/benefit of this procedure. *Pediatr Transplant*. 2008;12(2):137–145.
59. Kiliman MW. Glycogen storage disease due to phosphorylase kinase deficiency. In: Swallow DM, Edwards YH, editors. *Protein Dysfunction in Human Genetic Disease*. Oxford, United Kingdom: BIOS Scientific; 1997. pp. 57–75.
60. Chang S, Rosenberg MJ, Morton H, et al. Identification of a mutation in liver glycogen phosphorylase in glycogen storage disease type VI. *Hum Mol Genet*. 1998;7(5):865–870.
61. Willems PJ, Gerver WJ, Berger R, Fernandes J. The natural history of liver glycogenosis due to phosphorylase kinase deficiency: a longitudinal study of 41 patients. *Eur J Pediatr*. 1990;149(4):268–271.
62. Davit-Spraul A, Piraud M, Dobbelaere D, et al. Liver glycogen storage diseases due to phosphorylase system deficiencies: diagnosis thanks to non invasive blood enzymatic and molecular studies. *Mol Genet Metab*. 2011;104(1-2):137–143.
63. Nakai A, Shigematsu Y, Takano T, et al. Uncooked cornstarch treatment for hepatic phosphorylase kinase deficiency. *Eur J Pediatr*. 1994;153(8):581–583.
64. Burwinkel B, Rootwelt T, Kvittingen EA, et al. Severe phenotype of phosphorylase kinase-deficient liver glycogenosis with mutations in the PHKG2

gene. *Pediatr Res*. 2003;54(6):834–839.
65. Tsilianidis LA, Fiske LM, Siegel S, et al. Aggressive therapy improves cirrhosis in glycogen storage disease type IX. *Mol Genet Metabol*. 2013 [epub ahead of print March 21, 2013; PMID 23578772].
66. Nagai T, Matsua N, Tsuchiya Y, et al. Proximal renal tubular acidosis associated with glycogen storage disease, type 9. *Acta Paediatr Scand*. 1988;77(3):460–463.
67. Servidei S, Metlay LA, Chodosh, J, DiMauro S. Fatal infantile cardiopathy caused by phosphorylase b kinase deficiency. *J Pediatr*. 1988; 113(1 Pt 1):82–85.

Acute and Chronic Kidney Disease

51

Nutrition in Renal Disease

Nancy M. Rodig, MD

The following chapter will outline principles for the nutritional management of children with both acute and chronic disturbances of renal function. A significant decline in glomerular filtration rate will impair nitrogenous waste excretion; alter regulation of water, electrolyte, and acid–base homeostasis; and challenge the provision of optimal nutrition. This chapter will begin with a discussion of the nutritional management of acute kidney injury, which may be associated with a rapid decline in glomerular filtration rate and urine flow. Given the extreme physiologic abnormalities that may accompany severe acute kidney injury, the nutritional care of the critically ill child will be emphasized. The discussion will then turn to the care of children with chronic kidney disease, who are at increased risk for chronic malnutrition and growth impairment. Noting the shared issues faced by children with chronic kidney disease and those on maintenance dialysis, the nutritional management of these children will be discussed together. The nutritional care of children who have received renal transplantations will then be reviewed. As the range of clinical manifestations of both acute and chronic renal failure is broad, the need for individualized nutritional management is emphasized throughout.

ESTIMATION OF GLOMERULAR FILTRATION

RATE

Glomerular filtration rate (GFR) is equal to the sum of the filtration rates of all functioning nephrons and reflects functioning renal mass. Estimation of GFR allows clinicians to determine the severity and course of both acute and chronic disturbances of renal function and permits anticipation of necessary changes in therapy, including adjustments of nutritional support. The "gold standard" to measure GFR had been measurement of inulin clearance; however, this study is cumbersome and rarely employed in routine clinical practice. Alternatively, creatinine clearance based on a timed urine collection is readily available, though the collection is inconvenient, prone to error, and impractical in children who are not toilet trained. Estimation of GFR based on equations using serum creatinine and other variables such as age, height, and gender is convenient and easy to perform at the bedside. The Schwartz formula[1–3] (Table 51-1) and the Modification of Diet in Renal Disease (MDRD) formula[4] have gained widespread clinical use in North America for children and adults, respectively. Given the tubular secretion of creatinine and changes in methods used to measure creatinine, the Schwartz formula tends to overestimate GFR, and this overestimation increases with decreasing renal function.[5,6] More recently, data generated from the Chronic Kidney Disease in Children (CKiD) study have permitted the development of an updated and more accurate Schwartz formula, estimating GFR (creatinine clearance) from serum creatinine, the child's height, and a fixed proportionality constant[7]:

GFR (mL/min/1.73 m^2) = height (cm) × 0.413/serum creatinine (mg/dL)

This formula was developed from a study of children with mild to moderate CKD and is most useful in a GFR range of 15–75 mL/min/1.73 m^2. The updated Schwartz formula is less accurate in children with more normal renal funciton[8] and has not been validated in children with acute kidney injury. If an exact measurement of GFR is necessary, this can be determined by plasma disappearance of iohexol, a safe, nonionic, low-osmolar contrast agent. This study should be performed if renal function is believed to be stable, as the expense and effort would be not well spent if renal function is dynamic.

ACUTE KIDNEY INJURY

Acute kidney injury (AKI) is defined by a rapid and usually reversible

deterioration in GFR. Depending on the severity of the insult, AKI may be accompanied by impairment of acid–base homeostasis and loss of water and electrolyte regulation. In mild cases, nonoliguric AKI may be asymptomatic and only be detected when serum laboratory studies are performed. When severe, oliguric AKI may result in significant derangements of electrolyte and volume balance and necessitate the initiation of renal replacement therapy. Given the lack of a standardized definition of AKI, studies to identify preventive and therapeutic measures have been confounded. To address this classification, definitions have been proposed for adults, including the RIFLE[9] and AKIN criteria.[10] The RIFLE criteria consists of three levels of kidney dysfunction (Risk, Injury and Failure), and a pediatric modification of the RIFLE criteria (pRIFLE) has been validated and should facilitate the evaluation and treatment of children with AKI.[11]

The etiology of AKI has changed with advancement of medical and surgical practice, and this trend is most apparent in tertiary care centers. In referral centers, AKI due to primary renal disease is now less frequent than failure, secondary to ischemia, drug toxicity, or sepsis in children.[12] Underlying comorbidities or chronic conditions in critically ill children who develop AKI have been reported in as many two-thirds of these patients.[13,14] As in adults, the reported mortality for children with AKI is high, reaching 30%–40% and is greatest for those who require dialysis.[15–18] Preexisting or hospital acquired malnutrition has been identified as a contributing factor to the poor outcome of severe AKI,[19] and providing adequate nutrition should be viewed as a crucial element in the therapeutic effort.

TABLE 51-1 Estimation of GFR by the Schwartz Formula

$C_{cr} = k \times L/S_{cr}$

C_{cr} = creatinine clearance in mL/min/1.73 m^2

k = proportionality constant values:

Low birth infants during first year of life = 0.33
Full-term infants during first year of life = 0.45
Children and adolescent girls = 0.55
Adolescent boys = 0.7

L = length (cm)

S_{cr} = serum creatinine (mg/dL)

Source: Adapted from Schwartz GJ, Haycock GB, Edelmann CM Jr, Spitzer A. A simple estimate of

glomerular filtration rate in children derived from body length and plasma creatinine. *Pediatrics*. 1976;58:259–263; Schwartz GJ, Feld LG, Langford DJ. A simple estimate of glomerular filtration rate in full-term infants during the first year of life. *J Pediatr*. 1984;104:849–854; and Schwartz GJ, Gauthier B. A simple estimate of glomerular filtration rate in adolescent boys. *J Pediatr*. 1985;106:522–526.

Nutritional support obligates the administration of volume, either in the form enteral feeds, intravenous nutrition, or a combination of these. If AKI is accompanied by anuria or oligoanuria (urine flow < 0.5–1 mL/kg/h), nutrition will be limited or volume status will be worsened unless renal replacement therapy is initiated. The modality of renal replacement therapy is determined based on the clinical status of the patient and the expertise of the center. Options for renal replacement therapy include peritoneal dialysis and hemodialysis, and hemodialysis may be performed on an intermittent or continuous basis. For hemodynamically stable patients with AKI, intermittent hemodialysis is typically performed daily for 2–4 hours per session depending on the size of the patient. During the several hour treatment, ultrafiltration of the daily fluid requirement in additional to any other desired fluid removal is attempted as hemodynamically tolerated. If the patient is hemodynamically unstable, continuous renal replacement therapy (CRRT) allows for gradual ultrafiltration of fluid and is the modality of choice in such patients.[20] Two commonly used modalities of CRRT are continuous venovenous hemodiafiltration (CVVHD) and continuous venovenous hemofiltration (CVVH). CVVHD provides clearance by primarily diffusive mechanisms with some convective clearance, whereas continuous CVVH uses solely convective clearance.

AKI: Energy Requirements

Though an essential component of care, the nutritional management of children with AKI has been insufficiently studied. The need to provide adequate nutrition is of particular importance in critically ill children who have sustained AKI, as the high prevalence of malnutrition in this group is associated with excessive morbidity and mortality.[21,22] Timely initiation of appropriate nutrition is essential to optimize tissue healing and organ recovery, though the lack of sufficient data to propose evidence-based recommendations is notable and underscores the need for study. Careful assessment of energy requirements in children with AKI is essential, though challenging. Inadequate caloric provision will result in poor protein retention, and excessive nutrition increases the risk for metabolic derangement and fluid excess.

Total energy requirements include resting energy expenditure, energy needed for physical activity, and diet-induced thermogenesis. In patients with uncomplicated AKI, energy expenditure has been shown to be comparable to healthy subjects as measured by indirect calorimetry.[23] AKI associated with sepsis or multiorgan dysfunction results in increased energy expenditure, and the increase in expenditure is believed to be proportionate to the underlying process leading to the critical illness.[24] Predicting energy needs based on the severity of illness is often inaccurate.[25] When the resting energy expenditure of 33 critically ill children was measured by indirect calorimetry, 55% of subjects were hypometabolic and 17% were hypermetabolic.[26] One study of critically ill adult patients compared the energy expenditure of patients who required dialysis for severe AKI to those who had normal or only moderately impaired renal function. These patients demonstrated increased energy expenditure, though there was significant individual variation. The measured energy expenditure was greater for those who required only supportive measures than for those who required dialysis with average hypermetabolism measured at 42% and 28%, respectively. It was proposed that AKI reduced energy demand secondary to diminished renal metabolic activity.[27] When based on predictive equations, studies of critically ill children with AKI demonstrate a high prevalence of insufficient energy provision.[28] The use of indirect calorimetry allows nutrition to be tailored to the needs of the individual patient and is the preferred method when feasible, though lack of expertise and resource constraints may be limiting. When assessment by indirect calorimetry is not possible, energy expenditure can be predicted according to the Schofield prediction equation[29] and the American Society for Parenteral and Enteral Nutrition guidelines for critically ill children.[30] Given the dynamic nature of illness and the complexities of predicting metabolic state, the use of standard equations is prone to inaccuracies.

The general recommendation for adult patients with AKI is that energy intake be tailored to the individual patient, and an early study based on indirect calorimetry suggested that AKI and sepsis increased energy needs up to 30%.[23,31–34] Pediatric patients with AKI who are relatively well and nonoliguric are unlikely to have significantly increased energy requirements. In contrast, critically ill children will have expenditures concordant with the severity of their illness. However, given the potential risks for both under and overfeeding,[26] targeted indirect calorimetry should be considered for children at high risk for metabolic alterations,

such as those with prolonged ICU stay, unexpected fluctuations of weight, or evidence of persistent inflammatory state. The reader is referred to Chapter 62 for further guidance regarding energy requirements for the critically ill child.

Hyperglycemia is not uncommon in critically ill patients with AKI. The potential for altered carbohydrate metabolism must be considered given that standard nutrition provides the majority of calories from dextrose, and patients on dialysis are delivered additional dextrose from dialysate. Hyperglycemia is a consequence of both accelerated hepatic gluconeogenesis and insulin resistance. Increased hepatic gluconeogenesis results from conversion of amino acids released during protein catabolism.[35] The kidney plays a direct role in glucose homeostasis, which is regulated by insulin.[36] During AKI, insulin resistance may be more likely given the loss of the kidney as a major target organ for insulin action. The consequences of insulin resistance and hyperglycemia have been shown to be significant. Both were associated with increased mortality in critically ill adults with AKI.[37] Critically ill infants and children who demonstrated sustained hyperglycemia suffered a threefold increase in mortality over those with well-controlled glycemia.[38] Hyperglycemia has also been correlated with prolonged ventilator dependency and increased length of hospital stay.[39] Given the association of hyperglycemia with increased complications and worse prognosis,[40] efforts to maintain normoglycemia should be made, though there is insufficient data to recommend strict glycemic control in children. Furthermore, the risks of attempting strict glycemic control, such as hypoglycemia, must be acknowledged.

AKI: Protein Requirements

AKI is a state of increased catabolism, which parallels the severity of the underlying illness. During metabolic stress, the catabolism of skeletal muscle allows for the generation of glucose, which is the preferred substrate for the brain, red blood cells, and renal medulla. This stress response is an effective short-term adaptation, though a prolonged response is maladaptive and can result in reduction of lean body mass. Studies evaluating critically ill adults with AKI have reported high protein catabolic rates.[41] Virtually all nitrogen arising from amino acids liberated during protein degradation is converted to urea, and the extent of protein catabolism may be assessed by calculating urea nitrogen appearance (UNA). In dialysis patients, UNA equals the sum of the urea in the dialysis

ultrafiltrate, urine, and the change in the body nitrogen pool. The UNA of critically ill children with AKI maintained on CRRT was evaluated in an early study and measured to be >180 mg/kg/day.[42] This compares to a mean UNA of 103 mg/kg/day among children on maintenance (chronic) peritoneal dialysis who are at baseline health.[43] Increased protein catabolic rate was confirmed in a subsequent report of critically ill pediatric patients on CVVH and CVVHD with mean UNA of 291 and 245 mg/kg/day, respectively. Nitrogen balance, defined as the difference between nitrogen intake and UNA, was often negative in these patients despite the provision of 1.5 g/kg/day of protein.[24]

If the care of a patient with AKI requires dialysis, the modality of dialysis must be considered when determining protein requirements. Hemodialysis leads to nitrogen loss, as amino acids and small peptides are filtered across the dialysis membrane.[44,45] During continuous dialysis, there will be continuous nitrogen loss. One study of pediatric patients on CRRT measured amino acid clearance to be 12% and 11% of daily protein intake on CVVH and CVVHD, respectively.[24] A subsequent study measured amino acid clearance while on CVVHD to be 20% of protein intake.[46] Peritoneal dialysis also results in significant protein losses through the dialysis effluent, and the degree of protein loss varies inversely with the body surface area of the child. Peritoneal protein loss was highest in infants (277 ± 22 mg/kg/day) and lowest in children who were above 50 kg (91 ± 15 mg/kg/day).[47]

Adult studies suggest protein intake for patients supported on dialysis should be between 1.8 and 2.5 g/kg/day with the lower range appropriate for those on intermittent dialysis and the higher range needed to achieve positive nitrogen balance for those on CRRT.[48,49] Data supporting recommendations for protein intake in pediatric patients with AKI are lacking. A child with uncomplicated AKI should be provided protein in the amount recommended for age with adjustment to provide ongoing needs. While protein intake is increased, electrolytes, acid–base balance, and uremia should be assessed daily. Nutritional guidelines for critically ill children estimate protein requirements to be 2–3 g/kg/day for 0–2 years, 1.5–2 g/kg/day for 2–13 years, and 1.5 g/kg/day for 13–18 years.[30] Should the child require renal replacement therapy, particularly continuous modalities of hemodialysis, amino acid clearance up to 20% of protein intake should be accounted for. Adjustment of protein intake is recommended with a broad goal of maintaining blood urea nitrogen (BUN) >40 mg/dL and <80 mg/dL.

AKI: Micronutrient Requirements

Trace element requirements in AKI are not clearly defined. Most recommendations are extrapolated from data studying patients with chronic kidney disease. In addition, many of the findings regarding trace element metabolism in AKI may represent the effect of the acute phase response and not necessarily reflect specific effects induced by AKI. As trace elements have small molecular weights and critical illness results in altered protein binding, hemodialysis will alter homeostasis. In vitro data have demonstrated clearance of trace elements during CVVH, including selenium, chromium, copper, and zinc.[50] The ultrafiltration rate or rate of convective fluid removal, was a significant factor affecting clearance. However, other studies evaluating micronutrient losses in patients on CRRT have demonstrated varying results.[51,52] Variations likely stem from differences in patient population, CRRT method, and assays employed. One study confirmed clearance of selenium.[53] As selenium is cofactor for the antioxidant glutathione peroxidase enzymes, impaired defense against antioxidant stress is of concern. Further studies are needed to determine trace element requirements in renal failure.

Recommendations for vitamin supplementation during AKI are provided by the ASPEN guidelines.[54] Additional water-soluble vitamins should be administered if the patient is on hemodialysis to replace expected losses. Supplementation with folate, vitamin B_{12}, pyridoxine, thiamine, and riboflavin is recommended. Vitamin C is metabolized to oxalate, which can accumulate in patients with AKI and result in renal tubule toxicity. Oxalate accumulation has not been adequately studied in AKI and until further pediatric data are available, supplementation of vitamin C should not exceed the recommended daily intake. Fat-soluble vitamins may accumulate in CKD, as they are not removed with dialysis. Little data regarding fat-soluble vitamins in AKI are available, though deficiencies, with the exception of vitamin K, have been reported.[55] ASPEN guidelines recommend supplementation of fat-soluble vitamins if the duration of AKI is limited (i.e., <2 weeks).

AKI: Electrolyte Homeostasis

The kidneys are responsible for the normal homeostasis of many electrolytes, and AKI predisposes the patient to electrolyte and acid–base disorders. Full discussion of electrolyte physiology in the setting of altered renal function is beyond the scope of this chapter, though a limited discussion is provided to serve as a basis for nutritional planning.

Given the potential for life threatening arrhythmias, hyperkalemia is a dreaded consequence of AKI. Potassium is the most abundant intracellular cation, with less than 2% of total body potassium present in the extracellular fluid. As potassium is primarily renally excreted, decreased GFR and renal tubular damage may result in significant hyperkalemia. In addition to decreased excretion, hyperkalemia may result from transcellular shift induced by acidosis and increased extracellular load secondary to enhanced cell turnover, hemolysis, or transfusion of red blood cell products. Therefore, potassium should be withheld during evolving AKI until acceptable potassium homeostasis is assured. If dialysis is initiated, potassium clearance should be anticipated if the dialysate contains no or little potassium. While on CRRT, potassium will be cleared continuously. If clinically appropriate, potassium salts may be added to the dialysate (typically up to 3 mEq/L) to mitigate the tendency for hypokalemia.

Similar to hyperkalemia, hyperphosphatemia is a common complication of oliguric and anuric renal failure. Phosphorus is the major intracellular anion with less than 1% present in the extracellular space. Serum phosphorus occurs in two forms, organic and inorganic. The inorganic fraction is the principal circulating form and routinely assayed for clinical use. When GFR is within the normal range and renal tubular function is intact, approximately 90% of serum inorganic phosphorus is filtered, and ~80%–90% of filtered phosphorus is reabsorbed. In AKI, hyperphosphatemia may cause hypocalcemia as a result of calcium–phosphate precipitation, which typically begins when the calcium × phosphorus product is greater than 70. Treatment for hyperphosphatemia includes restricting or eliminating all sources of phosphorus and administrating oral phosphate binders with enteral feeds. Calcium carbonate and calcium acetate are commonly used phosphorus binders in this setting and will provide supplemental calcium, which may be beneficial if hypocalcemia has developed. If the patient is hypercalcemic, noncalcium containing binders such as sevelamer carbonate should be considered.

AKI: Routes of Nutrition

After estimating the nutritional needs of a child with AKI, the next challenge is to determine the appropriate route of nutrition. If the child's gastrointestinal tract is functioning, enteral nutrition is the preferred method of support for the critically ill patient. Proposed benefits of enteral nutrition include intestinal trophism, reducing bacterial translocation, and

cost effectiveness.[56,57] Early initiation of enteral nutrition has shown benefits in both adults[58] and children[59] with respect to decreased incidence of septic complications and improved prognosis.[60] A specific benefit of enteral feeds in the setting of oliguric AKI is the potential for providing concentrated nutrition and thereby limiting fluid intake.

Though enteral feeds are preferred, they may not be tolerated in critically ill patients. The underlying illness and need for vasoactive medications may compromise gastrointestinal perfusion. An additional concern in the setting of AKI is the detrimental effect of uremia on gastrointestinal motility. Nasogastric feeds were well tolerated when applied carefully to a population of adult patients with AKI without significant increase in major gastrointestinal complications.[61] To decrease the risk of aspiration and improve tolerance of feeds when gastric emptying is delayed, transpyloric feeds should be considered. When critically ill children with and without AKI were studied, continuous transpyloric feeds were associated with increased gastrointestinal complications, including abdominal distension and excessive gastric residue. However, transpyloric feeds were overall effective and well tolerated.[62,63] Presently there is insufficient evidence to support gastric versus postpyloric feedings, though post-pyloric feeds should be considered in patients at significant risk for aspiration and for those who fail gastric feedings.[30]

If enteral feeds are initiated, both standard formulas and formulas appropriate for decreased renal function are available. Similac PM60/40 is an infant formula with lower mineral content suitable for infants with acute reduction of renal function, though more commonly utilized in chronic kidney disease. Suplena and Nepro are specifically designed for patients with chronic kidney disease who are predialysis and dialysis dependent, respectively. A selection of formulas are highlighted in Table 51-2. to permit comparison of caloric and mineral content; full nutritional content is available in Appendix III. Feeds should be initiated with hypo- or normocaloric formula at low rates of 0.5–1 mL/kg/h with gradual increase to normal caloric delivery for age over 36–48 hours as tolerated. Gastric residuals and abdominal exams should be monitored closely with advancement.[63] If the patient is volume restricted, the rate should be increased to the target volume and then gradually fortified using modular components to meet nutritional needs. To optimize digestibility, similar proportions of fat, protein, and carbohydrate should be used. In patients on CRRT, amino acid and small peptide losses may challenge the ability to

supply adequate protein enterally. If indicated, administration of 10% amino acids may be considered to achieve desired goals.[61]

In critically ill patients with AKI, enteral feeds often cannot meet nutritional needs. Parenteral nutrition (PN) combined with enteral support or total PN may be necessary. A balanced PN regimen usually provides 10%–20% of total calories by amino acids, 50%–60% of calories by dextrose, and 20%–30% by lipid emulsion. Modification of PN should be performed based on amino acid losses and dextrose absorption from dialysis therapies, especially in CRRT. Electrolyte composition should be guided by regular assessment of serial laboratory studies.

CHRONIC KIDNEY DISEASE

Chronic kidney disease (CKD) is defined as kidney damage based on persistent structural or functional abnormalities, which may be associated with reduced or normal GFR. In children, CKD arises more commonly from congenital than acquired renal disease. When reviewing registry data of children with CKD, the most common primary diseases were obstructive uropathy (21.1%), renal aplasia, hypoplasia, and dysplasia (17.5%), and reflux nephropathy (8.4%). Focal segmental glomerulosclerosis, an acquired glomerulopathy, accounted for 8.7% of cases.[64] When kidney damage is significant, there is progressive loss of functioning nephron mass leading to end-stage renal disease (ESRD). In 2002, the Kidney Disease Outcomes Quality Initiative (K/DOQI) of the National Kidney Foundation (NKF) proposed definition criteria and a classification scheme defining the stages of CKD (Table 51-3).[65] Given the importance of early detection and intervention, guidelines to facilitate the detection of kidney disease in children and adults have been proposed.[65,66] When compared to adult onset CKD, persistent kidney disease during childhood poses unique challenges, including abnormal growth. Despite advances in medical management, growth in children with CKD remains suboptimal.

TABLE 51-2 Standard and Renal Specific Formulas and Supplements

		Protein	Na	K	Ca	P
Product (manufacturer)	**kcal/mL**	**g/1000 mL**	**mEq/1000 mL**		**mg/1000 mL**	
Infant Formulas						

Similac Advance (Abbott Nutrition)	0.67	14	7	18	528	284
Similac PM 60/40 (Abbott Nutrition)	0.67	15	7	14	379	189
Non Infant Formulas						
Pediasure (Abbott Nutrition)	1.0	30	17	34	1055	844
Nutren 2.0 (Nestle Health)	2.0	80	57	49	1340	1340
Suplena (Abbott)	1.8	45	35	29	1055	717
Nepro (Abbott)	1.8	81	46	27	1060	720

Sources: Adapted from Abbott Nutrition and Nestle Health Science 2013 Product Guides.

Recent data regarding growth outcomes of children with CKD is provided by the National Institute of Health sponsored CKiD study, a prospective study of North American children with mild to moderate CKD.[67] The median height standard deviation score (SDS) was −0.57, and 13% of the children had severe short stature as defined by a height SDS ≤ −1.88. Only ~25% of children with severe short stature were receiving recombinant human growth hormone (rhGH), a therapy proven safe and efficacious in children with growth failure associated CKD (CKiD paper). Lower levels of renal function and severe metabolic acidosis (CO_2 < 18 mEq/L) were associated with deficits of both height and weight. Of note, when compared to height growth, the weight of these children was relatively preserved, particularly in children with glomerular cause of kidney disease. The combined prevalence of overweight or obese children with CKD was comparable to the general population of children and adolescents in the United States when defined by age-based body mass index. Longitudinal studies of growth in the CKiD cohort are ongoing.

The youngest children with CKD comprise the most vulnerable population at risk for severe growth deficiency.[68] In the healthy state, one-third of total postnatal stature is achieved in the first two years of life. It is during this critical time that a significant loss of height potential can occur. The European Study Group for nutritional treatment of chronic renal failure in childhood identified infants with early onset chronic renal failure had lost approximately one standard deviation of height during fetal life and another standard deviation was lost by the third postnatal month.[69] This finding was consistent with reported average birth length of −0.74 SDS in 12 infants with chronic renal failure[70] and more recently corroborated in a report of the International Pediatric Peritoneal Dialysis Network.[71] In this study of over 150 children, the mean birth length was

−0.47 SDS, supporting a link between intrauterine growth and kidney disease.

An essential component of the care of children with CKD is the provision of adequate nutrition and regular evaluation of nutritional status. In 2008, the Kidney Disease Outcome Quality Initiative (KDOQI) Pediatric Clinical Practice Guideline for Nutrition in Chronic Renal Failure was revised and expanded.[72] While recognizing the lack of available data pertaining to nutrition in children with CKD, the recommendations were based on available evidence and expert opinion. The guideline provides recommendations for the evaluation of growth, nutritional status, and nutritional requirements for children with CKD stages 2–5 and includes recommendations for children on dialysis (CKD stage 5D). Though historically a focus has been directed at the recognition and treatment under nutrition, there is more recent evidence that obesity is an emerging problem in children with kidney disease.[67,73]

TABLE 51-3 Stages of Chronic Kidney Disease

CKD stage	GFR (mL/min/1.73 m^2)	Description: kidney damage with
1	>90	Normal or increased GFR
2	60–89	Mild decrease in GFR
3	30–59	Moderate decrease in GFR
4	15–29	Severe decrease in GFR
5	<15	Kidney failure
5D	<15	Kidney failure treated with dialysis

Source: Adapted from K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. *Am J Kidney Dis*. 2002;39:S1–S266.

If available, the support of a renal dietician is indispensable in optimizing the dietary management of children with CKD. The modification of food and fluid intake proves to be challenging for most. Every child with CKD and their careprovider should receive nutritional counseling based on individual needs. In addition to addressing the specifics of the renal disease process, education level and social influences of the family must be considered. The dietary care plan requires appropriate modification based on the changes in the child's age, nutritional status, renal function, treatment modality, and medication regimen. Depending on the disease time course, a renal dietician may support an individual through the various stages of renal insufficiency,

maintenance dialysis, and renal transplantation. Families often need continual guidance and support to establish and consistently enforce prescribed dietary guidelines. Nutritional specialists can ease the burden of restrictions and increase the likelihood of success by providing practical suggestions for modifying or replacing favorite foods or fluids.

Though inadequate nutrition will negatively impact growth in children with CKD, the variable response to nutritional support highlights the complexity of growth impairment in this population. Alterations in the growth hormone (GH)/insulin-like growth factor-1 (IGF-1) axis independently affect growth in the uremic milieu. Although serum GH and IGF-1 levels are usually normal or high in growth-retarded children with CKD, uremic serum has high IGF-1 binding capacity that results in low IGF-1 bioactivity.[74] In conjunction with optimal medical and nutritional management, recombinant human growth hormone (rhGH) therapy significantly improves linear growth.[75–77] Recommendations for evaluation and treatment of growth failure in children with CKD have been published and include recommendations for rhGH therapy,[78] though recent data suggest that only ~25% of children with CKD and severe short stature (height SDS ≤ –1.88) are prescribed this therapy.[67,79] Secondary hyperparathyroidism and chronic metabolic acidosis are additional factors that impair growth. To optimize the utilization of nutrition for growth and response to rhGH, appropriate correction of secondary hyperparathyroidism and metabolic acidosis is critical.

CKD: Assessment of Growth and Nutritional Status

The importance of nutritional status has been emphasized in numerous clinical practice guidelines regarding the care of patients with CKD.[80,81] Nutritional status is a complex concept, and no single measurement is comprehensive. Therefore a multistaged evaluation of body composition is necessary. Measurements of nutritional parameters are complicated in CKD given the potential for fluid overload and potential abnormalities in the distribution of fat and lean tissue. Children with CKD prior to and after renal transplantation have been found to have discordant body composition with relatively high fat mass and low lean mass.[82] In addition, assessment methods recommended for children with CKD were developed in healthy children, and the validity may be limited in CKD. A comprehensive review has been published summarizing the methods recommended for nutritional assessment of children with CKD and details the advantages and disadvantages of these methods.[83]

The most recently published KDOQI Clinical Practice Guideline for Nutrition in CKD recommends a three-day dietary intake and a variety of anthropometric measurements. The measures and suggested frequency are summarized in Table 51-4. Of note, body mass index (BMI) varies with age and pubertal status. Children with CKD are shorter and sexually immature,[79] and it is thus recommended that BMI might best be expressed relative to height–age in children in this population, as this measure more effectively compares children with CKD to children of the same height and stage of sexual development.[84] There are limitations using BMI-height-age as an assessment of growth, such as when short stature is disproportionate to the degree of delay in sexual maturation. For adolescents on dialysis, a monthly assessment of normalized protein catabolic rate (nPCR) to provide an objective measure of protein intake is recommended. Adolescents with nPCR <1 g/kg/day were at significantly increased risk for weight loss, though there was no association between nPCR and weight in younger children.[85] Reflecting growth challenges faced by infants, the frequency of nutritional and anthropometric assessment is increased in younger children. These guidelines outline the minimum frequency of assessment, and more frequent assessment may be appropriate and should be tailored to the individual patient. In particular, infants and children with comorbid conditions, worsening growth, or acute medical issues may warrant more frequent monitoring.

TABLE 51-4 Nutritional and Growth Assessment

	Minimal Interval of Measurement (months)									
	Age < 1 year			Age 1–3 years			Age > 3 years			
Measurement	CKD stage			CKD stage			CKD stage			
	2–3	4–5	5D	2–3	4–5	5D	2	3	4–5	5D
Dietary intake	0.5–3	0.5–3	0.5–2	1–3	1–3	1–3	6–12	6	3–4	3–4
Height or length-for-age % or SDS	0.5–1.5	0.5–1.5	0.5–1	1–3	1–2	1	3–6	3–6	1–3	1–3
Height or length velocity-for-age % or SDS	0.5–2	0.5–2	0.5–1	1–6	1–3	1–2	6	6	6	6
Estimated dry weight and weight-	0.5–	0.5–	0.25–	1–	1–	0.5–	3–	3–	1–	1–

for-age % or SDS	1.5	1.5	1	3	2	1	6	6	3	3
BMI-for-height-age % SDS	0.5–1.5	0.5–1.5	0.5–1	1–3	1–2	1	3–6	3–6	1–3	1–3
Head circumference–for-age % or SDS	0.5–1.5	0.5–1.5	0.5–1	1–3	1–2	1–2	Not applicable			

Source: Adapted from Group KW. KDOQI Clinical Practice Guideline for Nutrition in Children with CKD: 2008 update. Executive summary.[72]

CKD: Supplemental Feedings

Though the etiology of growth failure in CKD is multifactorial, inadequate nutrition will adversely affect growth outcome.[86,87] In addition to impacting somatic growth, malnutrition is associated with increased morbidity and mortality in children with CKD.[88,89] Several studies have demonstrated altered spontaneous nutritional intake in children with CKD and generally lower intake of total energy than that in healthy children.[90,91] Protein intake has been variable, with some studies demonstrating lower[91] and other studies reporting higher spontaneous intake of protein than recommended.[92,93]

The etiology of decreased spontaneous energy intake in children with CKD is multifactorial. Vomiting and gastroesophageal (GE) reflux are common in infants with CKD. GE reflux was identified in nearly 75% of 22 infants studied by 24-hour esophageal pH monitoring,[94] and a variety of forgut motor disorders have been identified, including delayed gastric motility and emptying.[95] In the CKD population, diminished appetite has been associated with higher serum concentrations of inflammatory markers, including tumor necrosis factor-α, interleukin-6, C-reactive protein, and leptin.[96–98] Among children with CKD, systemic inflammation and disturbances in appetite-regulating hormones, such as ghrelin, have been identified and places these children at risk for cachexia, a state of muscle wasting with or without the underutilization of fat.[99] Furthermore, poor appetite may be a consequence of multiple medications prescribed as part of a complex CKD regimen and a preference for water in children with polyuria and increased obligatory water needs.

As nutritional intake may be suboptimal in children with CKD, the use of supplemental enteral feedings is commonly implemented. Given that growth velocity is maximal during infancy and predominantly dependent on nutrition, most studies have focused on the younger child. Nasogastric and gastric feeds were initiated in 35 children up to age 5 years with estimated GFR <26 mL/min/1.73 m^2. The aim of supplemental feedings

was to provide at least 100% of estimated average requirement for energy and 100% of the reference nutrient intake for protein for height–age. The benefit was most apparent in children 0–2 years of age (n = 26) with improved mean weight SDS from −3.1 at the initiation of feeds to −1.4 at 2 years. Improvement in height SDS was also significant with mean height SDS −2.9 at initiation and −2.1 at 2 years.[100] A subsequent report from the same center studied growth outcomes of 81 children who presented with CKD (estimated GFR <20 mL/min/1.73 m^2) in the first six months of life. Supplemental enteral feeds were commenced in 81% of patients when growth became subnormal. The mean height SDS was within the normal range at 1 year of age and continued to improve thereafter.[101]

Though supplemental feeds were associated with improved growth in some studies,[102–105] others have not demonstrated the same benefit. Several studies have shown stabilization of linear growth without significant catch-up growth.[106,107] Abitbol et al. studied 12 infants with congenital renal insufficiency who were provided controlled feeding regimens starting at 3 months of age. Six of these infants received supplemental nasogastric or gastric tube feedings. The average energy intake was 95% of recommended daily allowance (RDA) with average protein intake of 141% RDA. The study revealed that the major height deficit was incurred during the first six months of life, as the length SDS fell from −0.7 at birth to approximately −1.9 at 6 months. There was no catch-up growth noted as indicated by the length SDS of −2.0 at 24 months.[70] A multicenter experience comes from the North American Pediatric Renal Trials and Collaborative Studies (NAPRTCS) survey, which reviewed the impact of supplemental feedings on growth and mortality in children <6 years of age at dialysis initiation. Questionnaires were completed on 137 patients, 70% of whom received supplemental feedings and outcome was compared to those who did not receive supplements. When compared to children between the ages of 2 and 5 years, significantly more children less than 2 years of age were provided supplemental feedings (~80% vs. 41%). Despite supplemental feedings, there was no difference in height and weight, and height and weight SDS did not improve over time with supplementation. In addition, mortality was unaffected.[108]

Another report from the International Pediatric Peritoneal Dialysis Network analyzed growth outcomes of 153 children in 18 countries initiating chronic peritoneal dialysis at <24 months of age.[71] During periods without enteral feedings BMI SDS uniformly decreased, whereas

nasogastric (NG) and gastrostomy feedings reversed this loss. Gastrostomy feeds were associated with superior growth outcome when compared to NG feeds. The proposed explanation for this difference was increased vomiting with NG feeds due to stenting of the gastroesophageal junction. Enteral feedings are an accepted standard of practice in the care of children with CKD who do not meet the RDA for energy and protein and are recommended in the KDOQI pediatric nutritional guidelines.[72] Studies thus far identify younger children as the group most likely to require this support, and the choice of nasogastric versus gastric feedings depends on the needs and comorbidities of the individual patient. Ideally, placement of a gastrostomy tube should occur prior to initiation of peritoneal dialysis (PD), and placement of percutaneous gastrostomy tube while on PD should be avoided given the risks for infection and PD failure. To encourage daytime oral intake and provide oral stimulation, continuous overnight feeds are generally preferred. Infants and toddlers who have been supported with tube feedings may experience oral aversion and feeding dysfunction and would benefit from the counseling of a multidisciplinary feeding team.[109,110]

CKD: Energy Requirements

Energy requirements for children with CKD should be considered to be the same as estimated for healthy children, as there is a lack of evidence to suggest needs are increased. When adjusted for lean body mass, the resting energy expenditure measured by indirect calorimetry of 25 children and adolescents on chronic hemodialysis was the same as for healthy controls.[111] Therefore, the energy intake should be 100% of the estimated energy requirements for chronologic age and adjusted for physical activity level and body size. As calculated energy requirements are estimates, all prescriptions should be individualized, and further adjustments should be made in response to weight loss or weight gain. Adequate energy intake is not only critical to allow optimal growth but prevents the utilization of protein stores as an energy source. Energy-malnourished children may require additional calories to restore normal growth and allow “catch up” growth if possible. With respect to macronutrient distribution, the recommended ranges for distribution of calories from protein, fat, and carbohydrate should not be different from that of the general pediatric population.[72] Given the increased cardiovascular risk of children with CKD,[112] preference for unsaturated fat and limiting saturated or trans fats should be counseled.

To increase energy intake and improve compliance, dietary modifications for children are typically less restrictive than those prescribed to adults. Dietary guidelines for adults often recommend specific amounts of a restricted nutrient, such as a “2 g sodium diet.” In pediatrics, restrictions are imposed only when clinically indicated and usually take the form of a diet low in a particular nutrient such as sodium, potassium, and phosphorus with education on which foods are high or low in that nutrient. Minimizing the number of dietary restrictions as allowable by metabolic balance will increase the likelihood of achieving energy goals. Individuals with poor appetites may respond to small, frequent meals and snacks. Energy can be added to foods by adding heart healthy oils or margarines, creams, syrups, or carbohydrate modules. For children who are underweight, low calorie or calorie free drinks should be restricted. If the child does not meet energy requirements, supplemental nutrition with energy-dense diet or commercial nutritional supplements should be provided. Oral supplementation is preferred, though enteral feedings may be required. Nonrenal pediatric feedings designed for children older than one year (Kindercal, Pediasure, Nutren Junior) have standard calcium, phosphorus, and potassium content, which may result in electrolyte disturbance depending on the volume required and the degree of renal insufficiency. Nutritionally complete renal formulas designed to be energy dense, high or low in protein, and low in electrolytes including phosphorus may be given to children older than one year. Suplena (Ross Products) is designed for CKD prior to dialysis dependence and Nepro (Abbott) is intended for patients on maintenance dialysis (Table 51-2). Serum magnesium should be monitored if used in younger children, as the magnesium content is higher in adult renal products than in infant or pediatric formulas.

Children with CKD may require volume restriction if urine output is limited. Volume restriction is most problematic if the intake of solid nutrition is limited due to age or oral aversion, which is of particular concern in infants and young children. If fluid restriction is indicated, fortification of feeds to increase caloric density will be necessary. Caloric density may be increased gradually by 2–4 kcal/ounce increments up to 50–60 kcal/ounce if needed, and the tempo of fortification will depend on individual tolerance. Both breast milk and commercial formulas may be fortified. It is not advisable to increase the energy density of formula by concentration alone due to the increased sodium, potassium, phosphorus, and overall renal solute load. Carbohydrate, protein, and fat modular products can be added to achieve the desired caloric density. Readily

available oils such as corn, canola, and safflower may be used for fat supplementation. The benefit of fat modular products is the process of emulsification, which prevents the fat from separating out of the formula during continuous feeds. When making a formula with increased energy density, an attempt should be made to preserve the proportionate distribution of energy from carbohydrate, protein, and fat as in the base formula.

Patients on maintenance dialysis may have specific nutritional issues related to renal replacement therapy. For children treated with peritoneal dialysis, early satiety or anorexia may result secondary to pressure exerted by the indwelling dialysate volume.[113,114] Intraperitoneal dialysate glucose absorption should also be considered when calculating total energy intake. Approximately 7–10 kcal/kg/day are absorbed by children on CAPD.[115] For patients on maintenance hemodialysis who are malnourished (BMI-for-height-age < 5%) and unable to meet nutritional goals via oral or enteral feedings, a trial of intradialytic parenteral nutrition (IDPN) has been suggested by some.[116] IDPN appears to be less effective therapy for psychosocial causes of malnutrition[117] and is not recommended in adult patients.[31] IDPN is infused continuously during the hemodialysis session through the venous limb of the circuit to prevent immediate clearance of trace elements and amino acids. Potential adverse outcomes include hyperglycemia, postinfusion hypoglycemia, and hyperlipidemia. IDPN should be utilized as a supplement to oral and enteral nutrition and not utilized as the sole or primary source of nutrition.

CKD: Protein Requirements

Care of CKD emphasizes supportive measures to retard disease progression. There has been concern that higher protein intake would induce intraglomerular hypertension and hyperfiltration injury, thereby hastening the progression of kidney disease. Given that a low protein diet was able to slow the deterioration of renal function in rodent studies,[118,119] prospective clinical studies prescribing low-protein diets were conducted in both adults[120–123] and children.[124] The results of prospective clinical studies in adults were contradictory. Overall the progression rate was not significantly influenced by protein restriction over a period of 2–3 years, though a low protein diet postponed the development of uremic symptoms and the start of dialysis for adult patients with advanced renal failure. In the pediatric study of nearly 200 children with CKD stages 3–4, a diet targeting 100% RDA for protein (0.8–1.1 g/kg/day based on ideal body

weight) was compared to a more liberal protein intake (mean 181% RDA). The diet with lower protein intake (achieving 120% of the RDA) did not affect the rate of decline in creatinine clearance or adversely affect growth.[124] Though these outcomes were not different, the increased phosphorus load of a higher protein diet must be considered, as studies have linked hyperphosphatemia and arteriopathy.[125,126] The association of high phosphorus load and cardiovascular risks provide the rationale for avoiding excessive protein intake in the CKD population.[127] Furthermore, avoiding excessive protein intake will limit accumulation of nitrogenous waste products.

Current recommendations for protein intake are based on the stage of CKD. For children with CKD stage 3, dietary protein intake should be 100%–140% of the daily recommended intake (DRI) for ideal body weight. For children with CKD stage 4–5, the recommended amount is 100%–120% of the DRI, as lower intake did not significantly delay dialysis initiation in the above cited pediatric trial. A diet targeting 100% of the DRI may be difficult for families to achieve and should be reconsidered if this restriction negatively impacts the child's total nutritional intake. For obese children, additional body protein is required for structural support of excess weight; estimation of protein requirements for obese children can be based on "adjusted" weight (i.e., adjusted weight = ideal weight for height + 25% × [actual weight – ideal weight]).[128] Ensuring adequate caloric needs overall is essential to reduce the inefficient use of dietary protein as a calorie source, resulting in increased urea generation. Even if protein intake exceeds recommended levels, protein malnutrition may occur if overall energy intake is low.

Of note, children on maintenance dialysis (CKD stage 5D) will incur dialytic nitrogen loss, and it is recommended to maintain dietary protein intake at 100% of the DRI for ideal body weight with an additional amount provided to account for protein and amino acid losses on dialysis (Table 51-5). Transperitoneal protein losses demonstrate significant variability and inverse correlation with body surface area. Infants were shown to have nearly twofold greater peritoneal protein losses per meter square body surface area than older children weighing more than 50 kg. Daily protein losses average 0.28 g/kg in the first year of life and decrease to < 0.1 g/kg in adolescents.[47] In addition, transperitoneal protein losses were twice as great in patients requiring maintenance PD due to steroid-resistant nephrotic syndrome when compared to children without nephrotic syndrome.[129] Additional dietary protein ranging from 0.15 to 0.35

mg/kg/day, dependent on patient age, should be provided to children on PD to account for losses. Amino acid and protein losses during HD vary according to the membrane characteristics of the dialysis filter and are inadequately studied in children. Based on adult data, it is recommended that additional 0.1 g/kg/day of dietary protein be provided to children on hemodialysis to compensate for losses. Given the significant variability in losses, regular assessment of growth, serum albumin, and urea levels is essential to determine if adjustment in protein intake is necessary. Regular assessment of predialysis urea levels may allow assessment of protein intake, though other clinical factors which affect blood urea nitrogen (BUN) must be considered. A predialysis BUN greater than 70–80 mmol/L may indicate excessive protein intake. However inadequate dialysis, recirculation of blood, and increased catabolism will also increase the predialysis BUN. Alternatively, persistently low predialysis BUN of <20 mmol/L may indicate inadequate protein intake.

TABLE 51-5 Recommended Daily Dietary Protein Intake in Children with CKD Stage 5D

Age	DRI (g/kg/d)	Children on HD (g/kg/d)	Children on PD (g/dk/d)
0–6 months	1.5	1.6	1.8
7–12 months	1.2	1.3	1.5
1–3 years	1.05	1.15	1.3
4–13 years	0.95	1.05	1.1
14–18 years	0.85	0.95	1.0

Source: Adapted from Group KW. KDOQI Clinical Practice Guideline for Nutrition in Children with CKD: 2008 update. Executive summary.[72]

CKD: Vitamin and micronutrient requirement

The specific vitamin requirements of children with CKD are poorly defined, underscoring the need for further study. The K/DOQI Pediatric Nutrition Guidelines recommends dietary of at least 100% of the DRI for thiamin (B_1), riboflavin (B_2), niacin (B_3), pantothenic acid (B_5), pyridoxin (B_6), biotin (B_8), cobalamin (B_{12}), ascorbic acid (C), retinol (A), α-tocopherol (E), vitamin K, folic acid, copper, and zinc for children with CKD stages 2–5D.[72] When dietary intake is considerably less than recommended, vitamin supplementation should be considered. Children on dialysis have additional risk for vitamin deficiency due to losses of water-

soluble vitamins from dialysis clearance. A study of children on peritoneal dialysis revealed the dietary intake of vitamins was lower than the DRI, and supplementation resulted in intakes that exceeded recommended amounts.[130–132] Children with a robust appetite for a variety of nutritious foods and children who receive a significant amount of nutrition from adult renal formulas will generally meet the DRI. If supplements are indicated, infants and small children can be given liquid or chewable multivitamin preparations at appropriate dosing, and adequate folic acid provision should be confirmed. Older children and adults can be provided B complex vitamins which generally contain 0.8–1 mg folic acid, such as Nephrocaps (Flemming & Company) and Nephrovite (R & D Laboratories, Inc.). Excessive amounts of vitamin A and vitamin C should be avoided. Excessive vitamin A can lead to hypercalcemia,[133] anemia, and hyperlipidemia. Vitamin C is metabolized to oxalate, which is renally excreted. Excessive vitamin C may result in elevated oxalate levels and complications of retention.[134]

In children with CKD, bone mineral metabolism and vitamin D homeostasis becomes disrupted. The most active form of vitamin D is 1,25-dihydroxycholecalciferol ($1,25[OH]_2D_3$), and the enzymatic step of 1α- hydroxylation occurs in the kidney. Impaired production of $1,25[OH]_2D_3$ in the context of CKD is multifactorial, including phosphate retention, loss of the renal enzyme, and inhibition of 1α- hydroxylation by fibroblast growth factor 23 (FGF-23).[135] With progression of renal disease, secondary hyperparathyroidism and defective bone mineralization may ensue due to inadequate $1,25(OH)_2D_3$ levels. Secondary hyperparathyroidism can begin as early as CKD stage 2. An early study of renal osteodystrophy in 29 children with moderate to severe CKD found that parathyroid hormone (PTH) levels were elevated in all children with GFR < 45 mL/min/1.73 m^2. “Early” renal osteodystrophy characterized by elevated PTH but normal serum chemistries was detected in one quarter of the patients.[136]

Deficiency of 25-hydroxyvitamin D has been well described in children with CKD and is associated with more advanced renal disease, dark complexions, and lower sunlight exposure in winter months.[137,138] The Kidney Disease: Improving Global Outcomes (KDIGO) and KDOQI guidelines recommend assessment of 25-cholecalciferol ($25[OH]D_3$) in CKD stages 2–5D be measured at least yearly and subsequent testing based on results and need for supplementation.[139] Supplementation with

vitamin D_2 (ergocalciferol) or vitamin D_3 (cholecalciferol) is suggested if the level is <30 ng/mL. Correction of vitamin D deficiency improved secondary hyperparathyroidism in children with CKD.[140] Guidelines for supplementation with ergocalciferol based on serum 25-cholecalciferol levels in children with CKD stages 2–4 have been provided by KDOQI (Table 51-6).[72,141] Given the potential effect of ergocalciferol therapy on calcium and phosphorus homeostasis, serum levels of calcium and phosphorus should be monitored at least monthly after initiation of therapy. Should hypercalcemia develop during supplementation, discontinuation of ergocalciferol should be considered. If hyperphosphatemia develops, interventions include restriction of dietary phosphate, initiation of a phosphate binder, and discontinuation of ergocalciferol supplementation. If indicated based on serum levels of PTH and vitamin D metabolites, supplementation with activated vitamin D should be provided. Calcitriol is commonly prescribed and dose adjusted to target-specific PTH levels based on renal function. The initiation and adjustment of calcitriol is typically the responsibility of a pediatric nephrologist. Due to potent effect on intestinal calcium absorption, calcitriol treatment can induce hypercalcemia and preclude its use at therapeutic doses. Newer vitamin D analogues have been developed which minimize intestinal calcium and phosphorus absorption while suppressing PTH levels as effectively as calcitriol.[142,143] Currently available active vitamin D sterols with less hypercalcemic effect include paracalcitol, alfacalcidiol, and doxercalciferol, though only paracalcitol has been studied in children.[144,145]

TABLE 51-6 Recommended Supplementation for Vitamin D Deficiency/Insufficiency in Children with CKD

Definition	Serum 25-hydroxyvitamin D (ng/mL)	Ergocalciferol (vitamin D_2) dose	Duration of therapy
Severe vitamin D deficiency	<5	8,000 IU daily (or 50,000 IU per week) × 4 weeks, then 4,000 IU daily (or 50,000 IU twice per month) × 2 months	3 months
Mild vitamin D deficiency	5-15	4,000 IU daily (or 50,000 IU every other week) × 12 weeks	3 months
Vitamin D insufficiency	16-30	2,000 IU daily (or 50,000 IU every 4 weeks) × 12 weeks	3 months

Sources: Adapted from Group KW. KDOQI Clinical Practice Guideline for Nutrition in Children with CKD: 2008 update. Executive summary. *Am J Kidney Dis*. 2009;53:S11–S104 and National Kidney Foundation. K/DOQI clinical practice guidelines for bone metabolism and disease in children with chronic kidney disease. *Am J Kidney Dis*. 2005;46:S1–S122.

Children with CKD are at risk for anemia secondary to erythropoietin deficiency and iron deficiency. The introduction of recombinant erythropoietin (rHuEPO) therapy in 1988 provided a highly effective treatment for correcting anemia associated with CKD.[146] Iron deficiency is the most common factor that limits the response to rHuEPO, and children on rHuEPO usually require large amounts of supplemental iron. For children on maintenance hemodialysis, it is strongly recommended that iron be given intravenously. For children who are pre-dialysis or on maintenance peritoneal dialysis, iron may be provided either orally or intravenously. Dosing of iron supplementation is guided by serum ferritin and transferrin saturation.[147]

Hyperhomocysteinemia is associated with folate deficiency and has been identified as an independent risk factor for cardiovascular disease in adults with chronic renal disease.[148,149] Hyperhomocysteinemia has also been found in children with chronic kidney disease, though a clear relation to vascular disease remains to be seen.[150] When children with and without CKD where studied, plasma homocysteine levels where found to increase with deterioration of renal function. Hyperhomocysteinemia was found in 5.2% of controls, 35.7% of non-dialyzed CKD, and 86.6% of children with ESRD on dialysis. Folate levels were also lower in patients than controls.[151] When children on peritoneal dialysis and hemodialysis were provided folic acid supplementation of 2.5 mg daily, plasma homocysteine concentrations were significantly reduced.[152] Currently there is no standard for monitoring homocysteine levels in children with CKD. Guidelines recommend that dietary intake for folic acid should achieve 100% the DRI. If dietary intake of folic acid is inadequate, supplementation should be considered.

CKD: Electrolyte Disturbances

Electrolyte disturbances commonly seen in children with CKD include hyperphosphatemia, hypocalcemia, and hyperkalemia. Though serum levels of sodium are typically within normal range, the inability to maintain sodium homeostasis will have significant clinical effect. Kidney disease associated with sodium retention may result in hypertension and edema. Alternatively, sodium losses from urine wasting in polyuric kidney

disease or losses from dialysis effluent may require sodium supplementation to ensure normal serum concentration and adequate growth. Dietary restrictions of electrolytes may pose significant challenges to the child and caregivers and contribute to inadequate energy intake overall. Restrictions should be applied only when indicated.

Hyperphosphatemia in CKD can lead to significant morbidity due to metastatic calcification, secondary hyperparathyroidism, and renal osteodystrophy.[153] More commonly seen when GFR falls <50 mL/min/1.73 m^2, hyperphosphatemia contributes to secondary hyperparathyroidism by lowering the serum ionized calcium, inhibiting the production of $1,25(OH)_2D_3$ and directly stimulating PTH secretion. Therefore, normal levels of serum phosphorus should be maintained to prevent renal osteodystrophy and poor growth. Guidelines recommend dietary phosphorus intake be restricted to 100% of the DRI for age when the serum PTH is above the target range for the stage of CKD and the serum phosphorus is within the normal range for age. Phosphorus intake should be restricted to 80% of the DRI if both the serum PTH and phosphorus levels are elevated.[72] Once a restriction of dietary phosphorus has been initiated, it is suggested that serum phosphorus concentrations be monitored at least every 3 months in children with CKD stages 3–4. For children with severe CKD or dialysis dependence (stages 5 and 5D), serum phosphorus concentrations should be followed at least monthly. In children who require phosphorus restriction, intake of milk, milk products, eggs, nuts, dried beans, peanut butter, whole grains, chocolate, and other high-phosphorus containing foods should be limited. Restriction of dietary phosphorus can challenge efforts to achieve adequate protein intake, as protein and phosphorus are often found in the same foods. Animal proteins have relatively favorable phosphorus to protein ratios when compared to the ratios in eggs, dairy products, legumes, and lentils.

In addition to limiting dietary phosphorus, intestinal absorption of phosphorus can be decreased with the use of phosphate binders. Options for phosphate binders include both calcium-based (calcium acetate, calcium carbonate) and noncalcium, nonmetal-containing binders (i.e., sevelamer carbonate, lanthanum carbonate).[154] Aluminum-containing binders are available, but long-term use has resulted in retention and toxicity.[155–157] First-line therapy for hyperphosphatemia in children is often a calcium-based binder given the wide availability of chewable and liquid preparations. However, children who receive calcium-based binders may experience hypercalcemia with associated risks of joint, vessel and

soft tissue metastatic calcification.[126] Sevelamer hydrochloride and carbonate have been shown to be effective and safe when studied in pediatric populations and reduce the likelihood of hypercalcemia,[158,159] though the hydrochloride salt has been associated with metabolic acidosis.[160] Lanthanum carbonate has not been studied in children. Calcium carbonate is readily available in a liquid form and sevelamer carbonate is available in packets, both suitable for younger children.

In formula-fed infants with CKD, one approach is to attempt feeds with standard formulas and use phosphate binders when indicated. Should hyperphosphatemia develop and persist despite prescription of phosphate binders, a low-phosphorus formula such as Similac PM 60/40 (Abbott Nutrition) may be provided. The nutrient content of Similac Advance (Abbott Nutrition), a standard infant formula, and Similac PM 60/40 is outlined in Table 51-2 for comparison. The calcium content of Similac PM 60/40 is lower than that of standard infant formulas. Therefore, ensuring adequate calcium intake and monitoring serum calcium, phosphorus, and parathyroid hormone is critical to guide therapy and promote normal bone mineralization. When indicated, calcium supplementation should be provided. In children with CKD stages 2–5D, it is suggested that the total calcium intake range 100%–200% of the DRI for age.[72] The calcium content of phosphate binders should be included in the daily calcium intake.

As with the development of hyperphosphatemia, progressive decline in kidney function predisposes to hyperkalemia. However, hyperkalemia does not typically occur in early stages of CKD due to adaptive mechanisms of individual nephrons as GFR declines. When hyperkalemia develops and persists, high-potassium containing foods should be limited. Unfortunately many high-potassium foods are favored by children and include potato products, tomato products, bananas, oranges, and chocolate. If necessary, potassium exchange resins, such as sodium polystyrene sulphonate, can be prescribed. Formula-fed infants who have significant CKD may require scheduled doses of sodium polystyrene daily, though this may result in increased spitting, aversion, sorbitol-induced diarrhea, and enteral tube obstruction. Methods to lower the potassium content of formula by pretreatment of the formula with sodium polystyrene sulphonate have been published.[161] One study demonstrated a ~60% reduction of potassium content, though the sodium content of the formula was disproportionately increased (~230%). The disproportionate effect was due, in part, to exchange for calcium and magnesium.[162] Increased

sodium content was seen in a subsequent report of formula pretreatment with sodium polystyrene sulfonate.[163] Should similar methods be employed, the potential complications of increased sodium intake and electrolyte abnormalities should be considered.

Sodium balance is directly linked to volume status. Kidney disease characterized by oliguria or anuria typically requires sodium and fluid restrictions to avoid clinical signs of volume excess, such as hypertension and edema. Children at risk for sodium retention are directed to limit dietary intake, including salty snacks, processed luncheon meats, canned soups, and packaged entrees. Food from fast-food restaurants should also be limited. Prior to the initiation of maintenance dialysis, diuretics may be used to improve sodium balance. After the initiation of maintenance dialysis, children continue to limit sodium intake, especially if hypertension is present. Infants on peritoneal dialysis may require sodium supplementation due to sodium losses incurred from ultrafiltrate, which is isotonic, coupled with the relatively low sodium concentration found in formula. To avoid hyponatremia and ensure adequate extracellular volume, sodium supplements may be needed (i.e., 2–5 mEq/kg/day) with close monitoring of electrolytes, volume balance, and growth.[164]

In addition to ensuring sufficient energy and protein intake, the effect of providing supplemental sodium and water to young children with polyuric, salt-wasting forms of chronic renal failure was studied. It was proposed that these children experience growth delay due to chronic intravascular depletion and negative sodium balance, and the provision of low caloric-density, high-volume, and sodium supplemented feeds would promote normal growth. The treatment group consisted of 24 children diagnosed with polyuric renal insufficiency (< 65 mL/min/1.73 m^2) in the first year of life and were compared to a historical population control and a literature control. The nutritional therapy prescribed consisted of an enteral formula diluted with water to a caloric density of 0.3–0.5 kcal/mL and supplemented with 2–4 mEq of sodium per 100 mL of formula. The average energy intake for the entire treatment group was 104 kcal/kg/day (102% RDA), and the average protein intake was 2.45 g/kg/day (153% RDA). None of the children received growth hormone. The patients provided this specified nutritional regimen maintained their height SDS at 1 and 2 years, with a net gain of height SDS of 0.15 during the two years. The change in SDS for height by regressional analysis was significantly greater in the treatment group than in the control groups.[165]

NUTRITIONAL MANAGEMENT AFTER RENAL TRANPLANATION

Renal transplantation is recognized as the preferred treatment for children with ESRD, conferring a survival benefit when compared to the survival of children maintained on chronic dialysis.[166] Furthermore, restoration of normal renal function can greatly improve a child's quality of life. If the kidney transplant functions well, normalization of fluid and dietary intake is permitted, though a no added salt diet is advised if the child is hypertensive. Children often have improvement in appetite after transplantation, though immunosuppressive drugs and other required medications may cause nausea, vomiting, constipation, diarrhea, and/or anorexia. These symptoms are typically treated symptomatically. Recent data demonstrate 5-year renal allograft survival of 84% and 78% for living and deceased donor source, respectively.[167] However, renal allograft function declines over time due to many factors including acute rejection episodes, chronic rejection, and nephrotoxic exposures. If a child has decreased allograft function, nutritional therapy should be based on guidelines for nutritional care of CKD as discussed above. The following section will discuss growth and metabolic disorders after renal transplantation.

Growth After Kidney Transplantation

The mean height of children at the time of kidney transplantation is approximately −1.8 SDS.[167] Though growth typically improves after transplantation, many children demonstrate abnormal growth overall.[168–170] The benefits of improved growth after transplantation are not equally realized in all children. Children less than 6 years of age demonstrate improved linear growth with an increase in height SDS of 0.68 two years after transplantation. Compared to younger children, older children have relatively stable height SDS. Though "catch up" growth has generally been restricted to younger patients, children who reach ESRD early in childhood incur the greatest height deficits overall.[167] Final adult height is positively associated with shorter duration of dialysis and height at the time of transplantation[171,172] and negatively associated with reduced renal funtion.[168] Given the known deleterious effects of steroids, including that of poorer linear growth, research efforts have focused on steroid sparing regimens utilizing early dose reduction, withdrawal, or avoidance of steroids.[173,174] Growth has improved with steroid minimization, though

the benefits have been primarily realized in young or prepubertal children.[171,175] If growth is poor, initiation of growth hormone should be considered.[147,176] Analysis of the NAPRTCS database comparing growth and allograft function in transplant recipients would suggest that this therapy is effective and safe[177] and is recommended by experts in the field.[178] The risks of growth hormone therapy in patient with recurrent rejection episodes should be considered.

Though linear growth may be suboptimal, registry data demonstrate a rapid increase in weight in children of all ages during the first six months after transplantation, with an average increase in weight of 0.81 SDS in the first year following transplantation.[167] This effect is compounded by an increase in the prevalence of pretransplant obesity.[179] Furthermore, studies have demonstrated worse 1-year allograft function in children who were obese at the time of transplanation[180,181] and worse graft outcome in those with rapid weight gain after transplantation.[182] Obesity is an independent risk factor for the development of systolic hypertension in children following renal transplantation.[183] Discordant body composition characterized by relatively low lean mass, high fat mass, and increased central adiposity after renal transplantation in children has been described.[82] Given the association of truncal obesity with increased cardiovascular morbidity in adults, this may suggest additional lifetime risks in these pediatric patients. These concerning trends underscore the need for dietary assessment and nutritional counseling after transplantation.

Posttransplant Dyslipidemia

Hyperlipidemia is a common and significant disorder after renal transplantation and occurs in up to 70% of adult recipients.[184,185] A known cause of cardiovascular disease, hyperlipidemia leads to increased morbidity and mortality in adult transplant recipients, a population which is already burdened with numerous cardiovascular risks.[186] Children also have a high incidence of lipid abnormalities after renal transplantation, reported in 15%–50% of those studied.[187,188] More recently steroid avoidance protocols have reported lower prevalence of elevated cholesterol compared to steroid-based regimens,[171] though another study found steroid minimization to be associated with lower HDL cholesterol[189] and underscores the need for further study. Though well documented in adults, the cardiovascular effects of hyperlipidemia in children are not

currently known.[190,191] In addition to dyslipidemia, children face numerous cardiovascular risks after transplantation, including diabetes, hypertension, and elevated BMI.[187] Furthermore, there are growing concerns that hyperlipidemia may promote allograft injury with increased risk of chronic transplant nephropathy and graft failure.[192,193]

Known risk factors for hyperlipidemia in adult renal transplant recipients include renal dysfunction, hypoalbuminemia, pretransplant hyperlipidemia, family history of hyperlipidemia, obesity, and specific diseases resulting in ESRD.[194] When risk factors for hypercholesterolemia or hypertriglyceridemia were studied in pediatric recipients, pretransplant hyperlipidemia, increased years since transplantation and reduced GFR correlated.[187] Immunosuppressive medications have also been associated with dyslipidemias, particularly steroids,[184,189,195] calcineurin inhibitors,[196,197] and sirolimus.[198,199] When comparing calcineurin inhibitors, the incidence of hyperlipidemia appears to be higher with cyclosporine than tacrolimus.[185,200,201] A longitudinal study of pediatric renal transplant recipients demonstrated that the prevalence of hyperlipidemia declined from 1 to 10 years posttransplant, and this was thought to be due to the reduction of immunosuppressive therapy. Elevated total cholesterol and total triglycerides declined from 70% to 35% and 46% to 15%, respectively.[202] Although diet may contribute to dyslipidemia, a study comparing healthy children to pediatric kidney transplant recipients did not find this assoication.[203] Both groups had similar intake of saturated fats, polyunsaturated fats, and cholesterol, though the serum levels of total cholesterol and triglycerides were higher in kidney transplant recipients. Of note, both groups took in low amounts of polyunsaturated fats and excessive amounts of saturated fats. Given additional risks for dyslipidemia burdening the transplant population, the need for counseling healthy diet and lifestyle is emphasized.

Posttransplant Diabetes Mellitus

Hyperglycemia and posttransplant diabetes mellitus (PTDM) are well known complications of solid organ transplantation. The reported incidence of PTDM in pediatric renal transplant recipients ranges from 1% to 7%, and risk factors include corticosteroid and calcineurin inhibitors. When compared to cyclosporine, tacrolimus-based regimens may present greater risk.[204–206] Corticosteroids alter carbohydrate metabolism by impairing peripheral utilization of glucose, elevating plasma glucagon concentration, and increasing gluconeogenesis.[207] Cyclosporine and

tacrolimus have been shown to inhibit insulin secretion from beta-cells.[208,209] Additional reported risk factors for PTDM include family history of type 2 DM, hyperglycemia in the two weeks after transplantation, African-American race, older age at transplantation (>13 years), and BMI ≥ 30 kg/m^2.[205,210,211] Increased weight and BMI were not identified as a risk factor.

In 2001, NAPRTCS published a report of 36 children who developed PTDM after renal transplantation.[210] The majority of patients (63.6%) presented within the first six months of transplantation, and the diagnosis was most frequently made by the detection of hyperglycemia on routine laboratory follow-up (64%). Three patients (9%) presented with diabetic ketoacidosis. The development of PTDM was a transient disorder in the majority, as less than one-third of patients remained on insulin therapy at follow-up. Another analysis of 16 children who developed PTDM noted that for patients who presented early after transplantation (mean 0.3 years), the disorder was more likely to be transient. Those who presented later (mean 2.7 years) were more likely to have persistent PTDM.[205]

Nutritional Management After Renal Transplantation

Guidelines for the nutritional management of pediatric renal transplant recipients underscore the nutritional challenges faced by these patients, including the side effects of medications and the potential for abnormal renal function. For children with reduced renal allograft function, dietary management with respect to protein, and phosphorus intake is similar as for children with CKD prior to transplantation. It is recommended that energy intake be 100% of the estimated energy requirements for chronological age with adjustment as needed to achieve desired growth outcomes. As many children are required to take in high quantities of fluids to ensure adequate intravascular volume and renal perfusion, this creates the potential for the intake of excessive calories. With the exception of underweight children, the majority of the fluid taken by children after transplant should be composed of water, fat-free or low-fat milk, and sugar-free drinks. The caloric source should be balanced among carbohydrate, protein, and unsaturated fats as recommended by the acceptable macronutrient distribution ranges. For those children who demonstrate glucose intolerance, hyperglycemia, or PTDM after transplantation, avoidance of simple sugars should be counseled. Derangements of glycemic control are typically associated with corticosteroid and calcineurin inhibitor therapy and during the first 3–6

months after transplantation when immunosuppressant dosing is highest. As the prevalence of posttransplant hypertension is high, restriction of dietary sodium should be recommended for those with hypertension or prehypertension. Given the risk factors for metabolic bone disease after transplantation, including corticosteroid therapy, preexisting metabolic bone disease, and residual hyperparathyroidism, calcium and vitamin D intakes of at least 100% of the DRI are recommended.

Pediatric renal transplant recipients are at increased risk for hyperlipidemia, and this risk will be lifelong given the current therapeutic options of dialysis and transplantation for treatment of ESRD. It is standard practice to monitor serum lipids after renal transplantation. Should hyperlipidemia develop, dietary modifications to reduce the intake of saturated fats should be employed. Healthy food preparation and heart-friendly fats such as canola oil, olive oil, and margarines should be emphasized. Delucci et al. offered the Step II American Heart Association diet (no more than 30% of total calories from fat, no more that 7% of calories from saturated fatty acids) to pediatric renal transplant patients with hyperlipidemia. Of 22 eligible patients, only 12 children were willing to participate. After 12 weeks of the diet, total cholesterol and LDL cholesterol declined modestly by 11% and 14%, respectively.[212] When the practical benefit of lifestyle change is limited, pharmacologic treatment should be considered. Lipid-lowering agents such as HMGCoA-reductase inhibitors have been used in pediatric renal transplant recipients with success.[213–216] However, these studies were small and larger controlled studies are needed to more clearly define efficacy and risk.

SUMMARY

Acute and chronic disturbances of kidney function in children require well-developed nutritional plans. Providing appropriate nutrition should be viewed as a critical element in the care of these children, as inadequate nutrition will negatively impact outcome, growth, and development. Alterations in renal function and clinical status during the disease course require repeated evaluations and adjustments of the nutritional care plan. If available, the assessment and counseling provided by a renal dietician is an indispensable resource for the child, caregivers, and clinicians.

REFERENCES

1. Schwartz GJ, Haycock GB, Edelmann CM Jr, Spitzer A. A simple estimate of

glomerular filtration rate in children derived from body length and plasma creatinine. *Pediatrics*. 1976;58:259–263.
2. Schwartz GJ, Feld LG, Langford DJ. A simple estimate of glomerular filtration rate in full-term infants during the first year of life. *J Pediatr*. 1984;104:849–854.
3. Schwartz GJ, Gauthier B. A simple estimate of glomerular filtration rate in adolescent boys. *J Pediatr*. 1985;106:522–526.
4. Levey AS, Bosch JP, Lewis JB, et al. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. Modification of Diet in Renal Disease Study Group. *Ann Intern Med*. 1999;130:461–470.
5. Springate JE, Christensen SL, Feld LG. Serum creatinine level and renal function in children. *Am J Dis Child*. 1992;146:1232–1235.
6. Seikaly MG, Browne R, Bajaj G, Arant BS Jr. Limitations to body length/serum creatinine ratio as an estimate of glomerular filtration in children. *Pediatr Nephrol*. 1996;10:709–711.
7. Schwartz GJ, Munoz A, Schneider MF, et al. New equations to estimate GFR in children with CKD. *J Am Soc Nephrol*. 2009;20:629–637.
8. Gao A, Cachat F, Faouzi M, et al. Comparison of the glomerular filtration rate in children by the new revised Schwartz formula and a new generalized formula. *Kidney Int*. 2013;83:524–530.
9. Bellomo R, Ronco C, Kellum JA, et al. Acute Dialysis Quality Initiative w. Acute renal failure - definition, outcome measures, animal models, fluid therapy and information technology needs: the Second International Consensus Conference of the Acute Dialysis Quality Initiative (ADQI) Group. *Crit Care*. 2004;8:R204–R212.
10. Mehta RL, Kellum JA, Shah SV, et al. Acute Kidney Injury Network: report of an initiative to improve outcomes in acute kidney injury. *Crit Care*. 2007;11:R31.
11. Akcan-Arikan A, Zappitelli M, Loftis LL, et al. Modified RIFLE criteria in critically ill children with acute kidney injury. *Kidney Int*. 2007;71:1028–1035.
12. Andreoli SP. Acute kidney injury in children. *Pediatr Nephrol*. 2009;24:253–263.
13. Hui-Stickle S, Brewer ED, Goldstein SL. Pediatric ARF epidemiology at a tertiary care center from 1999 to 2001. *Am J Kidney Dis*. 2005;45:96–101.
14. Kyle UG, Akcan-Arikan A, Orellana RA, Coss-Bu JA. Nutrition support among critically ill children with AKI. *Clin J Am Soc Nephrol*. 2013;8:568–574.
15. Vachvanichsanong P, Dissaneewate P, Lim A, McNeil E. Childhood acute renal failure: 22-year experience in a university hospital in southern Thailand. *Pediatrics*. 2006;118:e786–e791.
16. Plotz FB, Hulst HE, Twisk JW, et al. Effect of acute renal failure on outcome in children with severe septic shock. *Pediatr Nephrol*. 2005;20:1177–1181.
17. Maxvold NJ, Smoyer WE, Gardner JJ, Bunchman TE. Management of acute renal failure in the pediatric patient: hemofiltration versus hemodialysis. *Am J Kidney Dis*. 1997;30:S84–S88.
18. Zobel G, Ring E, Zobel V. Continuous arteriovenous renal replacement systems for critically ill children. *Pediatr Nephrol*. 1989;3:140–143.
19. Fiaccadori E, Lombardi M, Leonardi S, et al. Prevalence and clinical outcome associated with preexisting malnutrition in acute renal failure: a prospective cohort study. *J Am Soc Nephrol*. 1999;10:581–593.
20. Tolwani A. Continuous renal-replacement therapy for acute kidney injury. *N Engl J Med*. 2012;367:2505–2514.
21. Hulst JM, Joosten KF, Tibboel D, van Goudoever JB. Causes and consequences of inadequate substrate supply to pediatric ICU patients. *Curr Opin Clin Nutr Metab*

Care. 2006;9:297–303.
22. Zappitelli M, Goldstein SL, Symons JM, et al. Protein and calorie prescription for children and young adults receiving continuous renal replacement therapy: a report from the Prospective Pediatric Continuous Renal Replacement Therapy Registry Group. *Crit Care Med*. 2008;36:3239–3245.
23. Schneeweiss B, Graninger W, Stockenhuber F, et al. Energy metabolism in acute and chronic renal failure. *Am J Clin Nutr*. 1990;52:596–601.
24. Maxvold NJ, Smoyer WE, Custer JR, Bunchman TE. Amino acid loss and nitrogen balance in critically ill children with acute renal failure: a prospective comparison between classic hemofiltration and hemofiltration with dialysis. *Crit Care Med*. 2000;28:1161–1165.
25. Coss-Bu JA, Jefferson LS, Walding D, et al. Resting energy expenditure and nitrogen balance in critically ill pediatric patients on mechanical ventilation. *Nutrition*. 1998;14:649–652.
26. Mehta NM, Bechard LJ, Dolan M, et al. Energy imbalance and the risk of overfeeding in critically ill children. *Pediatr Crit Care Med*. 2011;12:398–405.
27. Soop M, Forsberg E, Thorne A, Alvestrand A. Energy expenditure in postoperative multiple organ failure with acute renal failure. *Clin Nephrol*. 1989;31:139–145.
28. Kyle UG, Jaimon N, Coss-Bu JA. Nutrition support in critically ill children: underdelivery of energy and protein compared with current recommendations. *J Acad Nutr Diet*. 2012;112:1987–1992.
29. Schofield WN. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr*. 1985;39 Suppl 1:5–41.
30. Mehta NM, Compher C, Directors ASPENBo. A.S.P.E.N. Clinical Guidelines: nutrition support of the critically ill child. *JPEN J Parenter Enteral Nutr*. 2009;33:260–276.
31. Brown RO, Compher C, American Society for P, Enteral Nutrition Board of D. A.S.P.E.N. clinical guidelines: nutrition support in adult acute and chronic renal failure. *JPEN J Parenter Enteral Nutr*. 2010;34:366–377.
32. Marin A, Hardy G. Practical implications of nutritional support during continuous renal replacement therapy. *Curr Opin Clin Nutr Metab Care*. 2001;4:219–225.
33. Druml W. Nutritional management of acute renal failure. *J Ren Nutr*. 2005;15:63–70.
34. Druml W. Nutritional management of acute renal failure. *Am J Kidney Dis*. 2001;37:S89–S94.
35. Cianciaruso B, Bellizzi V, Napoli R, et al. Hepatic uptake and release of glucose, lactate, and amino acids in acutely uremic dogs. *Metabolism*. 1991;40:261–269.
36. Stumvoll M, Chintalapudi U, Perriello G, et al. Uptake and release of glucose by the human kidney. Postabsorptive rates and responses to epinephrine. *J Clin Invest*. 1995;96:2528–2533.
37. Basi S, Pupim LB, Simmons EM, et al. Insulin resistance in critically ill patients with acute renal failure. *Am J Physiol Renal Physiol*. 2005;289:F259–F264.
38. Srinivasan V, Spinella PC, Drott HR, et al. Association of timing, duration, and intensity of hyperglycemia with intensive care unit mortality in critically ill children. *Pediatr Crit Care Med*. 2004;5:329–336.
39. Alaedeen DI, Walsh MC, Chwals WJ. Total parenteral nutrition-associated hyperglycemia correlates with prolonged mechanical ventilation and hospital stay in septic infants. *J Pediatr Surg*. 2006;41:239–244; discussion–44.
40. van den Berghe G, Wouters P, Weekers F, et al. Intensive insulin therapy in the critically ill patients. *N Engl J Med*. 2001;345:1359–1367.

41. Chima CS, Meyer L, Hummell AC, et al. Protein catabolic rate in patients with acute renal failure on continuous arteriovenous hemofiltration and total parenteral nutrition. *J Am Soc Nephrol*. 1993;3:1516–1521.
42. Kuttnig M, Zobel G, Ring E, et al. Nitrogen and amino acid balance during total parenteral nutrition and continuous arteriovenous hemofiltration in critically ill anuric children. *Child Nephrol Urol*. 1991;11:74–78.
43. Mendley SR, Majkowski NL. Urea and nitrogen excretion in pediatric peritoneal dialysis patients. *Kidney Int*. 2000;58:2564–2570.
44. Ikizler TA, Flakoll PJ, Parker RA, Hakim RM. Amino acid and albumin losses during hemodialysis. *Kidney Int*. 1994;46:830–837.
45. Hynote ED, McCamish MA, Depner TA, Davis PA. Amino acid losses during hemodialysis: effects of high-solute flux and parenteral nutrition in acute renal failure. *JPEN J Parenter Enteral Nutr*. 1995;19:15–21.
46. Zappitelli M, Juarez M, Castillo L, et al. Continuous renal replacement therapy amino acid, trace metal and folate clearance in critically ill children. *Intensive Care Med*. 2009;35:698–706.
47. Quan A, Baum M. Protein losses in children on continuous cycler peritoneal dialysis. *Pediatr Nephrol*. 1996;10:728–731.
48. Scheinkestel CD, Adams F, Mahony L, et al. Impact of increasing parenteral protein loads on amino acid levels and balance in critically ill anuric patients on continuous renal replacement therapy. *Nutrition*. 2003;19:733–740.
49. Scheinkestel CD, Kar L, Marshall K, et al. Prospective randomized trial to assess caloric and protein needs of critically Ill, anuric, ventilated patients requiring continuous renal replacement therapy. *Nutrition*. 2003;19:909–916.
50. Nakamura AT, Btaiche IF, Pasko DA, et al. In vitro clearance of trace elements via continuous renal replacement therapy. *J Ren Nutr*. 2004;14:214–219.
51. Wooley JA, Btaiche IF, Good KL. Metabolic and nutritional aspects of acute renal failure in critically ill patients requiring continuous renal replacement therapy. *Nutr Clin Pract*. 2005;20:176–191.
52. Story DA, Ronco C, Bellomo R. Trace element and vitamin concentrations and losses in critically ill patients treated with continuous venovenous hemofiltration. *Crit Care Med*. 1999;27:220–223.
53. Berger MM, Shenkin A, Revelly JP, et al. Copper, selenium, zinc, and thiamine balances during continuous venovenous hemodiafiltration in critically ill patients. *Am J Clin Nutr*. 2004;80:410–416.
54. Abitbol CL, Rossique M, Rios M. Nutritional support of the pediatric patient with acute renal failure. In: American Society of Parenteral and Enteral Nutrition, ed. *The ASPEN Nutrition Support Practice Manual*. 2nd ed. Silver Spring, MD: American Society of Parenteral and Enteral Nutrition; 2005:287–295.
55. Druml W, Schwarzenhofer M, Apsner R, Horl WH. Fat-soluble vitamins in patients with acute renal failure. *Miner Electrolyte Metab*. 1998;24:220–226.
56. Major K, Lefor AT, Wilson M. Route of nutrition support. *Nutrition*. 2002;18:445–446.
57. de Lucas C, Moreno M, Lopez-Herce J, et al. Transpyloric enteral nutrition reduces the complication rate and cost in the critically ill child. *J Pediatr Gastroenterol Nutr*. 2000;30:175–180.
58. Moore FA, Feliciano DV, Andrassy RJ, et al. Early enteral feeding, compared with parenteral, reduces postoperative septic complications. The results of a meta-analysis. *Ann Surg*. 1992;216:172–183.
59. Kawagoe JY, Segre CA, Pereira CR, et al. Risk factors for nosocomial infections in critically ill newborns: a 5-year prospective cohort study. *Am J Infect Control*.

2001;29:109–114.
60. Chellis MJ, Sanders SV, Webster H, et al. Early enteral feeding in the pediatric intensive care unit. *JPEN J Parenter Enteral Nutr*. 1996;20:71–73.
61. Fiaccadori E, Maggiore U, Giacosa R, et al. Enteral nutrition in patients with acute renal failure. *Kidney Int*. 2004;65:999–1008.
62. Lopez-Herce J, Sanchez C, Carrillo A, et al. Transpyloric enteral nutrition in the critically ill child with renal failure. *Intensive Care Med*. 2006;32:1599–1605.
63. Sanchez C, Lopez-Herce J, Carrillo A, et al. Early transpyloric enteral nutrition in critically ill children. *Nutrition*. 2007;23:16–22.
64. North American Pediatric Renal Trials and Collaborative Studies. NAPRTCS 2007 Annual Report: Renal Transplantation, Dialysis, Chronic Renal Insufficiency. https://webemmescom/study/ped/annlrept/annlrept2007.pdf. Accessed October 14, 2013.
65. K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. *Am J Kidney Dis*. 2002;39:S1–S266.
66. Hogg RJ, Furth S, Lemley KV, et al. National Kidney Foundation's Kidney Disease Outcomes Quality Initiative clinical practice guidelines for chronic kidney disease in children and adolescents: evaluation, classification, and stratification. *Pediatrics*. 2003;111:1416–1421.
67. Rodig NM, McDermott KC, Schneider MF, Hotchkiss HM et al. Growth in children with chronic kidney disease: a report from the Chronic Kidney Disease in Children Study. *Pediatr Nephrol*. 2014;29:1987–1995.
68. Seikaly MG, Salhab N, Gipson D, et al. Stature in children with chronic kidney disease: analysis of NAPRTCS database. *Pediatr Nephrol*. 2006;21:793–799.
69. Karlberg J, Schaefer F, Hennicke M, et al. Early age-dependent growth impairment in chronic renal failure. European Study Group for Nutritional Treatment of Chronic Renal Failure in Childhood. *Pediatr Nephrol*. 1996;10:283–287.
70. Abitbol CL, Zilleruelo G, Montane B, Strauss J. Growth of uremic infants on forced feeding regimens. *Pediatr Nephrol*. 1993;7:173–177.
71. Rees L, Azocar M, Borzych D, et al. Growth in very young children undergoing chronic peritoneal dialysis. *J Am Soc Nephrol*. 2011;22:2303–2312.
72. Group KW. KDOQI Clinical Practice Guideline for Nutrition in Children with CKD: 2008 update. Executive summary. *Am J Kidney Dis*. 2009;53:S11–S104.
73. Filler G, Reimao SM, Kathiravelu A, et al. Pediatric nephrology patients are overweight: 20 years' experience in a single Canadian tertiary pediatric nephrology clinic. *Int Urol Nephrol*. 2007;39:1235–1240.
74. Roelfsema V, Clark RG. The growth hormone and insulin-like growth factor axis: its manipulation for the benefit of growth disorders in renal failure. *J Am Soc Nephrol*. 2001;12:1297–1306.
75. Fine RN, Attie KM, Kuntze J, et al. Recombinant human growth hormone in infants and young children with chronic renal insufficiency. Genentech Collaborative Study Group. *Pediatr Nephrol*. 1995;9:451–457.
76. Hokken-Koelega A, Mulder P, De Jong R, et al. Long-term effects of growth hormone treatment on growth and puberty in patients with chronic renal insufficiency. *Pediatr Nephrol*. 2000;14:701–706.
77. Haffner D, Schaefer F, Nissel R, et al. Effect of growth hormone treatment on the adult height of children with chronic renal failure. German Study Group for Growth Hormone Treatment in Chronic Renal Failure. *N Engl J Med*. 2000;343:923–930.
78. Mahan JD, Warady BA. Assessment and treatment of short stature in pediatric

patients with chronic kidney disease: a consensus statement. *Pediatr Nephrol*. 2006;21:917–930.
79. Furth SL, Abraham AG, Jerry-Fluker J, et al. Metabolic abnormalities, cardiovascular disease risk factors, and GFR decline in children with chronic kidney disease. *Clin J Am Soc Nephrol*. 2011;6:2132–2140.
80. Clinical practice guidelines for nutrition in chronic renal failure. K/DOQI, National Kidney Foundation. *Am J Kidney Dis*. 2000;35:S1–S140.
81. Locatelli F, Fouque D, Heimburger O, et al. Nutritional status in dialysis patients: a European consensus. *Nephrol Dial Transplant*. 2002;17:563–572.
82. Rashid R, Neill E, Smith W, et al. Body composition and nutritional intake in children with chronic kidney disease. *Pediatr Nephrol*. 2006;21:1730–1738.
83. Foster BJ, Leonard MB. Measuring nutritional status in children with chronic kidney disease. *Am J Clin Nutr*. 2004;80:801–814.
84. Gao T, Leonard MB, Zemel B, et al. Interpretation of body mass index in children with CKD. *Clin J Am Soc Nephrol*. 2012;7:558–564.
85. Juarez-Congelosi M, Orellana P, Goldstein SL. Normalized protein catabolic rate versus serum albumin as a nutrition status marker in pediatric patients receiving hemodialysis. *J Ren Nutr*. 2007;17:269–274.
86. Simmons JM, Wilson CJ, Potter DE, Holliday MA. Relation of calorie deficiency to growth failure in children on hemodialysis and the growth response to calorie supplementation. *N Engl J Med*. 1971;285:653–666.
87. Holliday MA. Calorie deficiency in children with uremia: effect upon growth. *Pediatrics*. 1972;50:590–597.
88. Wong CS, Gipson DS, Gillen DL, et al. Anthropometric measures and risk of death in children with end-stage renal disease. *Am J Kidney Dis*. 2000;36:811–819.
89. Furth SL, Hwang W, Yang C, et al. Growth failure, risk of hospitalization and death for children with end-stage renal disease. *Pediatr Nephrol*. 2002;17:450–455.
90. Norman LJ, Coleman JE, Macdonald IA, et al. Nutrition and growth in relation to severity of renal disease in children. *Pediatr Nephrol*. 2000;15:259–265.
91. Ratsch IM, Catassi C, Verrina E, et al. Energy and nutrient intake of patients with mild-to-moderate chronic renal failure compared with healthy children: an Italian multicentre study. *Eur J Pediatr*. 1992;151:701–705.
92. Orejas G, Santos F, Malaga S, et al. Nutritional status of children with moderate chronic renal failure. *Pediatr Nephrol*. 1995;9:52–56.
93. Foreman JW, Abitbol CL, Trachtman H, et al. Nutritional intake in children with renal insufficiency: a report of the growth failure in children with renal diseases study. *J Am Coll Nutr*. 1996;15:579–585.
94. Ruley EJ, Bock GH, Kerzner B, et al. Feeding disorders and gastroesophageal reflux in infants with chronic renal failure. *Pediatr Nephrol*. 1989;3:424–429.
95. Ravelli AM, Ledermann SE, Bisset WM, et al. Foregut motor function in chronic renal failure. *Arch Dis Child*. 1992;67:1343–1347.
96. Kalantar-Zadeh K, Block G, McAllister CJ, et al. Appetite and inflammation, nutrition, anemia, and clinical outcome in hemodialysis patients. *Am J Clin Nutr*. 2004;80:299–307.
97. Daschner M, Tonshoff B, Blum WF, et al. Inappropriate elevation of serum leptin levels in children with chronic renal failure. European Study Group for Nutritional Treatment of Chronic Renal Failure in Childhood. *J Am Soc Nephrol*. 1998;9:1074–1079.
98. Mak RH, Cheung W, Cone RD, Marks DL. Leptin and inflammation-associated

cachexia in chronic kidney disease. *Kidney Int*. 2006; 69:794–797.
99. Mak RH, Cheung WW, Zhan JY, et al. Cachexia and protein-energy wasting in children with chronic kidney disease. *Pediatr Nephrol*. 2012;27:173–181.
100. Ledermann SE, Shaw V, Trompeter RS. Long-term enteral nutrition in infants and young children with chronic renal failure. *Pediatr Nephrol*. 1999;13:870–875.
101. Kari JA, Gonzalez C, Ledermann SE, et al. Outcome and growth of infants with severe chronic renal failure. *Kidney Int*. 2000;57:1681–1687.
102. Ledermann SE, Scanes ME, Fernando ON, et al. Long-term outcome of peritoneal dialysis in infants. *J Pediatr*. 2000;136:24–29.
103. Claris-Appiani A, Ardissino GL, Dacco V, et al. Catch-up growth in children with chronic renal failure treated with long-term enteral nutrition. *JPEN J Parenter Enteral Nutr*. 1995;19:175–178.
104. Strife CF, Quinlan M, Mears K, et al. Improved growth of three uremic children by nocturnal nasogastric feedings. *Am J Dis Child*. 1986;140:438–443.
105. Brewer ED. Growth of small children managed with chronic peritoneal dialysis and nasogastric tube feedings: 203-month experience in 14 patients. *Adv Perit Dial*. 1990;6:269–272.
106. Reed EE, Roy LP, Gaskin KJ, Knight JF. Nutritional intervention and growth in children with chronic renal failure. *J Ren Nutr*. 1998;8:122–126.
107. Ramage IJ, Geary DF, Harvey E, et al. Efficacy of gastrostomy feeding in infants and older children receiving chronic peritoneal dialysis. *Perit Dial Int*. 1999;19:231–236.
108. Ellis EN, Yiu V, Harley F, et al. The impact of supplemental feeding in young children on dialysis: a report of the North American Pediatric Renal Transplant Cooperative Study. *Pediatr Nephrol*. 2001;16:404–408.
109. Dello Strologo L, Principato F, Sinibaldi D, et al. Feeding dysfunction in infants with severe chronic renal failure after long-term nasogastric tube feeding. *Pediatr Nephrol*. 1997;11:84–86.
110. Schauster H, Dwyer J. Transition from tube feedings to feedings by mouth in children: preventing eating dysfunction. *J Am Diet Assoc*. 1996;96:277–281.
111. Marques de Aquino T, Avesani CM, Brasileiro RS, de Abreu Carvalhaes JT. Resting energy expenditure of children and adolescents undergoing hemodialysis. *J Ren Nutr*. 2008;18:312–319.
112. Brady TM, Schneider MF, Flynn JT, et al. Carotid intima-media thickness in children with CKD: results from the CKiD study. *Clin J Am Soc Nephrol*. 2012;7:1930–1937.
113. Ikizler TA, Hakim RM. Nutrition in end-stage renal disease. *Kidney Int*. 1996;50:343–357.
114. Wolfson M. Nutritional management of the continuous ambulatory peritoneal dialysis patient. *Am J Kidney Dis*. 1996;27:744–749.
115. Salusky IB, Fine RN, Nelson P, et al. Nutritional status of children undergoing continuous ambulatory peritoneal dialysis. *Am J Clin Nutr*. 1983;38:599–611.
116. Krause I, Shamir R, Davidovits M, et al. Intradialytic parenteral nutrition in malnourished children treated with hemodialysis. *J Ren Nutr*. 2002;12:55–59.
117. Orellana P, Juarez-Congelosi M, Goldstein SL. Intradialytic parenteral nutrition treatment and biochemical marker assessment for malnutrition in adolescent maintenance hemodialysis patients. *J Ren Nutr*. 2005;15:312–317.
118. Motomura K, Okuda S, Sanai T, et al. Importance of early initiation of dietary protein restriction for the prevention of experimental progressive renal disease. *Nephron*. 1988;49:144–149.
119. Baudoin P, Provoost AP. Effects of age at the time of unilateral nephrectomy and

dietary protein on long-term renal function in rats. *Pediatr Nephrol*. 1993;7:536–542.
120. Klahr S, Levey AS, Beck GJ, et al. The effects of dietary protein restriction and blood-pressure control on the progression of chronic renal disease. Modification of Diet in Renal Disease Study Group. *N Engl J Med*. 1994;330:877–884.
121. D'Amico G, Gentile MG, Fellin G, et al. Effect of dietary protein restriction on the progression of renal failure: a prospective randomized trial. *Nephrol Dial Transplant*. 1994;9:1590–1594.
122. Rosman JB, Langer K, Brandl M, et al. Protein-restricted diets in chronic renal failure: a four year follow-up shows limited indications. *Kidney Int Suppl*. 1989;27:S96–S102.
123. Rosman JB, ter Wee PM, Meijer S, et al. Prospective randomised trial of early dietary protein restriction in chronic renal failure. *Lancet*. 1984;2:1291–1296.
124. Wingen AM, Fabian-Bach C, Schaefer F, Mehls O. Randomised multicentre study of a low-protein diet on the progression of chronic renal failure in children. European Study Group of Nutritional Treatment of Chronic Renal Failure in Childhood. *Lancet*. 1997;349:1117–1123.
125. Civilibal M, Caliskan S, Adaletli I, et al. Coronary artery calcifications in children with end-stage renal disease. *Pediatr Nephrol*. 2006;21:1426–1433.
126. Goodman WG, Goldin J, Kuizon BD, et al. Coronary-artery calcification in young adults with end-stage renal disease who are undergoing dialysis. *N Engl J Med*. 2000;342:1478–1483.
127. Block GA, Hulbert-Shearon TE, Levin NW, Port FK. Association of serum phosphorus and calcium × phosphate product with mortality risk in chronic hemodialysis patients: a national study. *Am J Kidney Dis*. 1998;31:607–617.
128. Krenitsky J. Adjusted body weight, pro: evidence to support the use of adjusted body weight in calculating calorie requirements. *Nutr Clin Pract*. 2005;20:468–473.
129. Kopanati S, Baum M, Quan A. Peritoneal protein losses in children with steroid-resistant nephrotic syndrome on continuous-cycler peritoneal dialysis. *Pediatr Nephrol*. 2006;21:1013–1019.
130. Kriley M, Warady BA. Vitamin status of pediatric patients receiving long-term peritoneal dialysis. *Am J Clin Nutr*. 1991;53:1476–1479.
131. Warady BA, Kriley M, Alon U, Hellerstein S. Vitamin status of infants receiving long-term peritoneal dialysis. *Pediatr Nephrol*. 1994;8:354–356.
132. Pereira AM, Hamani N, Nogueira PC, Carvalhaes JT. Oral vitamin intake in children receiving long-term dialysis. *J Ren Nutr*. 2000;10:24–29.
133. Doireau V, Macher MA, Brun P, et al. [Vitamin A poisoning revealed by hypercalcemia in a child with kidney failure]. *Arch Pediatr*. 1996;3:888–890.
134. Pru C, Eaton J, Kjellstrand C. Vitamin C intoxication and hyperoxalemia in chronic hemodialysis patients. *Nephron*. 1985;39:112–116.
135. Saito H, Kusano K, Kinosaki M, et al. Human fibroblast growth factor-23 mutants suppress Na+-dependent phosphate co-transport activity and 1alpha,25-dihydroxyvitamin D3 production. *J Biol Chem*. 2003;278:2206–2211.
136. Norman ME, Mazur AT, Borden St, et al. Early diagnosis of juvenile renal osteodystrophy. *J Pediatr*. 1980;97:226–232.
137. Seeherunvong W, Abitbol CL, Chandar J, et al. Vitamin D insufficiency and deficiency in children with early chronic kidney disease. *J Pediatr*. 2009;154:906–911 e1.
138. Ali FN, Arguelles LM, Langman CB, Price HE. Vitamin D deficiency in children with chronic kidney disease: uncovering an epidemic. *Pediatrics*. 2009;123:791–

796.
139. Uhlig K, Berns JS, Kestenbaum B, et al. KDOQI US commentary on the 2009 KDIGO Clinical Practice Guideline for the Diagnosis, Evaluation, and Treatment of CKD-Mineral and Bone Disorder (CKD-MBD). *Am J Kidney Dis*. 2010;55:773–799.
140. Menon S, Valentini RP, Hidalgo G, et al. Vitamin D insufficiency and hyperparathyroidism in children with chronic kidney disease. *Pediatr Nephrol*. 2008;23:1831–1836.
141. National Kidney Foundation. K/DOQI clinical practice guidelines for bone metabolism and disease in children with chronic kidney disease. *Am J Kidney Dis*. 2005;46:S1–S122.
142. Salusky IB. Are new vitamin D analogues in renal bone disease superior to calcitriol? *Pediatr Nephrol*. 2005;20:393–398.
143. Slatopolsky E, Dusso A, Brown A. New analogs of vitamin D3. *Kidney Int Suppl*. 1999;73:S46–S51.
144. Greenbaum LA, Benador N, Goldstein SL, et al. Intravenous paricalcitol for treatment of secondary hyperparathyroidism in children on hemodialysis. *Am J Kidney Dis*. 2007;49:814–823.
145. Seeherunvong W, Nwobi O, Abitbol CL, et al. Paricalcitol versus calcitriol treatment for hyperparathyroidism in pediatric hemodialysis patients. *Pediatr Nephrol*. 2006;21:1434–1439.
146. Erslev AJ. Erythropoietin. *N Engl J Med*. 1991;324:1339–1344.
147. Ulinski T, Cochat P. Longitudinal growth in children following kidney transplantation: from conservative to pharmacological strategies. *Pediatr Nephrol*. 2006;21:903–909.
148. Chauveau P, Chadefaux B, Coude M, et al. Hyperhomocysteinemia, a risk factor for atherosclerosis in chronic uremic patients. *Kidney Int Suppl*. 1993;41:S72–S77.
149. Robinson K, Gupta A, Dennis V, et al. Hyperhomocysteinemia confers an independent increased risk of atherosclerosis in end-stage renal disease and is closely linked to plasma folate and pyridoxine concentrations. *Circulation*. 1996;94:2743–2748.
150. Poyrazoglu HM, Dusunsel R, Yikilmaz A, et al. Carotid artery thickness in children and young adults with end stage renal disease. *Pediatr Nephrol*. 2007;22:109–116.
151. Merouani A, Lambert M, Delvin EE, et al. Plasma homocysteine concentration in children with chronic renal failure. *Pediatr Nephrol*. 2001;16:805–811.
152. Schroder CH, de Boer AW, Giesen AM, et al. Treatment of hyperhomocysteinemia in children on dialysis by folic acid. *Pediatr Nephrol*. 1999;13:583–585.
153. Martin KJ, Gonzalez EA. Metabolic bone disease in chronic kidney disease. *J Am Soc Nephrol*. 2007;18:875–885.
154. Cannata-Andia JB, Rodriguez-Garcia M, Roman-Garcia P, et al. New therapies: calcimimetics, phosphate binders and vitamin D receptor activators. *Pediatr Nephrol*. 2010;25:609–616.
155. Salusky IB, Foley J, Nelson P, Goodman WG. Aluminum accumulation during treatment with aluminum hydroxide and dialysis in children and young adults with chronic renal disease. *N Engl J Med*. 1991;324:527–531.
156. Sedman AB, Miller NL, Warady BA, et al. Aluminum loading in children with chronic renal failure. *Kidney Int*. 1984;26:201–204.
157. Sedman AB, Wilkening GN, Warady BA, et al. Encephalopathy in childhood

secondary to aluminum toxicity. *J Pediatr*. 1984;105:836–838.
158. Salusky IB, Goodman WG, Sahney S, et al. Sevelamer controls parathyroid hormone-induced bone disease as efficiently as calcium carbonate without increasing serum calcium levels during therapy with active vitamin D sterols. *J Am Soc Nephrol*. 2005;16:2501–2508.
159. Gulati A, Sridhar V, Bose T, et al. Short-term efficacy of sevelamer versus calcium acetate in patients with chronic kidney disease stage 3-4. *Int Urol Nephrol*. 2010;42:1055–1062.
160. Ramos R, Moreso F, Borras M, et al. Sevelamer hydrochloride in peritoneal dialysis patients: results of a multicenter cross-sectional study. *Perit Dial Int*. 2007;27:697–701.
161. Thompson K, Flynn J, Okamura D, Zhou L. Pretreatment of formula or expressed breast milk with sodium polystyrene sulfonate (kayexalate((R))) as a treatment for hyperkalemia in infants with acute or chronic renal insufficiency. *J Ren Nutr*. 2013;23:333–339.
162. Bunchman TE, Wood EG, Schenck MH, et al. Pretreatment of formula with sodium polystyrene sulfonate to reduce dietary potassium intake. *Pediatr Nephrol*. 1991;5:29–32.
163. Rivard AL, Raup SM, Beilman GJ. Sodium polystyrene sulfonate used to reduce the potassium content of a high-protein enteral formula: a quantitative analysis. *JPEN J Parenter Enteral Nutr*. 2004;28:76–78.
164. Bunchman TE. Chronic dialysis in the infant less than 1 year of age. *Pediatr Nephrol*. 1995;9 Suppl:S18–S22.
165. Parekh RS, Flynn JT, Smoyer WE, et al. Improved growth in young children with severe chronic renal insufficiency who use specified nutritional therapy. *J Am Soc Nephrol*. 2001;12:2418–2426.
166. McDonald SP, Craig JC, Australian, New Zealand Paediatric Nephrology A. Long-term survival of children with end-stage renal disease. *N Engl J Med*. 2004;350:2654–2662.
167. North American Pediatric Renal Trials and Collaborative Studies. NAPRTCS 2010 Annual Transplant Report. https://webemmescom/study/ped/annlrept/2010_Report.pdf. Accessed October 14, 2013.
168. Tejani A, Fine R, Alexander S, et al. Factors predictive of sustained growth in children after renal transplantation. The North American Pediatric Renal Transplant Cooperative Study. *J Pediatr*. 1993;122:397–402.
169. Fine RN, Ho M, Tejani A. The contribution of renal transplantation to final adult height: a report of the North American Pediatric Renal Transplant Cooperative Study (NAPRTCS). *Pediatr Nephrol*. 2001;16:951–956.
170. Harmon WE, Jabs K. Factors affecting growth after renal transplantation. *J Am Soc Nephrol*. 1992;2:S295–S303.
171. Sarwal MM, Ettenger RB, Dharnidharka V, et al. Complete steroid avoidance is effective and safe in children with renal transplants: a multicenter randomized trial with three-year follow-up. *Am J Transplant*. 2012;12:2719–2729.
172. Jung HW, Kim HY, Lee YA, et al. Factors affecting growth and final adult height after pediatric renal transplantation. *Transplant Proc*. 2013;45:108–114.
173. Vidhun JR, Sarwal MM. Corticosteroid avoidance in pediatric renal transplantation. *Pediatr Nephrol*. 2005;20:418–426.
174. Sarwal MM, Vidhun JR, Alexander SR, et al. Continued superior outcomes with modification and lengthened follow-up of a steroid-avoidance pilot with extended daclizumab induction in pediatric renal transplantation. *Transplantation*.

2003;76:1331–1339.
175. Grenda R, Watson A, Trompeter R, et al. A randomized trial to assess the impact of early steroid withdrawal on growth in pediatric renal transplantation: the TWIST study. *Am J Transplant*. 2010;10:828–836.
176. Jabs K, Van Dop C, Harmon WE. Growth hormone treatment of growth failure among children with renal transplants. *Kidney Int Suppl*. 1993;43:S71–S75.
177. Fine RN, Stablein D. Long-term use of recombinant human growth hormone in pediatric allograft recipients: a report of the NAPRTCS Transplant Registry. *Pediatr Nephrol*. 2005;20:404–408.
178. Mehls O, Fine RN. Growth hormone treatment after renal transplantation: a promising but underused chance to improve growth. *Pediatr Nephrol*. 2013;28:1–4.
179. Hanevold CD, Ho PL, Talley L, Mitsnefes MM. Obesity and renal transplant outcome: a report of the North American Pediatric Renal Transplant Cooperative Study. *Pediatrics*. 2005;115:352–356.
180. Meier-Kriesche HU, Arndorfer JA, Kaplan B. The impact of body mass index on renal transplant outcomes: a significant independent risk factor for graft failure and patient death. *Transplantation*. 2002;73:70–74.
181. Mitsnefes MM, Khoury P, McEnery PT. Body mass index and allograft function in pediatric renal transplantation. *Pediatr Nephrol*. 2002;17:535–539.
182. Boschetti SB, Nogueira PC, Pereira AM, et al. Prevalence, risk factors, and consequences of overweight in children and adolescents who underwent renal transplantation—short- and medium-term analysis. *Pediatr Transplant*. 2013;17:41–47.
183. Denburg MR, Pradhan M, Shults J, et al. Longitudinal relations between obesity and hypertension following pediatric renal transplantation. *Pediatr Nephrol*. 2010;25:2129–2139.
184. Ong CS, Pollock CA, Caterson RJ, et al. Hyperlipidemia in renal transplant recipients: natural history and response to treatment. *Medicine (Baltimore)*. 1994;73:215–223.
185. Satterthwaite R, Aswad S, Sunga V, et al. Incidence of new-onset hypercholesterolemia in renal transplant patients treated with FK506 or cyclosporine. *Transplantation*. 1998;65:446–449.
186. Kasiske BL. Risk factors for accelerated atherosclerosis in renal transplant recipients. *Am J Med*. 1988;84:985–992.
187. Becker-Cohen R, Nir A, Rinat C, et al. Risk factors for cardiovascular disease in children and young adults after renal transplantation. *Clin J Am Soc Nephrol*. 2006;1:1284–1292.
188. Silverstein DM, Palmer J, Polinsky MS, et al. Risk factors for hyperlipidemia in long-term pediatric renal transplant recipients. *Pediatr Nephrol*. 2000;14:105–110.
189. Lau KK, Tancredi DJ, Perez RV, Butani L. Unusual pattern of dyslipidemia in children receiving steroid minimization immunosuppression after renal transplantation. *Clin J Am Soc Nephrol*. 2010;5:1506–1512.
190. Silverstein DM. Risk factors for cardiovascular disease in pediatric renal transplant recipients. *Pediatr Transplant*. 2004;8:386–393.
191. Parekh RS, Carroll CE, Wolfe RA, Port FK. Cardiovascular mortality in children and young adults with end-stage kidney disease. *J Pediatr*. 2002;141:191–197.
192. Roodnat JI, Mulder PG, Zietse R, et al. Cholesterol as an independent predictor of outcome after renal transplantation. *Transplantation*. 2000;69:1704–1710.
193. Massy ZA, Guijarro C, Wiederkehr MR, et al. Chronic renal allograft rejection: immunologic and nonimmunologic risk factors. *Kidney Int*. 1996;49:518–524.

194. Lowry RP, Soltys G, Mangel R, et al. Type II hyperlipoproteinemia, hyperapobetalipoproteinemia, and hyperalphalipoproteinemia following renal transplantation: prevalence and precipitating factors. *Transplant Proc*. 1987;19:2229–2232.
195. Vathsala A, Weinberg RB, Schoenberg L, et al. Lipid abnormalities in cyclosporine-prednisone-treated renal transplant recipients. *Transplantation*. 1989;48:37–43.
196. Kuster GM, Drexel H, Bleisch JA, et al. Relation of cyclosporine blood levels to adverse effects on lipoproteins. *Transplantation*. 1994;57:1479–1483.
197. Markell MS, Armenti V, Danovitch G, Sumrani N. Hyperlipidemia and glucose intolerance in the post-renal transplant patient. *J Am Soc Nephrol*. 1994;4:S37–S47.
198. Legendre C, Campistol JM, Squifflet JP, Burke JT, Sirolimus European Renal Transplant Study G. Cardiovascular risk factors of sirolimus compared with cyclosporine: early experience from two randomized trials in renal transplantation. *Transplant Proc*. 2003;35:151S–153S.
199. Hocker B, Feneberg R, Kopf S, et al. SRL-based immunosuppression vs. CNI minimization in pediatric renal transplant recipients with chronic CNI nephrotoxicity. *Pediatr Transplant*. 2006;10:593–601.
200. Pirsch JD, Miller J, Deierhoi MH, et al. A comparison of tacrolimus (FK506) and cyclosporine for immunosuppression after cadaveric renal transplantation. FK506 Kidney Transplant Study Group. *Transplantation*. 1997;63:977–983.
201. Ferraris JR, Ghezzi L, Waisman G, Krmar RT. Potential cardiovascular risk factors in paediatric renal transplant recipients. *Pediatr Nephrol*. 2006;21:119–125.
202. Milliner DS, Morgenstern BZ, Murphy M, et al. Lipid levels following renal transplantation in pediatric recipients. *Transplant Proc*. 1994;26:112–114.
203. Siirtola A, Virtanen SM, Ala-Houhala M, et al. Diet does not explain the high prevalence of dyslipidaemia in paediatric renal transplant recipients. *Pediatr Nephrol*. 2008;23:297–305.
204. Trompeter R, Filler G, Webb NJ, et al. Randomized trial of tacrolimus versus cyclosporin microemulsion in renal transplantation. *Pediatr Nephrol*. 2002;17:141–149.
205. Greenspan LC, Gitelman SE, Leung MA, et al. Increased incidence in post-transplant diabetes mellitus in children: a case-control analysis. *Pediatr Nephrol*. 2002;17:1–5.
206. El-Husseini AA, Foda MA, Osman YM, Sobh MA. Characteristics of long-term live-donor pediatric renal transplant survivors: a single-center experience. *Pediatr Transplant*. 2006;10:288–293.
207. Ruiz JO, Simmons RL, Callender CO, et al. Steroid diabetes in renal transplant recipients: pathogenetic factors and prognosis. *Surgery*. 1973;73:759–765.
208. Nielsen JH, Mandrup-Poulsen T, Nerup J. Direct effects of cyclosporin A on human pancreatic beta-cells. *Diabetes*. 1986;35:1049–1052.
209. Carroll PB, Boschero AC, Li MY, et al. Effect of the immunosuppressant FK506 on glucose-induced insulin secretion from adult rat islets of Langerhans. *Transplantation*. 1991;51:275–278.
210. Al-Uzri A, Stablein DM, R AC. Posttransplant diabetes mellitus in pediatric renal transplant recipients: a report of the North American Pediatric Renal Transplant Cooperative Study (NAPRTCS). *Transplantation*. 2001;72:1020–1024.
211. Burroughs TE, Swindle JP, Salvalaggio PR, et al. Increasing incidence of new-onset diabetes after transplant among pediatric renal transplant patients.

Transplantation. 2009;88:367–373.
212. Delucchi A, Marin V, Trabucco G, et al. Dyslipidemia and dietary modification in Chilean renal pediatric transplantation. *Transplant Proc*. 2001;33:1297–1301.
213. Butani L, Pai MV, Makker SP. Pilot study describing the use of pravastatin in pediatric renal transplant recipients. *Pediatr Transplant*. 2003;7:179–184.
214. Butani L. Prospective monitoring of lipid profiles in children receiving pravastatin preemptively after renal transplantation. *Pediatr Transplant*. 2005;9:746–753.
215. Krmar RT, Ferraris JR, Ramirez JA, et al. Use of atorvastatin in hyperlipidemic hypertensive renal transplant recipients. *Pediatr Nephrol*. 2002;17:540–543.
216. Argent E, Kainer G, Aitken M, et al. Atorvastatin treatment for hyperlipidemia in pediatric renal transplant recipients. *Pediatr Transplant*. 2003;7:38–42.

Inflammatory Bowel Disease

52

Francesca Penagini, MD
Rob Heuschkel, MBBS, MRCPCH

INTRODUCTION

From the earliest clinical descriptions of children with Crohn's disease (CD) it was clear that significant undernutrition is a major feature of the disease. Furthermore growth retardation, which may have a nutritional component, is a common clinical manifestation. While on occasion undernutrition and short stature may be a feature of chronic ulcerative colitis (UC) in children, these manifestations are less prominent in this disease.

Early reports of nutritional therapy in children with inflammatory bowel disease were thus aimed at the restoration of normal nutrition. More recently a body of evidence has shown that in children with CD, nutritional therapy has a specific anti-inflammatory action in addition to its ability to restore their nutritional status to normal. Unfortunately there is no such firm evidence in children with UC, yet here also the provision of adequate nutrition still remains important.

The value of nutritional therapy in CD chiefly centers on the use of exclusive enteral nutrition (EEN) in the treatment of active disease. Enteral nutrition may be defined as the provision of a liquid formula diet by mouth or by tube into the gastrointestinal tract.[1] Bottle feeding of normal infants is excluded from this definition. Although it is now clear that oral feeding may be highly effective, research began with liquid formulae administered by intragastric tubes. The technique of tube feeding goes back at least as far as 1872, with a report by Clouston in the *Lancet* on "Forcible Feeding,"[2] whereas the work of Elsie Widowson (1947) drew attention to the adverse effect of malnutrition on human disease in general.[3] The advent of effective parenteral nutrition then made clinicians aware how

dramatic the benefits of nutritional rehabilitation from malnutrition could be. The development of a range of complete formula diets of varying compositions, designed for a range of situations, has now made enteral nutrition a highly desirable and practical alternative to parenteral nutrition.

It was Giorgini et al. in 1973 who first suggested that treatment of malnutrition could lead to resolution of inflammation in a child with CD.[4] They described resolution of terminal ileal inflammation on barium follow-through after a period of elemental diet. In adults with CD, Logan et al. produced evidence that protein and lymphocyte loss from the gut decreased during feeding with an elemental diet.[5] Then in Quebec, Morin et al. described four children with Crohn's whose growth failure responded to an elemental diet and this was confirmed in larger studies.[6,7] In France, an elemental diet was given to children with CD by continuous nasogastric infusion. This was called constant rate enteral nutrition (CREN) but was a cumbersome technique not popular with the children. They used a complex regimen using oligopeptides, whereas others reported the use of a high osmolality amino acid–based elemental formula. At St. Bartholomew's Hospital in London, a semi-elemental diet using extensively hydrolyzed milk protein given intragastrically was shown in a small study to be as effective as conventional steroids in the induction of a clinical remission.[8] Most importantly though this study showed there was no short-term growth while on steroid therapy, whereas enteral nutrition lead to short-term growth acceleration. In subsequent follow-up studies it was shown that this could on occasion be sustained for some time if the child remained in remission even when back on a normal diet after the period of nutritional therapy.[9] Central to this approach to therapy was the concept of a period of six to eight weeks of enteral nutrition followed by a graded return to a normal diet.

ROLE OF DIET IN ETIOLOGY OF IBD

The etiology of IBD remains one of the greatest challenges in gastroenterology today. The presence of unexplained gut inflammation has inevitably led researchers to try and identify factors in our diet that may trigger, or even cause, IBD. Although it appears highly likely that there are several environmental factors involved in the etiopathogenesis of IBD,[10,11] the evidence that some of these may be dietary remains tempting. Although many studies claim that the intake of certain food components is associated with IBD, the specific effect of each component remains

questionable due to methodological limitations and numerous confounding variables. A recent meta-analysis confirmed that a high dietary intake of fat, polyunsaturated fatty acids (PUFAs), omega-6 fatty acids, and meat increased the risk of developing both CD and UC. The risk of developing CD appears reduced with high intake of fruit and dietary fiber, whereas a high vegetable intake decreased the risk of developing UC.[12] Whether nutrition has a direct effect on disease pathogenesis, or whether these effects are indirect via modulation of the intestinal microbiota, or both remains debated.[13]

It was initially suggested that the adoption of an urban lifestyle may lead to an increase in the incidence of IBD, as increases occurring much more rapidly than could be explained by genetic factors alone.[14,15]

Even though positive associations between high *protein* intake and IBD risk have been repeatedly reported in epidemiological studies, neither consistent data nor biologically plausible mechanisms are available. A possible exception is the consumption of animal protein, which might contribute to the etiology of IBD through its byproducts, which can be metabolized by the microbiota to substances that negatively affect colonic health. A recent prospective study conducted by Jantchou et al. demonstrated that a high protein intake, specifically animal protein, was associated with risk of IBD.[16]

With regards to *carbohydrates*, studies report conflicting outcomes.[12] Sugar was the first food component to be implicated in the development of CD in 1976,[17] yet its contribution to the pathophysiology of CD remains to be proven. A case control study conducted by Russel et al. demonstrated that patients with a recent diagnosis of CD were shown to consume greater quantities of sucrose than control groups,[18] although this was not confirmed in patients with UC.[19]

Recently, a large prospective cohort study (EPIC) found no association between UC and intake of sugar at baseline.[20] It has been hypothesized that excessive intake of highly fermentable but poorly absorbed short-chain carbohydrates and polyols may lead to bacterial overgrowth and next to increased intestinal permeability, which may confer risk of CD in genetically susceptible subjects.[21]

The positive effect attributed to dietary *fiber* has clear underlying mechanisms. Fermentable fiber can be metabolized by the microbiota to short-chain fatty acids; acetate, butyrate, and propionate. Butyrate has potentially important functions in the intestine as a source of energy for enterocytes and as anti-inflammatory mediator. Despite these potential

beneficial effects, which have been demonstrated in vitro, the results of retrospective studies on the relation between consumption of fiber and risk of IBD have been conflicting. High-fiber diet has been found to attenuate experimental colitis in animal models[22] and showed benefit in active IBD.[23] However, a large prospective cohort study (EPIC) found no protective effect against UC of a high intake of dietary fibers.[20]

The composition of the gut *microbiota* is thought to be a critical environmental factor in IBD, and recent evidence suggests a connection between diet and the intestinal bacteria.[13,24] Although it makes biological sense that different dietary regimens may affect IBD risk through affecting the composition of the gut microbiome, data are limited because this endpoint has not usually been included in human studies to date. In animal models, manipulation of both the type and level of both fat and carbohydrates have led to dramatic effects on the gut microbiota.[25] In humans, Werner[26] showed that depletion of luminal iron alters the gut microbiota and prevents some CD-like symptoms.

Patients with a recent diagnosis of CD have been shown to consume greater quantities of sucrose than control groups,[18] although this was not confirmed in patients with UC.[19] Whether or not a diet low in fiber leads to an increased risk of CD remains disputed.[27] Many of these studies have had methodological problems and as a result make it difficult to confirm that dietary differences antedated the development of disease. Reif et al.[28] however, again showed that a high level of sucrose intake (>92 g/day) occurred before symptom onset in patients with both UC and CD (OR 5.3 and 2.8, respectively), in contrast fructose and fiber consumption were both negatively associated with a risk for IBD. This carefully controlled study also reported an increase in fat consumption prior to a diagnosis of IBD. Omega-6 fatty acids such as linoleic acid can be metabolized to arachidonic acid (AA), which serves as a precursor for a family of proinflammatory signaling molecules, the so-called eicosanoids. In contrast, omega-3 fatty acids, particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) antagonize the formation of eicosanoid mediators from AA, suppressing inflammatory cytokines. Metabolites of AA are not only elevated in the mucosa of patients with UC,[29] but a prospective cohort study recently suggested that individuals with the highest relative concentrations of AA in adipose tissue, reflecting dietary intake, have a significantly increased risk of developing UC.[30] These studies concluded that up to 30% of adults with UC had a linoleic acid intake above the lowest quartile, with obvious implications for intervention

and potential symptom reduction.[29,30] These findings were mirrored by Geerling et al. who reported an increased risk of developing UC in those with a high consumption of monounsaturated and polyunsaturated (mainly n-6 PUFA) fats as well as vitamin B6.[31]

However overall, these findings suggest no more than that a low residue, highly refined diet is associated with the development of IBD, whereas a high fiber diet, rich in fruit and vegetables, and low in linoleic acid may be more protective. Large, prospective epidemiological studies will be necessary to adequately control for the many confounding variables that could affect this type of data.

Conflicting evidence is also available on the role of butyrate in the pathogenesis of UC. Data suggest that butyrate levels decrease in the distal colon,[32] with the ability of colonic mucosa to utilize butyrate being reduced even in macroscopically "normal" mucosa of patients with quiescent UC.[33] However, the response of distal UC and so-called diversion colitis to butyrate enemas has not been striking.[34,35] Dietary sulfates have been implicated in reducing the ability of colonocytes to utilize butyrate by their conversion to toxic sulfides like H_2S,[36] although luminal levels of sulfides do not appear to be significantly higher in patients with UC than in normal controls.[37] However, along with a putative role of oxidant injury in the pathogenesis of UC, all these mechanisms appear more likely to be involved as mediators of the injury in UC, rather than primary etiological factors.

It is also of note that researchers have been accumulating evidence over a number of years implicating the food borne pathogen *Mycobacterium avium* subsp. *paratuberculosis* in the pathogenesis of CD. Although detailed discussion of this is beyond the scope of this chapter, the presence of this organism in the food chain and its persistence in milk despite pasteurization, along with the characteristic Th1 immune response it evokes continues to hold interest.[38] However, well-controlled clinical trials have failed to show any benefit of antituberculous therapy.[39] Larger, population-based cohort studies are still needed to better define the importance of this and other specific dietary risk factors.

NUTRITIONAL COMPLICATIONS OF IBD

The chronic and relapsing nature of intestinal inflammation inevitably impacts on a patient's nutritional status. However, their nutritional state is not only determined by the dietary intake, but also by the patient's age,

type of disease, disease distribution as well as their disease activity. These factors all contribute to the wide range of nutritional complications seen; from almost none in a child with mild distal UC, to the most severe nutritional consequences of chronically active disease in a child with pan-enteric CD. Nutritional complications are rarer in children with UC, although a severe pancolitis may present with acute weight loss and significant iron-deficiency anemia.

Undernutrition in IBD may present in its different forms, most commonly as altered body composition, micronutrient deficiencies, and poor bone health; protein-energy malnutrition is exceedingly rare. Additionally, in pediatric IBD patients, growth failure and pubertal delay are associated with poor nutritional status. There are several factors that perturb the nutritional status in IBD patients. In particular, suboptimal intake, as a result of anorexia and food aversions, in combination with increased energy and nutrient losses, as a result of malabsorption and gut losses, can disturb the nutrient equilibrium and increase risk of undernutrition.[40] Moreover increased nutrient utilization and altered metabolism, as a result of activation of the immune system, tissue repair, and drug–nutrient interactions, may increase nutritional requirements well above those established for the general population (Figure 52-1).[41]

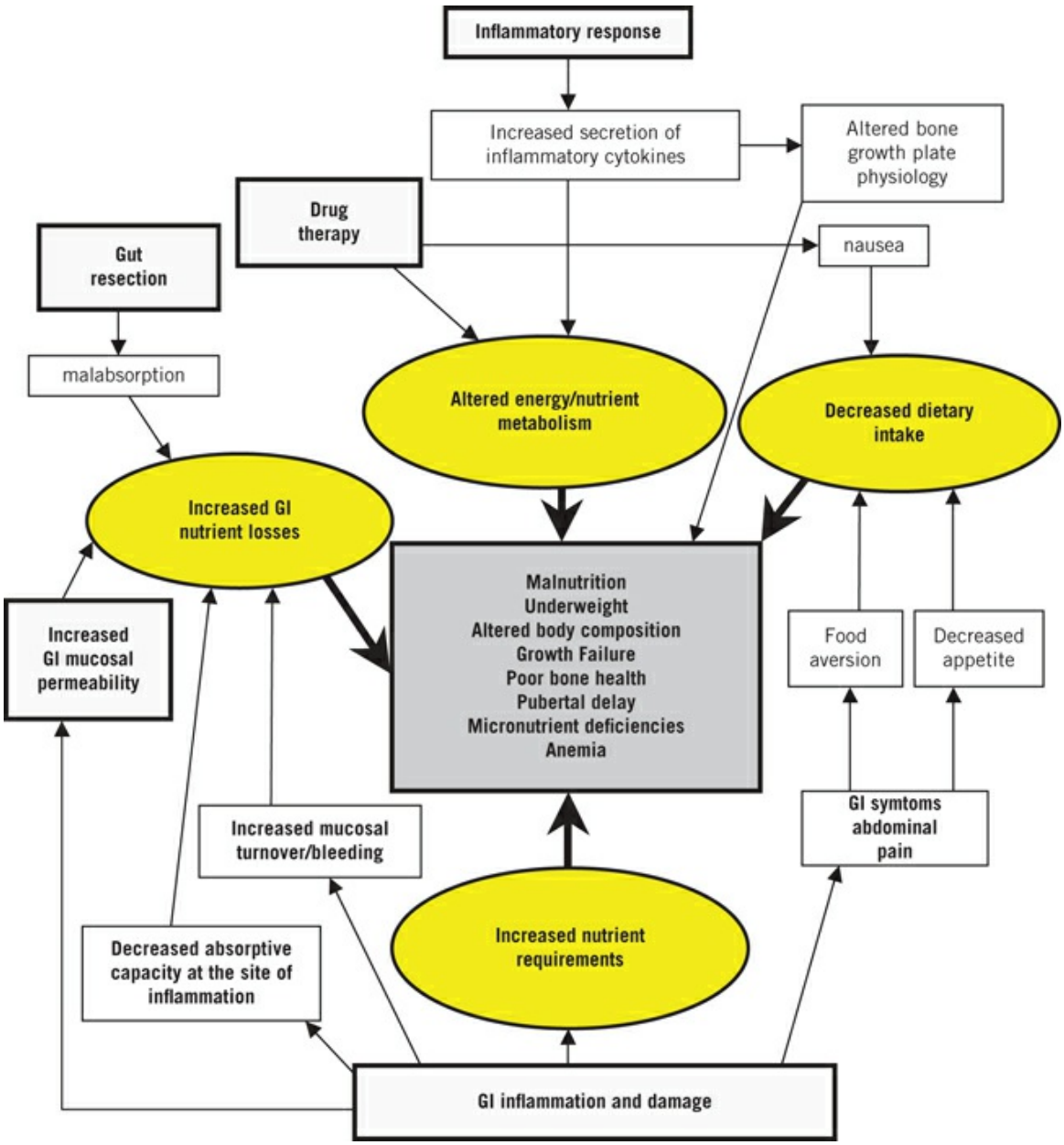


Figure 52-1 Mechanisms, inter-dependencies and key factors leading to nutritional compromise in patients with active Crohn's disease.

Caloric Deficiency and Growth Failure

The mechanisms surrounding the anorexia and undernutrition seen in IBD are complex and still poorly understood. As well as food avoidance as a result of abdominal pain or diarrhea, the reduction in caloric intake is due to the consequences of the systemic inflammatory response.[42,43]

Protein-energy malnutrition may occur as a result of unchecked disease

activity, with the associated anorexia being largely mediated by the proinflammatory cytokines IL-1 and TNF.[44] Activity of these cytokines leads to lean tissue loss,[45] reduced voluntary motor activity,[46] and protein synthesis, with a concomitant increase in protein catabolism.[47]

This state of increased need may not be met during active inflammation, and thus leads to chronic undernutrition. Winter et al. elegantly demonstrated the recovery of impaired pancreatic and gastric secretory activity with increasing body mass index (BMI) in severely malnourished (BMI ~14) adults with CD.[48] Many patients with IBD present features of protein energy undernutrition at the time of diagnosis, and this status fluctuates during the disease course. A history of weight loss is a common presenting feature of the newly diagnosed patients and accompanies almost every relapse, this was seen in approximately 60% of newly diagnosed CD children compared to 35% in UC patients.[49]

Protein intakes up to 1.5 g/kg body weight may be required per day to achieve adequate nutrition, with excessive supplementation perhaps even being counterproductive during the acute inflammatory state.[50] Increased protein oxidation and worsening of the nitrogen balance may result if overfeeding occurs during inflammation.[51] Children with IBD have abnormal anthropometry, whereas particularly those with CD present with low BMI, are underweight and thin compared to the national reference range or their healthy peer.[52]

Calorie intake in children with CD cannot be accurately determined by using estimates of average requirements (EARs), as four in five children will consume about 120% of EAR (range 100%–149%) while taking EEN as primary therapy at diagnosis.[53] Only the initial BMI was a predictor of subsequent weight gain on EEN, with a median of 11% being achieved over an eight-week course of treatment. Other authors have reported the intake in active disease to be 43%–82% of recommended values, yet with adequate calorie supplementation, improvements in linear growth can be achieved.[51,53–56] However, simple nutritional restitution may be insufficient to achieve adequate growth, suggesting that other factors are also important in mediating growth retardation.[57] An elegant study by Azcue et al.[58] demonstrated an inappropriately maintained resting energy expenditure (REE) in children with moderately active CD when compared to children with anorexia nervosa and similar degrees of malnutrition. This relative increase in energy expenditure, together with the voluntary reduction in intake further compromises nutritional status and hence optimal growth. Careful animal experiments, which did not look at body

composition, suggest that it is the reduced caloric intake that is largely responsible for the weight loss seen in active colitis. However, in this animal study, while able to restore calorie intake and body weight to normal, nutritional supplementation was unable to restore linear growth. At least in this animal model, 30%–40% of growth impairment is likely to be a direct result of the inflammatory process.[59]

Particularly in children with CD, active disease can have a profound and lasting effect on long-term growth,[60] and this is covered in greater detail later in the chapter. Although it is well documented that up to 90% of children with CD may present with impaired height velocity,[61] Hildebrand et al. also reported a subnormal growth velocity (<–2 SDS) in 24% of prepubertal children with UC, compared to 40% in those with CD.[62,60]

Body Composition

There are several reasons why body composition in IBD patients may differ from that of healthy individuals. The secretion of proinflammatory cytokines may alter energy metabolism, protein turnover, and energy substrate utilization, whereas the use of corticosteroids may increase body fat and have catabolic effects on lean mass (LM). There are few studies that have assessed body composition in children with IBD. LM has been consistently reported as being significantly lower in inflammatory bowel disease (IBD) patients compared to healthy control groups.[63] In a cross-sectional study,[64] it was found that children with longstanding CD had a 6% deficit in LM compared to healthy children after accounting for stature, age, pubertal stage, and race; however, fat mass (FM) was normal, suggesting nutritional cachexia. Thayu et al.[63] in a well-designed study of newly diagnosed children with CD, also reported gender associated differences in body composition. FM and LM relative to height (adjusted for age, race, and pubertal stage) were lower in female than in male patients. Compared to a cohort of healthy controls, body composition in girls was more consistent with wasting (low LM and FM), whereas in boys there was mostly preservation of FM and deficit in LM consistent with cachexia.

Osteoporosis

Owing to the fact that this complication is largely a problem of adult life, it is easy to forget that the accumulation of peak bone mass (PBM) occurs during adolescence and early adult life. The implications of a substantial

interruption in this process are only now becoming clearer as better long-term bone density data are becoming available. When disease activity is high, nutrient requirements increase as a result of malabsorption, enteric losses, and the metabolic cost of inflammation. Together these complications of inflammatory bowel disease also affect bone health by impairing normal bone mineral accrual, leading to reduced PBM, and in some cases, increased fracture risk due to early onset osteoporosis.

Much attention has focused on trying to tease out the relative importance of the factors that lead to reduction in bone mineral density (BMD) in patients with IBD. Inflammation with increased production of cytokines, growth failure, delayed puberty, malabsorption, decreased physical activity, and the use of glucocorticoids have been recognized as risk factors for suboptimal bone health in pediatric patients with CD or UC.[65] As the most important determinants of PBM appear to be age and genetic potential, these confound almost all studies in IBD trying to identify possible predictors such as nutritional status, disease duration, activity and distribution, and steroid therapy. Despite these shortcomings, poor bone health and reduced BMD has been commonly reported in adults with CD and is increasingly reported in children and young adult populations, both at diagnosis and over subsequent years. In a population-based study conducted by Schmidt et al.[66] 144 children and adolescents affected by IBD underwent a dual-energy X-ray absorptiometry of the whole body and spine. The entire IBD group showed significantly lower BMD Z-scores of the lumbar spine (L2–L4) in comparison to healthy controls. Low BMD with a Z-score ≤ –2 SD was present in 26.7% of the patients with CD and in 24.1% of the UC patients.[67] Despite this interpretation of BMD in the pediatric IBD population remains a problem given the significant pubertal and bone age delay, bone health remaining more complex than bone density alone. Bone mineral accrual during growth is highly related to muscle development, particularly during adolescence.[68,69]

In adults, it appears that up to 40% with established IBD have osteopenia (T ≤ –1.0; –1 to –2.5 SD from the mean), whereas around 10% have osteoporosis (T ≤ –2.5; >2.5 SD from the mean) when compared to age and sex matched controls.[70] The relative importance of disease activity, nutritional status, and cumulative steroid consumption remains debated. A large adult study did identify chronic steroid exposure as the only significant risk factor for the staggering 40% increased risk in osteoporotic fractures in adults with CD.[71,72] Furthermore, it appears that

BMD is no different to healthy age-matched adult controls within the first six months following symptom onset.[73]

However, up to 30% of children is still likely to have a significantly reduced BMD,[74,75] with children with CD tending to be more affected. Overall therefore, both unchecked inflammation and chronic steroid exposure lead to loss of bone mass.[76]

The impact of nutritional status is unclear, as the correlation with BMD may well be confounded by disease activity. Ahmed SF et al. suggest that the growth failure seen in children with CD leads to a reduction in the size/length of their bones, thereby leading to falsely low values of BMD. If adjustments for bone size are made, bone mass in children with active CD appears to be better than previously reported. If bone area is not considered, up to 65% of children with CD would be labeled with moderate to severe osteopenia, while this falls to 22% if appropriate corrections are made.[77] Although aggressive nutritional restitution is likely to minimize the chance of nutritionally dependent bone loss, whether or not supplementation with vitamin D and/or calcium provides added benefits remains to be determined by placebo-controlled studies. Identification of children at greatest risk is important, as urinary calcium excretion, plasma 25-OH vitamin D levels, and a baseline bone density scan allow monitoring and subsequent supplementation of documented deficiencies. There is great potential for bone remodeling if children are in good clinical remission. Following presentation with osteoporotic vertebral fractures and in a good nutritional state, a 16-year-old with CD displayed a dramatic recovery of vertebral height within three years of largely steroid–free therapy that controlled his disease activity.[78] Vitamin D deficiency is more common in adults and children with IBD, especially CD, than healthy controls.[79] Many factors are likely to contribute to this, including malabsorption secondary to mucosal disease or surgical resection, and reduced sunlight exposure, physical activity, and dietary intake.[80]

The fact that the consequences of osteoporosis occur so long after the diagnosis of IBD in childhood makes it imperative for pediatricians to be aware of the BMD data now accumulating on adults with IBD. Several larger series are now reporting the clear association with life-time steroid dose and the increased risk of osteoporotic fractures.[71] An ideal combination may be enteral nutrition and azathioprine/6-MP in the treatment of children with more severe CD. It is now clear that treatment regimens minimizing steroid therapy, and relying on nutritional therapy or other steroid-sparing agents, do not lead to the reduction in BMD seen in

patients receiving corticosteroids. Although the mechanism is still unclear, Infliximab has now been shown increase markers of bone formation.[81,82] Patients with a lifetime steroid exposure of 12.9 g have a significantly lower BMD than normal controls, whereas those with <2 g lifetime exposure are comparable to age-matched healthy controls.[83] However, it is also clear that osteopenia does occur in steroid-naïve patients.[84] Walther et al. demonstrated a similar proportion of osteoporosis in steroid-naive and steroid-treated patients.[85]

Besides corticosteroid therapy, which is known to influence bone integrity by inhibiting osteoblast proliferation and stimulating osteoclasts, the underlying systemic inflammation associated with CD has an independent detrimental influence on the bone health of children with CD. Inflammatory mediators such as TNF, IL-1β, and IL-6 may have a critical influence on mineral density and result in decreased bone formation, increased bone reabsorption, or both.[86]

Furthermore, unchecked inflammatory activity may lead to calcium and vitamin D malabsorption, which in turn increases parathyroid hormone secretion and further bone reabsorption.[87] Patients with small-bowel disease and/or resections are therefore also at risk of long-term osteoporosis.

Micronutrient Deficiencies

Identifying and treating macro- and micronutrient deficiencies are problematic for two main reasons. There is the initial difficulty of finding an accurate and reliable measure for each specific nutrient deficiency, followed by the inherent variability of a disease characterized by a remitting/relapsing course. There is also the impact of medical therapies and surgical interventions on nutrient status, for example, sulphasalazine inhibiting folate absorption and corticosteroids limiting calcium absorption. Clinically relevant micronutrient deficiencies in patients with IBD are unusual, and most are likely to occur when receiving long-term total parenteral nutrition (TPN).[88] Nonetheless, significant subclinical deficiencies in vitamins (B1, B6, C) and certain trace elements (magnesium, copper, and zinc) have been reported.[89]

Vitamin B12 deficiency occurs most often in patients with active terminal ileal CD, or following a resection of this part of the intestine. In addition to the usual consequences of these deficiencies, low serum B12 and folate levels may result in an elevation of homocysteine and hence contribute to a hypercoagulable state,[90] with folate supplementation

leading to a reduction in homocysteine levels.[91] Patients with IBD are also at risk of developing fat-soluble vitamin deficiency. A significant deficiency of both vitamins A and E has been documented in 16% of children with IBD, with moderate-to-severe disease activity leading to a deficiency in over 40% of children.[92] Whether or not this was a consequence of a poor dietary intake or disease activity was not made clear. The range of vitamin deficiencies in patients with long-standing IBD is wide. Two comprehensive studies documented deficiencies in biotin, folate, β-carotene, thiamine, and vitamins A, E, and C in 40%–90% of adults, despite inactive disease and adequate dietary intakes.[93,94]

Iron deficiency anemia is difficult to diagnose in children with CD. Both the anemia of chronic disease and the variability in serum ferritin as part of the acute phase response make clear guidelines impossible. Low values of serum ferritin (<15 μg/dL) certainly confirm iron deficiency, but this may also be present at much higher values. Combining the more stable serum transferrin receptor (sTfR) with serum ferritin has been of benefit in adults,[95] whereas a small pediatric study suggested basic red cell ferritin was a more reliable measure in the presence of inflammation.[96] Variable absorption and the common gastrointestinal adverse effects of oral iron also make the response to treatment difficult to interpret.[97] This, together with the potential exacerbation of colitis by the oxidative stress of oral iron supplementation, has led to successful trials of parenteral iron and erythropoietin use.[98,99]

Trace elements are reduced in up to 50% of patients with active IBD,[94] with zinc deficiency being the most common.[93] The latter has been implicated in the maintenance of intestinal barrier function by regulating tight-junction permeability, with supplementation leading to a reduction in gut permeability in both animal models of colitis and adults with active CD.[100] Authors have shown significantly reduced fractional zinc absorption in adolescents with stable CD compared to normal controls. This appears to be due to unusually high fecal zinc losses.[101]

Many micronutrients are also antioxidants (vitamins A, E, C and selenium), and thus, adequate supplementation may help counteract some of the oxidative stress that contributes to ongoing gut inflammation. Both serum selenium and erythrocyte glutathione peroxidase activity have been inversely correlated to plasma TNF-α levels in patients with active CD.[102]

Other Nutritional Consequences of IBD

Patients with long-standing IBD may of course also suffer the nutritional

consequences of their therapies. Sulphasalazine is most notable for its impact on folate metabolism by competitively inhibiting dihydropteroate reductase, but folate rarely requires supplementation. Patients with severe ileitis or a significant ileal resection may develop steatorrhoea, which in turn binds available free calcium, thus preventing calcium from binding to oxalate. High concentrations of luminal oxalates may lead to hyperoxaluria and renal calculi. A significant decrease in essential fatty acids has been correlated with a BMI < 50th centile and active CD,[103] irrespective of dietary intake.

INDICATIONS FOR NUTRITIONAL TREATMENT

The indications for nutritional intervention differ among the different types of IBD (Table 52-1). However, adequate nutritional support is an essential part of the long-term management of any chronic inflammatory disorder. In most cases, this involves the provision of nutritional supplements to complement an otherwise inadequate intake and may involve anything from overnight nasogastric feeds to long-term support on home parenteral nutrition. For UC, no exclusively nutritional therapy has been shown to be therapeutically effective. In contrast, over many years patients with active CD have responded well to EEN. There have only been anecdotal reports of patients with indeterminate colitis responding to EEN, although the responders are likely to be those with a more Crohn's-like phenotype.

TABLE 52-1 Indications for Enteral Nutrition in Crohn's Disease

- Exclusive enteral nutrition for active disease
- Perioperative parenteral nutrition and bowel rest
- Supplemental enteral nutrition for maintenance of disease remission
- Nutritional support to maintain adequate weight gain
- Dietary supplementation of vitamins/minerals

Ulcerative Colitis

There have been no studies showing that any form of nutritional intervention, used as sole therapy, can induce a remission in patients with active UC. Two randomized, prospective studies[104,105] clearly showed there was no role for exclusive total parenteral nutrition and bowel rest in

the management of acute UC. Although there were some nutritional advantages in the group on TPN, these were outweighed by the complications reported at the time.

Although often not as severe as in patients with CD, malnutrition does occur in children with UC.[106] However, although exclusively nutritional therapies are of no benefit in treating acute UC, nutritional support plays a vital role in minimizing further morbidity. Up to 10% of children with UC present with weight/height Z-scores ≤ –2,[107] and inevitably nutrition is compromised in acute disease, both by a reduced oral intake and by increased losses of both protein and blood from the gut mucosa.[108] Although the concept of continuing enteral feeds during an episode of acute colitis may raise concerns in some, the evidence clearly suggests that equal nutritional improvement occurs with TPN and total enteral nutrition (TEN) in active colitis.[109] Not only does the latter lead to fewer complications, it is less costly, more physiological, and much simpler to provide. However, there continue to be absolute contraindications to enteral nutrition (Table 52-2).

Although dietary supplementation plays a role in acute UC, there is no evidence that specific dietary exclusions are of benefit in the long-term. Well-balanced, healthy diets that include normal amounts of dietary fiber are to be encouraged. Short-term studies suggest improved gastrointestinal symptoms on isphaghula husk.[110] Initial data that suggested diets low in fiber were linked to an increased risk of colon cancer have now been refuted in large epidemiological surveys in women.[111] A study conducted by Sanchez-Fidalgo et al. demonstrated that dietary enrichment with extra virgin olive oil reduces the damage in acute colitis in murine models, by alleviating oxidative events and returning proinflammatory protein expression to basal levels. Extra virgin olive oil has demonstrated immunomodulatory and anti-inflammatory properties and in addition to its high monounsaturated fatty acid content evidences have accumulated on the favorable properties of minor, although highly bioactive, components present in the unsaponificable fraction (UF). Extra virgin olive oil UF diet might provide a basis for developing a new strategy in dietary supplementation for the prevention of UC.[112]

TABLE 52-2 Contraindications to Enteral Feeding

- Massive Hemorrhage
- Intestinal Obstruction
- Bowel perforation

- Toxic dilatation

Crohn's Disease

In children with CD, there are several reasons for nutritional intervention (Table 52-1).

Exclusive Enteral Nutrition

EEN is indicated as a first-line therapy in any child with acute CD, provided there are no absolute contraindications to enteral nutrition per se. The issue of whether or not enteral nutrition is as effective as steroid therapy at inducing a remission has been hotly debated for some time. However, what is clear from the pediatric literature over the last 20 years is that an exclusively nutritional therapy can be highly effective at inducing a remission from active disease.[113–115] The best results appear to be in those children with newly diagnosed disease,[116] although the pediatric evidence does remain limited in terms of the number of children reported. Studies have shown that EEN induces remission in up to 85% of children with newly diagnosed CD.[117,118] Large adult studies,[119,120] as well as some of the smaller pediatric studies,[115] suggest disease distribution does not affect the efficacy of the treatment. Colonic disease as well as terminal ileal disease appear to respond, with a similar reduction in inflammatory markers, disease activity, and improved mucosal histology.[114] More recent retrospective data however suggest that clinical remission with isolated colonic disease may only be about 50%, compared to about 75%–80% in the presence of any macroscopic ileal involvement.[121] Although there is only anecdotal evidence on the response of oral and perianal CD to EEN,[122] the authors have made use of EEN as an adjunct in treating severe perianal CD.

Adverse effects to using EEN in CD are very rare. However, we reported a case of refeeding syndrome in a child following her presentation with severe Crohn's colitis. A rapid loss of weight over the preceding four weeks, followed by treatment with exclusive polymeric nutrition, led to a dramatic fall in serum phosphate and signs of hypervolemia in the first few days of treatment.[123] It is important to remain aware of this complication when refeeding previously malnourished children with CD, as adequate supplementation in the first few days can prevent serious complications.

Perioperative Nutritional Support

Preoperative undernutrition has long been known to adversely affect surgical outcome. Although surgical outcome is related to the nutritional state of the patient[124] short-term preoperative use of TPN is insufficient to dramatically improve the nutritional status. Even 7–10 days of preoperative parenteral nutrition in malnourished "cancer" patients achieved only a 10% reduction in postoperative complications.[125] Parenteral nutrition may replace acute nutritional deficits in the immediate perioperative period and may also reduce length of small-bowel resection in adults with CD.[126,127] Uncontrolled studies in adults with IBD also suggest there is a reduction in postoperative complications following preoperative parenteral nutrition,[128] yet it appears most effective in patients with severe malnutrition and highly active disease.[129]

Improvements in the medical management of children with IBD are likely to continue reducing the need for surgery, yet recent studies still suggest that almost 60% of adults with CD will require an operation.[130] Older pediatric data showed that up to 80% of children with CD were needing surgery in the 15 years following diagnosis.[131] Emergency surgery may be unavoidable in a small number of children who present with fulminant colitis, however the majority will require surgery for treatment-resistant disease.

Although a child with acute, severe UC will require close monitoring of fluid and electrolytes in the perioperative period, they are less likely to have had a long period of active disease leading to chronic malnutrition.

Despite persistently active, treatment-resistant disease, surgery can often be planned some weeks in advance. In these circumstances, optimizing nutrition over a period of 4–6 weeks is likely to result in a much better postoperative course. Provided inflammatory activity and symptoms allow, several weeks of EEN in children with severe CD can lead to nutritional restitution prior to surgery. In isolated cases, the use of TPN for a several weeks may also be justified in preparation if enteral access is impossible. However, clinicians can be forced to compromise in the face of severe treatment-resistant disease, where disease activity does not allow time for any improvement in nutritional state and surgery should then not be delayed.

Postoperative support for all children, irrespective of the type of abdominal/perianal surgery should involve institution of enteral feeds as soon as possible. Adult evidence demonstrates no increase in complications when patients are fed enterally within 48 hours of colectomy.[132] Very early use of enteral supplements reduces early

postoperative complications even in undernourished adults, and leads to a better weight gain, as well as an improvement in physical and mental quality of life.[114] Routine use of postoperative TPN in well-nourished patients should be discouraged as it increases complication rates.[133] However, provision for TPN should still be made if enteral feeding is likely to be delayed for more than 1–3 days in any undernourished child who is likely to suffer a long convalescence without achieving full enteral feeds.

NUTRITIONAL TREATMENT OF ACTIVE CROHN'S DISEASE

In severe malnutrition, there is clear evidence of a secondary immune dysregulation.[134] Both T cell numbers and secretory IgA levels are consistently reduced, and there are also defects in bacterial phagocytosis, lysozyme, and interferon production. Although today's children with CD are not often severely malnourished, they are certainly well below their expected centile for weight.[135] It is therefore quite clear that this degree of malnutrition does little to attenuate tissue-damaging immune responses.

Parenteral Nutrition

The advent of parenteral nutrition in the early 1970s proved useful in managing intractable cases of CD[136] and its use became more widespread as an alternative primary therapy for severe disease.[137] As would later be clearly demonstrated for enteral nutrition,[60] it was parenteral nutrition that was first shown to reverse growth failure in children and adolescents with active CD.[55,138] However, as awareness of the septic and thrombotic complications associated with an indwelling line increased, the use of parenteral nutrition in CD declined. Although it still appears to be as effective as elemental nutrition in the short-term treatment of acute disease,[139] its complications, practical difficulties, and expense, limit its use. Furthermore, total parenteral nutrition induces mucosal atrophy and increases bacterial translocation,[140] something that is entirely preventable by luminal nutrition.

The combination of intravenous nutritional support and complete bowel rest has not been shown to be more effective in treating active disease than either EEN or TPN with oral feeding.[141] In a controlled study, 58%–71% of all groups achieved a clinical remission, indicating that

bowel rest per se has no added effect on the disease, but that improved nutrition in any form has clinical benefit. However, there have also been a number of case series suggesting that exclusive parenteral nutrition may be of significant benefit in severe Crohn's colitis,[126] with at least one study documenting endoscopic healing of mucosal ulcers on exclusive parenteral nutrition.[142] Despite this, the preference for nutritional support in most cases remains an enteral feed.

There has been limited success (35%) in achieving fistula closure on exclusive TPN,[143] with any improvement not being sustained once back on a normal diet. In view of other therapies, such as Octreotide and anti-TNFα antibody, TPN now has only a very limited role in the management of these fistulae.

Children only rarely have bowel resections large enough to require long-term home parenteral nutrition support. However, if in adults less than 100 cm of jejunum or less than 50 cm of small bowel with an intact colon remains, long-term parenteral nutrition is often necessary.[144] Yet over 70% of these patients will be on a full oral diet within 12 months of starting home parenteral nutrition.[145]

Enteral Nutrition

Elemental Diet

An elemental feed is a chemically defined diet whose protein source is amino acids or short-chain peptides, with short-chain carbohydrates and added fat, minerals, and vitamins. The National Aeronautics and Space Administration (NASA) had initially designed elemental diets for astronauts.[146] This was with the intention of providing a nutritionally complete diet of which as much as possible would be absorbed. However, although absorption was limited mainly to the upper small bowel, the diet did not prevent the production of stool as had been hoped.

Nutrition was initially used in inflammatory bowel disease as an adjunct in malnourished patients with growth failure. Elemental diets were developed and first used as sole therapy for IBD in adults in the 1970s.[147] It was then shown that nutrition had a beneficial effect on disease by reducing the increased gut permeability characteristic of CD.[5] Logan et al. studied seven adults with extensive jejuno–ileal CD. They showed a reduction in both gut protein and gut lymphocyte loss during a period of elemental feeding. This was the first report that an elemental diet could directly improve gut function, probably by reducing bowel inflammation.

In 1973, Giorgini et al.[4] reported the first successful use of enteral

nutrition in treating a child with acute CD. It was then shown that enteral nutrition was effective in treating a series of children with CD. Successful use in combination with drug therapy led on to a further study by the same group, which first showed exclusive CREN to be as effective as steroid therapy at inducing a remission.[148] While both these studies had few patients, they gave the first insights into the possible benefits of nutritional therapy as treatment for childhood CD. In addition to achieving disease remission, nutritional therapy was also found to have beneficial effects on inflammatory masses and fistulae.[6,148] Simultaneously, both Morin et al. and O'Morain et al.,[149] documented that an elemental diet improved linear growth in several children with active CD.

A variety of devices, formulas and regimens were then used to feed patients intragastrically. Continuous feeding[7,150] and then overnight feeds predominated[8] for the induction of remission. Supplementation of an elemental diet with glutamine, a gut-specific metabolic fuel, did not further improve efficacy, although the small numbers in this study make conclusions difficult.[151]

Semi-elemental Diet

Once elemental diets had achieved their first successes, short-chain peptide containing diets were suggested as a better nitrogen source than amino acids.[152] Silk et al. refuted previous evidence that free amino acids were better absorbed than di- and tripeptides. By using an intestinal perfusion technique in adults, he was able to demonstrate better absorption of amino acids from both casein and lactalbumin hydrolysates, than from an equimolar feed of free amino acids. Not only was there a more uniform absorption of amino acids, but the hydrolysates had a beneficial effect on jejunal absorption of water and electrolytes. There followed the first small randomized study of 7–8 children in each group, in which overnight nasogastric feeding of a semi-elemental diet was compared to prednisolone treatment in children with predominant small-bowel CD.[8] A 4–5-chain amino acid–based diet was as effective as steroids at achieving a remission in active CD. It again confirmed the North American finding that there was a clear acceleration in growth in the group not taking steroids.

Polymeric Diet

It therefore appeared that steroids may have a similar efficacy to semi-elemental feeds in the induction of remission. Several adult studies reported the efficacy of whole protein diet compared to both that of

elemental diets and to steroids. Raouf et al.[153] and others all found polymeric diets to be as effective as an elemental diet at inducing remission.

Polymeric diets were also shown to be as effective as conventional steroid treatment. Gonzalez-Huix et al.[109] confirmed this in adults, and Ruuska et al.[115] and Beattie et al.[154] in children. Ruuska et al. in a well-planned but small study (n = 19), showed a polymeric diet to be as effective as steroids in inducing a remission in children with acute CD. More recently Borrelli et al. demonstrated that a course of polymeric formula is equally effective to corticosteroids in inducing remission in children with active CD.[155] In this prospective 10-week open label trial, 37 children with active CD were randomized to receive oral polymeric formula alone or oral corticosteroids. At week 10, the proportion of patients achieving clinical remission was comparable between the two groups (polymeric formula: 15/19 [79%; 95% confidence interval (CI), 56%–92%]; corticosteroid group: 12/18 [67%; 95% CI, 44%–84%]; P = 0.4; not significant). However, the proportion of children showing mucosal healing was significantly higher in the polymeric (14/19; 74%; 95% CI, 51%–89%) than the corticosteroid group (6/18 [33%; 95% CI, 16%–57%]; $P < 0.05$). A further great advantage in using a polymeric diet then became clear. Although almost all children previously required feeding by intragastric tube, whole protein formulas such as AL110 (Nestlé-Clintec), used by Beattie et al.[113] and Nutrison Standard (Nutricia), used by Ruuska et al.[115] were palatable enough for daily oral consumption. Children rarely required a nasogastric tube to complete their entire nutritional needs. This provided a considerable improvement in the quality of life for children on several weeks of nutritional therapy.

The most recent and most definitive cohort study to date, by Fell et al.,[114] shows that a whole casein, polymeric diet (Modulen IBD, Nestlé Clinical Nutrition), rich in transforming growth factor β, is well tolerated and achieves a clinical and histological remission. Twenty-nine consecutive patients were treated with the exclusive enteral diet for an eight-week period. Although over half had mild disease, 12 had moderate-to-severe disease with a pediatric CD activity index (PCDAI) >30.[156] A nasogastric tube was only required in one patient for the first two weeks of treatment. Only 2 of 29 patients failed to show any clinical response, one with severe colonic disease, the other with an inflammatory mass requiring surgery. Twenty-three of 29 patients achieved a complete remission on PCDAI scoring. The PCDAI fell dramatically within two weeks of starting

the diet, but continued to fall until eight weeks of treatment. There was significant macroscopic and histological improvement after treatment, with mucosal healing occurring in the terminal ileum and colon of eight and two patients, respectively. Serum TNFα and mucosal mRNA for IL-1β and IL-8 were significantly reduced in both the terminal ileum and the colon after treatment. IFN-γ was significantly reduced and TGFβ was elevated in the terminal ileum alone. There is no direct evidence that the TGFβ in the enteral formula is responsible for the upregulation of mucosal TGFβ. Nonetheless, this study strengthens the findings by Breese at al.,[157] that polymeric enteral nutrition alone can achieve an improvement in histology and a complete normalization of some of the mucosal messenger RNA of proinflammatory cytokines involved in tissue damage.

Bannerjee et al. documented the most rapid reduction in proinflammatory cytokines to date, in children receiving EEN for active CD.[158] Within three days of starting an exclusive polymeric diet, there was a significant reduction in IL-6 and ESR. This, together with improvements in CRP, insulin-like growth factor-1 (IGF-1), and PCDAI by day seven, predated any measures of nutritional restituation (mid-upper arm circumference/triceps skinfold thickness). Further evidence showed that nutritional therapy in this situation is providing more than an optimal calorie intake.

The lipid composition of some polymeric diets has been held responsible for some of the variability in their efficacy.[159] High long chain triglycerides (LCT) concentrations have been associated with a poorer response in treating active CD, with suggestions that the high linoleic acid concentration may be responsible,[160] although high concentrations of medium chain triglycerides (MCT) in the feed does not affect its short-term efficacy.[161] However, the only two randomized studies have been unable to show a difference in efficacy between formulae containing either low or high amounts of long-chain triglyceride.[162] Only one randomized study appeared to show a significant difference in remission rates depending on the fatty acid composition of the polymeric feeds. Gassull et al. report a significantly better clinical remission rate in adults after four weeks of an exclusive enteral feed rich in n-6 fatty acids, compared to one high in monounsaturated fats (52% vs. 20%, respectively).[109] Why this conflicts with previous evidence remains unclear although the high percentage of synthetic oleate (79% of total fat) in this study may mask beneficial effects previously seen with formulas containing different fatty acid profiles.

There is limited evidence of milk intolerance in adults with active CD. True lactase deficiency is an unusual cause of symptoms in adults with active CD,[163] however, up to 46% complained of gastrointestinal symptoms related to milk intake. In contrast, exclusive enteral feeding with lactose-free, whole casein diets has not been associated with intolerance in children with active disease.

Although elemental or semi-elemental formula was initially used in EEN, available data does not show any significant difference in outcome based on the type of formula used.[164] A meta-analysis of 10 trials (334 adult patients) did not reveal any difference in efficacy between elemental and nonelemental formulations (odds ratio 1.10; 95% CI 0.69%–1.75%).[165] Polymeric formulas are generally less expensive and have better taste characteristics, permitting oral administration in many children. Moreover, the use of polymeric formula may be associated with better weight gain than an elemental diet.[164]

Steroids versus Enteral Nutrition

Contrary to these striking pediatric findings, large adult trials[166,119,167] as well as meta-analyses of the adult data, have found steroids to be more effective than enteral nutrition at inducing remission in active CD.[168] In the most recent Cochrane review comparing efficacy of induction between corticosteroids and EEN, meta-analyses of six trials (192 patients received EEN and 160 patients received steroids) favored corticosteroid therapy over enteral nutrition (95% confidence interval [CI] 0.21%–0.53%). The studies analyzed contained both adult and pediatric patients; however, many pediatric trials were excluded from this analysis owing to methodological weakness.[165] The meta-analysis by Griffiths et al., while including both children and adults, excluded most of the smaller pediatric studies. This large review (n = 413) reported that steroids were significantly more effective at achieving a remission than enteral diets (odds ratio 0.35; 95% CI 0.32%–0.58%). Like other analyses, this study also relies on clinical disease activity indices to document clinical remission rates, but these tend to favor steroid therapy by their reliance on a patient's general feeling of well-being. As a result, conclusions from these studies are frequently and inappropriately applied to children, despite there being clear differences between adult and pediatric patients. Most children have had much shorter disease duration and tend to be much more compliant with therapy. Furthermore, no comment is made on the differing adverse effects and abilities of the two treatments to heal gut mucosa, with

steroids having been well-documented to have limited effects on gut inflammation.[169] As yet no analysis has detected a significant difference in efficacy between elemental or polymeric diets.[170]

Despite the several small pediatric studies that suggest a useful role for enteral nutrition in active CD, the view has thus prevailed that enteral nutrition is less effective than steroid therapy. A meta-analysis of pediatric data was performed to maximize the available pediatric data.[171] Despite limited numbers of truly randomized children, sensitivity analyses allowed the authors to arrive at valid and important conclusions. The summary data clearly showed enteral nutrition to be as effective as steroids in the treatment of children with active CD. Additionally, a meta-analysis of pooled data from four randomized controlled trials in 144 children found no significant difference in remission rates at 8–10 weeks between EEN and corticosteroids (relative risk 0.97, 95% CI 0.7%–1.4%, random effects model).[172] Furthermore, to overturn this finding and demonstrate that steroids were significantly more effective than enteral nutrition would be close to impossible given the outcomes of the pediatric studies reported to date. All these studies principally address efficacy as their primary outcome measure, largely ignoring the very different side-effect profiles of each therapy. Corticosteroids have many significant adverse effects, whereas oral enteral nutrition has almost none. Prospective observational studies have also highlighted that even in the pediatric age group about 30% of children with CD, treated with corticosteroids within 30 days of diagnosis, are corticosteroid dependent at one year.[173]

Enteral Nutrition and Crohn's Colitis

There remains the further question of whether enteral nutrition is less effective at treating Crohn's colitis than Crohn's ileitis. Early use of enteral feed was limited to children with predominantly small-bowel disease,[174] with a suggestion that colonic disease was unresponsive to nutritional management.[175] More recent evidence from Thomas et al.,[176] Ruuska et al.,[115] and Fell et al.[114] however, supports the value of nutritional therapy in large-bowel and small-bowel disease. More recently Buchanan et al. retrospectively analyzed the remission rates of 114 children with CD treated with an eight week course of EEN and studied factors affecting treatment outcome, including disease phenotype. The analysis did not show any significant difference between the remission rates achieved by individuals with isolated small-bowel disease compared to those with isolated colonic disease [10⁄13 (77%) cf. 15⁄19 (79%)

respectively, $P = 0.88$], suggesting again that disease phenotype does not influence clinical remission.[177]

The improvement in colonic mucosal cytokine profiles after enteral nutrition,[114] provides hard evidence that there is an effect on colonic disease. Larger studies in adults also confirm that disease location does not appear to influence the response to treatment.[119,167,168] However, a more recent retrospective analysis of 60 children treated with EEN, suggested disease distribution may be relevant in the response to nutritional therapy. Children with any macroscopic ileal inflammation were significantly more likely to achieve a clinical remission than those children with colonic involvement alone (75% vs. 50%).[121,82] Despite this, many individuals with Crohn's colitis continue to respond extremely well, still making EEN the first choice for any child with CD, irrespective of their disease distribution.

Food Reintroduction

There has long been uncertainty as to the best way of reintroducing adults and children onto their "normal" diet after a period of EEN. The evidence for any of these different practices is very limited, with most approaches having been selected by experienced clinicians on the basis of theories prevalent at the time.

The best-described reintroduction program is based on the stepwise introduction foods, starting with the least allergenic.[178] One new food is introduced every 48 hours and if not tolerated, reintroduced at the end of the program. This systematic, but quite laborious program not only allows individual foods to be identified if causing immediate symptoms, but also gives the patients several more weeks on reasonable quantities of enteral nutrition while their normal diet is reestablished. The most frequently implicated foods causing discomfort in adults are cereals, dairy products, and yeast, although in children only a minority requires exclusion of specific items from their diet. In a two-year follow-up study of about 100 adults, the relapse rate was only just significantly lower in the group excluding dietary products that caused symptoms compared to those maintaining a normal diet.[179] There is thus insufficient evidence to routinely suggest exclusion of specific food items in children with CD.

Other units use less evidence based, but more practical reintroduction programs which range from the immediate introduction of a full diet, to a graded introduction over three weeks with the "ad libitum" diet increasing by 25% each week, along with the simultaneous reduction in enteral

nutrition.

ENTERAL NUTRITION AS MAINTENANCE THERAPY IN CROHN'S DISEASE

There has been much interest in whether dietary modification may prolong a remission in CD.

Numerous reports have considered and evaluated the role of partial enteral nutrition in the maintenance of remission and prevention of relapse. In addition to prolonging remission, such an approach may delay the requirement for further therapy (e.g., corticosteroids), while optimizing growth and nutrition. Partial enteral nutrition has been provided in various forms: overnight nasogastric (tube) (NG) feeds in conjunction with normal daytime eating, short course of NG feeds every few months, or as oral supplements in addition to oral eating throughout the day.

Initial studies suggested intermittent use of EEN for one month in every four could not only reduce steroid requirement and reduce disease activity, but also increase long-term growth rate in children with CD.[180] Although this intermittent use of EEN may have had a role in managing patients with otherwise intractable disease and a previous good response to enteral nutrition, repeated and prolonged use of exclusive enteral diets in most adolescents is difficult and mostly unnecessary. Given the availability of other, currently more effective maintenance agents, compliance with supplements of enteral nutrition is likely to be much greater if these can be shown to be effective.

Low-residue diets are not indicated in the maintenance of remission in CD. Unless required as a dietary modification in the presence of fibrosing/stricturing disease, they have not been shown to prevent disease relapse.[181]

A retrospective study in which a normal "ad libitum" diet of children with CD was supplemented with an additional 30% of the recommended daily calorie intake as polymeric feed, led to the halving of relapse rates within the first 12 months.[182] Small controlled studies also suggest an advantage to simple long-term calorie supplementation per se,[183] although larger, more definitive studies are still awaited. However, it is highly likely that a long-term nutritional supplement, in combination with a well-tolerated immunosuppressant, will provide the optimal maintenance therapy for most children with moderate-to-severe CD.

Several studies from Japan have also evaluated the role(s) of partial

enteral nutrition in adult populations. One study included 51 patients with CD in remission, who were randomized to receive half of their energy as elemental formula with normal diet (26 patients) or to have an unrestricted normal diet (with no additional supplements) (25 patients) for up to 24 months. Remission had previously been induced by EEN, parenteral nutrition, surgical resection, or corticosteroids. During the period of observation, the relapse rate of the treatment group was almost half of the free-diet group (34.6% vs. 64.0%; multivariate hazard ratio 0.40, 95% CI 0.16%–0.98%).[184]

Attempts at supplementing diets with specific anti-inflammatory agents continue, with fish oils receiving particular attention. Their effects on reducing mucosal eicosanoids (particularly leukotriene B4 and thromboxane A2)[178] have been widely reported. The n-3 fatty acids present in fish oils may also inhibit interleukin 1β and tumor necrosis factor production.[185] In addition, they also decrease platelet responsiveness,[186] and thus may act to reduce the multifocal gastrointestinal infarctions that have been reported as early pathogenic steps of CD.[187] Several large placebo-controlled, randomized studies have been performed on adults with both CD and UC,[188] but unfortunately much of the data remain conflicting. Although different methodology makes generalizations difficult, several important issues are raised. Although low-dose regimens cause no appreciable adverse effects,[189] compliance with high-dose fish oils in adults is poor, making long-term maintenance therapy in children even less attractive. Belluzzi et al.[188] found that supplementation with 2.7 g of n-3 fatty acids reduced the relapse rate in adults with CD, although another large study failed to confirm any benefit over placebo in using either >5 g/day of n-3 fatty acids, or maintaining a diet low in carbohydrate (84 g/day).[190] However, despite no clear-cut clinical benefits, significant changes are reported on a mucosal level. Colonic inflammatory cell infiltrates are reduced and the synthesis of LTB4 and TXA2 downregulated after supplementation with n-3 fatty acids.[190] It may therefore be that further refining of the composition, formulation, and dose is required before fish oil supplements become an accepted part of maintenance therapy. A small prospective study in children with CD (n = 38) suggests that adjunctive therapy with enteric coated omega-3 fatty acids and mesalazine (50 mg/kg/day) reduces relapse rates at 12 months compared to mesalazine alone.[191]

Other, more specific dietary interventions have been reported to prolong remission in CD. Although none of these have been assessed in

larger, placebo-controlled studies, they offer interesting clues to the role dietary modification may play in maintaining disease remission. A small study randomizing patients to a diet low in microparticles,[192] suggested significant benefits in disease activity and remission rates after four months when compared to 10 patients on a normal diet. Diets low in refined sugars have been not been found to prevent relapse rates in adults with CD.[193] Similarly, the presence of yeast in the diet of patients with CD may potentiate disease relapse, but larger studies are necessary before firm conclusions may be drawn.[194]

As the importance of gut microflora in the etiology of IBD has is increasingly recognized, so the prospect of modulating it by means of "functional foods" has gained much support. Although discussion is beyond the scope of this chapter, the mechanisms of action and potential implications of this exciting new field are great for patients with chronic intestinal disease driven by enteric flora. It is quite clear that over the next few years we are likely to see larger, placebo-controlled trials of both pro- and prebiotics. Prebiotics are the complex sugars such as fructo-oligosaccharides and inulin, which pass into the colon undigested where they may be selectively fermented by certain probiotic species.[195] Attempts to maintain remission in both UC and pouchitis with combined probiotic combinations has proved quite successful,[196] whereas *Lactobacillus GG* alone may have a role in improving gut barrier function and clinical status in children with CD.[197]

If long-term nutritional support is required, most children will tolerate a more palatable polymeric formula by mouth. However, some children will require supplementation by tube. Although short-term support can be given via a nasogastric tube, longer-term needs should be met via a more permanent device. Where nutritional support is needed for several months, placement of a percutaneous endoscopic gastrostomy (PEG) is ideal. Despite initial reservations about possible complications from fistulous tracts formation, PEG placement has now been documented to be uncomplicated in selected adults and children with IBD.[198] In patients receiving corticosteroids, adhesion of the gastric to the abdominal wall may be delayed, and thus early "pulling" of the PEG could lead to tract disruption.

THE ROLE OF ENTERAL NUTRITION ON GROWTH AND DEVELOPMENT

Epidemiology of Growth Failure

After the onset of symptoms, but before a diagnosis of CD is made, up to 46% of children will have a reduced height velocity, with only 12% having a normal height velocity up to the time of diagnosis.[61] In contrast only 3%–10% of children with UC may present with reduced height velocities at diagnosis.[62] Although much of this data are now over 10 years old, and an accurate diagnosis may now be made more quickly, it is clear that untreated CD has profound implications on a child's growth potential. Motil et al. reported in a cross-sectional study of their pediatric IBD patients that 23% had a Z-score < –1.64, with 50% of these having a bone age delayed by >1 year over their chronological age.[199] Physicians caring for children with IBD, particularly CD, therefore, have the unique opportunity to try and modify the impact that both disease and therapy has on the growth rate of their patients. Sawzcenko et al. recently reported that children with the IL6-174 promoter polymorphism are more growth retarded at diagnosis than those without.[200] Although levels of CRP were highest in children with this polymorphism, they appeared to be at no increased risk of developing CD than the other polymorphisms compared to 351 healthy controls. The authors speculate that IL-6 mediates growth failure in these children, and that genotyping at an early stage would allow the use of specific growth sparing/focused biological therapies.

A delay in presentation is associated with a significantly greater degree of growth impairment at diagnosis.[107] The length of this delay appears to correlate with a lower height Z-score at diagnosis, and hence with a lower final adult height.[201] A delay in diagnosing the more subtle presenting symptoms of jejunal CD may at least in part explain the more significant reduction in adult height seen in this particular group of children. Despite management within tertiary centers, 49%–65% of children with CD have a reduced height velocity in early puberty, with growth velocity remaining less than 4 cm/year during the first 1–2 years of therapy.[62,202] Hildebrand et al. report a mean Z-score for height of –0.6, two years after diagnosis in 46 consecutive children with CD.[62] Griffiths et al. reported that of 67 consecutively diagnosed children with CD followed to maturity, about two-third maintained or increased their Z-scores for height.[203]

The outcome of many of these studies reflects management where the mainstay of therapy was often corticosteroids, and so does not include the more recent move to the earlier use of steroid-sparing agents. However, although the most recent follow-up data show that the majority of children diagnosed with CD are closer to their expected adult height, almost 20%

will be at least 8 cm below the height expected by mid-parental centile.[201] Earlier diagnosis and identification of those children at risk of growth failure remain a significant problem for clinicians wanting to optimize growth outcomes.

Etiology of Growth Failure

The growth failure seen in children with IBD is multifactorial, with inflammation, undernutrition, and steroid therapy being its principal determinants.

Initial evidence suggested that chronic undernutrition was the main factor in the growth failure seen in children with IBD,[55] with studies confirming that simple nutritional restitution improved the linear growth of children with CD.[54,56] Children with active CD have a relative insufficiency of dietary intake when compared with children of the same age,[204] although they may compensate with an increased intake between disease relapses.[202] However, simply increasing the calorie intake, as seen in patients on steroids, does not confer the same benefit on growth as a lower calorie intake received on EEN.[176] The presence of low levels of IGF-1 in children with active CD strengthens this association, as the relationship between IGF-1 and malnutrition is now clear.[205] Both IGF-1 and its carrier protein IGF-binding protein-3, are increased significantly by treatment with EEN.[204,206] Further evidence that inflammation, and not only nutritional factors, plays a role in determining growth can be seen in studies of children undergoing surgery for active IBD. Although postoperative catch-up growth is not documented by all authors, several authors show a clear growth spurt occurring after the removal of an inflamed segment of bowel.[207] Although this is most striking in prepubertal children, the benefit is still substantial if surgery is carried out before the final stages of puberty.[135] This response may of course in part be the result of improved postoperative nutrition. However, Varille et al. neatly documented that even children undergoing localized resections for stricturing disease had a significant reduction in REE and increase in nitrogen utilization four weeks after surgery, despite maintaining preoperative nutritional and steroid regimens.[208]

There now seems little doubt that ongoing inflammatory activity plays a key role in the inhibition of growth.[209] Studies conducted in animal models and in humans show that proinflammatory cytokines have an effect on the hypothalamic–pituitary growth axis, influencing the growth hormone (GH)/insulin–like growth factor synthesis. In a colitis-induced rat

model, Ballinger et al.[210] showed that, despite nutritional supplementation to restore energy intake and to increase weight gain, inflammation contributed to 30%–40% of growth retardation. In rats with colitis, a negative correlation between plasma GH and IGF-1 levels was found, suggesting inflammation-induced hepatocyte resistance to GH stimulation. De Benedetti et al.[211] had previously suggested a direct effect of the proinflammatory cytokine interleukin (IL)-6 on the hypothalamic–pituitary growth axis.

Reported abnormalities of the somatotrophic axis in children with CD are variable. Some studies have reported low levels of IGF-1 but normal levels of stimulated or physiological GH secretion.[212,204] Farthing et al.[213] reported low-GH levels following insulin tolerance test in two out of five growth-retarded children with CD. None of the children was on corticosteroids and the GH insufficiency was attributed to the disease itself.[214] The link between systemic GH levels either from stimulation tests or physiological sampling, and serum IGF-1 levels and linear growth in IBD is still unclear although one group of investigators have reported an association between systemic IGF-1 level and height velocity in children with IBD.[204] The finding of relatively low serum IGF-1 levels with relatively normal GH levels in affected children and their association with short stature in affected cases points to the possibility of a state of GH resistance.

Downregulation of proinflammatory cytokines by enteral nutrition may partly improve growth by reducing IL-6, a potent inhibitor of IGF-1.[215] There is a dramatic reduction in CRP, with concomitant increase in IGF-1 and IGFBP-3, within 14 days of commencing EEN.[113,114] This clearly indicates that enteral nutrition also has a more rapid, direct anti-inflammatory action in addition to its longer-term nutritional benefits. As has been confirmed in children with rheumatoid arthritis,[160] systemic inflammation, as well as malnutrition, reduces IGF-1. Direct inhibitory effects of proinflammatory cytokines such as TNFα on developing growth plates[216] and a downregulation of IGF-1 by IL-6 are both likely to contribute to growth retardation.[211] The additional suppression of appetite by TNFα,[217] the failure to downregulate the REE during malnutrition in CD,[58] and the exacerbation of bowel symptoms by food intake, all add to the final reduction in height velocity.

Enteric losses during active disease can also contribute to the decline in nutritional state. Children with small-bowel CD have varying degrees of malabsorption, with some of the hypoalbuminemia occurring as a result of

enteric losses.[218] The most important determinant of hypoalbuminemia is the systemic inflammatory response. Although protein loss through an inflamed mucosa does occur, it is usually modest, nonspecific, and occurs in association with loss of globulins. The hypoalbuminemia seen in CD is more frequently associated with the hypergammaglobulinemia occurring as a result of the acute phase protein synthesis. The latter is principally mediated by IL-6.[219] Furthermore, a sustained protein intake of less than 0.4 g/kg/day[220] is necessary to achieve any reduction in serum albumin.

Bile acid malabsorption may not only occur in children with active terminal ileitis, as systemic effects of ileal inflammation but also lead to malabsorption in the proximal small bowel.[221] As a result fat malabsorption may occur in some children with CD. In addition, low concentrations of vitamins A, E, and zinc have been shown to correlate more with disease activity than nutritional state,[92,222] suggesting depletion occurs during the acute inflammation rather than as a result of chronic malabsorption.

On-going growth failure in the first years after diagnosis may be due to several factors. Growth velocity can be delayed well beyond nutritional restitution and normalization of inflammatory indices. This, together with the frequent use of high-dose steroids to achieve the initial remission, up to 85% of children in some studies,[223,203] may arrest normal growth for many months after diagnosis. However, the most recent data suggest that it is neither the disease distribution nor pubertal stage at diagnosis that determine final adult height,[224] but rather the use of corticosteroids during puberty. Children who were diagnosed with CD during puberty and were treated with corticosteroids were significantly shorter as adults when compared who did not receive corticosteroids. In this study, this was the only independent predictor of final adult height.

Glucocorticoids may impair growth through a variety of mechanisms including a local effect at the level of the growth plate by reducing cell number, cell proliferation, and proteoglycan content, and through a reduction of GH secretion and its action.[225] They can alter GH receptor expression and alter IGF-1 expression and sensitivity.[226,227] Clinical studies of the role of glucocorticoids in IBD and growth retardation report conflicting data. Although some studies have found that glucocorticoids use has no effect on growth retardation except when used in very high doses.[203,202] Markowitz et al.[57] and Burnham JM et al.[64] have reported a clear association of height with duration of glucocorticoid exposure and cumulative dose.

Nutritional Treatment of Growth Failure

Over 20 years ago, several small studies confirmed the central role of nutrition in the long-term management of growth failure in children with IBD. Motil et al.[199,228] concluded that neither low-grade chronic inflammation nor low-dose corticosteroid use reduced whole body protein synthesis, and that it was adequate dietary supplementation that led to a significant increase in growth velocity of children with growth failure. This was confirmed by others as nutritional therapies became more practical for the treatment of acute CD.[6,54] Sanderson et al. was the first to establish the benefit of enteral nutrition over steroid therapy on short-term growth in children with newly diagnosed CD.[8]

MECHANISM OF ENTERAL NUTRITION

Despite the wealth of information that exists about the benefits of enteral nutrition, the mechanisms of action remain largely unclear (Table 52-3).

The most frequently advanced theory is that the bacterial flora within the gut lumen is modified by enteral nutrition. The clinical evidence of any difference in flora between patients with CD and normal controls remains slim. It was shown in the late 1970s that there are higher bacterial counts within the terminal ileum of patients with active disease.[229] Studies at that time also suggested a reduction in fecal flora after enteral nutrition.[230] It is now clear that increasing severity of systemic disease is associated with an increase in the adherence of fecal bacteria to the enterocytes,[231] and this does not appear to be related to the degree of local mucosal inflammation. The response of acute CD to antibiotic therapy further implicates the bacterial flora in the disease pathogenesis,[232] although antibiotics such as the quinolones and metronidazole have other immunomodulatory effects in addition to their antimicrobial actions.[233] The organisms that appear increased in the lumen of patients with CD include Bacteroides, Eubacteria, and Peptostreptococcus.[234] Elegant work on mice that develop spontaneous colitis (TCRα-/-) has confirmed that feeding with an elemental diet prevents bowel inflammation.[235] Unlike the mice fed with the elemental diet, the mice fed with regular chow and that develop colitis are colonized by *Bacteroides vulgatus* in >80% of cases. Furthermore, instilling this strain into the rectum of the elementally fed mice, led to development of a typical Th-2 type, T cell–induced colitis. Further animal work has shown that elemental diet may reduce progression of granulomatous enteritis by modulating the activation of T cells, the

production of NO, and the generation of oxygen-free radicals.[235–237]

TABLE 52-3 Mechanisms of Enteral Nutrition

- Alteration in bacterial flora
- Reduction in antigenic load
- Whole body nutritional restitution
- Provision of enterocyte nutrition
- Direct immunoregulatory effect
- Increased concentrations of TGFβ
- Low lipid and fiber content

The reduction in antigenic load that accompanies EEN may also contribute, at least in part, to bowel rest. However, a whole protein diet, and even an ad libitum diet together with some parenteral nutrition, appear as effective as an exclusive elemental diet at inducing a remission.[114,115,141] The efficacy of polymeric diets and recent evidence that dietary supplementation with enteral nutrition may prolong a remission,[182] both suggest that reducing luminal antigens may only play a modest role in the efficacy of this therapy.

Whether enteral nutrition per se has either a direct and/or indirect immunoregulatory effect remains speculative. Degrees of moderate protein malnutrition have been associated with poor immune function. In rodents, protein deprivation leads to impairment of the mucosal immune response, as well as depleting a population of T cells that control oral tolerance.[238,239] This would suggest that poor nutrition inhibit T cells that downregulate the gut's response to foreign antigens. Enteral feeding may therefore have an indirect effect on the immune response by restoring an adequate nutritional status.

More recently, the direct influence of luminal content on immune function has been studied. Sanderson et al. provided evidence that luminal content can influence epithelial cell gene expression within the gut.[240] Short-chain fatty acids, such as butyrate, are bacterial metabolites from unabsorbed carbohydrates. Butyrate induces secretion of insulin growth factor–binding proteins (IGFBPs) by a complex process involving histone deacetylation.[241] Butyrate has also been shown to potentiate the secretion of IL-8 by intestinal epithelial cells (Caco-2) if these are stimulated with either lipopolysaccharide (LPS) or IL-1β. LPS was only able to induce IL-8 secretion if these cells were preincubated with butyrate, implying direct effects of the latter on gene regulation.[242] Epithelial cell gene regulation

by luminal products thus appears to be able to influence intestinal inflammation through release of inflammatory cytokines. Upregulation of the chemokine macrophage-inflammatory-protein-2 (MIP-2) increases local neutrophil recruitment.[243]

It has also been suggested that the presence or absence, of individual components of enteral feeds is important in immune regulation. The putative advantage of high TGF-β levels in both AL110 and CT3211 (~24 ppm) is based on the large body of experimental evidence that this cytokine has the ability to downregulate other proinflammatory cytokines.[244] Fell et al.[114] demonstrated mucosal upregulation of TGF-β within the terminal ileum after an eight-week course of TGF-β-rich CT3211. It is still unclear whether this is related to the increased luminal presence of TGF-β or is simply an epiphenomenon of tissue repair (Table 52-4).

It may also be that enteral nutrition plays a direct role in promoting the mechanisms involved in epithelial healing. There are numerous peptides involved in the restoration of a disrupted epithelial barrier. The ulcer-associated cell lineage (UACL)[245] secretes cytoprotective peptides that promote epithelial healing. Among them are epidermal growth factor (EGF), TGF-α, human spasmolytic peptide (hSP), and the family of trefoil peptides. The trefoil peptides in particular have been shown to be vital in protecting against mucosal damage.[246] Enteral nutrition may contribute toward the maintenance of mucosal integrity by boosting the proliferation of the UACL.[247]

Although the theory of "bowel rest" has its supporters, others continue to feel it is adequate nutrition alone that could induce remission and growth in these patients.[248] Kirschner et al.[54] demonstrated improved growth in children simply fed an extra 1000 kcal/day. The same group later confirmed that improved nutrition not only increased linear growth, but also returned previously low levels of IGF-1 to normal in children with active CD.[249] Thomas et al.[204] confirmed this finding with an elemental feed, which was as effective at increasing IGF-1 as prednisolone, yet better promoted linear growth.

TABLE 52-4 Data Showing mRNA Transcripts (103)/μg Total RNA from Biopsies Taken Before and After Treatment with CT3211 and Normal Controls

	Terminal Ileum			Colon		
	Control	Pretreatment	Posttreatment	Control	Pretreatment	Posttreatment
IL-1β	15**	350	16*	0.54**	130	18*
IFNγ	1**	8.6	1.1*	1.1*	3.5	1.2
IL-8	50	660	150	4.8*	1500	130*
IL-10	1.5	1.7	1.3	3.7**	1.9	1.2
TGF-β	16	4.2	3.4*	11	15	15

*Significant change ($p < 0.05$) with treatment assessed by *t*-test.

**Significant difference between pretreatment (diseased) and control values assessed by Mann–Whitney U-test.

There are likely to be many mechanisms responsible for the clinical efficacy of enteral nutrition. It is clear however, that the luminal environment is crucial to the expression of mucosal disease. Our ability to regulate specific aspects of this environment by nutritional or other means remains a great challenge. The multitude of variables that may be important in achieving a disease remission makes identification of single factors extremely difficult. Current attention is focused on modifying enteral formulas in line with the recent evidence on dietary fats, while continuing to ensure their clinical efficacy and tolerability.

Mucosal Healing

Attention has focused on the ability of treatments to achieve mucosal healing.[250] Breese et al.[157] gave an initial indication that enteral nutrition was able to downregulate intestinal mucosal inflammation. Enteral nutrition was as effective as cyclosporin and steroids in reducing the percentage of IL-2–secreting cells in the terminal ileum after treatment, while it appeared more effective than steroids at reducing the percentage of IFN-γ–secreting cells. Furthermore, it was only the enterally fed group that showed significant histological improvement.

Despite being only a small study, Breese et al. raised two important issues. First, that enteral nutrition may be able to heal mucosa, second, that mucosal cytokine analysis following treatment did not necessarily correlate with either clinical or histological indices of remission.

The mucosal cytokine responses of a much larger cohort of children were reported by Fell et al.[114] Although clear clinical and histological remission was achieved in over 70% of children, cytokine profiles also dramatically improved with a polymeric diet alone. The dramatic downregulation of the potent proinflammatory cytokines IL-1β, IFN-γ, and IL-8 is the most concrete evidence to date that enteral nutrition acts at the

mucosal level (Table 52-4).

The issue of whether clinical, endoscopic, histological, or immunological remission should be the gold standard remains a matter of personal practice. If we are to believe that the presence of chronic inflammation predisposes to long-term complications and malignancy,[251] it may be a state of immunological remission at the mucosal level, that should be achieved in children with a lifetime of CD ahead of them.

BODY COMPOSITION, ENERGY EXPENDITURE, AND PROTEIN METABOLISM

It has become clear that both in adults and in children, active gut inflammation is associated with alterations in body composition. Although substrate metabolism during active CD resembles that during starvation, these are more a consequence of malnutrition and may be easily reversed by treatment with EEN.[58,236] BMI, lean body mass (LBM), and fat mass are all decreased in children with IBD, and this may be related to poor calorie intake.[199] A higher BMI and LBM were both positively correlated with increased BMD, factors that again stress the importance of adequate nutrition in maximizing long-term BMD.

Enteral nutrition provides a simple but effective treatment for children with CD. Not only does it appear to influence disease activity directly, it also appears to have clear benefits on growth and nutrition. Gerasimidis et al. demonstrated that a 6–8-week course of EEN with a polymeric feed in children with active CD has a positive effect on body composition by improving LM but not fat mass.[252] In this study 17 children (male/female: 8/9; median age: 12.7 years) with active CD were treated exclusively for 6–8 weeks on a polymeric feed (Modulen IBD). Body impedance was measured at baseline, during EEN, and posttreatment on normal diet and converted to Z-scores of fat and LM. Lean but not fat mass improved at the end of EEN (initiation vs. end of EEN; fat mass [Z-score]: –0.5 vs. –0.3; $P = 0.141$; LM [Z-score]: –2.1 vs. –0.8; $P < 0.0001$). At baseline several children presented with suboptimal concentrations of carotenoids, trace elements, vitamin C, B6, and folate in plasma but not in erythrocytes. EEN improved concentrations for several nutrients, but more than 90% of patients had depleted concentrations of all carotenoids.

Early evidence suggested that REE was increased in patients with inactive CD.[253] It now appears however, that if the REE is calculated per unit of LBM, it is the same in active CD as it is in normal controls.[58] The

REE/LBM was found to be significantly lower in children with anorexia nervosa than in normal controls. When comparing these malnourished noninflammatory disease controls with children with active CD, the authors showed that the appropriate physiological response to starvation is lost during active inflammation. Thus this inappropriately elevated REE further contributes to weight loss. On treatment with elemental feed, their absolute REE did increase to above normal levels, but per unit of LBM only remained within the normal range. This study also showed that an elemental diet was significantly better than steroids at accruing lean body tissue and was again better at achieving linear growth.

Despite the documented increase in energy intake by children on steroids compared to those on an elemental diet,[176] the latter group has a significantly greater rise in their median height velocity standard deviation score (SDS) after four weeks on an elemental diet. Adequate calorie intake alone in the presence of steroid therapy (124% of RDA) is thus unable to achieve optimal growth. The short-term growth suppressant effects of prednisolone[254] probably override any advantage conferred by the improved calorie intake. IGF-1 levels appeared greater after steroid treatment than after enteral nutrition, yet a significant increase in IGF-1 was only associated with greater height velocity after an elemental diet.[204]

The rate of protein turnover in seven children with chronically active CD receiving steroids was considered to be similar to that of normal adolescent controls,[255] although their nitrogen balance was found to increase fourfold during nutritional supplementation. Another small study compared the effect of steroids (n= 6) and elemental feed (n = 6) after the first seven days of treatment in acute CD.[256] Labeled L-leucine studies showed an equal increase in protein turnover after seven days of treatment, whereas in the steroid treated group this was at the expense of total body protein stores. Clinical improvement was similar in each group. In a more recent study by Thomas et al.,[257] rates of protein turnover were also elevated in active CD. Labeled L-leucine studies and mass spectrometry analysis showed an equal reduction in protein turnover to normal levels, both by steroids (n = 4) and by elemental feed (n = 6). This study only assessed protein turnover after several weeks of treatment, and only after effective recovery and nutritional restitution were almost complete. Royall et al.[258] published evidence in 30 adults with active CD that proportionate increases in body fat, protein, and water occurred after 21 days of elemental nutrition. In this study, a sustained remission was only achieved in patients who showed a gain in total body nitrogen. Hannon et al.

demonstrated that in clinically stable adolescents with CD, enteral nutrition promotes anabolism by suppressing proteolysis and increasing protein synthesis.[259] In this study, leucine (Leu) and phenylalanine (Phe) kinetics were measured using stable isotopes under fasted and fed conditions during a single study visit in six adolescents with CD. In response to enteral nutrition, the endogenous rates of appearance of Leu and Phe (reflecting proteolysis) decreased significantly by 40%.

Further evidence that optimizing nutrition may contribute to a clinical remission arises from the study by Slonim et al.[260] The combination of a protein-rich diet (>2 g/day), together with subcutaneous GH led to a significant reduction in CDAI within four months, in a small group of adults with moderate-to-severe CD. Increased uptake of amino acids,[261] and improved intestinal and muscle protein synthesis are mechanism that are likely to be important in mediating this effect.[262] Direct effects of GH on IGF-1 are not felt to be as significant, as no correlation was found between IGF-1 levels and clinical response.

FUTURE DIRECTIONS

From the above discussion, it is abundantly clear that nutrition still has a major role to play in the management of children with IBD, and in particular those with CD. Although EEN is clearly effective in many children with active CD, it remains difficult for some children and their families to complete prolonged courses. Work is continuing to assess whether long-term supplementation of a normal diet prolongs remission.

Palatable whole protein formulae are being modified to include higher percentages of n-3 fatty acids, whereas the addition of pro- and prebiotics to feeds is also already underway.

Work also continues on isolating the specific factors within formulae that may directly act on mucosal inflammation. Upregulation of mucosal TGFβ may be a direct result of higher levels of TGFβ within a formula (Modulen IBD, Nestle), whereas further characterization of the gut microflora in children with IBD may allow specific supplementation with specific prebiotics, substrates that selectively stimulate the growth of certain favorable probiotic species.

CONCLUSIONS

Despite the ever-increasing choice of therapies available to children with inflammatory bowel disease, the role of nutrition remains central to their

optimal management. The impact of bowel inflammation on growth and development cannot be underestimated. As final adult height is determined during the pubertal growth spurt, it is crucial to minimize the impact that both the disease and its therapies may have on a child's growth potential. Advances in identifying children at particular risk of growth failure may, in the future, allow specific interventions to maximize growth.

We still strongly suggest that EEN remains the best primary therapy for the treatment of all children presenting with a new diagnosis of CD, apart from those with severe perianal disease. Thereafter, the challenge is to maintain a lasting remission, particularly during puberty. This is likely to be best achieved with early use of immunosuppressants such as Azathioprine/6-Mercaptopurine, hoping to minimize steroid use. Continued vigilance of undernutrition and appropriate use of dietary supplementation remains essential to an optimal outcome.

The consequences of developing a chronic inflammatory disease during childhood will be felt long after a child is handed over to our adult physician colleagues. Increased risks of osteoporotic fractures, high rates of surgery, and a reduced final height are only some of the areas where nutritional therapy is vitally important. The ability of therapies to achieve healing of the gut mucosa is of utmost importance in children who have a lifetime ahead of them. Although dietary therapies do not yet play a significant therapeutic role in maintenance therapy for IBD, it is likely that evidence about potential disease-modifying dietary supplements will continue to appear. It is important that as more potent immunological agents become available to treat these diseases, we do not forget the therapeutic role of nutrition in CD; its absence of adverse effects and its proven impact on growth and gut mucosa.

Although newer therapies may require less commitment from families and medical teams, their unknown long-term safety profile still makes enteral nutrition an excellent choice for children with CD for many years to come.

REFERENCES

1. Guidelines for use of home total parenteral nutrition. A.S.P.E.N. Board of Directors. American Society for Parenteral and Enteral Nutrition. *JPEN J Parenter Enteral Nutr*. 1987;11(4):342–344.
2. Clouston TS. Forcible feeding. *Lancet*. 1872;i:797–798.
3. Widdowson EM. A study of individual children's diets. *MRC Special Report*. 1947;257.
4. Giorgini GL, Stephens RV, Thayer WR Jr. The use of "medical by-pass" in the therapy of Crohn's disease: report of a case. *Am J Dig Dis*. 1973;18(2):153–157.

5. Logan RF, Gillon J, Ferrington C, Ferguson A. Reduction of gastrointestinal protein loss by elemental diet in Crohn's disease of the small bowel. *Gut*. 1981;22(5):383–387.
6. Morin CL, Roulet M, Roy CC, Weber A. Continuous elemental enteral alimentation in children with Crohn's disease and growth failure. *Gastroenterology*. 1980;79(6):1205–1210.
7. O'Morain C, Segal AW, Levi AJ. Elemental diet as primary treatment of acute Crohn's disease: a controlled trial. *Br Med J (Clin Res Ed)*. 1984;288(6434):1859–1862.
8. Sanderson IR, Udeen S, Davies PS, et al. Remission induced by an elemental diet in small bowel Crohn's disease. *Arch Dis Child*. 1987;62(2):123–127.
9. Savage MO, Beattie RM, Camacho-Hubner C, et al. Growth in Crohn's disease. *Acta Paediatr Suppl*. 1999;88(428):89–92.
10. Dalwadi H, Bo W, Braun J. Defining new pathogens and nonculturable infectious agents associated with inflammatory bowel disease. *Curr Opin Gastroenterol*. 2000;16(1):56–59.
11. Feeney MA, Murphy F, Clegg AJ, et al. A case-control study of childhood environmental risk factors for the development of inflammatory bowel disease. *Eur J Gastroenterol Hepatol*. 2002;14(5):529–534.
12. Hou JK, Abraham B, El-Serag H. Dietary intake and risk of developing inflammatory bowel disease: a systematic review of the literature. *Am J Gastroenterol*. 2011;106(4):563–573.
13. Albenberg LG, Lewis JD, Wu GD. Food and the gut microbiota in inflammatory bowel diseases: a critical connection. *Curr Opin Gastroenterol*. 2012;28(4):314–320.
14. Gilat T, Hacohen D, Lilos P, Langman MJ. Childhood factors in ulcerative colitis and Crohn's disease. An international cooperative study. *Scand J Gastroenterol*. 1987;22(8):1009–1024.
15. Mahmud N, Weir DG. The urban diet and Crohn's disease: is there a relationship? *Eur J Gastroenterol Hepatol*. 2001;13(2):93–95.
16. Jantchou P, Morois S, Clavel-Chapelon F, et al. Animal protein intake and risk of inflammatory bowel disease: The E3N prospective study. *Am J Gastroenterol*. 2010;105(10):2195–2201.
17. Miller B, Fervers F, Rohbeck R, Strohmeyer G. [Sugar consumption in patients with Crohn's disease]. *Verh Dtsch Ges Inn Med*. 1976;82 Pt 1:922–924.
18. Russel MG, Engels LG, Muris JW, et al. Modern life in the epidemiology of inflammatory bowel disease: a case-control study with special emphasis on nutritional factors. *Eur J Gastroenterol Hepatol*. 1998;10(3):243–249.
19. Thornton JR, Emmett PM, Heaton KW. Diet and ulcerative colitis. *Br Med J*. 1980;280(6210):293–294.
20. Hart AR, Luben R, Olsen A, et al. Diet in the aetiology of ulcerative colitis: a European prospective cohort study. *Digestion*. 2008;77(1):57–64.
21. Gibson PR, Shepherd SJ. Personal view: food for thought—western lifestyle and susceptibility to Crohn's disease. The FODMAP hypothesis. *Aliment Pharmacol Ther*. 2005;21(12):1399–1409.
22. Schrenk D. Dietary fiber, low-molecular-weight food constituents and colo-rectal inflammation in animal models—a review. *Mol Nutr Food Res*. 2009;53(10):1281–1288.
23. Yamamoto T, Nakahigashi M, Saniabadi AR. Review article: diet and inflammatory bowel disease—epidemiology and treatment. *Aliment Pharmacol Ther*. 2009;30(2):99–112.

24. Gentschew L, Ferguson LR. Role of nutrition and microbiota in susceptibility to inflammatory bowel diseases. *Mol Nutr Food Res*. 2012;56(4):524–535.
25. Greenblum S, Turnbaugh PJ, Borenstein E. Metagenomic systems biology of the human gut microbiome reveals topological shifts associated with obesity and inflammatory bowel disease. *Proc Natl Acad Sci USA*. 2012;109(2):594–599.
26. Werner T, Wagner SJ, Martinez I, et al. Depletion of luminal iron alters the gut microbiota and prevents Crohn's disease-like ileitis. *Gut*. 2011;60(3):325–333.
27. Persson PG, Ahlbom A, Hellers G. Diet and inflammatory bowel disease: a case-control study. *Epidemiology*. 1992;3(1):47–52.
28. Reif S, Klein I, Lubin F, et al. Pre-illness dietary factors in inflammatory bowel disease. *Gut*. 1997;40(6):754–760.
29. Tjonneland A, Overvad K, Bergmann MM, et al. Linoleic acid, a dietary n-6 polyunsaturated fatty acid, and the aetiology of ulcerative colitis: a nested case-control study within a European prospective cohort study. *Gut*. 2009;58(12):1606–1611.
30. de Silva PS, Olsen A, Christensen J, et al. An association between dietary arachidonic acid, measured in adipose tissue, and ulcerative colitis. *Gastroenterology*. 2010;139(6):1912–1917.
31. Geerling BJ, Dagnelie PC, Badart-Smook A, et al. Diet as a risk factor for the development of ulcerative colitis. *Am J Gastroenterol*. 2000;95(4):1008–1013.
32. Roediger WE, Heyworth M, Willoughby P, et al. Luminal ions and short chain fatty acids as markers of functional activity of the mucosa in ulcerative colitis. *J Clin Pathol*. 1982;35(3):323–326.
33. Chapman MA, Grahn MF. Ileal and colonic epithelial metabolism in quiescent ulcerative colitis. *Gut*. 1994;35(8):1152–1153.
34. Scheppach W, Muller JG, Boxberger F, et al. Histological changes in the colonic mucosa following irrigation with short-chain fatty acids. *Eur J Gastroenterol Hepatol*. 1997;9(2):163–168.
35. Steinhart AH, Hiruki T, Brzezinski A, Baker JP. Treatment of left-sided ulcerative colitis with butyrate enemas: a controlled trial. *Aliment Pharmacol Ther*. 1996;10(5):729–736.
36. Roediger WE, Duncan A, Kapaniris O, Millard S. Sulphide impairment of substrate oxidation in rat colonocytes: a biochemical basis for ulcerative colitis? *Clin Sci (Lond)*. 1993;85(5):623–627.
37. Pitcher MC, Beatty ER, Cummings JH. The contribution of sulphate reducing bacteria and 5-aminosalicylic acid to faecal sulphide in patients with ulcerative colitis. *Gut*. 2000;46(1):64–72.
38. Harris JE, Lammerding AM. Crohn's disease and Mycobacterium avium subsp. paratuberculosis: current issues. *J Food Prot*. 2001;64(12):2103–2110.
39. Thomas GA, Swift GL, Green JT, et al. Controlled trial of antituberculous chemotherapy in Crohn's disease: a five year follow up study. *Gut*. 1998;42(4):497–500.
40. Gerasimidis K, McGrogan P, Edwards CA. The aetiology and impact of malnutrition in paediatric inflammatory bowel disease. *J Hum Nutr Diet*. 2011;24(4):313–326.
41. Shamir R. Nutritional aspects in inflammatory bowel disease. *J Pediatr Gastroenterol Nutr*. 2009;48(suppl 2):S86–S88.
42. McCartney S, Ballinger A. Growth failure in inflammatory bowel disease. *Nutrition*. 1999;15(2):169–171.
43. Marcovecchio ML, Mohn A, Chiarelli F. Inflammatory cytokines and growth in childhood. *Curr Opin Endocrinol Diabetes Obes*. 2012;19(1):57–62.

44. Wong SC, Macrae VE, McGrogan P, Ahmed SF. The role of pro-inflammatory cytokines in inflammatory bowel disease growth retardation. *J Pediatr Gastroenterol Nutr*. 2006;43(2):144–155.
45. Ling PR, Schwartz JH, Bistrian BR. Mechanisms of host wasting induced by administration of cytokines in rats. *Am J Physiol*. 1997;272(3 Pt 1):E333–E339.
46. Smith BK, Conn CA, Kluger MJ. Experimental cachexia: effects of MCA sarcoma in the Fischer rat. *Am J Physiol*. 1993;265(2 Pt 2):R376–R384.
47. Flores EA, Bistrian BR, Pomposelli JJ, et al. Infusion of tumor necrosis factor/cachectin promotes muscle catabolism in the rat. A synergistic effect with interleukin 1. *J Clin Invest*. 1989;83(5):1614–1622.
48. Winter TA, O'Keefe SJ, Callanan M, Marks T. Impaired gastric acid and pancreatic enzyme secretion in patients with Crohn's disease may be a consequenece of a poor nutritional state. *Inflamm Bowel Dis.* 2004;10(5):618–625.
49. Sawczenko A, Sandhu BK. Presenting features of inflammatory bowel disease in Great Britain and Ireland. *Arch Dis Child*. 2003;88(11):995–1000.
50. Chikenji T, Elwyn DH, Gil KM, et al. Effects of increasing glucose intake on nitrogen balance and energy expenditure in malnourished adult patients receiving parenteral nutrition. *Clin Sci (Lond)*. 1987;72(4):489–501.
51. Macias WL, Alaka KJ, Murphy MH, et al. Impact of the nutritional regimen on protein catabolism and nitrogen balance in patients with acute renal failure. *JPEN J Parenter Enteral Nutr*. 1996;20(1):56–62.
52. Paerregaard A, Uldall Urne F. Anthropometry at the time of diagnosis in Danish children with inflammatory bowel disease. *Acta Paediatr*. 2005;94(11):1682–1683.
53. Gavin J, Anderson CE, Bremner AR, Beattie RM. Energy intakes of children with Crohn's disease treated with enteral nutrition as primary therapy. *J Hum Nutr Diet.* 2005;18(5):337–342.
54. Kirschner BS, Klich JR, Kalman SS, et al. Reversal of growth retardation in Crohn's disease with therapy emphasizing oral nutritional restitution. *Gastroenterology*. 1981;80(1):10–15.
55. Kelts DG, Grand RJ, Shen G, et al. Nutritional basis of growth failure in children and adolescents with Crohn's disease. *Gastroenterology*. 1979;76(4):720–727.
56. Aiges H, Markowitz J, Rosa J, Daum F. Home nocturnal supplemental nasogastric feedings in growth-retarded adolescents with Crohn's disease. *Gastroenterology*. 1989;97(4):905–910.
57. Markowitz J, Grancher K, Rosa J, et al. Growth failure in pediatric inflammatory bowel disease. *J Pediatr Gastroenterol Nutr*. 1993;16(4):373–380.
58. Azcue M, Rashid M, Griffiths A, Pencharz PB. Energy expenditure and body composition in children with Crohn's disease: effect of enteral nutrition and treatment with prednisolone. *Gut.* 1997;41(2):203–208.
59. Ballinger AB, Camacho-Hubner C, Croft NM. Growth failure and intestinal inflammation. *QJM.* 2001;94(3):121–125.
60. Walker-Smith JA. Management of growth failure in Crohn's disease. *Arch Dis Child*. 1996;75(4):351–354.
61. Kanof ME, Lake AM, Bayless TM. Decreased height velocity in children and adolescents before the diagnosis of Crohn's disease. *Gastroenterology*. 1988;95(6):1523–1527.
62. Hildebrand H, Karlberg J, Kristiansson B. Longitudinal growth in children and adolescents with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr.* 1994;18(2):165–173.
63. Thayu M, Shults J, Burnham JM, et al. Gender differences in body composition

deficits at diagnosis in children and adolescents with Crohn's disease. *Inflamm Bowel Dis*. 2007;13(9):1121–1128.
64. Burnham JM, Shults J, Semeao E, et al. Body-composition alterations consistent with cachexia in children and young adults with Crohn disease. *Am J Clin Nutr*. 2005;82(2):413–420.
65. Pappa H, Thayu M, Sylvester F, et al. Skeletal health of children and adolescents with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr*. 2011;53(1):11–25.
66. Schmidt S, Mellstrom D, Norjavaara E, et al. Low bone mineral density in children and adolescents with inflammatory bowel disease: a population-based study from Western Sweden. *Inflamm Bowel Dis*. 2009;15(12):1844–1850.
67. Schmidt S, Mellstrom D, Norjavaara E, et al. Longitudinal assessment of bone mineral density in children and adolescents with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr*. 2012;55(5):511–518.
68. Bailey DA. The Saskatchewan Pediatric Bone Mineral Accrual Study: bone mineral acquisition during the growing years. *Int J Sports Med*. 1997;18(suppl 3):S191–S194.
69. Hill RJ, Brookes DS, Davies PS. Bones in pediatric Crohn's disease: a review of fracture risk in children and adults. *Inflamm Bowel Dis*. 2011;17(5):1223–1228.
70. Compston JE. Review article: osteoporosis, corticosteroids and inflammatory bowel disease. *Aliment Pharmacol Ther*. 1995;9(3):237–250.
71. Bernstein CN, Blanchard JF, Leslie W, et al. The incidence of fracture among patients with inflammatory bowel disease. A population-based cohort study. *Ann Intern Med*. 2000;133(10):795–799.
72. Arden NK, Major P, Poole JR, et al. Size at birth, adult intestinal calcium absorption and 1,25(OH)(2) vitamin D. *QJM*. 2002;95(1):15–21.
73. Schoon EJ, van Nunen AB, Wouters RS, et al. Osteopenia and osteoporosis in Crohn's disease: prevalence in a Dutch population-based cohort. *Scand J Gastroenterol Suppl*. 2000(232):43–47.
74. Herzog D, Bishop N, Glorieux F, Seidman EG. Interpretation of bone mineral density values in pediatric Crohn's disease. *Inflamm Bowel Dis*. 1998;4(4):261–267.
75. Gokhale R, Favus MJ, Karrison T, et al. Bone mineral density assessment in children with inflammatory bowel disease. *Gastroenterology*. 1998;114(5):902–911.
76. Picado C, Luengo M. Corticosteroid-induced bone loss. Prevention and management. *Drug Saf*. 1996;15(5):347–359.
77. Ahmed SF, Horrocks IA, Patterson T, et al. Bone mineral assessment by dual energy X-ray absorptiometry in children with inflammatory bowel disease: evaluation by age or bone area. *J Pediatr Gastroenterol Nutr*. 2004;38(3):276–280.
78. Thearle M, Horlick M, Bilezikian JP, et al. Osteoporosis: an unusual presentation of childhood Crohn's disease. *J Clin Endocrinol Metab*. 2000;85(6):2122–2126.
79. Levin AD, Wadhera V, Leach ST, et al. Vitamin D deficiency in children with inflammatory bowel disease. *Dig Dis Sci*. 2011;56(3):830–836.
80. Garg M, Lubel JS, Sparrow MP, et al. Review article: vitamin D and inflammatory bowel disease—established concepts and future directions. *Aliment Pharmacol Ther*. 2012;36(4):324–344.
81. Abreu MT, Geller JL, Vasiliauskas EA, et al. Treatment with infliximab is associated with increased markers of bone formation in patients with Crohn's disease. *J Clin Gastroenterol*. 2006;40(1):55–63.

82. Pazianas M, Rhim AD, Weinberg AM, et al. The effect of anti- TNF-alpha therapy on spinal bone mineral density in patients with Crohn's disease. *Ann N Y Acad Sci.* 2006;1068:543–556.
83. Dear KL, Compston JE, Hunter JO. Treatments for Crohn's disease that minimise steroid doses are associated with a reduced risk of osteoporosis. *Clin Nutr*. 2001;20(6):541–546.
84. Habtezion A, Silverberg MS, Parkes R, et al. Risk factors for low bone density in Crohn's disease. *Inflamm Bowel Dis*. 2002;8(2):87–92.
85. Walther F, Fusch C, Radke M, et al. Osteoporosis in pediatric patients suffering from chronic inflammatory bowel disease with and without steroid treatment. *J Pediatr Gastroenterol Nutr*. 2006;43(1):42–51.
86. Ghishan FK, Kiela PR. Advances in the understanding of mineral and bone metabolism in inflammatory bowel diseases. *Am J Physiol Gastrointest Liver Physiol*. 2011;300(2):G191–G201.
87. Jahnsen J, Falch JA, Mowinckel P, Aadland E. Vitamin D status, parathyroid hormone and bone mineral density in patients with inflammatory bowel disease. *Scand J Gastroenterol*. 2002;37(2):192–199.
88. Quercia RA, Korn S, O'Neill D, et al. Selenium deficiency and fatal cardiomyopathy in a patient receiving long-term home parenteral nutrition. *Clin Pharm*. 1984;3(5):531–535.
89. Filippi J, Al Jaouni R, Wiroth JB, et al. Nutritional deficiencies in patients with Crohn's disease in remission. *Inflamm Bowel Dis.* 2006;12(3):185–191.
90. Chowers Y, Sela BA, Holland R, et al. Increased levels of homocysteine in patients with Crohn's disease are related to folate levels. *Am J Gastroenterol*. 2000;95(12):3498–3502.
91. Mahmud N, Molloy A, McPartlin J, et al. Increased prevalence of methylenetetrahydrofolate reductase C677T variant in patients with inflammatory bowel disease, and its clinical implications. *Gut*. 1999;45(3):389–394.
92. Bousvaros A, Zurakowski D, Duggan C, et al. Vitamins A and E serum levels in children and young adults with inflammatory bowel disease: effect of disease activity. *J Pediatr Gastroenterol Nutr*. 1998;26(2):129–135.
93. Geerling BJ, Badart-Smook A, Stockbrugger RW, Brummer RJ. Comprehensive nutritional status in patients with long-standing Crohn disease currently in remission [see comments]. *Am J Clin Nutr.* 1998;67(5):919–926.
94. Fernandez-Banares F, Mingorance MD, Esteve M, et al. Serum zinc, copper, and selenium levels in inflammatory bowel disease: effect of total enteral nutrition on trace element status. *Am J Gastroenterol*. 1990;85(12):1584–1589.
95. Thomas C, Thomas L. Biochemical markers and hematologic indices in the diagnosis of functional iron deficiency. *Clin Chem*. 2002;48(7):1066–1076.
96. Maherzi A, Galan P, Cezard JP, et al. Assessment of iron status in children and adolescents with Crohn's disease: value of basic red cell ferritin. *Ann Nutr Metab*. 1996;40(6):331–335.
97. Oldenburg B, Koningsberger JC, Van Berge Henegouwen GP, et al. Iron and inflammatory bowel disease. *Aliment Pharmacol Ther*. 2001;15(4):429–438.
98. Gasche C, Dejaco C, Waldhoer T, et al. Intravenous iron and erythropoietin for anemia associated with Crohn's disease. A randomized, controlled trial. *Ann Intern Med*. 1997;126(10):782–787.
99. Schreiber S, Howaldt S, Schnoor M, et al. Recombinant erythropoietin for the treatment of anemia in inflammatory bowel disease [see comments]. *N Engl J Med.* 1996;334(10):619–623.
100. Sturniolo GC, Di LV, Ferronato A, et al. Zinc supplementation tightens "leaky

gut" in Crohn's disease. *Inflamm Bowel Dis.* 2001;7(2):94–98.
101. Griffin IJ, Kim SC, Hicks PD, et al. Zinc metabolism in adolescents with Crohn's disease. *Pediatr Res.* 2004;56(2):235–239.
102. Reimund JM, Hirth C, Koehl C, et al. Antioxidant and immune status in active Crohn's disease. A possible relationship. *Clin Nutr*. 2000;19(1):43–48.
103. Trebble TM, Wootton SA, May A, et al. Essential fatty acid status in paediatric Crohn's disease: relationship with disease activity and nutritional status. *Aliment Pharmacol Ther.* 2003;18(4):433–442.
104. Dickinson RJ, Ashton MG, Axon AT, et al. Controlled trial of intravenous hyperalimentation and total bowel rest as an adjunct to the routine therapy of acute colitis. *Gastroenterology*. 1980;79(6):1199–1204.
105. McIntyre PB, Powell-Tuck J, Wood SR, et al. Controlled trial of bowel rest in the treatment of severe acute colitis. *Gut.* 1986;27(5):481–485.
106. Seidman EG. Nutritional management of inflammatory bowel disease. *Gastroenterol Clin North Am*. 1989;18(1):129–155.
107. Spray C, Debelle GD, Murphy MS. Current diagnosis, management and morbidity in paediatric inflammatory bowel disease. *Acta Paediatr.* 2001;90(4):400–405.
108. Powell-Tuck J. Protein metabolism in inflammatory bowel disease. *Gut*. 1986;27 Suppl 1:67–71.
109. Gonzalez-Huix F, Fernandez-Banares F, Esteve-Comas M, et al. Enteral versus parenteral nutrition as adjunct therapy in acute ulcerative colitis. *Am J Gastroenterol*. 1993;88(2):227–232.
110. Hallert C, Kaldma M, Petersson BG. Ispaghula husk may relieve gastrointestinal symptoms in ulcerative colitis in remission. *Scand J Gastroenterol*. 1991;26(7):747–750.
111. Fuchs CS, Giovannucci EL, Colditz GA, et al. Dietary fiber and the risk of colorectal cancer and adenoma in women. *N Engl J Med*. 1999;340(3):169–176.
112. Sanchez-Fidalgo S, Cardeno A, Sanchez-Hidalgo M, et al. Dietary unsaponificable fraction from extra virgin olive oil supplementation attenuates acute ulcerative colitis in mice. *Eur J Pharm Sci*. 2013;48(3):572–581.
113. Beattie RM, Schiffrin EJ, Donnet-Hughes A, et al. Polymeric nutrition as the primary therapy in children with small bowel Crohn's disease. *Aliment Pharmacol Ther.* 1994;8(6):609–615.
114. Fell JM, Paintin M, Arnaud-Battandier F, et al. Mucosal healing and a fall in mucosal pro-inflammatory cytokine mRNA induced by a specific oral polymeric diet in paediatric Crohn's disease. *Aliment Pharmacol Ther.* 2000;14(3):281–289.
115. Ruuska T, Savilahti E, Maki M, et al. Exclusive whole protein enteral diet versus prednisolone in the treatment of acute Crohn's disease in children. *J Pediatr Gastroenterol Nutr*. 1994;19(2):175–180.
116. Seidman E, Griffiths A, Jones A, Issenman R. Semi-elemental diet vs prednisolone in pediatric Crohn's disease. *Gastroenterology*. 1993;104(4):778.
117. Day AS, Whitten KE, Lemberg DA, et al. Exclusive enteral feeding as primary therapy for Crohn's disease in Australian children and adolescents: a feasible and effective approach. *J Gastroenterol Hepatol.* 2006;21(10):1609–1614.
118. Knight C, El-Matary W, Spray C, Sandhu BK. Long-term outcome of nutritional therapy in paediatric Crohn's disease. *Clin Nutr*. 2005;24(5):775–779.
119. Lochs H, Steinhardt HJ, Klaus-Wentz B, et al. Comparison of enteral nutrition and drug treatment in active Crohn's disease. Results of the European Cooperative Crohn's Disease Study. IV [see comments]. *Gastroenterology*. 1991;101(4):881–888.
120. Malchow H, Ewe K, Brandes JW, et al. European Cooperative Crohn's Disease

Study (ECCDS): results of drug treatment. *Gastroenterology*. 1984;86(2):249–266.
121. Afzal NA, Davies S, Paintin M, et al. Colonic Crohn's disease in children does not respond well to treatment with enteral nutrition if the ileum is not involved. *Dig Dis Sci*. 2005;50(8):1471–1475.
122. Lim S, Dohil R, Meadows N, et al. Treatment of orofacial and ileo-colonic Crohn's disease with total enteral nutrition [In Process Citation]. *J R Soc Med*. 1998;91(9):489–490.
123. Afzal NA, Addai S, Fagbemi A, et al. Refeeding syndrome with enteral nutrition in children: a case report, literature review and clinical guidelines. *Clin Nutr*. 2002;21(6):515–520.
124. Dempsey DT, Mullen JL, Buzby GP. The link between nutritional status and clinical outcome: can nutritional intervention modify it? *Am J Clin Nutr*. 1988;47(2 suppl):352–356.
125. Klein S, Simes J, Blackburn GL. Total parenteral nutrition and cancer clinical trials. *Cancer*. 1986;58(6):1378–1386.
126. Shiloni E, Coronado E, Freund HR. Role of total parenteral nutrition in the treatment of Crohn's disease. *Am J Surg*. 1989;157(1):180–185.
127. Lashner BA, Evans AA, Hanauer SB. Preoperative total parenteral nutrition for bowel resection in Crohn's disease. *Dig Dis Sci*. 1989;34(5):741–746.
128. Rombeau JL, Barot LR, Williamson CE, Mullen JL. Preoperative total parenteral nutrition and surgical outcome in patients with inflammatory bowel disease. *Am J Surg*. 1982;143(1):139–143.
129. Jacobson S, Kallner A. Effect of total parenteral nutrition on serum concentrations of eight proteins in Crohn's disease. *Am J Gastroenterol*. 1984;79(7):501–505.
130. Loftus EV Jr, Sandborn WJ. Epidemiology of inflammatory bowel disease. *Gastroenterol Clin North Am*. 2002;31(1):1–20.
131. Puntis J, McNeish AS, Allan RN. Long term prognosis of Crohn's disease with onset in childhood and adolescence. *Gut*. 1984;25(4):329–336.
132. Beier-Holgersen R, Boesby S. Influence of postoperative enteral nutrition on postsurgical infections. *Gut*. 1996;39(6):833–835.
133. Perioperative total parenteral nutrition in surgical patients. The Veterans Affairs Total Parenteral Nutrition Cooperative Study Group. *N Engl J Med*. 1991;325(8):525–532.
134. Chandra RK. Interactions of nutrition, infection and immune response. Immunocompetence in nutritional deficiency, methodological considerations and intervention strategies. *Acta Paediatr Scand*. 1979;68(1):137–144.
135. Seidman E, LeLeiko N, Ament M, et al. Nutritional issues in pediatric inflammatory bowel disease. *J Pediatr Gastroenterol Nutr*. 1991;12(4):424–438.
136. Anderson DL, Boyce HW Jr. Use of parenteral nutrition in treatment of advanced regional enteritis. *Am J Dig Dis*. 1973;18(8):633–640.
137. Fischer JE, Foster GS, Abel RM, et al. Hyperalimentation as primary therapy for inflammatory bowel disease. *Am J Surg*. 1973;125(2):165–175.
138. Layden T, Rosenberg F, Nemchausky G, et al. Reversal of growth arrest in adolescents with Crohn's disease after parenteral alimentation. *Gastroenterology*. 1976;70(6):1017–1021.
139. Wright RA, Adler EC. Peripheral parenteral nutrition is no better than enteral nutrition in acute exacerbation of Crohn's disease: a prospective trial. *J Clin Gastroenterol*. 1990;12(4):396–399.
140. Deitch EA, Xu D, Naruhn MB, et al. Elemental diet and IV- TPN-induced bacterial translocation is associated with loss of intestinal mucosal barrier function

against bacteria [see comments]. *Ann Surg*. 1995;221(3):299–307.
141. Greenberg GR, Fleming CR, Jeejeebhoy KN, et al. Controlled trial of bowel rest and nutritional support in the management of Crohn's disease. *Gut*. 1988;29(10):1309–1315.
142. Fuchigami T, Ohgushi H, Imamura K, et al. Effects of total parenteral nutrition on colonic lesions in Crohn's disease: radiographic and endoscopic study. *Gastroenterol Jpn*. 1982;17(6):521–529.
143. Himal HS, Allard JR, Nadeau JE, et al. The importance of adequate nutrition in closure of small intestinal fistulas. *Br J Surg*. 1974;61(9):724–726.
144. Spiliotis J, Briand D, Gouttebel MC, et al. Treatment of fistulas of the gastrointestinal tract with total parenteral nutrition and octreotide in patients with carcinoma. *Surg Gynecol Obstet*. 1993;176(6):575–580.
145. Howard L, Heaphey L, Fleming CR, et al. Four years of North American registry home parenteral nutrition outcome data and their implications for patient management. *JPEN J Parenter Enteral Nutr*. 1991;15(4):384–393.
146. Greenstein JP, Birnbaum SM, Winitz M, Otey M. Quantitative nutritional studies with water soluble chemically defined diets. 1. Growth, reproduction and lactation in rats. *Arch Biochem Biophys*. 1957;72:396–416.
147. Voitk AJ, Echave V, Feller JH, et al. Experience with elemental diet in the treatment of inflammatory bowel disease. Is this primary therapy? *Arch Surg*. 1973;107(2):329–333.
148. Navarro J, Vargas J, Cezard JP, et al. Prolonged constant rate elemental enteral nutrition in Crohn's disease. *J Pediatr Gastroenterol Nutr*. 1982;1(4):541–546.
149. O'Morain C. Crohn's disease treated by elemental diet. *J R Soc Med*. 1982;75(2):135–136.
150. Morin CL, Roulet M, Roy CC, et al. Continuous elemental enteral alimentation in the treatment of children and adolescents with Crohn's disease. *JPEN J Parenter Enteral Nutr*. 1982;6(3):194–199.
151. Akobeng AK, Miller V, Stanton J, et al. Double-blind randomized controlled trial of glutamine-enriched polymeric diet in the treatment of active Crohn's disease. *J Pediatr Gastroenterol Nutr*. 2000;30(1):78–84.
152. Silk DB, Fairclough PD, Clark ML, et al. Use of a peptide rather than free amino acid nitrogen source in chemically defined "elemental" diets. *JPEN J Parenter Enteral Nutr*. 1980;4(6):548–553.
153. Raouf AH, Hildrey V, Daniel J, et al. Enteral feeding as sole treatment for Crohn's disease: controlled trial of whole protein v amino acid based feed and a case study of dietary challenge. *Gut*. 1991;32(6):702–707.
154. Beattie RM, Walker-Smith JA. Treatment of active Crohn's disease by exclusion diet. *J Pediatr Gastroenterol Nutr*. 1994;19(1):135–136.
155. Borrelli O, Cordischi L, Cirulli M, et al. Polymeric diet alone versus corticosteroids in the treatment of active pediatric Crohn's disease: a randomized controlled open-label trial. *Clin Gastroenterol Hepatol*. 2006;4(6):744–753.
156. Hyams JS, Ferry GD, Mandel FS, et al. Development and validation of a pediatric Crohn's disease activity index. *J Pediatr Gastroenterol Nutr*. 1991;12(4):439–447.
157. Breese EJ, Michie CA, Nicholls SW, et al. The effect of treatment on lymphokine-secreting cells in the intestinal mucosa of children with Crohn's disease. *Aliment Pharmacol Ther*. 1995;9(5):547–552.
158. Bannerjee K, Camacho-Hubner C, Babinska K, et al. Anti-inflammatory and growth-stimulating effects precede nutritional restitution during enteral feeding in Crohn disease. *J Pediatr Gastroenterol Nutr*. 2004;38(3):270–275.

159. Gassull MA, Fernandez-Banares F, Cabre E, et al. Fat composition may be a clue to explain the primary therapeutic effect of enteral nutrition in Crohn's disease: results of a double blind randomised multicentre European trial. *Gut*. 2002;51(2):164–168.
160. Miura S, Tsuzuki Y, Hokari R, Ishii H. Modulation of intestinal immune system by dietary fat intake: relevance to Crohn's disease. *J Gastroenterol Hepatol*. 1998;13(12):1183–1190.
161. Sakurai T, Matsui T, Yao T, et al. Short-term efficacy of enteral nutrition in the treatment of active Crohn's disease: a randomized, controlled trial comparing nutrient formulas. *JPEN J Parenter Enteral Nutr*. 2002;26(2):98–103.
162. Leiper K, Woolner J, Mullan MM, et al. A randomised controlled trial of high versus low long chain triglyceride whole protein feed in active Crohn's disease. *Gut*. 2001;49(6):790–794.
163. von Tirpitz C, Kohn C, Steinkamp M, et al. Lactose intolerance in active Crohn's disease: clinical value of duodenal lactase analysis. *J Clin Gastroenterol*. 2002;34(1):49–53.
164. Ludvigsson JF, Krantz M, Bodin L, et al. Elemental versus polymeric enteral nutrition in paediatric Crohn's disease: a multicentre randomized controlled trial. *Acta Paediatr*. 2004;93(3):327–335.
165. Zachos M, Tondeur M, Griffiths AM. Enteral nutritional therapy for induction of remission in Crohn's disease. *Cochrane Database Syst Rev*. 2007(1):CD000542.
166. Gonzalez-Huix F, de Leon R, Fernandez-Banares F, et al. Polymeric enteral diets as primary treatment of active Crohn's disease: a prospective steroid controlled trial. *Gut*. 1993;34(6):778–782.
167. Malchow H, Steinhardt HJ, Lorenz-Meyer H, et al. Feasibility and effectiveness of a defined-formula diet regimen in treating active Crohn's disease. European Cooperative Crohn's Disease Study III. *Scand J Gastroenterol*. 1990;25(3):235–244.
168. Griffiths AM, Ohlsson A, Sherman PM, Sutherland LR. Meta-analysis of enteral nutrition as a primary treatment of active Crohn's disease. *Gastroenterology*. 1995;108(4):1056–1067.
169. Modigliani R, Mary JY, Simon JF, et al. Clinical, biological, and endoscopic picture of attacks of Crohn's disease. Evolution on prednisolone. Groupe d'Etude Therapeutique des Affections Inflammatoires Digestives. *Gastroenterology*. 1990;98(4):811–818.
170. Zachos M, Tondeur M, Griffiths AM. Enteral nutritional therapy for inducing remission of Crohn's disease. *Cochrane Database Syst Rev.* 2001(3):CD000542.
171. Heuschkel RB, Menache CC, Megerian JT, Baird AE. Enteral nutrition and corticosteroids in the treatment of acute Crohn's disease in children. *J Pediatr Gastroenterol Nutr*. 2000;31(1):8–15.
172. Dziechciarz P, Horvath A, Shamir R, Szajewska H. Meta-analysis: enteral nutrition in active Crohn's disease in children. *Aliment Pharmacol Ther*. 2007;26(6):795–806.
173. Markowitz J, Hyams J, Mack D, et al. Corticosteroid therapy in the age of infliximab: acute and 1-year outcomes in newly diagnosed children with Crohn's disease. *Clin Gastroenterol Hepatol.* 2006;4(9):1124–1129.
174. Murch SH, Walker-Smith JA. Nutrition in inflammatory bowel disease. *Baillieres Clin Gastroenterol*. 1998;12(4).
175. Rigaud D, Cosnes J, Le Quintrec Y, et al. Controlled trial comparing two types of enteral nutrition in treatment of active Crohn's disease: elemental versus polymeric diet. *Gut.* 1991;32(12):1492–1497.

176. Thomas AG, Taylor F, Miller V. Dietary intake and nutritional treatment in childhood Crohn's disease. *J Pediatr Gastroenterol Nutr*. 1993;17(1):75–81.
177. Buchanan E, Gaunt WW, Cardigan T, et al. The use of exclusive enteral nutrition for induction of remission in children with Crohn's disease demonstrates that disease phenotype does not influence clinical remission. *Aliment Pharmacol Ther*. 2009;30(5):501–507.
178. Riordan AM, Hunter JO, Cowan RE, et al. Treatment of active Crohn's disease by exclusion diet: East Anglian multicentre controlled trial [see comments]. *Lancet*. 1993;342(8880):1131–1134.
179. Pearson M, Teahon K, Levi AJ, Bjarnason I. Food intolerance and Crohn's disease. *Gut*. 1993;34(6):783–787.
180. Polk DB, Hattner JA, Kerner JA Jr. Improved growth and disease activity after intermittent administration of a defined formula diet in children with Crohn's disease. *JPEN J Parenter Enteral Nutr*. 1992;16(6):499–504.
181. Levenstein S, Prantera C, Luzi C, D'Ubaldi A. Low residue or normal diet in Crohn's disease: a prospective controlled study in Italian patients. *Gut*. 1985;26(10):989–993.
182. Wilschanski M, Sherman P, Pencharz P, et al. Supplementary enteral nutrition maintains remission in paediatric Crohn's disease. *Gut*. 1996;38(4):543–548.
183. Verma S, Brown S, Kirkwood B, Giaffer MH. Polymeric versus elemental diet as primary treatment in active Crohn's disease: a randomized, double-blind trial. *Am J Gastroenterol.* 2000;95(3):735–739.
184. Takagi S, Utsunomiya K, Kuriyama S, et al. Effectiveness of an "half elemental diet" as maintenance therapy for Crohn's disease: a randomized-controlled trial. *Aliment Pharmacol Ther*. 2006;24(9): 1333–1340.
185. Endres S, Ghorbani R, Kelley VE, et al. The effect of dietary supplementation with n-3 polyunsaturated fatty acids on the synthesis of interleukin-1 and tumor necrosis factor by mononuclear cells. *N Engl J Med*. 1989;320(5):265–271.
186. Jaschonek K, Clemens MR, Scheurlen M. Decreased responsiveness of platelets to a stable prostacyclin analogue in patients with Crohn's disease. Reversal by n-3 polyunsaturated fatty acids. *Thromb Res*. 1991;63(6):667–672.
187. Wakefield AJ, Sawyerr AM, Dhillon AP, et al. Pathogenesis of Crohn's disease: multifocal gastrointestinal infarction [see comments]. *Lancet*. 1989;2(8671):1057–1062.
188. Belluzzi A, Brignola C, Campieri M, et al. Effect of an enteric-coated fish-oil preparation on relapses in Crohn's disease [see comments]. *N Engl J Med.* 1996;334(24):1557–1560.
189. Middleton SJ, Naylor S, Woolner J, Hunter JO. A double-blind, randomized, placebo-controlled trial of essential fatty acid supplementation in the maintenance of remission of ulcerative colitis. *Aliment Pharmacol Ther*. 2002;16(6):1131–1135.
190. Lofberg R, Rutgeerts P, Malchow H, et al. Budesonide prolongs time to relapse in ileal and ileocaecal Crohn's disease. A placebo controlled one year study. *Gut*. 1996;39(1):82–86.
191. Romano C, Cucchiara S, Barabino A, et al. Usefulness of omega-3 fatty acid supplementation in addition to mesalazine in maintaining remission in pediatric Crohn's disease: a double-blind, randomized, placebo-controlled study. *World J Gastroenterol.* 2005;11(45):7118–7121.
192. Lomer MC, Harvey RS, Evans SM, et al. Efficacy and tolerability of a low microparticle diet in a double blind, randomized, pilot study in Crohn's disease. *Eur J Gastroenterol Hepatol*. 2001;13(2):101–106.

193. Ritchie JK, Wadsworth J, Lennard-Jones JE, Rogers E. Controlled multicentre therapeutic trial of an unrefined carbohydrate, fibre rich diet in Crohn's disease. *Br Med J (Clin Res Ed)*. 1987;295(6597):517–520.
194. Barclay GR, McKenzie H, Pennington J, et al. The effect of dietary yeast on the activity of stable chronic Crohn's disease. *Scand J Gastroenterol*. 1992;27(3):196–200.
195. Gibson GR. Dietary modulation of the human gut microflora using the prebiotics oligofructose and inulin. *J Nutr*. 1999;129(7 suppl):1438S–1441S.
196. Gionchetti P, Rizzello F, Venturi A, et al. Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis: a double-blind, placebo-controlled trial. *Gastroenterology*. 2000;119(2):305–309.
197. Gupta P, Andrew H, Kirschner BS, Guandalini S. Is lactobacillus GG helpful in children with Crohn's disease? Results of a preliminary, open-label study. *J Pediatr Gastroenterol Nutr.* 2000;31(4):453–457.
198. Israel DM, Hassall E. Prolonged use of gastrostomy for enteral hyperalimentation in children with Crohn's disease. *Am J Gastroenterol*. 1995;90(7):1084–1088.
199. Motil KJ, Grand RJ, Maletskos CJ, Young VR. The effect of disease, drug, and diet on whole body protein metabolism in adolescents with Crohn disease and growth failure. *J Pediatr*. 1982;101(3):345–351.
200. Sawczenko A, Azooz O, Paraszczuk J, et al. Intestinal inflammation-induced growth retardation acts through IL-6 in rats and depends on the -174 IL-6 G/C polymorphism in children. *Proc Natl Acad Sci USA*. 2005;102(37):13260–13265.
201. Sawczenko A, Ballinger AB, Savage MO, Sanderson IR. Clinical features affecting final adult height in patients with pediatric-onset Crohn's disease. *Pediatrics*. 2006;118(1):124–129.
202. Motil KJ, Grand RJ, Davis-Kraft L, et al. Growth failure in children with inflammatory bowel disease: a prospective study. *Gastroenterology*. 1993;105(3):681–691.
203. Griffiths AM, Nguyen P, Smith C, et al. Growth and clinical course of children with Crohn's disease. *Gut*. 1993;34(7):939–943.
204. Thomas AG, Holly JM, Taylor F, Miller V. Insulin like growth factor-I, insulin like growth factor binding protein-1, and insulin in childhood Crohn's disease. *Gut*. 1993;34(7):944–947.
205. Thissen JP, Underwood LE, Ketelslegers JM. Regulation of insulin-like growth factor-I in starvation and injury. *Nutr Rev.* 1999;57(6):167–176.
206. Beattie RM, Camacho-Hubner C, Wacharasindhu S, et al. Responsiveness of IGF-I and IGFBP-3 to therapeutic intervention in children and adolescents with Crohn's disease. *Clin Endocrinol (Oxf)*. 1998;49(4):483–489.
207. Davies G, Evans CM, Shand WS, Walker-Smith JA. Surgery for Crohn's disease in childhood: influence of site of disease and operative procedure on outcome. *Br J Surg.* 1990;77(8):891–894.
208. Varille V, Cezard JP, de Lagausie P, et al. Resting energy expenditure before and after surgical resection of gut lesions in pediatric Crohn's disease. *J Pediatr Gastroenterol Nutr*. 1996;23(1):13–19.
209. Murch SH, Lamkin VA, Savage MO, et al. Serum concentrations of tumour necrosis factor alpha in childhood chronic inflammatory bowel disease. *Gut*. 1991;32(8):913–917.
210. Ballinger AB, Azooz O, El-Haj T, et al. Growth failure occurs through a decrease in insulin-like growth factor 1 which is independent of undernutrition in a rat model of colitis. *Gut*. 2000;46(5):694–700.
211. De Benedetti F, Alonzi T, Moretta A, et al. Interleukin 6 causes growth

impairment in transgenic mice through a decrease in insulin-like growth factor-I. A model for stunted growth in children with chronic inflammation. *J Clin Invest*. 1997;99(4):643–650.
212. Chong SK, Grossman A, Walker-Smith JA, Rees LH. Endocrine dysfunction in children with Crohn's disease. *J Pediatr Gastroenterol Nutr*. 1984;3(4):529–534.
213. Farthing MJ, Campbell CA, Walker-Smith J, et al. Nocturnal growth hormone and gonadotrophin secretion in growth retarded children with Crohn's disease. *Gut*. 1981;22(11):933–938.
214. McCaffery TD Jr, Nasr K, Lawrence AM, Kirsner JB. Effect of administered human growth hormone on growth retardation in inflammatory bowel disease. *Am J Dig Dis*. 1974;19(5):411–416.
215. Mingrone G, Greco AV, Benedetti G, et al. Increased resting lipid oxidation in Crohn's disease. *Dig Dis Sci*. 1996;41(1):72–76.
216. Koniaris SG, Fisher SE, Rubin CT, Chawla A. Experimental colitis impairs linear bone growth independent of nutritional factors. *J Pediatr Gastroenterol Nutr*. 1997;25(2):137–141.
217. Braegger CP, Nicholls S, Murch SH, et al. Tumour necrosis factor alpha in stool as a marker of intestinal inflammation [see comments]. *Lancet*. 1992;339(8785):89–91.
218. Beeken WL, Busch HJ, Sylwester DL. Intestinal protein loss in Crohn's disease. *Gastroenterology*. 1972;62(2):207–215.
219. Moshage H. Cytokines and the hepatic acute phase response. *J Pathol*. 1997;181(3):257–266.
220. Gersovitz M, Munro HN, Udall J, Young VR. Albumin synthesis in young and elderly subjects using a new stable isotope methodology: response to level of protein intake. *Metabolism*. 1980;29(11):1075–1086.
221. Stelzner M, Somasundaram S, Khakberdiev T. Systemic effects of acute terminal ileitis on uninflammed gut aggravates bile acid malabsorption. *J Surg Res*. 2001;99(2):359–364.
222. Schoelmerich J, Becher MS, Hoppe-Seyler P, et al. Zinc and vitamin A deficiency in patients with Crohn's disease is correlated with activity but not with localization or extent of the disease. *Hepatogastroenterology*. 1985;32(1):34–38.
223. Markowitz J, Daum F. Growth impairment in pediatric inflammatory bowel disease. *Am J Gastroenterol*. 1994;89(3):319–326.
224. Alemzadeh N, Rekers-Mombarg LT, Mearin ML, et al. Adult height in patients with early onset of Crohn's disease. *Gut*. 2002;51(1):26–29.
225. Mushtaq T, Ahmed SF. The impact of corticosteroids on growth and bone health. *Arch Dis Child*. 2002;87(2):93–96.
226. Jux C, Leiber K, Hugel U, et al. Dexamethasone impairs growth hormone (GH)-stimulated growth by suppression of local insulin-like growth factor (IGF)-I production and expression of GH- and IGF-I-receptor in cultured rat chondrocytes. *Endocrinology*. 1998;139(7):3296–3305.
227. Gabrielsson BG, Carmignac DF, Flavell DM, Robinson IC. Steroid regulation of growth hormone (GH) receptor and GH-binding protein messenger ribonucleic acids in the rat. *Endocrinology*. 1995;136(1):209–217.
228. Motil KJ, Grand RJ. Nutritional management of inflammatory bowel disease. *Pediatr Clin North Am*. 1985;32(2):447–469.
229. Peach S, Lock MR, Katz D, et al. Mucosal-associated bacterial flora of the intestine in patients with Crohn's disease and in a control group. *Gut*. 1978;19(11):1034–1042.
230. Axelsson CK, Justesen T. Studies of the duodenal and fecal flora in

gastrointestinal disorders during treatment with an elemental diet. *Gastroenterology*. 1977;72(3):397–401.
231. Swidsinski A, Ladhoff A, Pernthaler A, et al. Mucosal flora in inflammatory bowel disease. *Gastroenterology*. 2002;122(1):44–54.
232. Rutgeerts P, Hiele M, Geboes K, et al. Controlled trial of metronidazole treatment for prevention of Crohn's recurrence after ileal resection [see comments]. *Gastroenterology*. 1995;108(6):1617–1621.
233. Pourtaghi N, Radvar M, Mooney J, Kinane DF. The effect of subgingival antimicrobial therapy on the levels of stromelysin and tissue inhibitor of metalloproteinases in gingival crevicular fluid. *J Periodontol*. 1996;67(9):866–870.
234. Linskens RK, Huijsdens XW, Savelkoul PH, et al. The bacterial flora in inflammatory bowel disease: current insights in pathogenesis and the influence of antibiotics and probiotics. *Scand J Gastroenterol Suppl*. 2001(234):29–40.
235. Kishi D, Takahashi I, Kai Y, et al. Alteration of V beta usage and cytokine production of CD4+ TCR beta beta homodimer T cells by elimination of Bacteroides vulgatus prevents colitis in TCR alpha-chain-deficient mice. *J Immunol.* 2000;165(10):5891–5899.
236. Tanaka S, Miura S, Kimura H, et al. Elemental diet reduces progression of chronic inflammation and alters nitric oxide metabolism in a rat model of granulomatous colitis. *Gastroenterology*. 1999;106:A780.
237. Tanaka S, Miura S, Kimura H, et al. Amelioration of chronic inflammation by ingestion of elemental diet in a rat model of granulomatous enteritis. *Dig Dis Sci*. 1997;42(2):408–419.
238. Koster F, Pierce NF. Effect of protein deprivation on immunoregulatory cells in the rat mucosal immune response. *Clin Exp Immunol*. 1985;60(1):217–224.
239. Lamont AG, Gordon M, Ferguson A. Oral tolerance in protein-deprived mice. II. Evidence of normal "gut processing" of ovalbumin, but suppressor cell deficiency, in deprived mice. *Immunology*. 1987;61(3):339–343.
240. Sanderson IR. Dietary regulation of genes expressed in the developing intestinal epithelium [In Process Citation]. *Am J Clin Nutr*. 1998;68(5):999–1005.
241. Nishimura A, Fujimoto M, Oguchi S, et al. Short-chain fatty acids regulate IGF-binding protein secretion by intestinal epithelial cells. *Am J Physiol*. 1998;275(1 Pt 1):E55–E63.
242. Fusunyan RD, Quinn JJ, Ohno Y, et al. Butyrate enhances interleukin (IL)-8 secretion by intestinal epithelial cells in response to IL-1beta and lipopolysaccharide. *Pediatr Res*. 1998;43(1):84–90.
243. Ohtsuka Y, Lee J, Stamm DS, Sanderson IR. MIP-2 secreted by epithelial cells increases neutrophil and lymphocyte recruitment in the mouse intestine. *Gut*. 2001;49(4):526–533.
244. Kulkarni AB, Karlsson S. Transforming growth factor-beta 1 knockout mice. A mutation in one cytokine gene causes a dramatic inflammatory disease. *Am J Pathol*. 1993;143(1):3–9.
245. Wright NA, Poulsom R, Stamp G, et al. Trefoil peptide gene expression in gastrointestinal epithelial cells in inflammatory bowel disease. *Gastroenterology*. 1993;104(1):12–20.
246. Babyatsky MW, deBeaumont M, Thim L, Podolsky DK. Oral trefoil peptides protect against ethanol- and indomethacin-induced gastric injury in rats [see comments]. *Gastroenterology*. 1996;110(2):489–497.
247. Beattie RM, Bentsen BS, MacDonald TT. Childhood Crohn's disease and the efficacy of enteral diets. *Nutrition.* 1998;14(4):345–350.

248. Kirschner BS. Nutritional consequences of inflammatory bowel disease on growth. *J Am Coll Nutr*. 1988;7(4):301–308.
249. Kirschner BS, Sutton MM. Somatomedin-C levels in growth-impaired children and adolescents with chronic inflammatory bowel disease. *Gastroenterology*. 1986;91(4):830–836.
250. Walker-Smith JA. Mucosal healing in Crohn's disease [letter; comment]. *Gastroenterology*. 1998;114(2):419–420.
251. Munkholm P, Langholz E, Davidsen M, Binder V. Intestinal cancer risk and mortality in patients with Crohn's disease. *Gastroenterology*. 1993;105(6):1716–1723.
252. Gerasimidis K, Talwar D, Duncan A, et al. Impact of exclusive enteral nutrition on body composition and circulating micronutrients in plasma and erythrocytes of children with active Crohn's disease. *Inflamm Bowel Dis*. 2012;18(9):1672–1681.
253. Zoli G, Katelaris PH, Garrow J, et al. Increased energy expenditure in growing adolescents with Crohn's disease. *Dig Dis Sci*. 1996;41(9):1754–1759.
254. Friedman M, Strang LB. Effect of long-term corticosteroids and corticotrophin on the growth of children. *Lancet*. 1966;2(7463):569–572.
255. Motil KJ, Grand RJ, Matthews DE, et al. Whole body leucine metabolism in adolescents with Crohn's disease and growth failure during nutritional supplementation. *Gastroenterology*. 1982;82(6):1359–1368.
256. O'Keefe SJ, Ogden J, Rund J, Potter P. Steroids and bowel rest versus elemental diet in the treatment of patients with Crohn's disease: the effects on protein metabolism and immune function. *JPEN J Parenter Enteral Nutr*. 1989;13(5):455–460.
257. Thomas AG, Miller V, Taylor F, et al. Whole body protein turnover in childhood Crohn's disease. *Gut*. 1992;33(5):675–677.
258. Royall D, Greenberg GR, Allard JP, et al. Total enteral nutrition support improves body composition of patients with active Crohn's disease [see comments]. *JPEN J Parenter Enteral Nutr*. 1995;19(2):95–99.
259. Hannon TS, Dimeglio LA, Pfefferkorn MD, Denne SC. Acute effects of enteral nutrition on protein turnover in adolescents with Crohn disease. *Pediatr Res*. 2007;61(3):356–360.
260. Slonim AE, Bulone L, Damore MB, et al. A preliminary study of growth hormone therapy for Crohn's disease. *N Engl J Med*. 2000;342(22):1633–1637.
261. Inoue Y, Copeland EM, Souba WW. Growth hormone enhances amino acid uptake by the human small intestine. *Ann Surg*. 1994;219(6): 715–722; discussion 22–24.
262. Lo HC, Ney DM. GH and IGF-I differentially increase protein synthesis in skeletal muscle and jejunum of parenterally fed rats. *Am J Physiol*. 1996;271(5 Pt 1):E872–E878.

Celiac Disease

53

Dascha C. Weir, MD and Ciarán P. Kelly, MD

Celiac disease (CD) is an immune-mediated enteropathy secondary to permanent sensitivity to wheat gluten and related proteins in rye and barley. It results in characteristic histologic changes consisting of inflammation, crypt hyperplasia and villous atrophy of the small intestine in genetically susceptible individuals. Significant variability in the clinical presentation of CD in the pediatric population complicates recognition of the disease in many patients. Treatment for CD consists of a lifelong strict gluten-free diet (GFD). Adherence to this diet is associated with resolution of most related signs and symptoms and a decreased risk of related complications. With an explosion of new knowledge over the span of two decades, our understanding of CD has changed dramatically. CD has gone from a rare condition causing gastrointestinal symptoms in children of European origin to a common disorder causing symptoms that affect multiple organ systems in all ages virtually worldwide.[1]

The overall prevalence of CD is similar in Europe and North America affecting up to 1% of the population.[2,3] A large multicenter study in the United States, using serologic screening with biopsy confirmation to identify cases of CD, showed a prevalence of CD of 1:133 in individuals with no evident risk factors. Prevalence of CD in symptomatic patients was 1:56. The prevalence of diagnosed CD is much lower, especially in the United States. This reflects under-recognition and under-diagnosis related to the wide clinical spectrum of disease and the presence of silent disease.[4] In the United States, the prevalence of CD in people of non-European decent is not known. Studies suggest that, in the United States, Hispanic and African American populations have a lower prevalence of CD than non-Hispanic Caucasians.[5] For example, a study in Colorado showed that the incidence of positive serology in Hispanic children was three times lower than in non-Hispanic whites.[6] African Americans also represent a small number of patients currently diagnosed with CD. A large

center in New York City reported African Americans comprised 1.3% of their patients with biopsy-proven CD.[7] This is likely partially because of genetic differences in these populations. However, given the increasing recognition of CD in non-Caucasian populations internationally, serious consideration should be given to the possibility of under-diagnosis in minority populations in the United States.

CD is increasingly being diagnosed in other parts of the world, involving populations that were not traditionally thought to be effected by the disease. Reports from Mexico, South America, North India, the Middle East, Turkey and North Africa, have changed previous assumptions that CD is a European affliction.[8–17] CD is rare in people of Japanese, Chinese, and purely African-Caribbean decent.[18]

CD is associated with several autoimmune conditions and genetic disorders and often presents atypically or silently in these populations. In children, it is associated with autoimmune thyroid disease and with type I diabetes mellitus with a 7.8% and 4.5% prevalence of CD in affected children.[19,20] Up to 7% of patients with a selective IgA deficiency also have CD.[21] Patients with trisomy 21, Turner's syndrome and Williams syndrome have also been found to be at higher risk of CD than the general population.[21–24]

PATHOGENESIS

Environmental, immunologic, and genetic factors are all important contributors to the pathogenesis of CD. Clearly, enteric exposure to specific proteins in wheat, rye, and barley are essential to disease activation in a genetically susceptible person. Wheat, rye, and barley have common ancestry and are all derived from the Triticaeae tribe of the grass family. Broadly termed "gluten," the specific proline and glutamine-rich proteins that activate disease are gliadins in wheat, secalins in rye and hordeins in barley. Gluteins, the second major protein fraction of wheat gluten, is likely involved to a lesser degree. Multiple toxic antigenic epitopes have been described (Figure 53-1).

These peptides activate immune cells causing a chain of events that result in tissue damage. The mechanisms for this activation are incompletely understood but involve both adaptive and innate immunity. Partially because of their high proline content, these large cereal peptides are resistant to digestion by intestinal proteases and pass through the epithelium and into the lamina propria in an intact state. The mechanisms

of this passage through the intestinal barrier are not well understood. Tissue transglutaminase deamidates these peptides in the lamina propria, causing them to become negatively charged as the glutamine residues are converted to glutaminc acid. Deamidated peptides bind with far greater affinity to specific positively- charged residues on the peptide-binding groove of HLA-DQ2 or HLA-DQ8 major histocompatibility complex Class II molecules. These peptides are then presented by HLA-DQ2 or HLA-DQ8 to activate gliadin-specific CD4+ T cells thereby activating an intestinal inflammatory response leading to the characteristic histopathological changes of CD. Intraepithelial lymphocytes, T cells that are markedly increased in the intestinal biopsies of patients with active CD, are also involved. It is now evident that gluten can stimulate interleukin-15 production, leading to the transformation of some T cells into natural killer cells and ultimately to enterocyte destruction (Figure 53-2).[1,26,27] Recent studies suggest that both pathogenic and commensal microbiota may also play a role in the development of CD.[28–32]

Genetics are clearly a factor in the development of CD. Among monozygous twins, there is a 70%–75% CD concordance rate and first-degree relatives have a 10% risk of disease.[34,35] CD is strongly associated with HLA class II genes that map to the DQ locus. The HLA-DQ2 heterodimer is expressed in up to 95% of celiac patients with most of the remaining 5% expressing HLA-DQ8 heterodimer. These HLA types are present in 30%–40% of the general population. Only about 4% of people with these HLA types develop CD.[36–38] Additionally, CD is concordant in only 30%–40% of HLA matched siblings. Clearly, these HLA types are necessary but not sufficient for CD to develop and other genes are involved. Candidate susceptibility genes include myosin IXB (MY09B) and regions of chromosome 5, 6, 11 and 19.[1,39] In fact, up to 40 CD risk loci (including HLA) have been identified through genotyping.[28,40]

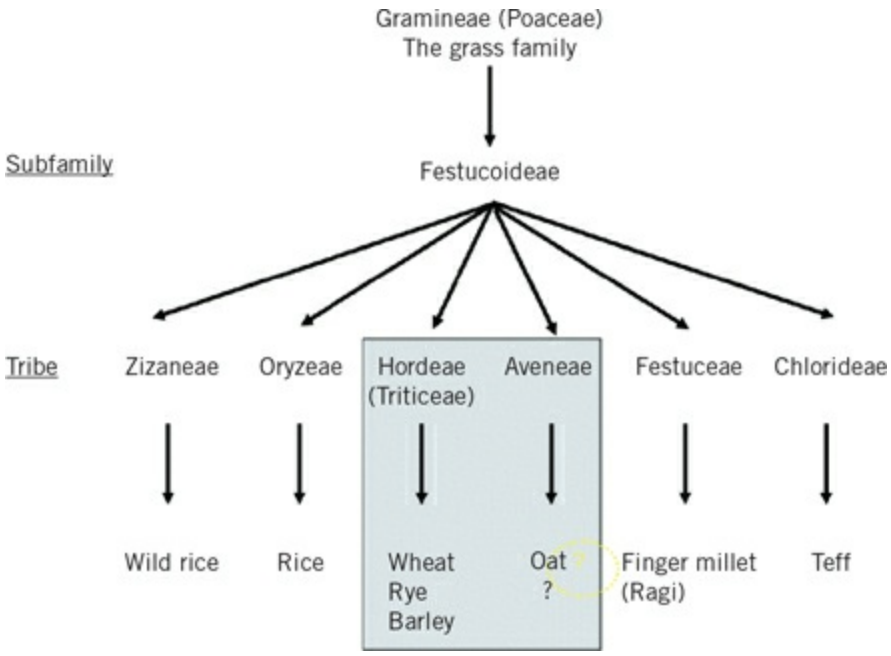


Figure 53-1 The grass family: grains in the tribe Triticeae mediate CD. Species of interest in the Festucoideae subfamily of the grass family are depicted. It seems likely that, for most patients with CD, only grains in the tribe Triticeae mediate CD.[25]

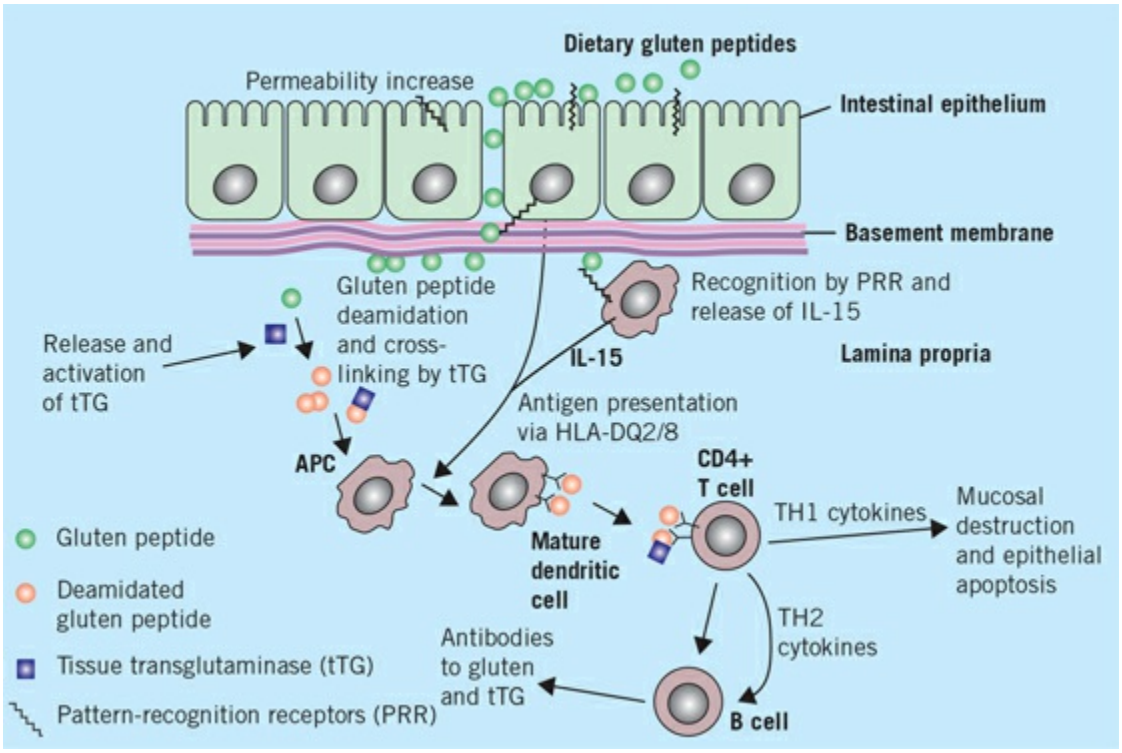


Figure 53-2 Adaptive and innate immune system responses in CD. Crosslinking of gluten by tTG potentiates presentation of peptides by antigen-presenting cells (APC); deamidation of gluten improves binding to HLA-DQ2 or HLA-DQ8, triggering adaptive T-cell responses (inflammation and tissue remodeling [Th1] or antibody production [Th2]). Cells of the innate system recognize gluten peptides directly via pattern recognition receptors, and release cytokines (IL-15) that drive adaptive responses. Dendritic and T cells circulate to mesenteric lymph nodes (where they encounter T cells) and back to lamina propria.[33]

Environmental factors, beyond exposure to gluten, also seem to play an important role in determining which genetically susceptible individuals develop CD. Epidemiologic studies suggest infant feeding practices can impact risk. Both the timing of gluten introduction and the amount of gluten exposure appear to affect risk of developing CD. Specifically, there is evidence that early exposure to gluten before 3 months of age or later initial exposure after 7 months of age increase risk of CD autoimmunity in high risk children. Larger amounts of gluten, introduced in early life, also seem to increase risk. Breastfeeding, particularly at the time of gluten introduction, appears to significantly reduce risk. It is not clear if this protective effect is permanent or if breastfeeding only slows the onset of disease.[38,41–47] Nonetheless, current recommendations suggest introduction of small amounts of gluten between 4–6 months of age ideally while the child is still breastfeeding.[48,49]

Exposure to multiple infections early in life, season of birth, birth by cesarean section, and alterations in the composition of intestinal microbiota have also been identified as possible risk factors in the development of CD.29–32,44,50–56

CLINICAL PRESENTATION

Traditionally, CD was thought to present primarily with gastrointestinal symptoms in the young. This “classical” presentation consists of poor growth, chronic diarrhea, abdominal distention, muscle wasting, poor appetite, and irritability typically seen in patients of 6–24 months of age. Edema, pallor and emesis are also seen. Rarely, children can present in celiac crisis with copious watery diarrhea, marked abdominal distension, dehydration, electrolyte imbalances, hypotension, and lethargy.[57] As serologic testing and our understanding of CD have improved,

"nonclassical" or "atypical" presentations of CD are increasingly being recognized. In pediatric patients, this form of presentation is characterized by delayed onset with gastrointestinal complaints such as nausea, bloating, recurrent abdominal pain, or constipation.[21] Many patients have no gastrointestinal complaints but have extraintestinal manifestations such as short stature, osteopenia, joint pains, pubertal delay, iron deficiency anemia, dental enamel defects, neurologic disorders, or elevated transaminases. Some patients, with classic histologic changes on intestinal biopsy, are entirely asymptomatic and are described as having "silent" CD.[18,57] A comprehensive study based in the United States reported that gastrointestinal symptoms were prominent in pediatric patients diagnosed before 3 years of age. However, in children diagnosed over the age of 3 years, the majority presented with nongastrointestinal manifestations. This study reported the average age of diagnosis in children to be approximately 9 years of age.[58] Other North American and European studies have shown similar trends.[59–64]

CD is increasingly being diagnosed in adults. In fact, the mean age of diagnosis is now in midadult life (circa 45 years of age).[1] Many of these patients have likely had CD since childhood. Adults can present with gastrointestinal symptoms such as diarrhea, flatulence, abdominal bloating, and discomfort but often present with extraintestinal manifestations such as iron deficiency anemia, macrocytic anemia, osteopenia, malaise, infertility, obstetrical complications, and neurologic conditions. Dermatitis herpetiformis, a pruritic blistering skin disorder, is also seen with CD but rarely occurs in pediatric patients (Table 53-1 and Figure 53-3).[18,57]

DIAGNOSIS

While the pathogenesis of CD and the genetic factors involved are still being elucidated, the resulting inflammation and tissue damage causes characteristic and well-recognized histopathologic changes. Small intestinal biopsy histology remains the gold standard in the diagnosis of CD. The spectrum of histopathologic changes in CD have been classified by Marsh with additional more recent modifications.[65] Marsh I lesions, an early but often nonspecific finding, are characterized by increased intraepithelial lymphocytes (>30 IEL per 100 enterocytes) and normal villous architecture. Marsh II lesions consist of crypt hyperplasia in the setting of increased numbers of intraepithelial lymphocytes (>30 per 100 epithelial cells). Marsh III lesions, type a–c, are characterized by partial to

total villous atrophy in addition to crypt hyperplasia and increased numbers of intraepithelial lymphocytes. Because of patchy distribution of these histopathologic changes, multiple biopsies taken from the duodenal mucosa are indicated.[63,66–68] Additionally, CD can involve the duodenal bulb only. It is therefore important that duodenal bulb biopsies are also obtained.[63,69] To avoid misinterpretation, biopsies (especially of the duodenal bulb) are best reviewed by an experienced gastrointestinal pathologist (Figure 53-4).

The development of accurate serologic markers has markedly improved our ability to identify patients with untreated CD and to monitor them after diagnosis. Serologic testing, coupled with clinical assessment and histopathologic confirmation, has become a cornerstone of diagnosis. IgA endomysial antibodies (EMA IgA) can be detected by indirect immunofluorescence using human umbilical cord or monkey esophagus. In pediatric patients, the sensitivity ranges from 89%–94% and the specificity ranges from 99%–100%.[70,71] However, this test is expensive and heavily operator dependent. IgA tissue transglutaminase (TTG IgA) is a less expensive, less operator-dependent assay. Its sensitivity ranges from 84.6%–100% and specificity ranges from 76%–97.4% in the pediatric population.[70,72,73] These tests perform less well in pediatric patient compared to adults, especially in patients under the age of 2 years. False negatives can also occur in the setting of mild enteropathy and in patients with IgA deficiency. TTG IgA and EMA IgA have largely replaced the older IgA and IgG antigliadin antibody tests. Antigliadin antibodies have poor test performance, with low specificity. Newer serologic markers, IgA and IgG deamidated gliadin peptide (DGP) antibodies perform better than the standard antigliadin antibody tests. DGP sensitivity ranges from 85.6–89.9 % and its specificity ranges from 92.5–95.5%.[74] However, DGP IgA and IgG do not perform better in pediatric patients compared to the TTG IgA and EMA IgA, even in patients under 2 years of age.[75,76] There is also a role for DGP IgG in patients with IgA deficiency, though in pediatric patients TTG IgG may also be used.[77,78]

TABLE 53-1 Clinical Manifestation of Celiac Disease in Children

Manifestations secondary to untreated CD	Associated diseases (or diseases secondary to untreated CD)
CD with classic symptoms	***Autoimmune diseases***

– Abdominal distension
– Anorexia
– Chronic or recurrent diarrhea
– Failure to thrive /weight loss
– Irritability
– Muscle wasting
– Celiac crisis (rare)

CD with nonclassic symptoms

– Arthritis
– Aphthous stomatitis
– Constipation
– Dental enamel defects
– Dermatitis herpetiformis
– Hepatitis
– Iron-deficient anemia
– Pubertal delay
– Recurrent abdominal pain
– Short stature
– Vomiting
– Type 1 diabetes
– Thyroiditis
– Sjogren's syndrome

Neurologic and psychologic disturbances

– Ataxia
– Depression
– Epilepsy with intracranial calcifications

Other disorders

– IgA nephropathy
– Osteopenia/osteoporosis

Genetic disorders

– Down syndrome
– Turner syndrome
– Williams syndrome
– IgA deficiency

Adapted with permission from Fasano, A.[57]

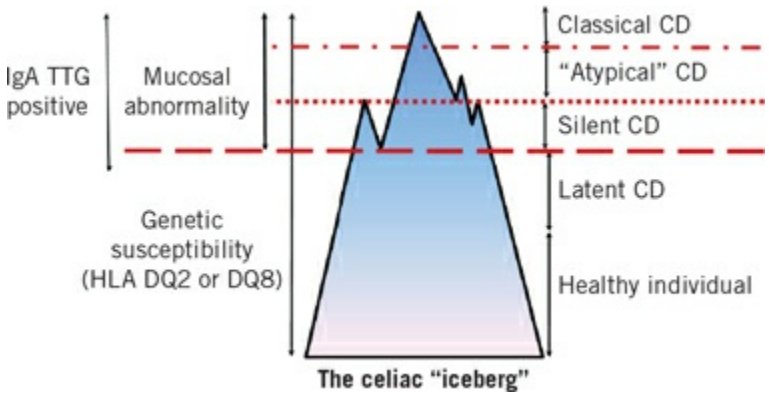


Figure 53-3 Clinical manifestations of CD—the celiac iceberg. CD presents with a wide range of symptoms. Classical CD with prominent diarrhea and malabsorption is now less common than atypical CD

presenting with iron deficiency anemia or other disease manifestations with mild or absent gastrointestinal symptoms. Many individuals have silent CD with no symptoms or disease manifestations despite positive tTG serology and abnormal small bowel histology. Only a subset of individuals with CD has been diagnosed. This subgroup is often referred to as the “tip of the celiac iceberg.” Most individuals with genetic susceptibility (HLA DQ2 or DQ8 positive) do not develop CD.

HLA typing has a limited role in the evaluation of pediatric CD, but may be useful in situations where diagnosis is difficult.[76] Because almost all celiac patients have either HLA DQ2 or DQ8, the absence of these markers effectually excludes the diagnosis.[79–82] Conversely, the presence of celiac HLA types has a low positive predictive value, as their presence is widespread in the non-celiac general population as discussed above.

New guidelines for the diagnosis of CD in children were developed and published by the European Society for Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) in 2012. For the asymptomatic child in a high celiac risk group, either by family history or associated condition, HLA typing is proposed as the first step of evaluation to guide who needs further serologic screening. Additionally, these recommendations suggest that a subset of children do not need biopsy confirmation of CD if they have: (a) symptoms compatible with CD, (b) a highly elevated TTG IgA (greater than 10X the upper limit of normal), (c) a positive EMA IgA on a second blood sample, and (d) HLA susceptibility for CD.[77] These recommendations have not yet been widely adopted in the United States, but will likely impact clinical practice into the future.

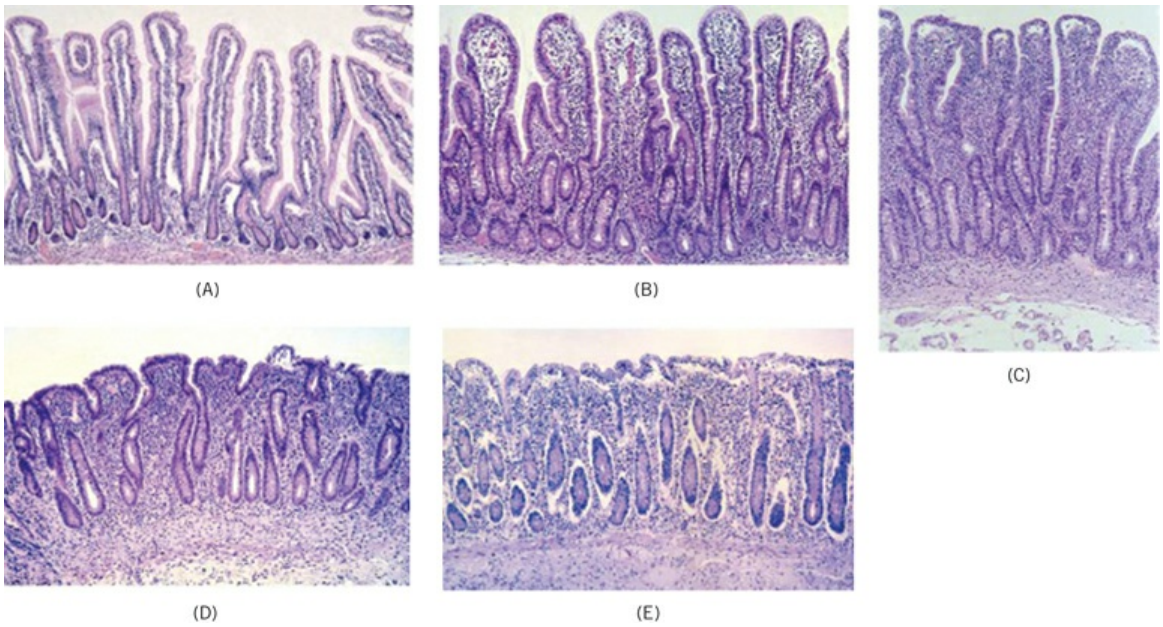


Figure 53-4 The spectrum of intestinal damage in CD, Marsh classification. A, Marsh I: increased intraepithelial lymphocytes; B, Marsh II: increased intraepithelial lymphocytes and crypt hyperplasia; C, Marsh IIIA: partial villious atrophy; D, Marsh IIIB: subtotal villious atrophy; E, Marsh III C: total villous atrophy. (Reproduced with permission from Green PH, Rostami K, Marsh MN.[201])

Serologic markers also have a role in monitoring disease control once a GFD has been initiated. TTG IgA gradually normalizes within 3–12 months on a GFD depending upon the initial antibody concentration. TTG IgA has greater sensitivity than EMA IgA in monitoring for continued gluten intake, but may not be sensitive enough to detect occasional or minor dietary errors.[70,83]

Positive TTG or EMA serology and/or resolution of symptoms on a GFD are not sufficient to make a definitive diagnosis of CD and, in most cases, do not replace the need for duodenal biopsies to make the initial diagnosis. However, repeat biopsies are no longer routinely recommended. Historically, repeat intestinal biopsies were used to confirm diagnosis and to monitor disease control. However, the availability of more accurate serologic markers has greatly reduced the use of repeat biopsies in patients with CD. A second biopsy is now typically limited to patients with poor clinical response to a strict GFD. Gluten rechallenge, once a mainstay of diagnosis, has also largely fallen by the wayside, especially in the pediatric

population. Rechallenge is now indicated only in patients on a GFD in whom the diagnosis remains in doubt (Figure 53-5).[18]

First-degree relatives of people with CD have a 10% risk of disease. In family members, symptoms may be subtle or differ significantly from those in the effected child. A careful review of symptoms or signs of possible CD in first-degree relatives should be sought routinely when a child or adolescent is diagnosed with CD. Serologic screening of all first-degree family members of a patient diagnosed with CD is recommended by some celiac specialists, but the cost to benefit ratio of this intervention is unclear.[84–86]

EFFECT OF CD ON GROWTH AND BODY COMPOSITION

Short stature is a common presenting sign of CD, especially in the pediatric population. It may be the only manifestation.[87–91] The pathophysiology of CD growth retardation is not fully defined, but is traditionally thought to be secondary to associated malnutrition. With the initiation of a GFD and nutritional rehabilitation, children with CD undergo significant growth acceleration. This catch-up growth appears to be most evident within the first year on a gluten- free diet.[92,93] There is some evidence that, even when diagnosed early, monitored closely and kept on a strict GFD, many children do not catch up completely and remain slightly below average height for age and skeletal age into adolescence.[94,95] New evidence suggests overlap of CD with growth hormone deficiency.[96–98] When catch-up growth on a GFD is not observed, further evaluation and possible growth hormone replacement is warranted.[99]

In addition to linear growth, body composition is also significantly affected by CD. Longitudinal study of children and adolescents with newly diagnosed CD has shown significant decreased weight, fat mass, and muscle mass compared to controls.[100] Decreased fat mass has also been described in children presenting with asymptomatic disease.[101] Similar findings have also been described in the adult population.[102] After one year on a GFD, affected children have been found to have complete restoration of body composition with no significant differences in weight, fat mass, or lean mass of the limbs compared to healthy controls. This restoration appears to occur faster and more completely when the GFD is initiated in childhood or adolescence compared to adulthood. It also

appears to be a lasting effect, as body composition in young adults who had initiated a GFD in childhood or adolescence has been shown to be normal.[100] Even without strict adherence, long-term maintenance of the diet through adolescence was associated with improvements in body composition.[103] As nonclassical forms of CD are increasingly being recognized, these characteristic changes in body composition may not be found consistently. Up to 39% of adults with CD are overweight or obese (specifically defined as a body mass index (BMI) > 25) at the time of diagnosis.[104–106] Overweight and obese status in children has also been noted in up to 19% of children at the time of diagnosis.[58,107,108]

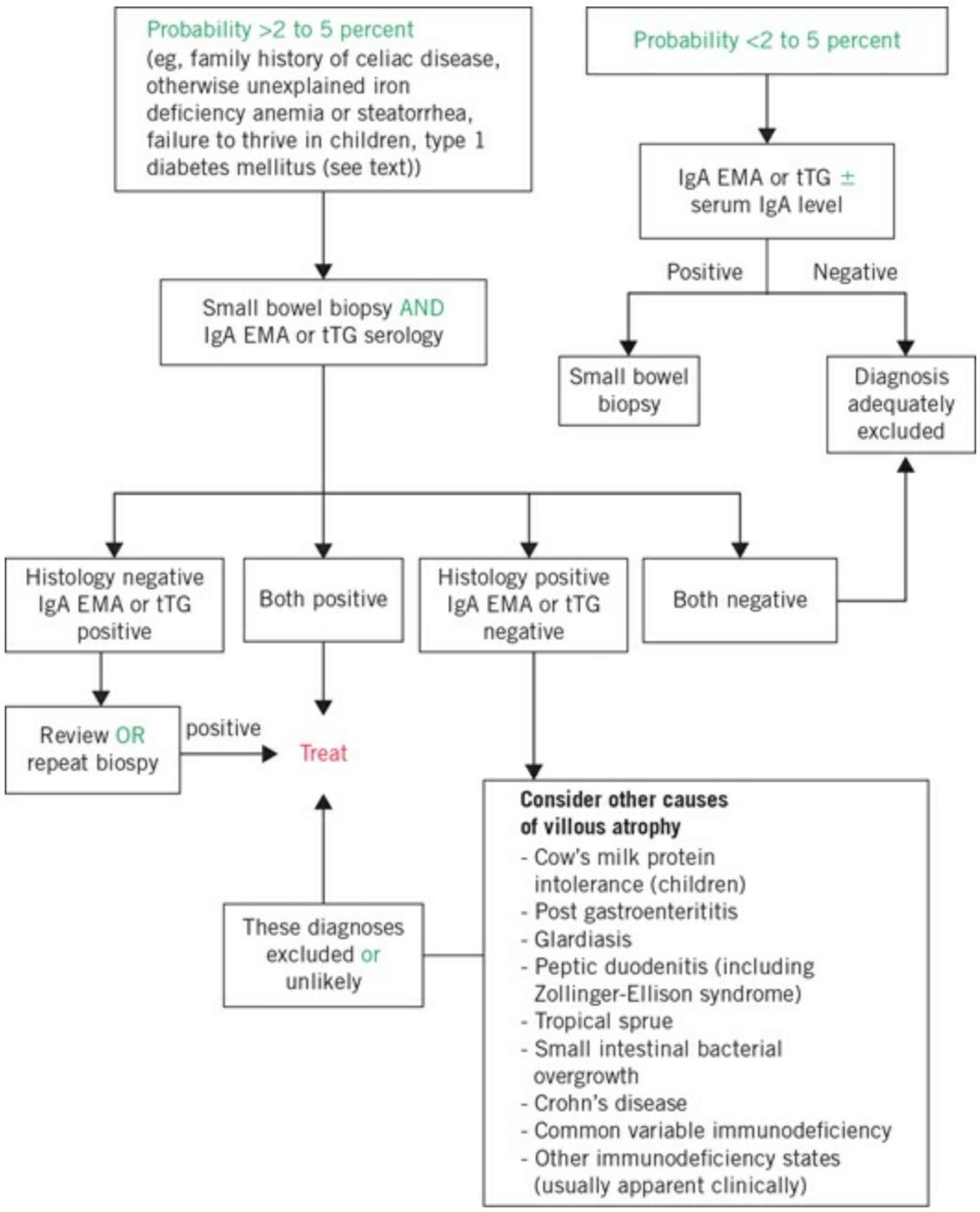


Figure 53-5 Making the diagnosis of CD.

Some patients with CD gain excessive weight on a gluten-free diet. A large study of adults in the United States, showed that up to 16% of patients who were underweight or normal weight at the time of celiac diagnosis transitioned to an overweight BMI class on a gluten-free diet. 22% of patients who were overweight at diagnosis gained additional weight on the diet.[106] However, other adults' studies have shown 18%–

54% of overweight celiac patients lost weight on the gluten-free diet.[109,110] Similar patterns have been noted in children with CD. Excessive weight gain has been shown to occur in children with CD on the gluten-free diet.[107] However, there is also data that shows overweight and obese children diagnosed with CD significantly lower or even normalize their BMI on the gluten-free diet.[108] Ongoing involvement of a dietician specialized in the gluten-free diet is essential (Figure 53-6 and 53-7).



Figure 53-6 A toddler with "classic" presentation of CD. A, At the time of the diagnosis; B, at the time of diagnosis (2); and C, after 1 year on a GFD.

Osteoporosis, rickets, and osteomalacia are all seen with CD. The specific mechanism of bone disease associated with CD is unclear, but is likely related to malabsorption of calcium and vitamin D and a possible effect of increased interleukins and other inflammatory mediators. Decreased bone mineral density is frequently seen in patients with CD, even in patients with clinically silent CD.[111] In untreated adult celiac patients, the reported prevalence of decreased bone mineral content is 22%–80%. In children and adolescents with CD, bone mineral density is often decreased at the time of diagnosis (3–39% prevalence).[112] However, after one year on a strict GFD, the bone mineral density of children and

adolescents with CD appears to normalize and reach the level of healthy controls. After 4 years of a GFD, these patients continue to have normal values.[111,113] Again, this restoration appears to be most consistent in the setting of early recognition and treatment.[114] Interestingly, patients who are poorly compliant did not appear to achieve peak bone mineralization.[113] Assessment of bone mineral density in children with CD is recommended at one year after diagnosis. Assessment of bone mineral density is also recommended in patients with poor adherence to the gluten-free diet.[115] The effect of the GFD on bone mineral density in adult studies has shown conflicting results. Some studies report improvement of bone mineral density on the diet and others show no such effect.[102,116–118] The age at diagnosis and initiation of the GFD seems to be a determining factor in bone restoration perhaps because of the increased activity of bone metabolism in childhood and adolescence.[119] There are conflicting data, however, on whether fracture risk is increased in adult patients with CD, so the clinical significance of this osteoporosis is not clear.[120,121]

Another characteristic change associated with CD involves dentition. Dental enamel defects of both deciduous and permanent teeth involving symmetric and chronologic distribution are well recognized in CD affecting up to 96% of children with CD and 83% of adults.[122,123]

NUTRITIONAL DEFICIENCIES

Nutritional deficiencies associated with CD are well recognized and should be an important consideration at the time of diagnosis and in continued management. Abnormal folate, iron and vitamin B12 levels are found in children, adolescents and adults with newly diagnosed or untreated CD. This appears to be true in patients with clinically silent disease as well as those with more “classic” manifestations. A study of screening-detected adolescent cases of CD in Finland (30% asymptomatic) found that 1/3 had abnormal folate or iron status.[124] European studies in adults have shown decreased levels of vitamin B12 in 11%–41% of patients with CD.[125,126] Additionally, there is some evidence that untreated adult patients with CD have reduced endogenous pools of zinc.[127] Children with active gastrointestinal CD have also been shown to have lower serum zinc levels than healthy controls and children with celiac on the GFD.[128] Fat soluble vitamins such as vitamin E, vitamin D and vitamin K levels may also be low.[129,130]

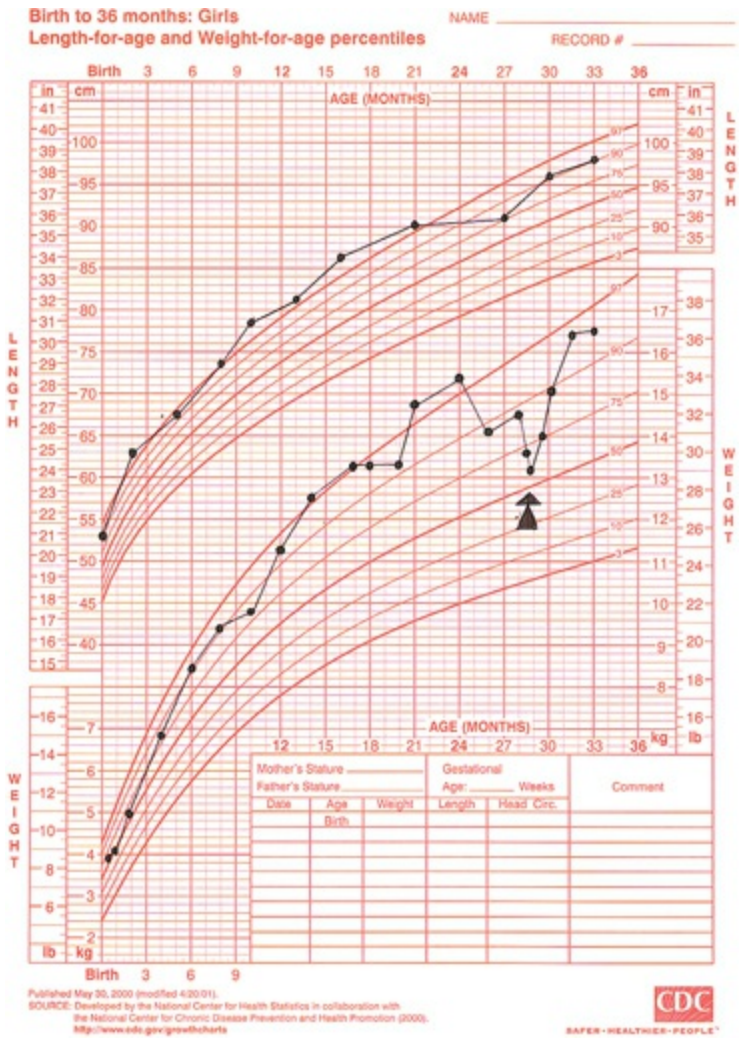


Figure 53-7 Growth chart of a classically presenting toddler with CD. The arrow indicates time of diagnosis and initiation of a GFD.

Iron deficiency anemia is the most common clinical presentation of CD in adults, but is also seen in children.[131,132] This is likely secondary to decreased absorption of iron in the proximal small bowel but may also be related to inadequate dietary intake as a result of CD associated anorexia or chronic inflammation. Megoblastic anemia due to folate or vitamin B12 deficiency is also seen. Thrombocytopenia, leukopenia and prolonged coagulation have also been reported in the pediatric population.[133] Notably, these abnormalities appear to resolve with treatment. For example, up to 95% of patients have correction of their anemia after 1 year on the GFD.[134]

OTHER EXTRAINTESTINAL MANIFESTATIONS

Hepatic involvement is seen in adult and pediatric patients with CD. In a European study of 114 consecutive pediatric patients with CD, mild hypertransaminasemia was observed in 32% of patients at the time of diagnosis. In 5 patients (4.3%), it was the only manifestation of CD. In these children, aminotransferases normalized within one year of initiating a GFD either before or at the same time as serologic marker normalization. Hypertransaminasemia does not seem to be related to malnutrition in these patients and the mechanism of hepatic injury is not clear.[135] Similar findings have been reported in the adult celiac population.[136] Severe liver disease that resolved with a GFD has also been reported in adults with CD.[137]

Pubertal delay can occur in both boys and girls with CD. Hypogonadism, infertility, and sexual dysfunction have all been described in the literature. Boys have been found to have an abnormal pattern of androgen resistance. Girls with untreated CD have been shown to have delayed menarche compared to their mothers and to girls with CD who are on a GFD.[138,139] Increased risk for recurrent miscarriage, premature delivery, intrauterine growth retardation, and lower birth weight babies have each been reported in women with untreated CD.[140–145]

Neurologic complications are a rare but recognized manifestation of CD. In fact, there are a growing number of reports linking neurologic symptoms with CD, sometimes as the presenting manifestation. Cerebellar ataxia, peripheral neuropathy and epilepsy have been described primarily in the adult celiac population. Depression and anxiety have also been described to affect up to a third of patients with CD.[146–148]

There are limited studies on other neurologic manifestations of CD in children where the risk of neurologic complications appears lower than in adults.[149] Headache is the most common neurologic complaint in children affecting almost a third of patients. Neurologic disorders such as hypotonia, developmental delay, learning disabilities, headache, and ataxia have been described in up to 52% of patients with CD diagnosed in childhood which was significantly higher than a healthy control group. There is conflicting data about the association of CD and attention deficit hyperactivity disorder (ADHD).[150,151] Focal white matter lesions in the brain have been reported in up to 20% of pediatric patients with CD.[152] The syndrome of bilateral occipital cerebral calcifications and seizures, also known as CD, epilepsy and cerebral calcifications syndrome (CEC syndrome), has also been well described and presents primarily in

childhood.[153] Epilepsy unrelated to CEC syndrome has been reported with CD in children.[154] There is no increase the incidence of tics in patients with CD.[155] Despite previous reports to the contrary, an association between CD and autism has not been confirmed.[57,148,156,157]

The spectrum of neurologic complications responds variably to a GFD. Up to half of children experiencing headaches improved on the diet. High rates of response in transient infantile hypotonia associated with classical early onset CD have also been reported, perhaps because of resolution of nutritional deficiencies.[155] In cases of epilepsy, improved seizure control has been reported but not seizure resolution. Depressive symptoms and anxiety may improve with a GFD in some patients but depression remains common in the treated celiac population. A few reports describe improvement of neuropathy and ataxia on a GFD, but for most patients these neurologic problems persist or progress despite gluten avoidance.[148]

NATURAL HISTORY

Overall mortality in patients with CD has been described as two times that of the general population. The risk of increased mortality appears higher in patients with malabsorptive symptoms with no increased risk in patients with minor or no symptoms. Increased mortality has also been observed in association with delayed diagnosis and with poor adherence to the GFD.[158–160]

Specifically, an increased risk of malignancy has been associated with CD since the 1960s. Patients with CD appear to be at increased risk for non-Hodgkin lymphoma, especially small-intestinal T cell lymphoma, small intestinal adenocarcinoma, squamous cell carcinomas of the esophagus, mouth and pharynx, and other gastrointestinal tract tumors. However, the absolute risk for malignancy remains small and recent data suggest that the risk while present is less than previously estimated and is low in patients with clinically silent CD.[159,161–165] The mechanism for this increased risk of cancer is not well understood but is perhaps related to chronic inflammation, increased exposure to environmental carcinogens secondary to intestinal permeability, immune surveillance abnormalities, or nutritional deficits.

Increased mortality and malignancy appear to be issues that primarily affect adult patients with CD. Few cases of cancer in children and adolescents with CD have been reported and increased risk of malignancy in this population has not been shown clearly.[162,164,166,167] Nonetheless,

prompt identification of CD and appropriate treatment likely has long-term effects into adulthood. Adult patients with CD who were diagnosed in childhood appear to have a normal mortality rate presumably a benefit of long-term adherence to a GFD.[159] Additionally, patients who adhere to a GFD appear to have decreased malignancy risk that approaches that of the general population.[164,165,168]

The prevalence of autoimmune disorders is higher in adolescents and adults with CD and in their relatives as compared to the general population.[169–171] This association is likely related to the common genetic background including HLA type.[172] It has been suggested that the risk of autoimmunity is related to the duration of gluten exposure and the age of diagnosis.[169] However, other studies have disputed this theory.[173,174] Specifically, the risk of thyroid autoimmune disorders is increased in patients with CD even when treated with the GFD.[175–177]

TREATMENT

Gluten and related proteins in rye and barley are clearly implicated in the pathogenesis of CD and eliminating these proteins from the diet is currently the mainstay of treatment. Adherence to a strict GFD leads to histopathologic remission, serologic normalization, resolution of most of the related symptoms, and likely prevention of long-term complications.

However, the GFD is often challenging and sometimes overwhelming to people with CD. Wheat-based foods are a major staple of the American diet and finding palatable gluten-free alternatives can be difficult and expensive. Additionally, gluten is a hidden ingredient in many foods and medications and labeling can be confusing. The initiation of a strict GFD can have significant implications on quality of life. In fact, both adults and children with CD report negative feelings towards the diet and report difficulties with adherence especially related to eating out of the home, traveling, and social events.[178,179] Thus it is not surprising that adherence to the GFD is a major issue in CD management. This is true across all ages especially in adolescents and young adults. Strict adherence with a GFD in this age group has been reported to be between 45% and 65%.[180,181] A very young age of diagnosis is associated with higher rates of adherence.[182] Screening detected disease, with either atypical or silent features, is associated with poorer compliance.[183] Patient and family education and support help the transition to a GFD and increase adherence. Intensive support is best given via a team approach with involvement of

both a physician and a dietitian experienced in CD. Involvement in support groups is also helpful.[178,179]

The role of oats in the GFD has been subject to change. Several studies now show that the inclusion of pure oats is safe for the majority of children and adults with CD. Rarely, in a small number of individuals, the avenin proteins in oats can trigger a response similar to gluten.[184–188] However, high levels of cross-contamination with wheat in the North American oats supply has been problematic and, in the past, led to recommendations for patients with CD to avoid all oats. As uncontaminated sources of oats have now become more readily available, the introduction of oats with clinical follow up may be attempted in patients with well-treated CD.[189]

Lactose intolerance is common in both children and adults with untreated CD. In fact, up to half of newly diagnosed patients are affected. Lactose intolerance in the setting of active CD is likely secondary to low lactase enzyme activity in the damaged small intestinal mucosa. Lactose containing foods may need to be avoided initially in newly diagnosed patients with significant symptoms. Fortunately, most patients can tolerate dairy intake within 3–6 months after initiation of a GFD.[190,191] However, primary lactose intolerance can coexist with CD.[192]

While general nutritional status improves with treatment, a lifelong GFD may not be nutritionally well balanced. Few gluten-free products in the United States are enriched and most are lower in fiber than their wheat-containing counterparts.[184] In fact, adult and adolescent patients on a GFD have been shown to have decreased fiber intake.[193,194] Mean daily intakes of folate and vitamin B12 were significantly lower in adults with CD on a GFD as compared to healthy controls.[195] Children on a GFD were also found to consume only 60% of the recommended daily allowance of iron.[196] Another nutritional study of children with CD found that those children on a strict diet were more likely to have a nutritionally unbalanced diet and were more likely to be overweight than children who were not adherent to the diet.[197] A study in adult celiac patients after 8–12 years of GFD with biopsy-proven remission showed that 56% showed signs of poor vitamin status, specifically with lower plasma levels of folate, vitamin B6 and elevated levels of homocysteine (a metabolic marker of folate, vitamin B6 and vitamin B12 deficiency).[195] Thus the use of a daily gluten-free multivitamin supplement is often recommended.[184]

Please refer to Chapter 72, Special Diets, for additional information about a gluten-restricted diet and celiac resources and support groups.

BEYOND THE GFD

While highly effective, a GFD is not an ideal therapeutic option for many individuals with CD because of the challenges associated with diet adherence and the negative impact on quality of life. Developing grains, through selective breeding and transgenic technology, that have low or nontoxic peptide sequences and acceptable baking qualities is technically challenging. Fortunately, as our understanding of the pathophysiology of CD has grown, several potential targets for new treatments have been identified. Alternatives to the GFD are currently being investigated.

Intraluminal therapies in the form of endoproteolytic enzymes, such as ALV003 or AN-PEP, may help degrade gluten peptides in the intestinal tract thereby rendering them nontoxic. Larazotide acetate affects tight junctions and acts to strengthen the intestinal epithelial barrier and reduce the passage of toxic gluten peptides into the lamina propria. Evidence suggests that larazotide reduces symptoms and gluten-induced immune reactivity in patients with CD on a gluten challenge.[198] Development of a desensitizing vaccine is also underway.[199] Other potential strategies target recognition and presentation of the offending gluten antigens via blockade of dendritic cells and other antigen presenting cells, specific antagonization of HLA-DQ peptide binding or by inhibition of TTG. Interfering with the subsequent inflammatory response by cytokine therapy and selective adhesion molecule inhibition is another potential intervention that is currently being explored in other disease processes.[200]

Many of these potential new treatments, while promising, are in the early phases of investigation.

Ideally, a safe and effective treatment will be developed that can replace the GFD and allow patients with CD to eat freely. However, a more realistic goal may be to develop therapies that allow patients with CD increased flexibility with their diet. A treatment that allows individuals to tolerate small or moderate amounts of gluten in their diets would clearly have a positive impact on the lives of people with CD.

REFERENCES

1. Treem WR. Emerging concepts in celiac disease. *Curr Opin Pediatr*. 2004;16:552–559.
2. Maki M, Mustalahti K, Kokkonen J, et al. Prevalence of Celiac disease among children in Finland. *N Engl J Med*. 2003;348:2517–2524.
3. Carlsson AK, Axelsson IE, Borulf SK, Bredberg AC, Ivarsson SA. Serological screening for celiac disease in healthy 2.5-year-old children in Sweden. *Pediatrics*. 2001;107:42–45.

4. Fasano A, Berti I, Gerarduzzi T, et al. Prevalence of celiac disease in at-risk and not-at-risk groups in the United States: a large multicenter study. *Arch Intern Med*. 2003;163:286–292.
5. Rubio-Tapia A, Ludvigsson JF, Brantner TL, Murray JA, Everhart JE. The prevalence of celiac disease in the United States. *Am J Gastroenterol*. 2012;107:1538–1544; quiz 7, 45.
6. Hoffenberg EJ, MacKenzie T, Barriga KJ, et al. A prospective study of the incidence of childhood celiac disease. *J Pediatr*. 2003;143:308–314.
7. Brar P, Lee AR, Lewis SK, Bhagat G, Green PH. Celiac disease in African-Americans. *Dig Dis Sci*. 2006;51:1012–1015.
8. Remes-Troche JM, Ramirez-Iglesias MT, Rubio-Tapia A, Alonso-Ramos A, Velazquez A, Uscanga LF. Celiac disease could be a frequent disease in Mexico: prevalence of tissue transglutaminase antibody in healthy blood donors. *J Clin Gastroenterol*. 2006;40:697–700.
9. Melo SB, Fernandes MI, Peres LC, Troncon LE, Galvao LC. Prevalence and demographic characteristics of celiac disease among blood donors in Ribeirao Preto, State of Sao Paulo, Brazil. *Dig Dis Sci*. 2006;51:1020–1025.
10. Sood A, Midha V, Sood N, Avasthi G, Sehgal A. Prevalence of celiac disease among school children in Punjab, North India. *J Gastroenterol Hepatol*. 2006;21:1622–1625.
11. Rawashdeh MO, Khalil B, Raweily E. Celiac disease in Arabs. *J Pediatr Gastroenterol Nutr*. 1996;23:415–418.
12. Masjedizadeh R, Hajiani E, Hashemi J, Shayesteh AA, Moula K, Rajabi T. Celiac disease in South-West of Iran. *World J Gastroenterol*. 2006;12:4416–4419.
13. Ertekin V, Selimoglu MA, Kardas F, Aktas E. Prevalence of celiac disease in Turkish children. *J Clin Gastroenterol*. 2005;39:689–691.
14. Catassi C, Ratsch IM, Gandolfi L, et al. Why is coeliac disease endemic in the people of the Sahara? *Lancet*. 1999;354:647–648.
15. Bdioui F, Sakly N, Hassine M, Saffar H. Prevalence of celiac disease in Tunisian blood donors. *Gastroenterol Clin Biol*. 2006;30:33–36.
16. al-Tawaty AI, Elbargathy SM. Coeliac disease in north-eastern Libya. *Ann Trop Paediatr*. 1998;18:27–30.
17. Al-Hussaini A, Sulaiman N, Al-Zahrani M, Alenizi A, El Haj I. High prevalence of celiac disease among Saudi children with type 1 diabetes: a prospective cross-sectional study. *BMC gastroenterol*. 2012;12:180.
18. Farrell RJ, Kelly CP. Celiac sprue. *N Engl J Med*. 2002;346:180–188.
19. Larizza D, Calcaterra V, De Giacomo C, et al. Celiac disease in children with autoimmune thyroid disease. *J Pediatr*. 2001;139:738–740.
20. Holmes GK. Screening for coeliac disease in type 1 diabetes. *Arch Dis Child*. 2002;87:495–498.
21. Hill ID, Dirks MH, Liptak GS, et al. Guideline for the diagnosis and treatment of celiac disease in children: recommendations of the North American Society for Pediatric Gastroenterology, Hepatology and Nutrition. *J Pediatr Gastroenterol Nutr*. 2005;40:1–19.
22. Bonamico M, Pasquino AM, Mariani P, et al. Prevalence and clinical picture of celiac disease in Turner syndrome. *J Clin Endocrinol Metab*. 2002;87:5495–5498.
23. Ivarsson SA, Carlsson A, Bredberg A, et al. Prevalence of coeliac disease in Turner syndrome. *Acta Paediatr*. 1999;88:933–936.
24. Giannotti A, Tiberio G, Castro M, et al. Coeliac disease in Williams syndrome. *J Med Genet*. 2001;38:767–768.
25. Jabri B, Kasarda DD, Green PH. Innate and adaptive immunity: the yin and yang

of celiac disease. *Immunol Rev.* 2005;206:219–231.
26. Kagnoff MF. Overview and pathogenesis of celiac disease. *Gastroenterology.* 2005;128:S10–S18.
27. Hourigan CS. The molecular basis of coeliac disease. *Clin Exp Med.* 2006;6:53–59.
28. Kupfer SS, Jabri B. Pathophysiology of celiac disease. *Gastrointest Endosc Clin N Am.* 2012;22:639–660.
29. Sanz Y, De Pama G, Laparra M. Unraveling the ties between celiac disease and intestinal microbiota. *Int Rev Immunol*. 2011;30:207–218.
30. Decker E, Engelmann G, Findeisen A, et al. Cesarean delivery is associated with celiac disease but not inflammatory bowel disease in children. *Pediatrics.* 2010;125:e1433–e1440.
31. Sellitto M, Bai G, Serena G, et al. Proof of concept of microbiome-metabolome analysis and delayed gluten exposure on celiac disease autoimmunity in genetically at-risk infants. *PloS one* 2012;7:e33387.
32. Kalliomaki M, Satokari R, Lahteenoja H, et al. Expression of microbiota, Toll-like receptors, and their regulators in the small intestinal mucosa in celiac disease. *J Pediatr Gastroenterol Nutr.* 2012;54:727–732.
33. Schuppan D, Esslinger B, Dieterich W. Innate immunity and coeliac disease. *Lancet*. 2003;362:3–4.
34. Greco L, Romino R, Coto I, et al. The first large population based twin study of coeliac disease. *Gut*. 2002;50:624–628.
35. van Heel DA, Hunt K, Greco L, Wijmenga C. Genetics in coeliac disease. *Best Pract Res Clin Gastroenterol*. 2005;19:323–339.
36. Sollid LM, Lie BA. Celiac disease genetics: current concepts and practical applications. *Clin Gastroenterol Hepatol*. 2005;3:843–851.
37. Petronzelli F, Bonamico M, Ferrante P, et al. Genetic contribution of the HLA region to the familial clustering of coeliac disease. *Annals of Human Genetics*. 1997;61:307–317.
38. Silano M, Agostoni C, Guandalini S. Effect of the timing of gluten introduction on the development of celiac disease. *World J Gastroenterol*. 2010;16:1939–1942.
39. Stepniak D, Koning F. Celiac disease—sandwiched between innate and adaptive immunity. *Hum Immunol*. 2006;67:460–468.
40. Trynka G, Hunt KA, Bockett NA, et al. Dense genotyping identifies and localizes multiple common and rare variant association signals in celiac disease. *Nat Genet.* 2011;43:1193–1201.
41. Norris JM, Barriga K, Hoffenberg EJ, et al. Risk of celiac disease autoimmunity and timing of gluten introduction in the diet of infants at increased risk of disease. *JAMA*. 2005;293:2343–2351.
42. Ivarsson A, Myleus A, Norstrom F, et al. Prevalence of childhood celiac disease and changes in infant feeding. *Pediatrics*. 2013;131:e687–e694.
43. Carlsson A, Agardh D, Borulf S, Grodzinsky E, Axelsson I, Ivarsson SA. Prevalence of celiac disease: before and after a national change in feeding recommendations. *Scand J Gastroenterol* 2006;41:553–558.
44. Myleus A, Hernell O, Gothefors L, et al. Early infections are associated with increased risk for celiac disease: an incident case-referent study. *BMC Pediatr*. 2012;12:194.
45. Persson LA, Ivarsson A, Hernell O. Breast-feeding protects against celiac disease in childhood—epidemiological evidence. *Adv Exp Med Biol.* 2002;503:115–123.
46. Akobeng AK, Ramanan AV, Buchan I, Heller RF. Effect of breast feeding on risk of coeliac disease: a systematic review and meta-analysis of observational studies.

Arch Dis Child. 2006;91:39–43.
47. Ludvigsson JF, Fasano A. Timing of introduction of gluten and celiac disease risk. Annals of nutrition & metabolism 2012;60 Suppl 2:22–29.
48. Agostoni C, Decsi T, Fewtrell M, et al. Complementary feeding: a commentary by the ESPGHAN Committee on Nutrition. *J Pediatr Gastroenterol Nutr.* 2008;46:99–110.
49. Szajewska H, Chmielewska A, Piescik-Lech M, et al. Systematic review: early infant feeding and the prevention of coeliac disease. *Aliment Pharmacol Ther.* 2012;36:607–618.
50. Stene LC, Honeyman MC, Hoffenberg EJ, et al. Rotavirus infection frequency and risk of celiac disease autoimmunity in early childhood: a longitudinal study. *Am J Gastroenterol.* 2006;101:2333–2340.
51. Pavone P, Nicolini E, Taibi R, Ruggieri M. Rotavirus and celiac disease. *Am J Gastroenterol.* 2007;102:1831.
52. Tanpowpong P, Obuch JC, Jiang H, et al. Multicenter Study on Season of Birth and Celiac Disease: Evidence for a New Theoretical Model of Pathogenesis. *J Pediatr*. 2013;162:501–504.
53. Lewy H, Meirson H, Laron Z. Seasonality of birth month of children with celiac disease differs from that in the general population and between sexes and is linked to family history and environmental factors. *J Pediatr Gastroenterol Nutr.* 2009;48:181–185.
54. Sjoberg V, Sandstrom O, Hedberg M, Hammarstrom S, Hernell O, Hammarstrom ML. Intestinal T-cell responses in celiac disease—impact of celiac disease associated bacteria. *PloS one.* 2013;8:e53414.
55. Pozo-Rubio T, Olivares M, Nova E, et al. Immune development and intestinal microbiota in celiac disease. *Clin Dev Immunol.* 2012;2012:654143.
56. Sanchez E, Ribes-Koninckx C, Calabuig M, Sanz Y. Intestinal Staphylococcus spp. and virulent features associated with coeliac disease. *J Clin Pathol.* 2012;65:830–834.
57. Fasano A. Clinical presentation of celiac disease in the pediatric population. *Gastroenterology*. 2005;128:S68–S73.
58. Telega G, Bennet TR, Werlin S. Emerging new clinical patterns in the presentation of celiac disease. Archives of pediatrics & adolescent medicine 2008;162:164–168.
59. Balamtekin N, Uslu N, Baysoy G, et al. The presentation of celiac disease in 220 Turkish children. *Turk J Pediatr*. 2010;52:239–244.
60. Roma E, Panayiotou J, Karantana H, et al. Changing pattern in the clinical presentation of pediatric celiac disease: a 30-year study. *Digestion*. 2009;80:185–191.
61. McGowan KE, Castiglione DA, Butzner JD. The changing face of childhood celiac disease in north america: impact of serological testing. *Pediatrics.* 2009;124:1572–1578.
62. Lurz E, Scheidegger U, Spalinger J, Schoni M, Schibli S. Clinical presentation of celiac disease and the diagnostic accuracy of serologic markers in children. *Eur J Pediatr*. 2009;168:839–845.
63. Weir DC, Glickman JN, Roiff T, Valim C, Leichtner AM. Variability of histopathological changes in childhood celiac disease. *Am J Gastroenterol.* 2010;105:207–212.
64. Tanpowpong P, Broder-Fingert S, Katz AJ, Camargo CA, Jr. Age-related patterns in clinical presentations and gluten-related issues among children and adolescents with celiac disease. *Clin Transl Gastroenterol.* 2012;3:e9.

65. Marsh MN. Gluten, major histocompatibility complex, and the small intestine. A molecular and immunobiologic approach to the spectrum of gluten sensitivity (‘celiac sprue’). *Gastroenterology*. 1992;102:330–354.
66. Bonamico M, Mariani P, Thanasi E, et al. Patchy villous atrophy of the duodenum in childhood celiac disease. *J Pediatr Gastroenterol Nutr*. 2004;38:204–207.
67. Prasad KK, Thapa BR, Nain CK, Singh K. The frequency of histologic lesion variability of the duodenal mucosa in children with celiac disease. *World J Pediatr*. 2010;6:60–64.
68. Ravelli A, Villanacci V, Monfredini C, Martinazzi S, Grassi V, Manenti S. How patchy is patchy villous atrophy?: distribution pattern of histological lesions in the duodenum of children with celiac disease. *Am J Gastroenterol.* 2010;105:2103–2110.
69. Rashid M, MacDonald A. Importance of duodenal bulb biopsies in children for diagnosis of celiac disease in clinical practice. *BMC Gastroenterol*. 2009;9:78.
70. Baudon JJ, Johanet C, Absalon YB, Morgant G, Cabrol S, Mougenot JF. Diagnosing celiac disease: a comparison of human tissue transglutaminase antibodies with antigliadin and antiendomysium antibodies. *Arch Pediatr Adolesc Med*. 2004;158:584–588.
71. Tonutti E, Visentini D, Bizzaro N, et al. The role of antitissue transglutaminase assay for the diagnosis and monitoring of coeliac disease: a French-Italian multicentre study. *J Clin Pathol*. 2003;56:389–393.
72. Barker CC, Mitton C, Jevon G, Mock T. Can tissue transglutaminase antibody titers replace small-bowel biopsy to diagnose celiac disease in select pediatric populations? *Pediatrics*. 2005;115:1341–1136.
73. Hansson T, Dahlbom I, Rogberg S, et al. Recombinant human tissue transglutaminase for diagnosis and follow-up of childhood coeliac disease. *Pediatr Res.* 2002;51:700–705.
74. Lewis NR, Scott BB. Meta-analysis: deamidated gliadin peptide antibody and tissue transglutaminase antibody compared as screening tests for coeliac disease. *Aliment Pharmacol Ther.* 2010;31:73–81.
75. Giersiepen K, Lelgemann M, Stuhldreher N, et al. Accuracy of diagnostic antibody tests for coeliac disease in children: summary of an evidence report. *J Pediatr Gastroenterol Nutr.* 2012;54:229–241.
76. Olen O, Gudjonsdottir AH, Browaldh L, et al. Antibodies against deamidated gliadin peptides and tissue transglutaminase for diagnosis of pediatric celiac disease. *J Pediatr Gastroenterol Nutr.* 2012;55: 695–700.
77. Husby S, Koletzko S, Korponay-Szabo IR, et al. European Society for Pediatric Gastroenterology, Hepatology, and Nutrition guidelines for the diagnosis of coeliac disease. *J Pediatr Gastroenterol Nutr.* 2012;54:136–160.
78. Villalta D, Alessio MG, Tampoia M, et al. Testing for IgG class antibodies in celiac disease patients with selective IgA deficiency. A comparison of the diagnostic accuracy of 9 IgG anti-tissue transglutaminase, 1 IgG anti-gliadin and 1 IgG anti-deaminated gliadin peptide antibody assays. *Clin Chim Acta.* 2007;382:95–99.
79. Lundin KE, Gjertsen HA, Scott H, Sollid LM, Thorsby E. Function of DQ2 and DQ8 as HLA susceptibility molecules in celiac disease. Human immunology 1994;41:24–7.
80. Hadithi M, von Blomberg BM, Crusius JB, et al. Accuracy of serologic tests and HLA-DQ typing for diagnosing celiac disease. *Ann Intern Med*. 2007;147:294–302.
81. Sollid LM, Markussen G, Ek J, Gjerde H, Vartdal F, Thorsby E. Evidence for a

primary association of celiac disease to a particular HLA-DQ alpha/beta heterodimer. *J Exp Med*. 1989;169:345–350.
82. Wolters VM, Wijmenga C. Genetic background of celiac disease and its clinical implications. *Am J Gastroenterol.* 2008;103:190–195.
83. Bazzigaluppi E, Roggero P, Parma B, et al. Antibodies to recombinant human tissue-transglutaminase in coeliac disease: diagnostic effectiveness and decline pattern after gluten-free diet. *Dig Liver Dis*. 2006;38:98–102.
84. Fasano A, Catassi C. Clinical practice. Celiac disease. *N Engl J Med.* 2012;367:2419–2426.
85. Goldberg D, Kryszak D, Fasano A, Green PH. Screening for celiac disease in family members: is follow-up testing necessary? *Digest Dis Sci*. 2007;52:1082–1086.
86. Dolinsek J, Urlep D, Karell K, Partanen J, Micetic-Turk D. The prevalence of celiac disease among family members of celiac disease patients. *Wien Klin Wochenschr*. 2004;116 Suppl 2:8–12.
87. Bonamico M, Scire G, Mariani P, et al. Short stature as the primary manifestation of monosymptomatic celiac disease. *J Pediatr Gastroenterol Nutr*. 1992;14:12–16.
88. Rosenbach Y, Dinari G, Zahavi I, Nitzan M. Short stature as the major manifestation of celiac disease in older children. *Clin Pediatr* (Phila). 1986;25:13–16.
89. Knudtzon J, Fluge G, Aksnes L. Routine measurements of gluten antibodies in children of short stature. *J Pediatr Gastroenterol Nutr*. 1991;12:190–194.
90. Groll A, Candy DC, Preece MA, Tanner JM, Harries JT. Short stature as the primary manifestation of coeliac disease. *Lancet* 1980;2: 1097–1099.
91. Cacciari E, Salardi S, Lazzari R, et al. Short stature and celiac disease: a relationship to consider even in patients with no gastrointestinal tract symptoms. *J Pediatr*. 1983;103:708–711.
92. Bosio L, Barera G, Mistura L, Sassi G, Bianchi C. Growth acceleration and final height after treatment for delayed diagnosis of celiac disease. *J Pediatr Gastroenterol Nutr*. 1990;11:324–329.
93. Zung A, Kori M. Lack of association between seroconversion and catch-up growth in children with celiac disease. *J Pediatr Endocr Met*. 2012;25:131–137.
94. Gemme G, Vignolo M, Naselli A, Garzia P. Linear growth and skeletal maturation in subjects with treated celiac disease. J *Pediatr Gastroenterol Nutr*. 1999;29:339–342.
95. Patwari AK, Kapur G, Satyanarayana L, et al. Catch-up growth in children with late-diagnosed coeliac disease. *Br J Nutr*. 2005;94:437–442.
96. Giovenale D, Meazza C, Cardinale GM, et al. The prevalence of growth hormone deficiency and celiac disease in short children. *Clin Med Res.* 2006;4:180–183.
97. Salardi S, Cacciari E, Volta U, et al. Growth and adult height in atypical coeliac patients, with or without growth hormone deficiency. *J Pediatr Endocr Met*. 2005;18:769–775.
98. Nemet D, Raz A, Zifman E, Morag H, Eliakim A. Short stature, celiac disease and growth hormone deficiency. *J Pediatr Endocr Met*. 2009;22:979–983.
99. Meazza C, Pagani S, Laarej K, et al. Short stature in children with coeliac disease. *Pediatr Endocr Rev*. 2009;6:457–463.
100. Barera G, Mora S, Brambilla P, et al. Body composition in children with celiac disease and the effects of a gluten-free diet: a prospective case-control study. *Am J Clin Nutr*. 2000;72:71–75.
101. Mazure RM, Vazquez H, Gonzalez D, et al. Early changes of body composition in

asymptomatic celiac disease patients. *Am J Gastroenterol*. 1996;91:726–730.
102. Bode S, Hassager C, Gudmand-Hoyer E, Christiansen C. Body composition and calcium metabolism in adult treated coeliac disease. *Gut*. 1991;32:1342–1355.
103. Carbone MC, Pitzalis G, Ferri M, et al. Body composition in coeliac disease adolescents on a gluten-free diet: a longitudinal study. *Acta Diabetol*. 2003;40 Suppl 1:S171–S173.
104. Brar P, Egbuna I, Ramakrishnan S, Holleran S, Green PH. BMI in the celiac disease patients of United States—Abstract. XII. Paper presented at: International Celiac Disease Symposium 2006; 2006;D–325.
105. Dickey W, Kearney N. Overweight in celiac disease: prevalence, clinical characteristics, and effect of a gluten-free diet. *Am J Gastroenterol.* 2006;101:2356–2359.
106. Kabbani TA, Goldberg A, Kelly CP, et al. Body mass index and the risk of obesity in coeliac disease treated with the gluten-free diet. *Aliment Pharmacol Ther*. 2012;35:723–729.
107. Valletta E, Fornaro M, Cipolli M, Conte S, Bissolo F, Danchielli C. Celiac disease and obesity: need for nutritional follow-up after diagnosis. *Eur J Clin Nutr*. 2010;64:1371–1372.
108. Reilly NR, Aguilar K, Hassid BG, et al. Celiac disease in normal-weight and overweight children: clinical features and growth outcomes following a gluten-free diet. *J Pediatr Gastroenterol Nutr.* 2011;53: 528–531.
109. Ukkola A, Maki M, Kurppa K, et al. Changes in body mass index on a gluten-free diet in coeliac disease: a nationwide study. European journal of internal medicine 2012;23:384–388.
110. Cheng J, Brar PS, Lee AR, Green PH. Body mass index in celiac disease: beneficial effect of a gluten-free diet. *J Clin Gastroenterol*. 2010;44:267–271.
111. Kavak US, Yuce A, Kocak N, et al. Bone mineral density in children with untreated and treated celiac disease. *J Pediatr Gastroenterol Nutr*. 2003;37:434–436.
112. Bernstein CN, Leslie WD, Leboff MS. AGA technical review on osteoporosis in gastrointestinal diseases. *Gastroenterology* 2003;124: 795–841.
113. Mora S, Barera G, Beccio S, et al. A prospective, longitudinal study of the long-term effect of treatment on bone density in children with celiac disease. *J Pediatr.* 2001;139:516–521.
114. Scotta MS, Salvatore S, Salvatoni A, et al. Bone mineralization and body composition in young patients with celiac disease. *Am J Gastroenterol*. 1997;92:1331–1334.
115. Fouda MA, Khan AA, Sultan MS, Rios LP, McAssey K, Armstrong D. Evaluation and management of skeletal health in celiac disease: position statement. *Can J Gastroenterol.* 2012;26:819–829.
116. Caraceni MP, Molteni N, Bardella MT, Ortolani S, Nogara A, Bianchi PA. Bone and mineral metabolism in adult celiac disease. *Am J Gastroenterol.* 1988;83:274–277.
117. Molteni N, Caraceni MP, Bardella MT, Ortolani S, Gandolini GG, Bianchi P. Bone mineral density in adult celiac patients and the effect of gluten-free diet from childhood. *Am J Gastroenterol*. 1990;85:51–53.
118. McFarlane XA, Bhalla AK, Robertson DA. Effect of a gluten free diet on osteopenia in adults with newly diagnosed coeliac disease. *Gut*. 1996;39:180–184.
119. Ciacci C, Maurelli L, Klain M, et al. Effects of dietary treatment on bone mineral density in adults with celiac disease: factors predicting response. *Am J Gastroenterol*. 1997;92:992–996.

120. Moreno ML, Vazquez H, Mazure R, et al. Stratification of bone fracture risk in patients with celiac disease. *Clin Gastroenterol Hepatol* 2004;2:127–134.
121. Thomason K, West J, Logan RF, Coupland C, Holmes GK. Fracture experience of patients with coeliac disease: a population based survey. *Gut* 2003;52:518–522.
122. Aine L, Maki M, Collin P, Keyrilainen O. Dental enamel defects in celiac disease. *J Oral Pathol Med*. 1990;19:241–245.
123. Ortega Paez E, Junco Lafuente P, Baca Garcia P, Maldonado Lozano J, Llodra Calvo JC. Prevalence of dental enamel defects in celiac patients with deciduous dentition: a pilot study. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2008;106:74–78.
124. Haapalahti M, Kulmala P, Karttunen TJ, et al. Nutritional status in adolescents and young adults with screen-detected celiac disease. *J Pediatr Gastroenterol Nutr*. 2005;40:566–570.
125. Dickey W. Low serum vitamin B12 is common in coeliac disease and is not due to autoimmune gastritis. *Eur J Gastroenterol Hepatol*. 2002;14:425–427.
126. Dahele A, Ghosh S. Vitamin B12 deficiency in untreated celiac disease. *Am J Gastroenterol*. 2001;96:745–750.
127. Crofton RW, Aggett PJ, Gvozdanovic S, Gvozdanovic D, Mowat NA, Brunt PW. Zinc metabolism in celiac disease. *Am J Clin Nutr*. 1990;52:379–382.
128. Naveh Y, Lightman A, Zinder O. A prospective study of serum zinc concentration in children with celiac disease. *J Pediatr*. 1983;102:734–736.
129. Villanueva J, Maranda L, Nwosu BU. Is vitamin D deficiency a feature of pediatric celiac disease? *J Pediatr Endocr Met*. 2012;25:607–610.
130. Mager DR, Qiao J, Turner J. Vitamin D and K status influences bone mineral density and bone accrual in children and adolescents with celiac disease. *Eur J Clin Nutr*. 2012;66:488–495.
131. Bonamico M, Vania A, Monti S, et al. Iron deficiency in children with celiac disease. *J Pediatr Gastroenterol Nutr*. 1987;6:702–706.
132. Kalayci AG, Kanber Y, Birinci A, Yildiz L, Albayrak D. The prevalence of coeliac disease as detected by screening in children with iron deficiency anaemia. *Acta Paediatr*. 2005;94:678–681.
133. Fisgin T, Yarali N, Duru F, Usta B, Kara A. Hematologic manifestation of childhood celiac disease. *Acta Haematol*. 2004;111:211–214.
134. Green PHR, Stavropoulos SN, Panagi SG, et al. Characteristics of adult celiac disease in the USA: results of a national survey. *Am J Gastroenterol*. 2001;96:126–131.
135. Farre C, Esteve M, Curcoy A, et al. Hypertransaminasemia in pediatric celiac disease patients and its prevalence as a diagnostic clue. *Am J Gastroenterol*. 2002;97:3176–3181.
136. Bardella MT, Fraquelli M, Quatrini M, Molteni N, Bianchi P, Conte D. Prevalence of hypertransaminasemia in adult celiac patients and effect of gluten-free diet. *Hepatology*. 1995;22:833–836.
137. Kaukinen K, Halme L, Collin P, et al. Celiac disease in patients with severe liver disease: gluten-free diet may reverse hepatic failure. *Gastroenterology*. 2002;122:881–888.
138. Rujner J. [Age at menarche in girls with celiac disease]. *Ginekol Pol*. 1999;70:359–362.
139. Bona G, Marinello D, Oderda G. Mechanisms of abnormal puberty in coeliac disease. *Horm Res* 2002;57 Suppl 2:63–65.
140. Sher KS, Mayberry JF. Female fertility, obstetric and gynaecological history in coeliac disease: a case control study. *Acta Paediatr Suppl*. 1996;412:76–77.

141. Smecuol E, Maurino E, Vazquez H, et al. Gynaecological and obstetric disorders in coeliac disease: frequent clinical onset during pregnancy or the puerperium. *Eur J Gastroenterol Hepatol*. 1996;8:63–89.
142. Ludvigsson JF, Montgomery SM, Ekbom A. Celiac disease and risk of adverse fetal outcome: a population-based cohort study. *Gastroenterology*. 2005;129:454–463.
143. Freeman HJ. Reproductive changes associated with celiac disease. *World J Gastroentero.* 2010;16:5810–5814.
144. Shah S, Leffler D. Celiac disease: an underappreciated issue in women's health. *Women's Health* (London, England). 2010;6:753–766.
145. Kumar A, Meena M, Begum N, et al. Latent celiac disease in reproductive performance of women. *Fertil Steril.* 2011;95:922–927.
146. Hadjivassiliou M, Grunewald RA, Kandler RH, et al. Neuropathy associated with gluten sensitivity. *J Neurol Neurosurg Psychiatry.* 2006.
147. Muller AF, Donnelly MT, Smith CM, Grundman MJ, Holmes GK, Toghill PJ. Neurological complications of celiac disease: a rare but continuing problem. *Am J Gastroenterol.* 1996;91:1430–1435.
148. Bushara KO. Neurologic presentation of celiac disease. *Gastroenterology.* 2005;128:S92–S927.
149. Lionetti E, Francavilla R, Pavone P, et al. The neurology of coeliac disease in childhood: what is the evidence? A systematic review and meta-analysis. *Dev Med Child Neurol.* 2010;52:700–707.
150. Niederhofer H, Pittschieler K. A preliminary investigation of ADHD symptoms in persons with celiac disease. *J Atten Disord*. 2006;10:200–204.
151. Gungor S, Celiloglu OS, Ozcan OO, Raif SG, Selimoglu MA. Frequency of celiac disease in attention-deficit/hyperactivity disorder. *J Pediatr Gastroenterol Nutr.* 2013;56:211–214.
152. Kieslich M, Errazuriz G, Posselt HG, Moeller-Hartmann W, Zanella F, Boehles H. Brain white-matter lesions in celiac disease: a prospective study of 75 diet-treated patients. *Pediatrics.* 2001;108:E21.
153. Gobbi G, Bouquet F, Greco L, et al. Coeliac disease, epilepsy, and cerebral calcifications. The Italian Working Group on Coeliac Disease and Epilepsy. *Lancet.* 1992;340:439–443.
154. Ludvigsson JF, Zingone F, Tomson T, Ekbom A, Ciacci C. Increased risk of epilepsy in biopsy-verified celiac disease: a population-based cohort study. *Neurology*. 2012;78:1401–1407.
155. Zelnik N, Pacht A, Obeid R, Lerner A. Range of neurologic disorders in patients with celiac disease. *Pediatrics*. 2004;113:1672–1676.
156. Pavone L, Fiumara A, Bottaro G, Mazzone D, Coleman M. Autism and celiac disease: failure to validate the hypothesis that a link might exist. *Biol Psychiatry.* 1997;42:72–75.
157. Batista IC, Gandolfi L, Nobrega YK, et al. Autism spectrum disorder and celiac disease: no evidence for a link. *Arquivos de neuro-psiquiatria.* 2012;70:28–33.
158. Corrao G, Corazza GR, Bagnardi V, et al. Mortality in patients with coeliac disease and their relatives: a cohort study. *Lancet.* 2001;358:356–361.
159. Logan RF, Rifkind EA, Turner ID, Ferguson A. Mortality in celiac disease. *Gastroenterology*. 1989;97:265–271.
160. Rubio-Tapia A, Rahim MW, See JA, Lahr BD, Wu TT, Murray JA. Mucosal recovery and mortality in adults with celiac disease after treatment with a gluten-free diet. *Am J Gastroenterol.* 2010;105:1412–1420.
161. Swinson CM, Slavin G, Coles EC, Booth CC. Coeliac disease and malignancy.

Lancet. 1983;1:111–115.
162. Askling J, Linet M, Gridley G, Halstensen TS, Ekstrom K, Ekbom A. Cancer incidence in a population-based cohort of individuals hospitalized with celiac disease or dermatitis herpetiformis. *Gastroenterology.* 2002;123:1428–1435.
163. Mearin ML, Catassi C, Brousse N, et al. European multi-centre study on coeliac disease and non-Hodgkin lymphoma. *Eur J Gastroenterol Hepatol.* 2006;18:187–194.
164. Catassi C, Bearzi I, Holmes GK. Association of celiac disease and intestinal lymphomas and other cancers. *Gastroenterology.* 2005;128:S79–S86.
165. Ludvigsson JF. Mortality and malignancy in celiac disease. *Gastrointest Endosc Clin N Am*. 2012;22:705–722.
166. Cereda S, Cefalo G, Spreafico F, et al. Celiac disease and childhood cancer. *J Pediatr Hematol Oncol.* 2006;28:346–349.
167. Schweizer JJ, Oren A, Mearin ML. Cancer in children with celiac disease: a survey of the European Society of Paediatric Gastroenterology, Hepatology and Nutrition. *J Pediatr Gastroenterol Nutr.* 2001;33:97–100.
168. Holmes GK, Prior P, Lane MR, Pope D, Allan RN. Malignancy in coeliac disease—effect of a gluten free diet. *Gut*. 1989;30:333–338.
169. Ventura A, Magazzu G, Greco L. Duration of exposure to gluten and risk for autoimmune disorders in patients with celiac disease. SIGEP Study Group for Autoimmune Disorders in Celiac Disease. *Gastroenterology.* 1999;117:297–303.
170. Petaros P, Martelossi S, Tommasini A, Torre G, Caradonna M, Ventura A. Prevalence of autoimmune disorders in relatives of patients with celiac disease. *Dig Dis Sci*. 2002;47:1427–1431.
171. Cataldo F, Marino V. Increased prevalence of autoimmune diseases in first-degree relatives of patients with celiac disease. *J Pediatr Gastroenterol Nutr*. 2003;36:470–473.
172. Ch'ng CL, Jones MK, Kingham JG. Celiac disease and autoimmune thyroid disease. *Clin Med Res*. 2007;5:184–192.
173. Sategna Guidetti C, Solerio E, Scaglione N, Aimo G, Mengozzi G. Duration of gluten exposure in adult coeliac disease does not correlate with the risk for autoimmune disorders. *Gut.* 2001;49:502–505.
174. Viljamaa M, Kaukinen K, Huhtala H, Kyronpalo S, Rasmussen M, Collin P. Coeliac disease, autoimmune diseases and gluten exposure. *Scand J Gastroenterol.* 2005;40:437–443.
175. Ansaldi N, Palmas T, Corrias A, et al. Autoimmune thyroid disease and celiac disease in children. *J Pediatr Gastroenterol Nutr*. 2003;37:63–66.
176. Sategna-Guidetti C, Volta U, Ciacci C, et al. Prevalence of thyroid disorders in untreated adult celiac disease patients and effect of gluten withdrawal: an Italian multicenter study. *Am J Gastroenterol.* 2001;96:751–757.
177. Metso S, Hyytia-Ilmonen H, Kaukinen K, et al. Gluten-free diet and autoimmune thyroiditis in patients with celiac disease. A prospective controlled study. *Scand J Gastroenterol.* 2012;47:43–48.
178. Case S. The gluten-free diet: how to provide effective education and resources. *Gastroenterology.* 2005;128:S128–S134.
179. Pietzak MM. Follow-up of patients with celiac disease: achieving compliance with treatment. *Gastroenterology.* 2005;128:S135–S1341.
180. Mayer M, Greco L, Troncone R, Auricchio S, Marsh MN. Compliance of adolescents with coeliac disease with a gluten free diet. *Gut.* 1991;32:881–885.
181. Bardella MT, Molteni N, Prampolini L, et al. Need for follow up in coeliac disease. *Arch Dis Child*. 1994;70:211–213.

182. Hogberg L, Grodzinsky E, Stenhammar L. Better dietary compliance in patients with coeliac disease diagnosed in early childhood. *Scand J Gastroenterol.* 2003;38:751–754.
183. Fabiani E, Catassi C, Villari A, et al. Dietary compliance in screening-detected coeliac disease adolescents. *Acta Paediatr Suppl.* 1996;412:65–67.
184. Kupper C. Dietary guidelines and implementation for celiac disease. *Gastroenterology.* 2005;128:S121–S127.
185. Janatuinen EK, Kemppainen TA, Julkunen RJ, et al. No harm from five year ingestion of oats in coeliac disease. *Gut.* 2002;50:332–325.
186. Janatuinen EK, Pikkarainen PH, Kemppainen TA, et al. A comparison of diets with and without oats in adults with celiac disease. *N Engl J Med.* 1995;333:1033–1037.
187. Holm K, Maki M, Vuolteenaho N, et al. Oats in the treatment of childhood coeliac disease: a 2-year controlled trial and a long-term clinical follow-up study. *Aliment Pharmacol Ther.* 2006;23:1463–1472.
188. Koskinen O, Villanen M, Korponay-Szabo I, Lindfors K, Maki M, Kaukinen K. Oats do not induce systemic or mucosal autoantibody response in children with coeliac disease. *J Pediatr Gastroenterol Nutr.* 2009;48:559–565.
189. Pulido OM, Gillespie Z, Zarkadas M, et al. Introduction of oats in the diet of individuals with celiac disease: a systematic review. *Adv Food Nutr Res.* 2009;57:235–285.
190. Bode S, Gudmand-Hoyer E. Incidence and clinical significance of lactose malabsorption in adult coeliac disease. *Scand J Gastroenterol.* 1988;23:484–488.
191. Roggero P, Ceccatelli MP, Volpe C, et al. Extent of lactose absorption in children with active celiac disease. *J Pediatr Gastroenterol Nutr.* 1989;9:290–294.
192. Basso MS, Luciano R, Ferretti F, et al. Association between celiac disease and primary lactase deficiency. *Eur J Clin Nutr.* 2012;66:1364–1365.
193. Kemppainen T, Uusitupa M, Janatuinen E, Jarvinen R, Julkunen R, Pikkarainen P. Intakes of nutrients and nutritional status in coeliac patients. *Scand J Gastroenterol.* 1995;30:575–579.
194. Hopman EG, le Cessie S, von Blomberg BM, Mearin ML. Nutritional management of the gluten-free diet in young people with celiac disease in The Netherlands. *J Pediatr Gastroenterol Nutr.* 2006;43:102–108.
195. Hallert C, Grant C, Grehn S, et al. Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. *Aliment Pharmacol Ther.* 2002;16:1333–1339.
196. Hjelt K, Krasilnikoff PA. The impact of gluten on haematological status, dietary intakes of haemopoietic nutrients and vitamin B12 and folic acid absorption in children with coeliac disease. *Acta Paediatr Scand.* 1990;79:911–919.
197. Mariani P, Viti MG, Montuori M, et al. The gluten-free diet: a nutritional risk factor for adolescents with celiac disease? *J Pediatr Gastroenterol Nutr.* 1998;27:519–523.
198. Kelly CP, Green PH, Murray JA, et al. Larazotide acetate in patients with coeliac disease undergoing a gluten challenge: a randomised placebo-controlled study. *Aliment Pharmacol Ther.* 2013;37: 252–262.
199. Crespo Perez L, Castillejo de Villasante G, Cano Ruiz A, Leon F. Non-dietary therapeutic clinical trials in coeliac disease. *Eur J Intern Med.* 2012;23:9–14.
200. Sollid LM, Khosla C. Future therapeutic options for celiac disease. *Nat Clin Pract Gastroenterol Hepatol.* 2005;2:140–147.
201. Green PH, Rostami K, Marsh MN. Diagnosis of coeliac disease. Best Pract Res Clin Gastroenterol. 2005;19(3):389–400.

Food Allergies

54

Julie Wang, MD, Jennifer M. Maloney, MD, and Hugh A. Sampson, MD

DEFINITION

The term “allergy” is subject to a wide range of interpretations. Many conditions are labeled as allergic disorders; however, a clear immunologic basis for disease is not present. As a result, a clear definition is necessary before delving into the subject of food allergy.

Adverse reactions to food include a wide spectrum of food allergic and nonallergic reactions (see Figure 54-1). Food allergies result from immunologic responses to foods, through either IgE or non-IgE-mediated mechanisms, that occur reproducibly on exposure to the triggering food.[1] Nonallergic adverse food reactions include intolerances that result from host factors,[1] such as enzyme deficiencies, for example, lactase deficiency, which results in the bloating, abdominal pain, and occasionally diarrhea of lactose intolerance. Toxic food reactions represent a separate category, which result from inherent toxicities or properties of the food, for example, histamine-mediated symptoms due to scromboid fish poisoning.[1]

PREVALENCE

Although 10%–50% of the population believes they have adverse reactions to foods, not all of these reactions are immune mediated.[1] Many studies have investigated the prevalence of food allergies, however, few population studies have used double-blind, placebo-controlled food challenges (DBPCFCs) to confirm the diagnosis of food allergy, which can lead to an overestimate of prevalence. Rona et al. performed a meta-analysis focusing on milk, egg, peanut, and seafood allergy, and determined the prevalence of food allergies to be approximately 3.5%.[2] The majority of the studies included in this meta-analysis used self-

reported food allergy, many used skin prick testing (SPT) and food-specific IgE levels to confirm sensitization to the food allergens, and some employed DBPCFCs. A US study that used several national health databases and health-care surveys to determine food allergy prevalence concluded that 3.9% of US children report having food allergy, which is an 18% increase in prevalence between 1997 and 2007.[3] More recently, higher estimates have been reported in the United States (8.0%, based on self-report),[4] in Canada (7.14%, based on self-report),[5] and in Australia (>10% of a general infant cohort had challenge-proven food allergy).[6]

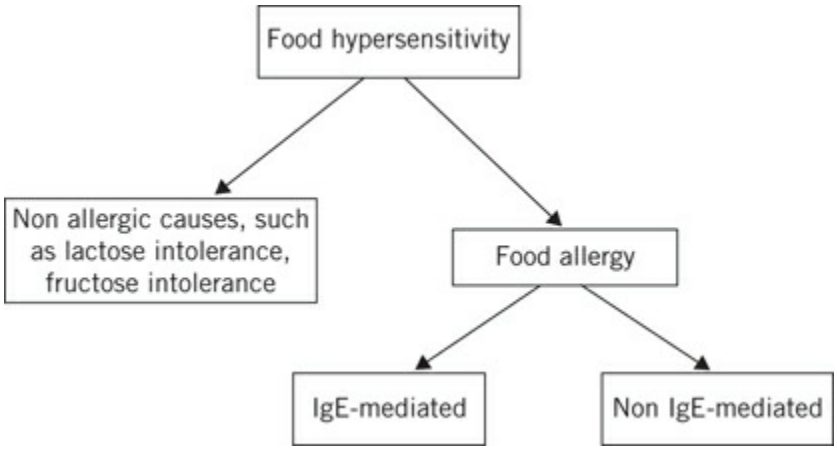


Figure 54-1 Classification of food hypersensitivities.

Cow milk allergy affects approximately 1%–2% of infants in their first year of life.[1] IgE-mediated processes account for approximately 60% of cases. Hypersensitivity to egg affects approximately 1.6% of children,[1] and peanut affects over 0.06%–5.9% of children.[1] Interestingly, over an 11-year period, the rate of peanut allergy in the United States appears to have tripled in young children.[7]

The persistence of symptomatic food allergy varies depending on the food. It used to be believed that most children outgrew their milk and egg allergies by school-age, however, more recent reports indicate that it is taking longer for children to outgrow their milk and egg allergy, with most developing tolerance by their mid-teenage years.[8,9] In contrast, only 20% of children with peanut allergy and 9% with tree nut allergy will develop tolerance.[10,11]

Pollen food allergy syndrome (PFAS, also known as oral allergy

syndrome) is a common symptom complex, which usually provokes isolated oral symptoms following exposure to heat-labile proteins in uncooked fruits and vegetables. Pollen allergic individuals are at risk due to the homology between specific pollen proteins and proteins in certain foods. Of patients with allergic rhinitis, it has been estimated that 23%–76% of patients experience oral symptoms to at least one food.[12] The cooked version of the foods is typically tolerated due to disruption of the protein conformation or tertiary structure.

Food allergy commonly presents in patients with or who will develop other atopic diseases. Approximately one-third of children and adolescents with moderate-severe atopic dermatitis have IgE-mediated food allergy.[1] In addition, the presence of food allergy increases the probability of developing allergic airway disease.[1] Despite the concomitant existence of food allergy and asthma, wheezing is rarely an isolated finding in a food allergic reaction.[13]

PATHOPHYSIOLOGY

Food allergy results from an atypical response of the mucosal immune system to orally consumed antigens. The gastrointestinal (GI) mucosa is an extensive structure responsible for digestion and absorption of nutrients as well as protection from pathogenic organisms. A nonspecific physical barrier works with immune and nonimmune cells and cytokines to maintain immunologic homeostasis.

The physical constituents of the GI mucosa comprise epithelial cells, the mucin glycoprotein lining, trefoil factors, proteolytic enzymes, and bile salts. The epithelial cells are attached by intercellular tight junctions. The mucin lining traps large particles due to its viscous nature and functions as a reservoir for secretory IgA (sIgA), which entraps bacteria and viruses. Trefoil peptides maintain barrier function by restoring its integrity if defects occur. Proteolytic enzymes, bile salts, and exposures to wide ranges of pH gradients work to eliminate pathogenic organisms and modify the immunogenicity of proteins due to proteolysis and emulsification.[14]

Furthermore, the mucosal immune system provides an active line of defense through both the innate and adaptive arms. Peyer’s patches (organized lymphoid structures of the small intestine and rectum), sIgA, dendritic cells, antigen-presenting macrophages, MHC class I and II bearing T lymphocytes, intestinal epithelia cells, as well as other cytokine-

producing cells allow for immunologic responsiveness.[15] Despite the complex interplay of the mucosal system, approximately 2% of intact food proteins are absorbed through the mature GI tract and reach the lymphatic and portal circulation. The intestinal permeability is increased during infancy, and young children's vulnerability is further augmented by the nature of their immature mucosal barrier, which has decreased gastric acid production and reduced pancreatic and intestinal enzymatic activity.[16] Consequently, there is increased absorption of intact food proteins, which may cause stimulation of the immune system and generation of IgE antibody.[16]

Oral tolerance allows individuals to encounter immense quantities of dietary protein and commensal bacteria without inciting a dynamic immune response. Antigenic factors and host factors are involved in the generation of oral tolerance, see Table 54-1. Antigenic factors, which influence oral tolerance, include the form and the dose of the antigen. Soluble antigens are more tolerogenic than particulate antigens. Antigenic dose is also a key component to tolerance development. Experiments in mice have shown that oral tolerance can be induced by the administration of a single high dose of allergen or repeated low doses.[17] High-dose oral tolerance may occur through Fas-mediated apoptosis or through the generation of anergy. Anergy occurs when T cells encounter antigens in the absence of co-stimulatory signaling. Low-dose tolerance develops as a result of regulatory T cell activation. The best categorized regulatory T cells are CD4+ cells (TH3 via TGF-β, TR1 via IL10, CD4+CD25+ cells possibly via surface bound TGF-β), CD8+ suppressor cells, and NK1.1+T cells.[15,18]

TABLE 54-1 Factors Influencing the Development of Oral Tolerance

Antigenic Factors

- Form – soluble antigens are more tolerogenic than particulate antigens
- Dose – tolerance is induced by single high dose or repeated low doses

Host Factors

- Age – ability to generate oral tolerance increases with age
- Genetics – genetic composition influence oral tolerance
- Gut Flora – contributes to the development of oral tolerance

Alternate routes of exposure (i.e., via skin or respiratory tract) can lead to loss of oral tolerance. Peanut exposure via the epicutaneous or epidermal routes in a murine model with slight inflammation of the skin induces Th2 immune responses and promotes allergic sensitization.[19] In humans, increased rates of peanut allergy were found in children with atopic dermatitis, who used topical creams containing peanut oil (OR 6.8).[20] Specific aeroallergen exposure through the respiratory route leads to PFAS.[12]

HOST FACTORS

Host factors involved in the generation of oral tolerance include age, genetics, and GI flora. The ability to become tolerant to food antigens appears to become more effective with age. Neonates develop a stronger immunologic response to dietary proteins in their first few months of life, as demonstrated by rapid increases in circulating antibodies when fed milk or soy formulas,[21] and prevalence of food allergies is higher in young children as compared to adults.[4] However, a few recent studies suggest early introduction of foods may be beneficial in some cases. A comparative study of Jewish children in Israel and the United Kingdom found a 10-fold lower incidence of peanut allergy in Israel where infants are fed peanut proteins [Bamba®, Osem Corporation, Israel] starting at an early age as compared to the United Kingdom where peanut is not introduced until children are much older.[22] The Learning Early About Peanut Allergies (LEAP) study is currently exploring the effect of early peanut allergen exposure on the development of peanut allergy. Similarly, introduction of cow's milk in the first two weeks of life was associated with the lowest incidence of cow's milk allergy in a large cohort of Israeli children.[23]

Host genetic composition influences the development of oral tolerance or food hypersensitivity, as demonstrated in murine models and human disease. Induction of oral tolerance in mice differs depending on the strain of mouse.[24] In humans, a higher frequency of three HLA class II genotypes has been found in peanut allergic patients.[25] In addition, monozygotic twins have been shown to have a high concordance rate of peanut allergy,[26] supporting the interplay of genetic influence on oral tolerance and food hypersensitivity.

Commensal gut flora take residence in the GI tract within 24 hours

after birth at a concentration that has been estimated to be between 10^{12} and 10^{14} bacteria per gram of colon tissue.[14] The impact of gut flora on oral tolerance is supported by the finding that mice raised in germfree environments following birth are unable to develop tolerance to orally administered OVA.[27] Furthermore, both mice with TLR 4 deficiency plus normal GI flora and mice who have intact TLR 4 signaling but are raised in germfree conditions are susceptible to the induction of allergy,[28] suggesting that cooperation between the innate immune system and the gut flora is essential for oral tolerance induction. A recent study demonstrated that mice with food allergy have a microbiota signature that is distinct from congenic, but food allergy-resistant wild-type control mice.[29] Transfer of microbiota from food allergic mice to naïve, germfree mice resulted in transmission of food allergy susceptibility. Furthermore, the study demonstrated that the microbiota signature could be reprogrammed to a tolerance-associated signature after treatment with allergen-specific T regulatory cells. Therefore, the potential for probiotics in the prevention and treatment of human allergic disease has generated great interest. Results from several studies have demonstrated effectiveness of probiotics in the prevention of eczema in certain populations; however, benefits for food allergy prevention have not yet been reported.[30–32]

Other factors that can influence the clinical response of food allergy include comorbid asthma, exercise, viral infections, and concurrent use of alcohol or medications. Asthma is known to be a risk factor for more severe anaphylaxis.[33] Exercise, use of alcohol, or medications (aspirin, beta-blockers, angiotensin converting enzyme inhibitors, monoamine oxidase inhibitors, tricyclic antidepressants), and concurrent infection may increase the severity of anaphylactic reactions or diminish the efficacy of epinephrine.[34]

FOOD ALLERGEN FACTORS

Despite the wide range of foods that humans consume, surprisingly few foods account for the majority of food allergies. Milk, egg, peanut, wheat, and soy are responsible for most food-induced reactions in children, whereas peanut, tree nuts, fish, and shellfish cause most reactions in adults.[1]

Two forms of IgE-mediated food allergy have been proposed. Class 1 food allergy results from sensitization through the GI tract.[35] The class 1 food allergens are generally 10–70 kd in size and highly stable when

subjected to heat, acid, or proteases.[16] Examples of class 1 food allergens include milk (caseins), peanut (vicilins), egg (ovomucoid), and nonspecific lipid transfer proteins.

Class 2 food allergy results from sensitization to inhalant allergens that are partially homologous to proteins in certain fruits and vegetables, and principally occurs in adolescents and adults. Class 2 allergens are heat labile and susceptible to digestive processes.[35] Consequently, symptoms occur when the food is ingested in the raw form, but not when cooked. For example, Bet v 1, a major allergen of birch pollen, shares homology with class 2 allergens in various fruits and vegetables (e.g., apple, Mal d 1; carrot, and Dau c 1). A similarity in the three-dimensional structures (rather than the overall sequence identity) between birch pollen epitopes and the epitopes of specific foods has been demonstrated.[36] IgE antibody recognition of structurally related epitopes among pollens and foods likely results in PFAS.

Different phenotypes of food allergy may be distinguished by the presence of specific IgE to sequential or conformational epitopes. Transient allergy to milk and egg is primarily due to IgE antibody binding to conformational epitopes whereas persistent allergy to milk and egg is characterized by significant binding to sequential epitopes.[37,38] Recent studies show that most milk and egg allergic individuals are able to tolerate extensively heated or baked forms of these foods,[39,40] suggesting that these individuals have specific IgE to conformational epitopes that are disrupted by heating. In addition, different patterns of epitope recognition or epitope diversity may provide additional insight on the clinical manifestations of allergic reactions to peanut and milk.[41,42] Although heating can reduce the allergenicity of milk and egg proteins, it is important to know that heating peanut proteins at high temperatures (i.e., roasting) increases the allergenicity of peanut allergens.[43]

REACTION MECHANISMS

IgE-Mediated Reactions

Food allergic reactions involving IgE antibodies occur when IgE binds to and cross-links the high affinity IgE receptor (FcεRI) on mast cells and basophils. Following mast cell and basophil activation, preformed and newly synthesized mediators are released. The mediators include histamine, leukotrienes, numerous cytokines, chemokines, and proteases.[44]

Perivascular mast cells are present in significant concentrations in

areas that border the external environment, such as skin and the respiratory and GI tracts. When activated, mediator release produces vasodilation, bronchoconstriction, and mucus production, which leads to the symptoms of an immediate hypersensitivity reaction. Six to 24 hours after the initial allergen encounter, a late-phase reaction may occur which is characterized by edema and an inflammatory cell influx.[44]

IgE-mediated reactions usually occur within minutes to one hour following ingestion of the causal food. Multiple organ systems may be involved including the skin, respiratory tract, GI tract, and cardiovascular system.

Non IgE-Mediated Reactions

Several food hypersensitivity disorders exist that are not solely IgE-mediated. Cell-mediated hypersensitivity reactions, orchestrated by antigen-sensitized T cells, may be responsible for the manifestations of certain conditions. A few diseases are thought to result from a combination of cell-mediated and IgE-mediated mechanisms, for example, atopic dermatitis, allergic eosinophilic esophagitis.[1]

CLINICAL FOOD ALLERGIC DISORDERS

Food-Induced Anaphylaxis

Food-induced anaphylaxis is a severe, potentially fatal, systemic allergic reaction that occurs rapidly after exposure to an allergy-causing food.[45] The majority of anaphylactic events are characterized by cutaneous symptoms, such as urticaria, angioedema, and flushing, although the absence of skin involvement does not exclude the diagnosis. For example, cardiovascular collapse with resultant shock may occur without cutaneous symptoms due to decreased blood supply to the skin.[34] In fact, in a report of six food-induced, fatal and seven near-fatal, anaphylactic reactions, 38% of reactions were not accompanied by cutaneous symptoms.[33]

As with any IgE-mediated reaction, there may be a late-phase response associated with anaphylaxis. The incidence of such reactions is described to occur in up to 20% of food-induced events.[45] The only published series of anaphylactic episodes in a pediatric population, reports the incidence of biphasic reactions to be 6%.[46] With biphasic reactions, a period of recovery after the initial reaction is followed by a recurrence of symptoms. Late-phase reactions are variable, may be severe and severity cannot be predicted based on earlier symptoms.

Food-associated exercise-induced anaphylaxis usually occurs when exercise follows the ingestion of a specific food (such as wheat, shellfish, celery) by approximately 2–4 hours.[1] A less common form of food-associated exercise-induced anaphylaxis occurs with the ingestion of any food prior to exercise. The pathogenesis of food-dependent exercise-induced anaphylaxis is unclear.

Cutaneous Food Hypersensitivity Reactions

Acute urticaria and angioedema are the most common manifestations of food-induced allergic reactions (Table 54-2).[1] The onset of symptoms may occur minutes after ingestion. Urticaria also frequently develops as a result of direct skin contact with food allergens. Contact reactions are typically localized to areas of direct exposure and are unlikely to provoke systemic reactions, unless inadvertent ingestion occurs.[47] Chronic urticaria and angioedema are defined as the presence of symptoms for more than six weeks and are infrequently due to food allergy.

Atopic dermatitis is a mixed IgE-mediated and non-IgE-mediated disorder. It is a chronic skin condition that often starts in childhood and is characterized by a relapsing and remitting course. Acute lesions are intensely pruritic and appear as erythematous macular and papulovesicular lesions. Scratching results in excoriation with crusting and exudation. The rash may progress to a subacute form with erythematous, scaling papules and further advances with the chronic changes of skin thickening or lichenification.[48]

The cutaneous lesions of atopic dermatitis usually occur on the face, scalp, and extensor surfaces in young infants. The distribution tends to shift as children get older, affecting the flexural surfaces of the extremities.[48] Atopic dermatitis skin lesions are characterized by a Th2 cytokine profile (acute lesions—IL-4, IL-5, IL-13; chronic lesions—IL-5, IL-13). These cytokines result in an inflammatory state with upregulation of adhesion molecules on vascular endothelial cells, upregulation of the high-affinity IgE receptor on Langerhan's cells and other antigen presenting cells, recruitment of inflammatory cells to the skin, and local production of IgE.

TABLE 54-2 Cutaneous Hypersensitivity Disorders

Cutaneous manifestations of food allergy
IgE mediated
Urticaria and angioedema, skin erythema, morbilliform rash

Mixed IgE and cell mediated
Atopic dermatitis
Cell mediated
Contact dermatitis, dermatitis herpetiformis

Approximately one-third of children and adolescents with moderate to severe atopic dermatitis have IgE-mediated food hypersensitivity.[1] When patients with food allergy-related atopic dermatitis undergo food challenges, positive challenges are usually hallmarked by cutaneous symptoms such as pruritic, morbilliform, or macular eruptions with a tendency to occur in skin areas affected by atopic dermatitis. In addition to affecting the skin, the majority of positive food challenges trigger other organ system involvement (GI or respiratory). Egg, milk, wheat, peanut, tree nuts, and soy are the most frequent foods to which patients are clinically reactive,[1] with egg affecting approximately two-thirds of patients with food allergy and atopic dermatitis.[49] If the history reveals food-related symptoms and tests for specific IgE to the food are positive, elimination is the best initial approach. However, caution is needed because although removal of a causal food may improve the skin disease, the patient develops an increased probability of an acute allergic reaction, even anaphylaxis, with reintroduction of the food.[50]

Dermatitis herpetiformis is a polymorphous, pruritic rash characterized by erythematous, urticarial plaques, papules, and vesicles. It develops as a result of gluten sensitivity, usually in association with gluten-sensitive enteropathy (celiac disease). Patients with dermatitis herpetiformis have serum IgA autoantibodies to epidermal transglutaminase.[51]

Contact dermatitis is a non-IgE-mediated skin condition, which may be provoked by food exposures.[1]

Respiratory Food Hypersensitivity Reactions

Food-induced respiratory symptoms usually occur in conjunction with other organ system reactions (Table 54-3).[13] Respiratory symptoms consist of rhinoconjunctivitis, laryngeal edema, cough, and/or bronchospasm. Comorbid asthma is a risk factor for more severe food allergic reactions, including fatal and near-fatal anaphylaxis.[33] In addition, having a food allergy has been shown to be associated with life-threatening asthma. In a case-control study, it was demonstrated that children requiring intubation at the time of asthma exacerbation were 5.9 times more likely to have a food allergy compared with matched children who had milder asthma

exacerbations.[52]

TABLE 54-3 Respiratory Hypersensitivity Disorders

Respiratory manifestations of food allergy
IgE mediated
Bronchospasm, cough, rhinitis, congestion, rhinorrhea, sneeze
Mixed IgE and cell mediated
Asthma
Cell mediated
Heiner's syndrome

Rhinitis and nasal symptoms are commonly seen in patients who have reactions during food challenges. Just as it is uncommon to have isolated food-induced asthmatic symptoms, it is very unusual to have nasal symptoms as the only manifestation of an acute food-allergic reaction.[53]

Allergic reactions can develop as a result of inhalation of airborne food proteins. This modality of exposure is more significant when vapors or steams (from foods such as fish or eggs) are being emitted during the cooking process.[54] Inhalation reactions secondary to peanuts, such as particulate matter from peanut dust when shells are opened, have also been reported.[55] In contrast, exposure of peanut allergic patients to the smell of peanut butter, due to aromatic pyrazines, has not been shown to result in systemic or respiratory symptoms.[47]

Food-induced pulmonary hemosiderosis (Heiner's syndrome) is a rare non-IgE-mediated hypersensitivity reaction primarily to milk, which results in the development of recurrent pneumonia, hemosiderosis, GI blood loss, iron deficiency anemia, and failure to thrive. Removal of milk from the diet results in symptom resolution.[1]

Gastrointestinal Food Hypersensitivity Disorders

Food-induced reactions affecting the GI tract may be IgE-mediated, non-IgE-mediated, or due to an interplay of IgE and non-IgE-mediated reactivity (Table 54-4). GI anaphylaxis is an IgE-mediated reaction that includes symptoms of acute nausea, abdominal pain, colic, vomiting, and/or diarrhea. More indolent symptoms may occur in infants with atopic dermatitis and food allergy who are chronically ingesting the allergen. In these patients, intermittent vomiting and failure to thrive may be part of the symptom constellation.[16]

PFAS, also known as oral allergy syndrome, is also an IgE-mediated reaction that occurs in pollen allergic individuals due to homologous proteins shared between foods and specific airborne pollens.[12] Patients with birch pollen allergy may develop oropharyngeal symptoms with raw apple, carrot, potato, celery, pear, kiwi, stone fruits, hazelnut, almond, and peanut; patients with ragweed pollen allergy may be sensitive to various melons and banana; patients with grass pollen allergy might become symptomatic with tomato, melons, and kiwi; and patients with mugwort pollen allergy may develop symptoms with celery, carrots, and spices. Symptoms of PFAS are usually confined to the oropharyngeal cavity, and the foods are usually tolerated in the cooked form because the allergens do not retain their tertiary structure at high temperatures.

TABLE 54-4 Gastrointestinal Hypersensitivity Disorders

Gastrointestinal manifestations of food allergy
IgE mediated
Nausea, abdominal pain, colic, vomiting, diarrhea
Pollen food allergy syndrome ⇒ usually only oropharygeal symptoms
Mixed IgE and cell mediated
Allergic eosinophilic esophagitis, eosinophilic gastroenteritis
Cell mediated
Food protein induced enterocolitis, proctocolitis, celiac disease

Eosinophilic esophagitis (EoE) and gastroenteritis are disorders believed to be due to Th2-mediated mechanisms. Eosinophils infiltrate the walls of the esophagus, stomach, and small intestine. Peripheral eosinophilia is found in approximately half of the patients.[56] When evaluating these patients, other known causes of eosinophilia must be excluded, such as GI reflux, drug reactions, parasitic infections, and malignancy. Patients with GI eosinophilic disorders have a high incidence of concomitant atopy, with many having sensitizations to foods and environmental allergens.

In a disease-free state, eosinophils are usually not present in the esophagus. EoE may occur at any age from infancy through adolescence and into adulthood. Symptoms include abdominal pain, spitting-up or vomiting, food refusal, and gastroesophageal reflux symptoms in younger patients, and midsternal chest pain, dysphagia, and food impaction in adolescents and adults. Diagnosis is defined by multiple criteria: evidence

of esophageal dysmotility, histology showing at least 15 esophageal intraepithelial eosinophils per 40× high power field, persistence of esophageal eosinophilia despite proton pump inhibitor therapy, and remission of disease with dietary intervention or topical corticosteroid therapy.[57] Histological features include eosinophils in the proximal and distal esophagus, thickened esophageal mucosa, papillary elongation, and basal zone hyperplasia. Endoscopic findings include furrowing (vertical, linear creases), mucosal rings, strictures, ulcerations, and whitish papules that are secondary pockets of subepithelial eosinophils.[57]

Allergic eosinophilic gastroenteritis is much less common than EoE and may occur at any age. Most notably, patients frequently present with weight loss and failure to thrive.[56] Additional symptoms include abdominal pain, emesis, nausea, and diarrhea. Especially in young infants, presentation may consist of gastric outlet obstruction or symptoms resembling pyloric stenosis. Diagnosis is made through biopsies, which reveal significant eosinophilic infiltration of the gastric and/or duodenal mucosa, with no other identifiable medical cause (i.e., infection, inflammatory bowel disease, drug reaction). A subgroup of patients develop anemia and hypoalbuminemia, secondary to fecal loss of blood and a protein-losing enteropathy.[58]

Allergic proctocolitis is a non-IgE-mediated, eosinophilic disorder that typically presents in the first few weeks to months of life.[1] Infants appear in good health; however, they have blood and mucus in the stool, which increases in frequency unless the causal allergen is removed from the diet. Growth delay and poor weight gain are not usual presenting features.

Proctocolitis commonly occurs in breast-fed infants (as many as 60% of cases), with milk and soy formulas being the causative foods in the majority of the remaining cases.[1] For infants who develop proctocolitis while breast-feeding, it is believed that cow's milk proteins consumed by the mother are the triggering agents in breast milk, and it has been demonstrated that β-lactoglobulin (one of the major allergens in milk) is detectable in the majority of breast milk samples following maternal cow milk consumption.[59] Elimination of cow's milk from the mother's diet usually will bring about gradual resolution of symptoms; however, if cessation of bleeding does not occur with maternal dietary manipulation, a casein hydrolysate formula, or in rare instances, an amino acid–based formula[60] eliminates gross symptoms, typically within 48–72 hours. For formula-fed infants drinking cow milk formula, a large percentage who have milk-induced proctocolitis, will also become symptomatic to other

foods if introduced in the first six months of life, for example, soy ingestion.[1] A casein hydrolysate or amino acid formula is the recommended treatment.

Food protein-induced enterocolitis syndrome (FPIES) is a non-IgE-mediated food allergy, which usually occurs in formula-fed infants, although it has been reported to a variety of foods (especially cereal grains) in older infants, and is characterized by irritability, excessive, vigorous vomiting, and diarrhea.[1] Diarrhea may have occult blood and stool smears may reveal leukocytes and eosinophils. The stool may also be positive for reducing substances. Depending on the severity of symptoms, patients may develop dehydration with accompanying hypotension and lethargy. Metabolic derangements such as acidosis and hyponatremia can occur as does leukocytosis and a "left-shift." In severe cases, children may even present with methemoglobinemia.

Symptoms may follow a more indolent course for children who are fed the causal food from birth or shortly thereafter. Failure to thrive and hypoalbuminemia in conjunction with chronic emesis and diarrhea may be presenting features. When the causal food is removed and later reintroduced, repetitive vomiting within 2–3 hours and late-onset diarrhea occur.[1] Milk and soy are most frequently the causative foods in food-protein–induced enterocolitis; however, solid foods (such as oat, barley, rice, chicken, and turkey) may be responsible.[61] Approximately 50% of patients reactive to milk also develop reactivity to soy, necessitating treatment with a casein hydrolysate formula or, in rare instances, an amino acid–based formula.[62]

Celiac disease is a chronic inflammatory disorder of the small intestine. An interplay between genetic risk factors and dietary triggers are thought to result in the manifestations of this disease, which include intestinal villus atrophy, malabsorption, and chronic inflammation of the small intestinal mucosa. The major associated haplotypes are HLA-DQ2 and HLA-DQ8, with approximately 95% of patients having HLA-DQ2. Celiac disease is strongly associated with dermatitis herpetiformis, and patients with celiac disease are at increased risk for other autoimmune conditions, such as type 1A diabetes mellitus and autoimmune thyroiditis.[63]

As a result of the immunologic imbalance, patients with celiac disease react to peptide sequences of gluten proteins. Responsible gluten proteins are present in wheat, rye, and barley. Unlike IgE-mediated allergy to wheat, barley, or rye, the T cell cytokine profile from patients with celiac

disease has a T_H1 bias. Removal of gluten from the diet leads to symptom resolution. Patients with untreated disease are at increased risk for T cell non-Hodgkin's lymphoma.[63]

The diagnostic gold standard for celiac disease is the intestinal biopsy, which demonstrates a loss of villi, lymphocytic, and plasma cell infiltration of the lamina propria and intraepithelial compartments and crypt lengthening. Detection of IgA anti-endomysial and transglutaminase-2 antibodies are accurate for the diagnosis of celiac disease.[64] Importantly, testing for celiac disease, either by biopsy or through serologic studies, must be performed while the patient has a diet that includes gluten; otherwise, false-negative results may occur.

DIAGNOSIS

The diagnosis of food allergies requires a comprehensive history, specifically focusing on adverse reactions to particular foods, the time frame of such reactions, whether other contributing factors were present (i.e., exercise), the extent of organ system involvement, and required treatment.[1] Attention to accompanying atopic conditions in the patient and family history of allergy are important. Diet diaries may be helpful in recognizing patterns of reactivity, in identifying commonly consumed allergens, and in identifying foods that may have "hidden allergens," which have provoked the reaction.

The history may be more helpful in situations when food ingestions are causally linked to a reaction, such as the development of urticaria and cough immediately after cashew consumption. For chronic disorders, such an atopic dermatitis, asthma, and GI disorders, the history has a poor predictive value. This point is highlighted by a study in which double-blind placebo-controlled food challenges were conducted in 38 children with severe persistent asthma, where the asthma symptoms were attributed to food allergy. Only 20% of the food challenges (70 challenges conducted) resulted in symptoms. Furthermore, the challenges elicited symptoms predominantly affecting the GI tract despite the chronic respiratory complaints.[65]

Diagnostic Modalities for IgE-Mediated Disorders

SPT is a rapid screening method for IgE-mediated food hypersensitivity. A small amount of food allergen extract is placed on the skin and then subjected to a prick or puncture, which break the skin integrity. A negative

control (saline) and a positive control (histamine) are also placed on the skin. Wheals which are 3 mm larger than the negative control are considered positive.

Unfortunately, the positive predictive value of a positive SPT is less than 50% when compared to results of double-blind placebo-controlled food challenges.[1] Therefore, the medical history remains important because SPTs must be interpreted in the context of the clinical situation. Positive SPTs may be helpful especially when a clear cut history of food reactivity is present. In addition, when hypersensitivity is suspected to milk, egg, or peanut, "diagnostic SPT mean wheal diameters" have been defined (cow milk ≥8 mm, egg ≥7 mm, peanut ≥8 mm; for children ≤2 years of age, cow milk ≥6 mm, egg ≥5 mm, peanut ≥4 mm).[66] However, test results are highly dependent on reagents used (which are not standardized), application technique, and interpretation by the physician. Negative skin tests essentially exclude IgE-mediated allergy.

Intradermal skin testing has no place in the diagnosis of food allergy (Table 54-5).[1] In addition to having poor specificity and positive predictive value, fatalities have been reported with the use of this diagnostic modality.

Detection of food-specific IgE antibodies (i.e., ImmunoCAP, Thermo Fisher Scientific; Waltham, MA) has been shown to be predictive in symptomatic IgE-mediated food allergy.[67] The ImmunoCAP system contrasts with qualitative radioallergosorbent test (RAST) assays because standardized allergens are used, the dose response curve is calibrated against the World Health Organization IgE standard and the matrix binds more antigen, which provides a steep dose response curve. Diagnostic "decision points" have been generated for common food allergens, namely milk, egg, peanut, and fish (see Figure 54-2). The decision points indicate reaction likelihood (a >95% probability of reaction), but do not predict reaction severity. As serum-specific IgE levels decrease to undetectable, the likelihood of reaction decreases. However, reactions may still occur for a small subset of patients when serum-specific IgE levels are undetectable.

TABLE 54-5 Methods of Diagnosing Food Allergy

Diagnosis of Food Allergy
History ± diet diary
Oral food challenge
IgE mediated and mixed disease
Prick skin testing

Serum specific IgE testing

Cell mediated and mixed disease

± Patch testing

Whereas SPT and serum-specific IgE tests are instrumental in the diagnosis of IgE-mediated food allergy, for non-IgE-mediated and mixed disorders, their diagnostic utility has little or no value. Patch testing is a modality being considered for diagnosing delayed contact hypersensitivity reactions where T lymphocytes are the major effector cell. For testing, the food allergen is placed on a small area of the skin under an occlusive dressing. The patch is removed after 48 hours, and the area is examined at 48 hours and again at 72 hours. Positive tests demonstrate erythema, edema, papules, vesicles, and/or bullae, and are graded based on severity of reactivity. Although patch testing is a common diagnostic practice for cutaneous contact allergy due to chemicals, its use is not typically used for the diagnosis of food-related conditions.[1] Patch testing may have some utility in the evaluation of food allergy in those with atopic dermatitis or EoE, but this remains to be established with further studies.[1]

The DBPCFC remains the gold standard for diagnosing food hypersensitivities.[1] For the DBPCFC, neither the patient nor the physician knows whether the challenge material is the allergen or the placebo. This approach reduces both patient and observer bias. Single-blind challenges and open challenges are other options best utilized for foods that are unlikely to produce a reaction. For single-blind challenges, the physician knows the content of the challenge material and the patient is blinded, and for open challenges, all participants know the challenge content. The decision to pursue a food challenge should be based on the clinical history in conjunction with SPT and serum-specific IgE results. Food challenges are not benign procedures. Positive challenges may produce mild, moderate, or severe symptoms, and often require treatment[68]; therefore, a properly equipped environment is essential.

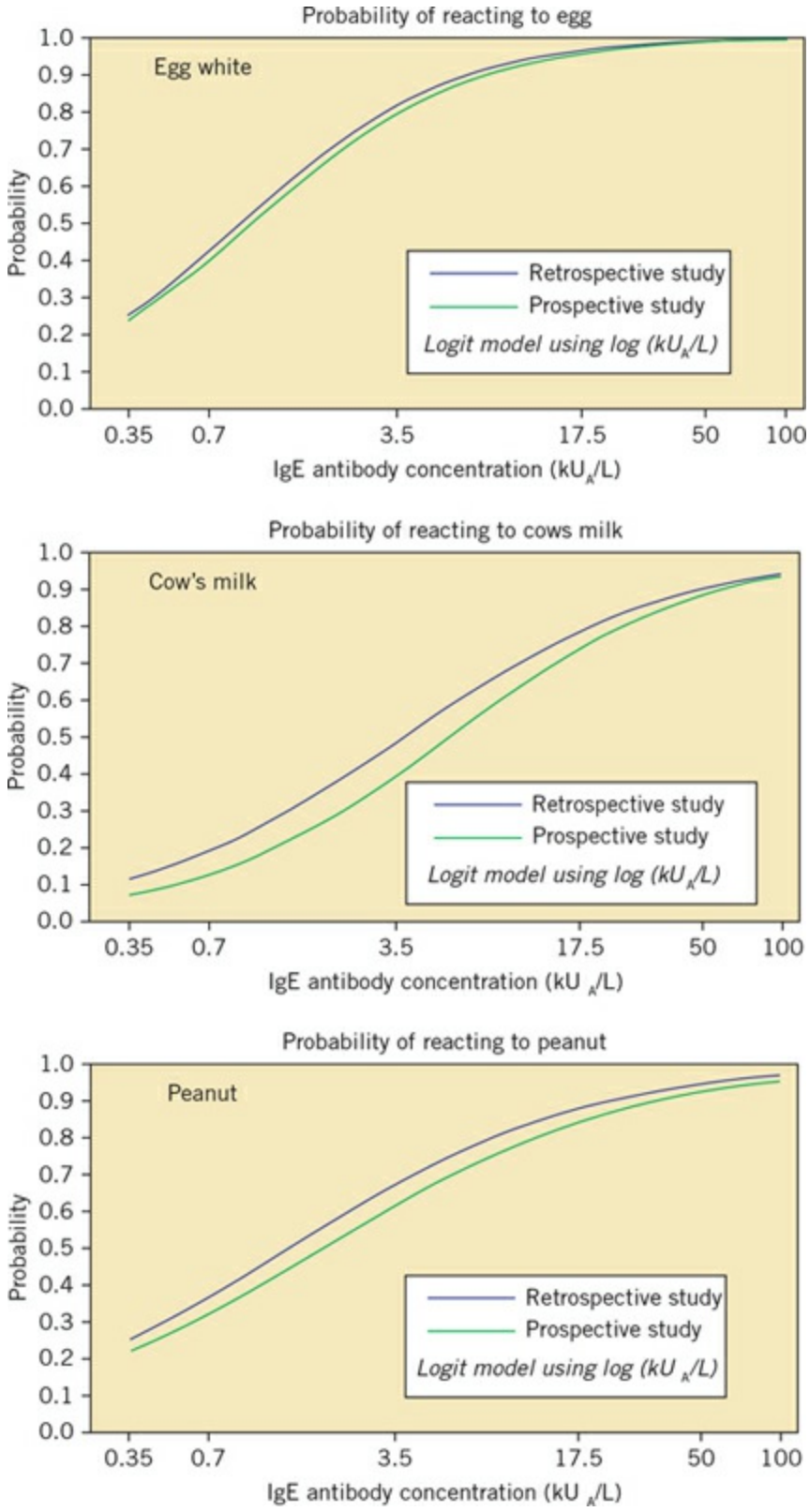


Figure 54-2 Diagnostic decision points for specific foods. (Reproduced with authorization from Sampson HA, Utility of food-

specific IgE concentrations in predicting symptomatic food allergy. *J Allergy Clin Immunol*. 2001;107(5):891–896.)

Prior to food challenges, patients should be instructed to avoid medications that may interfere with interpretation of results, such as antihistamines and beta-adrenergic bronchodilators. For IgE-mediated food allergies, the aim is for the patient to eat the equivalent of approximately 8–10 g of dry (dehydrated) food mixed within a vehicle.[68] A negative-blinded challenge must be followed by an open challenge to assure that the patient can ingest the food safely in natural, meal-sized portions.

For non-IgE-mediated food protein–induced enterocolitis, 0.15–0.3 mg protein/kg body weight, up to a maximum of 10 g is administered in three doses.[68] A reaction typically occurs 1–3 hours later, but when symptoms occur, they may be severe with excessive vomiting and possibly result in hypotension; therefore, intravenous access is recommended prior to the start of the challenge. For other conditions such as the eosinophilic GI disorders, several feedings of the food over a few days may be necessary to induce symptoms.

MANAGEMENT OF FOOD ALLERGY

Dietary avoidance of the causal food allergen is the key element for management of food hypersensitivity at this time (Table 54-6).[1] To practice strict avoidance, patients and their caregivers must be supplied with appropriate education materials to prevent ingestion of foods that may have had inadvertent cross-contact with allergens or foods that have hidden allergens. One study illustrated that for 156 reactions to peanut and tree nuts, 50% of accidental ingestions in food establishments were due to "hidden ingredients," such as an ingredient in a sauce or dressing.[69] Although just one study, it illustrates the difficulties that food allergic patients face with daily experiences, such as purchasing prepared foods or eating in restaurants. The Food Allergen Labeling and Consumer Protection Act (FALCPA) in the United States requires food manufacturers to clearly label their products that contain any of the eight major food allergens (milk, egg, peanut, tree nuts, wheat, soy, shell fish, and fish). This benefits patients allergic to these foods; however, the labeling does not necessarily include labeling for all other food allergens. An excellent educational resource for patients and families affected by

food allergies is the Food Allergy Resource and Education organization (www.foodallergy.org).

For patients with IgE-mediated food allergy, accidental ingestions may trigger potentially life-threatening allergic reactions. Therefore, patients and caregivers must be prepared to respond in case of an allergic reaction. Written anaphylaxis action plans should explicitly state the symptoms of anaphylaxis and when self-injectable epinephrine should be administered. Antihistamines are useful in treating mild symptoms, particularly cutaneous and oral symptoms, but will not reverse systemic reactions. Epinephrine is the treatment of choice for acute anaphylaxis (1:1000 dilution, 0.01 mg/kg with a maximum of 0.3 mg). Fatalities from anaphylactic events often appear to result from the late administration of epinephrine and subsequent irreversible cardiopulmonary complications.[34]

The treatment of eosinophilic disorders requires allergen avoidance.[56,57] Frequently, multiple foods must be eliminated from the diet. Many patients with eosinophilic GI disorders have evidence of sensitization to foods (by skin prick tests and/or serum-specific IgE assays); however, immediate reactions to foods are infrequently reported. Dietary elimination based on SPT and serum-specific IgE results alone is generally insufficient to promote disease resolution of eosinophilic disorders. Multiple foods may be causative in the same patient, with the most common ones including milk, soy, egg, and wheat.[56,57] A six food elimination diet has been proposed for the initial evaluation of food-induced symptoms; resolution of symptoms reportedly occurs in about two-thirds of patients. Another approach is to place the patient on an elemental (L-amino acid) formula and slowly add foods back into the diet when symptoms have resolved. Several weeks on an elemental formula is usually required for diminution of symptoms and for regression of the GI eosinophilic infiltrate. Open challenges with the causative foods, following elimination, have been shown to frequently provoke symptoms indistinguishable to those experienced before the elimination diet.

In cases where dietary intervention is not feasible or effective, administration of anti-inflammatory drugs (systemic or topical corticosteroids) can result in clinical and histopathologic improvement[56,57]; however, the side effects that accompany prolonged corticosteroid therapy is unacceptable, and the disease typically returns with discontinuation of the medication.

For proctocolitis, progression to a normal diet, including the eliminated allergen, is usually possible by 1–2 years of age.[1] If the prick skin tests and serum-specific IgE levels are negative, gradual introduction typically takes

place at home. For FPIES, action plans must be in place in the event of accidental ingestions. First-line treatment is fluid resuscitation rather than epinephrine and antihistamines (although these medications should be administered if there is concomitant IgE-mediated disease).[70] Corticosteroids should be considered, especially when past history reveals serious reactions. Corticosteroids likely suppress a presumed T cell immune response. When food reintroduction is considered, challenges in a properly equipped environment are necessary. The high risk of developing FPIES to multiple foods, for example, milk and soy, multiple cereal grains, often necessitates multiple food challenges. Up to 50% of patients reactive to milk may react to soy; up to 65% of patients reactive to grains may react to milk or soy; up to 50% of patients reactive to one grain may react to another grain.[70] More than half of the patients with FPIES due to milk become tolerant by age 3 years. Resolution of FPIES varies by age and is influenced by the primary causative food and the presence of concomitant IgE-mediated disease.[70]

NOVEL THERAPIES

Much recent attention has focused on means of altering the immune system to either eliminate food allergy or increase tolerance to allergenic food. A very limited number double-blind, randomized, placebo-controlled studies of oral immunotherapy (OIT) performed indicated that desensitization was induced in the majority of patients treated and a state of "sustained unresponsiveness" in a subset of subjects.[71–73] However, it is unclear why certain subjects benefit from this therapy whereas others do not, what the duration of sustained unresponsiveness is for those who do experience it, long-term consequences of this therapy, and the underlying mechanisms responsible for these outcomes. Furthermore, adverse reactions during OIT are common. Reactions have ranged from mild localized reactions to systemic reactions requiring epinephrine.[74,75] There are also reports of subjects developing GI symptoms similar to EoE while undergoing OIT.[74,76] At this time, OIT is not approved by regulatory bodies and must be considered experimental.

Sublingual immunotherapy (SLIT)[77] and epicutaneous immunotherapy (EPIT)[78] are alternate routes of administration that are currently being explored.

The type of allergen used can also play a role in the safety and efficacy of immunotherapy. OIT with heat-denatured proteins may accelerate the

development of tolerance to regular milk and egg in those who are allergic, but can tolerate baked milk or egg products in their diet.[79,80] In these studies, the addition of the baked milk and egg products into the diets was safe and well-tolerated. Modified recombinant vaccines with altered IgE-binding epitopes, peptide immunotherapy containing T cell epitopes, use of immunostimulatory sequences as adjuvants, and allergen gene immunization are other potential therapies that are under investigation for peanut allergy.[81]

TABLE 54-6 Treatment Approach to Food Allergy

Treatment of Food Allergy
Food avoidance
Anaphylaxis action plan—epinephrine, anti-histamines, corticosteroids
FPIES action plan—intravenous fluids, ± corticosteroids
Possible Future Therapies
Immunotherapy—oral, sublingual, epicutaneous, heated denatured proteins, recombinant proteins
Monoclonal anti-IgE
Traditional Chinese medicine

Allergen nonspecific forms of treatment for food allergy include anti-IgE therapy and traditional Chinese medicine. Humanized IgG_1 monoclonal antibody against IgE (TNX-901) was tested in peanut-allergic subjects and was shown to enable subjects to tolerate increased quantities of peanut following therapy. Although not a cure for food allergy, the medication offered protection in the event of inadvertent ingestion.[82] Investigation of another anti-IgE preparation, omalizumab (Xolair®, Genentech), for the treatment of peanut allergy was initiated, but discontinued for safety concerns related to the pretreatment oral peanut challenge.[83] Based on the limited data from 14 subjects who completed the study, the omalizumab-treated subjects were able to tolerate increased amounts of peanut as compared to the placebo group. Anti-IgE therapy is now being explored in combination with OIT. A pilot study for milk allergy demonstrated that combination therapy resulted in desensitization within 7–11 weeks.[84]

The potential value of traditional Chinese medicine, which has been used in Asia for centuries, as an effective form of treatment for allergic disease is being explored. Experiments administering a Chinese herbal medicine formula have been able to block anaphylaxis in a murine model

of peanut allergy and multiple food allergy.[85] Phase 1 clinical trials have demonstrated the formula to be safe and well-tolerated by food allergic individuals.[85] A phase 2 clinical trial is currently underway.

SUMMARY

Food allergy is a general term that includes a wide range of conditions. The pathogenesis involves IgE-mediated and/or non-IgE-mediated processes with varying manifestations depending on the mechanisms of disease. Diagnosis requires a skillful history, physical exam, and targeted diagnostic testing. Treatment requires avoidance of the causal allergen(s). Hopefully in the future, diagnostic modalities will be more fine-tuned and additional treatment options will be available.

REFERENCES

1. NIAID-Sponsored Expert Panel, Boyce JA, Assa'ad A, Burks AW, et al. Guidelines for the diagnosis and management of food allergy in the United States: report of the NIAID-sponsored expert panel. *J Allergy Clin Immunol*. 2010;126(6 Suppl):S1–S58.
2. Rona RJ, Keil T, Summers C, et al. The prevalence of food allergy: a meta-analysis. *J Allergy Clin Immunol*. 2007;120:638–646.
3. Branum AM, Lukacs SL. Food allergy among children in the United States. *Pediatrics*. 2009;124:1549–1555.
4. Gupta RS, Springston EE, Warrier MR, et al. The prevalence, severity, and distribution of childhood food allergy in the United States. *Pediatrics*. 2011;128:e9–e17.
5. Soller L, Ben-Shoshan M, Harrington DW, et al. Overall prevalence of self-reported food allergy in Canada. *J Allergy Clin Immunol*. 2012;130:986–988.
6. Osborne NJ, Koplin JJ, Martin PE, et al. Prevalence of challenge-proven IgE-mediated food allergy using population-based sampling and predetermined challenge criteria in infants. *J Allergy Clin Immunol*. 2011;127:668–676.e1–e2.
7. Sicherer SH, Muñoz-Furlong A, Godbold JH, et al. US prevalence of self-reported peanut, tree nut, and sesame allergy: 11-year follow-up. *J Allergy Clin Immunol*. 2010;125:1322–1326.
8. Skripak JM, Matsui EC, Mudd K, et al. The natural history of IgE-mediated cow's milk allergy. *J Allergy Clin Immunol*. 2007;120:1172–1177.
9. Savage JH, Matsui EC, Skripak JM, et al. The natural history of egg allergy. *J Allergy Clin Immunol*. 2007;120:1413–1417.
10. Skolnick HS, Conover-Walker MK, Koerner CB, et al. The natural history of peanut allergy. *J Allergy Clin Immunol*. 2001;107:367–374.
11. Fleischer DM, Conover-Walker MK, Matsui EC, et al. The natural history of tree nut allergy. *J Allergy Clin Immunol*. 2005;116:1087–1093.
12. Sicherer SH. Clinical implications of cross-reactive food allergens. *J Allergy Clin Immunol*. 2001;108:881–890.
13. James JM. Respiratory manifestations of food allergy. *Pediatrics*. 2003;111(6 Pt

3):1625–1630.
14. Mayer L. Mucosal immunity. *Pediatrics*. 2003;111:1595–1600.
15. Chehade M, Mayer L. Oral tolerance and its relation to food hypersensitivities. *J Allergy Clin Immunol*. 2005;115:3–12.
16. Sampson HA. Food allergy. Part 1: immunopathogenesis and clinical disorders. *J Allergy Clin Immunol*. 1999;103:717–728.
17. Friedman A, Weiner HL. Induction of anergy or active suppression following oral tolerance is determined by antigen dosage. *Proc Natl Acad Sci USA*. 1994 ;91:6688–6692.
18. Ko J, Mayer L. Oral tolerance: lessons on treatment of food allergy. *Eur J Gastroenterol Hepatol*. 2005;17:1299–1303.
19. Strid J, Thomson M, Hourihane J, et al. A novel model of sensitization and oral tolerance to peanut protein. *Immunology*. 2004;113:293–303.
20. Lack G, Fox D, Northstone K, et al. Factors associated with the development of peanut allergy in childhood. *N Engl J Med*. 2003;348:977–985.
21. Eastham EJ, Lichauco T, Grady MI, et al. Antigenicity of infant formulas: role of immature intestine on protein permeability. *J Pediatr*. 1978;93:561–564.
22. Du Toit G, Katz Y, Sasieni P, et al. Early consumption of peanuts in infancy is associated with a low prevalence of peanut allergy. *J Allergy Clin Immunol*. 2008;122:984–991.
23. Katz Y, Rajuan N, Goldberg MR, et al. Early exposure to cow's milk protein is protective against IgE-mediated cow's milk protein allergy. *J Allergy Clin Immunol*. 2010;126:77–82.e1.
24. Morafo V, Srivastava K, Huang CK, et al. Genetic susceptibility to food allergy is linked to differential TH2-TH1 responses in C3H/HeJ and BALB/c mice. *J Allergy Clin Immunol*. 2003;111:1122–1128.
25. Howell WM, Turner SJ, Hourihane JO, et al. HLA class II DRB1, DQB1 and DPB1 genotypic associations with peanut allergy: evidence from a family-based and case-control study. *Clin Exp Allergy*. 1998;28:156–162.
26. Sicherer SH, Furlong TJ, Maes HH, et al. Genetics of peanut allergy: a twin study. *J Allergy Clin Immunol*. 2000;106:53–56.
27. Sudo N, Sawamura S, Tanaka K, et al. The requirement of intestinal bacterial flora for the development of an IgE production system fully susceptible to oral tolerance induction. *J Immunol*. 1997;159:1739–1745.
28. Bashir ME, Louie S, Shi HN, et al. Toll-like receptor 4 signaling by intestinal microbes influences susceptibility to food allergy. *J Immunol*. 2004;172:6978–6987.
29. Rivas MN, Burton OT, Wise P, et al. A microbiotia signature associated with experimental food allergy promotes allergic sensitization and anaphylaxis. *J Allergy Clin Immunol*. 2013;131:201–212.
30. Kalliomaki M, Salminen S, Poussa T, et al. Probiotics during the first 7 years of life: a cumulative risk reduction of eczema in a randomized, placebo-controlled trial. *J Allergy Clin Immunol*. 2007;119:1019–1021.
31. Kuitunen M, Kukkonen K, Juntunen-Backman K, et al. Probiotics prevent IgE-associated allergy until age 5 years in cesarean-delivered children but not in the total cohort. *J Allergy Clin Immunol*. 2009;123:335–341.
32. West CE, Hammarström ML, Hernell O. Probiotics during weaning reduce the incidence of eczema. *Pediatr Allergy Immunol*. 2009;20: 430–437.
33. Sampson HA, Mendelson L, Rosen JP. Fatal and near-fatal anaphylactic reactions to food in children and adolescents. *N Engl J Med*. 1992;327:380–384.
34. Lieberman P, Nicklas RA, Oppenheimer J, et al. The diagnosis and management

of anaphylaxis practice parameter: 2010 update. *J Allergy Clin Immunol*. 2010;126:477–480.e1-42.
35. Breiteneder H, Ebner C. Molecular and biochemical classification of plant-derived food allergens. *J Allergy Clin Immunol*. 2000;106:27–36.
36. Jenkins JA, Griffiths-Jones S, Shewry PR, et al. Structural relatedness of plant food allergens with specific reference to cross-reactive allergens: an in silico analysis. *J Allergy Clin Immunol*. 2005;115:163–170.
37. Jarvinen KM, Beyer K, Vila L, et al. B-cell epitopes as a screening instrument for persistent cow's milk allergy. *J Allergy Clin Immunol*. 2002;110:293–297.
38. Jarvinen KM, Beyer K, Vila L, et al. Specificity of IgE antibodies to sequential epitopes of hen's egg ovomucoid as a marker for persistence of egg allergy. *Allergy*. 2007;62:758–765.
39. Lemon-Mule H, Sampson HA, Sicherer SH, et al. Immunologic changes in children with egg allergy ingesting extensively heated egg. *J Allergy Clin Immunol*. 2008;122:977–983.
40. Nowak-Wegrzyn A, Bloom KA, Sicherer SH, et al. Tolerance to extensively heated milk in children with cow's milk allergy. *J Allergy Clin Immunol*. 2008;122:342–347.
41. Flinterman AE, Knol EF, Lencer DA, et al. Peanut epitopes for IgE and IgG4 in peanut-sensitized children in relation to severity of peanut allergy. *J Allergy Clin Immunol*. 2008;121:737–743.
42. Wang J, Lin J, Bardina L, et al. Correlation of IgE/IgG_4 milk epitopes and affinity of milk-specific IgE antibodies with different phenotypes of clinical milk allergy. *J Allergy Clin Immunol*. 2010;125:695–702.
43. Beyer K, Morrow E, Li XM, et al. Effects of cooking methods on peanut allergenicity. *J Allergy Clin Immunol*. 2001;107:1077–1081.
44. Prussin C, Metcalfe DD. IgE, mast cells, basophils, and eosinophils. *J Allergy Clin Immunol*. 2003;111:S486–S494.
45. Sampson HA, Munoz-Furlong A, Campbell RL, et al. Second symposium on the definition and management of anaphylaxis: summary report—Second National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network symposium. *J Allergy Clin Immunol*. 2006;117:391–397.
46. Lee JM, Greenes DS. Biphasic anaphylactic reactions in pediatrics. *Pediatrics*. 2000;106:762–766.
47. Simonte SJ, Ma S, Mofidi S, et al. Relevance of casual contact with peanut butter in children with peanut allergy. *J Allergy Clin Immunol*. 2003;112:180–182.
48. Boguniewicz M, Leung DY. Atopic dermatitis: a disease of altered skin barrier and immune dysregulation. *Immunol Rev*. 2011;242:233–246.
49. Sampson HA. Food sensitivity and the pathogenesis of atopic dermatitis. *J R Soc Med*. 1997;90 Suppl 30:2–8.
50. Flinterman AE, Knulst AC, Meijer Y, et al. Acute allergic reactions in children with AEDS after prolonged cow's milk elimination diets. *Allergy*. 2006;61:370–374.
51. Karpati S. Dermatitis herpetiformis: close to unravelling a disease. *J Dermatol Sci*. 2004;34:83–90.
52. Roberts G, Patel N, Levi-Schaffer F, et al. Food allergy as a risk factor for life-threatening asthma in childhood: a case-controlled study. *J Allergy Clin Immunol*. 2003;112:168–174.
53. James JM, Bernhisel-Broadbent J, Sampson HA. Respiratory reactions provoked by double-blind food challenges in children. *Am J Respir Crit Care Med*. 1994;149:59–64.

54. Roberts G, Golder N, Lack G. Bronchial challenges with aerosolized food in asthmatic, food-allergic children. *Allergy*. 2002;57:713–717.
55. Sicherer SH, Furlong TJ, DeSimone J, et al. Self-reported allergic reactions to peanut on commercial airliners. *J Allergy Clin Immunol*. 1999;104:186–189.
56. Rothenberg ME. Eosinophilic gastrointestinal disorders (EGID). *J Allergy Clin Immunol*. 2004;113:11–28.
57. Liacouras CA, Furuta GT, Hirano I, et al. Eosinophilic esophagitis: updated consensus recommendations for children and adults. *J Allergy Clin Immunol*. 2011;128:3–20.
58. Chehade M, Magid MS, Mofidi S, et al. Allergic eosinophilic gastroenteritis with protein-losing enteropathy: intestinal pathology, clinical course, and long-term follow-up. *J Pediatr Gastroenterol Nutr*. 2006;42:516–521.
59. Kilshaw PJ, Cant AJ. The passage of maternal dietary proteins into human breast milk. *Int Arch Allergy Appl Immunol*. 1984;75:8–15.
60. Vanderhoof JA, Murray ND, Kaufman SS, et al. Intolerance to protein hydrolysate infant formulas: an underrecognized cause of gastrointestinal symptoms in infants. *J Pediatr*. 1997;131:741–744.
61. Nowak-Wegrzyn A, Sampson HA, Wood RA, et al. Food protein-induced enterocolitis syndrome caused by solid food proteins. *Pediatrics*. 2003;111:829–835.
62. Kelso JM, Sampson HA. Food protein-induced enterocolitis to casein hydrolysate formulas. *J Allergy Clin Immunol*. 1993;92:909–910.
63. Meresse B, Malamut G, Cerf-Bensussan N. Celiac disease: an immunological jigsaw. *Immunity*. 2012;36:907–919.
64. Giersiepen K, Lelgemann M, Stuhldreher N, et al. Accuracy of diagnostic antibody tests for coeliac disease in children: summary of an evidence report. *J Pediatr Gastroenterol Nutr*. 2012;54:229–241.
65. May CD. Objective clinical and laboratory studies of immediate hypersensitivity reactions to foods in asthmatic children. *J Allergy Clin Immunol*. 1976;58:500–515.
66. Sporik R, Hill DJ, Hosking CS. Specificity of allergen skin testing in predicting positive open food challenges to milk, egg and peanut in children. *Clin Exp Allergy*. 2000;30:1540–1546.
67. Sampson HA. Utility of food-specific IgE concentrations in predicting symptomatic food allergy. *J Allergy Clin Immunol*. 2001;107:891–896.
68. Nowak-Wegrzyn A, Assa'ad AH, Bahna SL, et al. Work Group report: oral food challenge testing. *J Allergy Clin Immunol*. 2009;123:S365–S383.
69. Furlong TJ, DeSimone J, Sicherer SH. Peanut and tree nut allergic reactions in restaurants and other food establishments. *J Allergy Clin Immunol*. 2001;108:867–870.
70. Sicherer SH. Food protein-induced enterocolitis syndrome: case presentations and management lessons. *J Allergy Clin Immunol*. 2005;115:149–156.
71. Skripak JM, Nash SD, Rowley H, et al. A randomized, double-blind, placebo-controlled study of milk oral immunotherapy for cow's milk allergy. *J Allergy Clin Immunol*. 2008;122:1154–1160.
72. Burks AW, Jones SM, Wood RA, et al. Oral immunotherapy for treatment of egg allergy in children. *N Engl J Med*. 2012;367:233–243.
73. Varshney P, Jones SM, Scurlock AM, et al. A randomized controlled study of peanut oral immunotherapy: clinical desensitization and modulation of the allergic response. *J Allergy Clin Immunol*. 2011;127:654–660.
74. Narisety SD, Skripak JM, Steele P, et al. Open-label maintenance after milk oral

immunotherapy for IgE-mediated cow's milk allergy. *J Allergy Clin Immunol*. 2009;124:610–612.
75. Jones SM, Pons L, Roberts JL, et al. Clinical efficacy and immune regulation with peanut oral immunotherapy. *J Allergy Clin Immunol*. 2009;124:292–300.
76. Sánchez-García S, Rodríguez Del Río P, Escudero C, et al. Possible eosinophilic esophagitis induced by milk oral immunotherapy. *J Allergy Clin Immunol*. 2012;129:1155–1157.
77. Keet CA, Frischmeyer-Guerrerio PA, Thyagarajan A, et al. The safety and efficacy of sublingual and oral immunotherapy for milk allergy. *J Allergy Clin Immunol*. 2012;129:448–455, 455.e1–455.e5.
78. Dupont C, Kalach N, Soulaines P, et al. Cow's milk epicutaneous immunotherapy in children: a pilot trial of safety, acceptability, and impact on allergic reactivity. *J Allergy Clin Immunol*. 2010;125:1165–1167.
79. Kim JS, Nowak-Węgrzyn A, Sicherer SH, et al. Dietary baked milk accelerates the resolution of cow's milk allergy in children. *J Allergy Clin Immunol*. 2011;128:125–131.e2.
80. Leonard SA, Sampson HA, Sicherer SH, et al. Dietary baked egg accelerates resolution of egg allergy in children. *J Allergy Clin Immunol*. 2012;130:473–480.e1.
81. Wang J, Sampson HA. Treatments for food allergy: how close are we? *Immunol Res*. 2012;54:83–94.
82. Leung DY, Sampson HA, Yunginger JW, et al. Effect of anti-IgE therapy in patients with peanut allergy. *N Engl J Med*. 2003;348:986–993.
83. Sampson HA, Leung DYM, Burks AW, et al. A phase II, randomized, double-blind, parallel group, placebo-controlled oral food challenge trial of Xolair (omalizumab) in peanut allergy. *J Allergy Clin Immun*. 2011;127:1309–1310.
84. Nadeau KC, Schneider LC, Hoyte L, et al. Rapid oral desensitization in combination with omalizumab therapy in patients with cow's milk allergy. *J Allergy Clin Immunol*. 2011;127:1622–1623.
85. Wang J, Li XM. Chinese herbal therapy for the treatment of food allergy. *Curr Allergy Asthma Rep*. 2012;12:332–338.

Exocrine Pancreas

55

Kevin Gaskin, MD, FRACP

EXOCRINE PANCREATIC DISEASE

There are a variety of childhood exocrine pancreatic disorders (EPDs) of an inherited or acquired nature in terms of their etiology. The acquired disorders include primary protein energy malnutrition, tropical calcific pancreatitis, and nongenetic chronic pancreatitis. From a global perspective, primary protein energy malnutrition is possibly the commonest EPD, but not surprisingly there is a dearth of evidence-based pancreatic studies in such populations as compared to the far less commonly inherited/genetic diseases described in the western world. The present chapter will discuss the clinical phenotypes and the molecular basis/pathophysiology of the genetic disorders, highlight the mechanisms of fat and protein maldigestion and malabsorption common to all patients with severe exocrine pancreatic dysfunction, and the nutritional consequences and management of children with exocrine pancreatic dysfunction.

Molecular Basis and Clinical Features of Inherited Exocrine Pancreatic Disorders

The inherited genetic disorders are uncommon or rare and are summarized in Table 55-1, according to the molecular defects and functional effects thereof. The commonest, cystic fibrosis (CF), occurs in 1:2500–1:4000 live births in Western countries.[1] The second commonest, Shwachman–Diamond syndrome (SDS),[2] has a much lower incidence of 1:76,000 globally, approximately 3% of the CF population.[3] The remaining disorders, including Pearson's Marrow Pancreas syndrome (PMPS),[4] Johanson–Blizzard syndrome (JBS),[5] and pancreatic agenesis (PA),[6] are rare, with less than 100, 30, and 50 cases, respectively, reported

worldwide. There are only anecdotal reports of enzyme deficiency syndromes, but although very rare their recognition has emphasized the important role these enzymes/coenzymes have in fat and protein digestion.

TABLE 55-1 Hereditary Exocrine Pancreatic Disorders

Disorder	Genetics Mutations of	Functional Defect
Cystic fibrosis	CFTR gene 7q31	Impaired anion transport via CFTR
Shwachman–Diamond syndrome	Shwachman, Bodian, Diamond syndrome gene 7q11	Possible defects in ribosomal maturation structure, function
Pearson's Marrow Pancreas syndrome	Mitochondrial DNA deletion	Abnormal oxidative phosphorylation, severe sideroblastic anemia, pancreatic insufficiency
Johanson–Blizzard syndrome	UBR-1 gene chromosome 15q13	Impaired synthesis of ubiquitin ligase with dysregulation of protein degradation, progressive inflammation, fibrous tissue proliferation
Pancreatic agenesis	i. PDX1 gene at 13q12, which encodes IPF-1 transcription factor ii. PTF-1a gene 10p12 iii. GATA-binding factor on 18q11	Interference with protein transcription factors, formation and function
Hereditary pancreatitis	Protease serine-1 gene 7q34	Premature activation of trypsinogen in pancreas
Congenital enteropeptidase (enterokinase) deficiency	Proenteropeptidase, PRSS-7 gene 21q21	Failure to produce enteropeptidase and lack of trypsinogen activation to trypsin
Isolated lipase colipase deficiency	No mutations described	Fat malabsorption

The molecular basis of the genetic disorders has been elucidated over the last 20 years and particularly in CF has provided insight into the pathophysiology of the disorder. Each of these conditions will be discussed in terms of their pathophysiology and clinical manifestations.

Cystic Fibrosis

The molecular/genetic basis of CF was unraveled in 1989 with the discovery of the cystic fibrosis transmembrane conductance regulator (CFTR) gene[7,8] and the subsequent recognition that CFTR protein is a cyclic adenosine monophosphate (c-AMP)- dependent anion transport channel in epithelial cell membranes.[9] Mutations of CFTR cause a multiplicity of electrolyte transport defects as, for instance, poor reabsorption of Cl^- from the sweat duct resorptive epithelium[10] and poor Cl^- and HCO_3^- secretion from both respiratory and pancreatic duct epithelium.[11,12] In vivo, the poor reabsorption of sweat duct Cl^- leads to excessive sweat losses of Cl^- and is the basis of sweat Cl^- testing as the gold standard diagnostic marker of CF. The in vitro work on CF pancreatic cell lines demonstrating the impairment of pancreatic duct Cl^- and HCO_3^- secretion[11,12] also confirms the earlier in vivo work on CF patients using pancreatic stimulation tests.[13,14] These studies demonstrated that CF patients had significantly impaired Cl^- and HCO_3^- secretion in comparison with normal and disease controls, irrespective of the degree of destruction or lack of development of pancreatic acinar tissue.

CFTR/c-AMP-dependent HCO_3^- secretion has more recently been found, in vitro, to be impaired in a variety of epithelial CF tissues in addition to the above, including biliary tract, gut, and uterine/cervical epithelia, with a coimpairment of mucin secretion.[12] Mucin appears to aggregate in glandular structures and an inflammatory, fibrotic process ensues. Even impairing HCO_3^- secretion in the absence of CFTR mutations can induce a similar process. In a similar vein, in a CF mouse model,[15] poor alkalinization of pancreatic fluid leads to intraduct aggregation of glycoprotein-2 obstructing small ductules consistent with postmortem studies on CF infants and children, and at least in part contributing to the fibrotic process observed so characteristically in this disease.

To date, over 1900 CF-causing mutations have been recognized. Considering this large number, over the last 15 years these mutations have been classified into five classes according to their biosynthetic defects and dysfunction,[16,17] as per Table 55-2. In terms of pancreatic disease, Classes I, II, and III mutations are associated with severe pancreatic disease with fat malabsorption, the pancreatic insufficient (PI) phenotype, whereas Classes IV and V mutations are associated with less severe impairment of

and even normal pancreatic enzyme secretion with normal fat absorption, the pancreatic sufficient (PS) phenotype (see the section *Maldigestion and Malabsorption*). The two phenotypes, PI and PS, are regarded as classic forms of CF with sweat chloride levels over 60 mmol/L and characterized by multisystem involvement as outlined in Table 55-3. PS patients also have milder lung disease and longer survival, a lower sweat chloride, and compared to their PI counterparts experience less liver or gut disease.[18] Although rare reports of meconium ileus, distal intestinal obstruction syndrome, or cholelithiasis have occurred in PS patients, along with fibrosing colonopathy (FC) and intussusception, these complications almost exclusively occur in the PI phenotype.

TABLE 55-2 Classification of CFTR Mutations Reflecting Biosynthetic Defects and Function

Class I	• Defective protein production • Frameshift and nonsense mutations • Unstable messenger RNA
Class II	• Defective CFTR processing (includes ΔF508 as most common mutation)
Class III	• Defective regulation of CFTR gene • Mutations can dramatically impair channel function, G551D
Class IV	• Defective channel conduction, e.g., R117H
Class V	• Abnormal splicing of CFTR gene • Reduction in normally functioning channels

In those patients with classical CF, particularly the 85%–90% with the PI phenotype, over 95% of the mortality is related to the inexorable progression of the lung disease. Early colonization with *Pseudomonas* and *Staphylococcus aureus*, bronchiectasis, and obstructive respiratory disease ultimately lead to respiratory failure and the need for lung transplantation. Recent studies suggest survival has improved dramatically over the last 30 years.[19–21] In the early 1980s, the best median survival was up to 30 years. This has now improved to over 45 years and over 90% of patients are expected to survive into adulthood. Although the improvement is multifactorial, delivery of care in specialist multidisciplinary CF clinics,[22] early diagnosis through neonatal screening[23] and protocol-driven antibiotic regimens, segregation of patients with lung *Pseudomonas* colonization to

prevent early lung damage, and maintenance of normal nutrition have no doubt contributed. In one of the Australian newborn screening programs there was a definite survival advantage to the screened population. CF infants in the first 3 years of the program were compared to a retrospective nonscreened group for the 3 years prior to the commencement of screening. The screened infants maintained a significant average advantage of a 10% higher forced expiratory volume in 1 second (FEV_1) percent predicted at 10[23] and 15[24] years of age, increasing to 16% at 18 years.[25] The mortality in the nonscreened group (66%) was over twice the mortality of the screened group (32%) by 25 years of age,[25] confirming two U.S. CF Database reports of longer survival in those diagnosed and treated from early in life.[26a,26b]

In non-neonatal CF patients, general genotyping for the commonest mutations (ΔF508, a Class II mutation) in patients of Northern European ethnicity reveals approximately 50% are ΔF508 homozygotes, 40%–45% are ΔF508 compound heterozygotes (with ΔF508 mutation and a non-ΔF508 mutation), and 5%–10% have two non-ΔF508 mutations.[27] Virtually all ΔF508 homozygotes, 70% of ΔF508 compound heterozygotes, but only 30%–35% with two non-ΔF508 mutations, were PI. Those with PS have at least one Class IV or V non-ΔF508 mutation. In patients of Southern European (SE) ethnicity, ΔF508 homozygotes represent less than 30% of their total CF population[28] but, again, in non-neonatal populations, there is a similar proportion of PI and PS, approximately 85% and 15%, respectively, indicating the Classes I–III (severe pancreatic mutations) are still predominant in these older SE populations.

Neonatal screening provides a different perspective on the pancreatic phenotype/genotype relationship as, at the time of neonatal diagnosis, the PS proportion has varied from 25%[29] to 37%[30] of the population. Over the first 10 years of life 25% of these PS patients developed PI and to date patients in this category have had two Class I–III mutations. Some 10% of patients with at least one Class IV or V mutation are at risk from recurrent pancreatitis, which may lead to the patient becoming PI during their adult years.

TABLE 55-3 Clinical Manifestations of Cystic Fibrosis

Pulmonary disease	• Recurrent infections: – *Staphylococcus*

	– *Pseudomonas* – *Burkholderia* – Fungal • Bronchiectasis • Lung fibrosis • Pneumothorax • Pulmonary hemorrhage • Emphysema • Respiratory failure
Pancreatic disease	• Pancreatic insufficiency • Pancreatic sufficiency ± pancreatitis
Gut disease	• Gastroesophageal reflux • Esophagitis • Giardiasis • Celiac disease • Neonatal meconium ileus • Distal intestinal obstruction syndrome • Intussusception • Appendicitis • Appendiceal mucocoele ± abscess • Fibrosing colonopathy • Crohn's disease • Fecal retention, megacolon • Rectal prolapse
Biliary tract/liver disease	• Microgallbladder • Nonfunctioning gallbladder • Obstructed cystic duct • Cholelithiasis • Common bile duct stenosis • Common bile duct stones • Hepatosteatosis • Focal biliary fibrosis • Multilobular biliary cirrhosis ± portal hypertension ± liver failure
Dermatological	• Salt wasting • Hyponatremic dehydration

Some CF mutations are associated with minimal organ involvement or mono organ presentations, including sinusitis, pancreatitis, and congenital bilateral or unilateral absence of the vas deferens. Other mutations have been found through population research studies or neonatal screening and appear unassociated with any disease to date, but many in this category

have only had limited follow-up. They are sometimes referred to as having CFTR disease, but this title may prove inappropriate, with longer follow-up studies.

Shwachman–Diamond Syndrome

SDS is a multisystem autosomal recessive disease[2,3,31] associated with mutations of the SBDS (Shwachman, Bodian, Diamond Syndrome) gene with a pericentrometric location on 7q.[3] It has 5 exons and spans less than 9 Kb encoding for a protein of 250 amino acid residues. The function of this protein is currently unknown, but it is suspected to be involved with fundamental aspects of RNA maturation, metabolism, and/or ribosomal structure and function. The SBDS gene is the only one currently associated with SDS. In patients with SDS over 90% have at least one of the mutant SBDS alleles caused by gene conversion. The three most common gene conversions on exon 2 account for over 76% of the disease alleles and for over 62% of affected individuals for the two predominant disease causing mutations.[32]

Classical clinical features of SDS include pancreatic hypoplasia with PI from infancy, bone marrow dysfunction, particularly neutropenia, metaphyseal dysostosis, and short stature, as outlined in Table 55-4, and a normal sweat chloride test. The EPD is associated with severe lipomatosis replacing acinar tissue of the exocrine pancreas, as can be delineated by ultrasound, computed tomography, or magnetic resonance imaging studies, but, as opposed to CF, maintaining normal ductal function in terms of water and electrolyte secretion and normal islet function in terms of insulin secretion. It is also evident that pancreatic function may improve with age, with up to 50% of patients becoming PS in the first 5–10 years of life.[33] Morbidity is related to severe neutropenia with associated sepsis, thrombocytopenia, and red cell aplasia. Mortality in early childhood is associated with severe sepsis and in older children/young adults to acute myeloid leukemia which appears 15%–22% higher in SDS compared to the general population,[34] leading to the recommendation for regular 1–3 yearly bone marrow examinations for SDS patients. Nearly all SDS patients with proven SBDS mutations have bony metaphyseal dysplasia,[35] which likely contributes to the short stature and is a useful diagnostic aid if overtly present.

Pearson's Marrow Pancreas Syndrome

PMPS is a rare, multisystem mitochondrial DNA deletion syndrome

belonging to the group of mitochondrial cytopathies.[4] A common mutation is a 4.9 Kb deletion, but the deletion size can extend up to 14 Kb.[36] The size of the deletion does not correlate with clinical features or severity, particularly the development of the Kearns-Sayre neurodegenerative syndrome in those who survive postinfancy. The disease is characterized by a severe transfusion-dependent macrocytic anemia with bone marrow features of megaloblastosis and vacuolation of bone marrow cells, and the presence of ringed sideroblasts. The pancreas, in comparison with SDS, is fibrotic and small.

TABLE 55-4 Non-cystic Fibrosis Pancreatic Diseases

Shwachman–Diamond syndrome	• Pancreatic acinar cell hypoplasia with lipomatosis • Bone marrow dysfunction: – Neutropenia – Thrombocytopenia – Red cell hypoplasia – Myelodysplastic syndrome – Acute myeloid leukemia • Bone disease: – Metaphyseal dysplasia – Shortened splayed ribs – Short stature • Miscellaneous: – Developmental delay – Eczematous skin lesions – Hepatosteatosis – Renal tubular dysfunction
Pearson's Marrow Pancreas Syndrome	• Pancreatic fibrosis with pancreatic insufficiency • Severe macrocytic anemia (often transfusion dependent) • Vacuolated cells, megaloblastosis and ringed sideroblasts in bone marrow aspirates • Variable neutropenia and thrombocytopenia • Developmental delay • Kearns-Sayre disease in later life • Pigmenting retinopathy • Ophthalmoplegia
Johanson–Blizzard syndrome	• Pancreatic lipomatosis with pancreatic insufficiency • Aplasia or hypoplasia of alae nasi • Congenital sensorineural deafness • Hypothyroidism • Short stature + growth hormone deficiency

	• Imperforate anus • “Upsweep” frontal hair pattern • Scalp defect/cutis aplasia • Absence of permanent teeth • Congenital heart disease • Intellectual disability • Lacrimal duct anomalies
Pancreatic Agenesis	• Pancreatic insufficiency • Diabetes mellitus • Cerebellar agenesis • Polysplenia

Johanson–Blizzard syndrome

JBS is the rarest of the multisystem-inherited EPDs[5] and is an autosomal recessive disease caused by mutations of the ubiquitin protein ligase E3 component n-recognin-1 (UBR-1) gene located on chromosome 15 at position q13.[37] Ubiquitin is a universally expressed protein in all cells and plays a normal role of tagging other proteins that are mismatched, damaged, dysfunctional, or unneeded for degradation. Ubiquitin ligase commences this process by attaching ubiquitin to the lysine side chain of the tagged protein. This process is fundamental for the normal cellular processes: cell division, surface receptor function, signaling, apoptosis, quality control of proteins, and inflammatory cascades. Mutations of the UBR-1 gene lead to the progressive congenital damage due to a deranged inflammatory response, failure of normal apoptosis, and persistence of abnormal protein leading to the skeletal and other structural or functional abnormalities seen in JBS. Of the various missense, nonsense, and splice site mutations identified to date no specific genotype/phenotype have been found.

Similar to SDS, JBS patients have severe lipomatosis of their exocrine pancreas and PI from early infancy. Very few of these patients have undergone formal pancreatic stimulation tests. Prognosis is poor if the PI is not recognized. Diagnosis of the disorder is facilitated by the combination of features as per Table 55-4, including flaring of the alae nasi, deafness, imperforate anus, PI, and short stature.

Hereditary Pancreatitis (HP)

The autosomal dominant form of inherited pancreatitis is caused by mutations of the protease serine-1 (PRSS-1)[38] gene, which normally

produces cationic trypsinogen. The latter is inactive in the pancreas and is transported to the duodenum where it undergoes cleavage by enterokinase to form the active enzyme, trypsin. PRSS-1 is located at 7q34. Over 40 mutations have now been found to cause HP and the commonest, R122H, is a point mutation exchanging the amino acid arginine with histidine. It remains uncertain as to how these mutations cause premature intrapancreatic trypsinogen activation, but of recent interest is the role of chymotrypsin C (CTRC).[39] CTRC promotes degradation of wild-type trypsinogens and trypsin. However, trypsinogen mutants causing classic HP, in the presence of CTRC, are autoactivated at increased rates and produce greater active trypsin concentrations than the wild type trypsinogens, suggesting there is a CTRC dysregulation involved with trypsinogens mutations and the occurrence of HP. It is noted that the average PRSS-1 gene mutation frequency in over 1000 patients with idiopathic pancreatitis was only 1.9%, although it has been reported in up to 10% in smaller studies.[40] The major clinical manifestations include recurrent acute pancreatitis commencing in childhood, and complications thereof, including intrapancreatic stones, cysts and strictures, and the early occurrence of pancreatic adenocarcinoma. Endoscopic retrograde cholangiopancreatography techniques for removing duct stones, and dilating and stenting strictures can markedly improve the patient's quality of life, but surgery using Peustow procedures or similar operations may have to be undertaken. Debate continues about the need for total pancreatectomy to prevent the occurrence of pancreatic cancer after the age of 50.

Pancreatic Agenesis

This rare autosomal recessively inherited disorder has typically been found in neonates[6] and is associated with mutations in the PDX1 gene at 13q12, which encodes for the insulin-promoting factor-1 transcription factor.[41] A variant disorder, also inherited as an autosomal recessive disorder associated with cerebellar agenesis, is caused by missense mutations of the PTF-1a gene at 10p12.[42] Recently, 15/27 (56%) of patients with PA were reported as having heterozygous mutations of GATA6, a gene that produces a protein—the transcription factor known as GATA-binding factor 6—and is located on 18q11.[43] Clinically, PA presents in the neonatal period with diabetes mellitus and pancreatic insufficiency.

Congenital Enteropeptidase (Enterokinase) Deficiency

(CED)

CED is a rare autosomal recessive disorder. Enteropeptidase, a serine protease, is a duodenal brush border enzyme required for cleaving of the proenzyme pancreatic trypsinogen to form active trypsin. Enteropeptidase is in fact secreted as a proenzyme, proenteropeptidase, which requires cleavage by a recently discovered enzyme “duodenase” also expressed in the duodenum.[44]

The human gene for proenteropeptidase, PRSS-7, has been mapped to 21q21. Of the 13 or more families with recognized CED, at least two have undergone molecular analysis.[45] In one family, patients demonstrated nonsense mutations in exons 18 and 22 and the parents were heterozygous for one or other of these mutations. In the second family, the index patient had a nonsense mutation in exon 8 and a frameshift mutation in exon 23. Clinically, CED presents in early infancy with severe diarrhea, failure to thrive, and associated severe hypoalbuminemia and edema. Diagnosis is confirmed by obtaining duodenal juice from a pancreatic stimulation test and measuring trypsin before and after adding exogenous enterokinase.

Fat and protein maldigestion/malabsorption occurs in CED and the fat maldigestion has been attributed to the lack of conversion of procolipase to active colipase in the absence of trypsin activation. Patients require pancreatic enzyme replacement therapy (PERT) and this appears very efficacious in correcting the deficiency as many of the original patients diagnosed over 25 years ago are still alive today. However, considering the standard pancreatic enzyme powder does not contain enterokinase (a small bowel mucosal enzyme), it is likely that at least some of the trypsinogens has been activated during the manufacturing process from pork pancreas. Moreover, some adult patients who were diagnosed in early infancy and who still have persistently low enterokinase levels seem to be able to cease their enzyme replacement therapy, leading to the hypothesis that they are able to autoactivate their endogenous trypsinogen.

Although there were earlier papers during the 1960–1970s on congenital isolated trypsinogen deficiency, there have been no subsequent papers and no further molecular analysis.

Congenital Lipase/Colipase Deficiency

There are a limited number of reports on isolated lipase/colipase deficiencies reported in five children up until 1990. Although the genes for pancreatic lipase at 10q24 and colipase at 6p21 have been found, no further work on the above patients has been published to confirm the

presence of specific mutations of either gene to explain these deficiencies. They present with oily stools in early infancy and in the few cases observed fecal fat averaged 50% of oral fat intake. One case had a quantitative pancreatic stimulation test showing both lipase and colipase secretion values were less than 2% of average normal values, but had normal protease and amylase secretion.[46] Other cases have had absent lipase[47] or colipase secretion.[48] Patients thrive well on oral enzyme replacement therapy, but there are no recent data published attesting to their well-being as adults.

FAT AND PROTEIN MALDIGESTION AND MALABSORPTION IN EPD

Normal Fat Digestion

Over 95% of fat ingested by humans is in the chemical form of triglycerides and, except for coconut oil, 98%–100% are long-chain triglycerides (LCTs) with 14 or more carbon atoms per fatty acid ester side chains. Medium-chain triglycerides (MCTs) as, for example, in natural coconut oil, contain 6–12 carbon atoms per fatty acid side chain. Both LCT and MCT require digestion by pancreatic lipase before absorption occurs, but medium-chain fatty acid esters are water soluble and do not require micellar solubilization which is an essential part of the process in transferring insoluble long-chain fatty acids to the enterocyte. The process of fat digestion is complex given there are two major pancreatic lipases—lipase and phospholipase A_2 (PLA_2)—and the lipase cofactor, colipase. Although phospholipids comprise less than 2% of dietary fat (2 g in a diet of 100 g of triglyceride daily), at least 12–15 g are cosecreted with bile acids from the liver on a daily basis. The role of the enzymes and colipase in digesting triglycerides and their interaction with bile acids and phospholipids is shown in the figure 55-1a. In vivo, in the small intestine, the long-chain fatty acids are insoluble in water and can only pass through the unstirred water layer covering the enterocyte if solubilized by incorporation into mixed micelles comprised of bile acids and phospholipids. In Figure 55-1b, in an in vitro experiment with buffer and the SCT, with a simple micelle with a bile acid concentration above the critical micellar concentration, the bile acids prevent adsorption of the lipase to the lipid surface, thus effectively completely inhibiting lipase activity. As per Figure 55-1c, this inhibition can be relieved by adding the smaller and less charged molecule, colipase, which can interdigitate

between the bile acids and then with its specific affinity for lipase bring that molecule into apposition with the surface of the lipid droplet and allow lipolysis to occur.[49–51] Although this concept readily explains the role of colipase in relieving the inhibition of pancreatic lipase activity by bile acids, it does not account for what happens with a mixed micelle with both bile acids and phospholipids present. Figures 55-1d and 55-e depict that the mixed micelles containing bile acid and phosphatidylcholine (PC) actually prevent apposition of colipase with the lipid droplet, completely inhibiting lipolysis.[52] However, as shown in Figure 55-1f, following hydrolysis of the common phospholipid in bile, PC, by PLA_2 to lysoPC, which is water soluble and enters the aqueous phase of the medium, colipase can then gain access to the lipid droplet surface and then activate triglyceride hydrolysis by pancreatic lipase.

Fat Digestion and Absorption in EPDs

How do these concepts apply to EPDs? In congenital lipase/colipase deficiency near absent lipase and colipase secretion during pancreatic stimulation tests was associated with up to 50% fat malabsorption.[46] However, to date, there has not been a report of congenital pancreatic PLA_2 deficiency to confirm its role in triglyceride hydrolysis. In this regard, two novel enzyme replacement preparations, the recombinant breast milk bile salt stimulated lipase (BSSL) and the recombinant microbial lipase have both demonstrated limited efficacy in relieving steatorrhea in CF patients, noting breast-fed CF PI infants at diagnosis, prior to enzyme supplements, have similar fat excretions to bottle-fed infants[29] and those older patients receiving the microbial lipase[53] had an average fat excretion near 40%, which was very similar to the 39% fat excretion documented previously in CF PI patients when not receiving enzyme supplements.[54] Both the new preparations lack PLA_2 present in the conventional microspheric pork pancreas replacement therapy which on average reduces fecal fat to 12%–15% of fat intake. PLA_2 supplementation is essential for the action of pancreatic lipase in the presence of mixed micelles containing PC and one has to question whether the same principle applies to the above nonpancreatic lipases.

Classical pancreatic phenotypes of both CF and SDS include those who are PI with fat malabsorption (fecal fat >7% of oral fat intake) and those who are PS (fecal fat ≤7% of oral fat intake). These definitions have arisen out of a study[55] assessing lipase and colipase secretion rates

measured during quantitative marker perfusion based pancreatic stimulation tests and comparing them with fecal fat excretion. Not surprisingly, colipase secretion matched closely with fecal fat excretion as, in the presence of bile acids, lipase activity is solely dependent on the amount of colipase activity present. Patients with colipase activity in excess of 100 U/kg/h (or 1% of average normal values ≈10,000 U/kg/h) had normal fecal fats, ≤7% of daily fat intake, that is, these patients had sufficient endogenous colipase/lipase activity to maintain normal fat absorption. In contrast, those with colipase secretion less than 100 U/kg/h had insufficient colipase/lipase secretion resulting in fat malabsorption.

It is evident from these data PS patients have varying degrees of pancreatic dysfunction in terms of enzyme/coenzyme secretion from above 1% to 100% of average normal values, identifying the large reserve of exocrine pancreatic function in humans of all ages. Of further interest, among PI patients there are varying degrees of fat malabsorption from over 7% and up to 80% of daily fat intake. Although some of this variation could be attributed to residual pancreatic enzyme secretion, others have proposed that variable secretion of pregastric or lingual lipases could also contribute to this finding. Others have proposed that inadequate bile acid levels from bile acid malabsorption, bacterial contamination of the small intestine, and decreased enterocyte fatty acid uptake and transport are impaired. Although the latter has been attributed to a defect in fatty acid transport related to CFTR dysfunction, it is obviously not present in PS patients who maintain normal fat absorption.

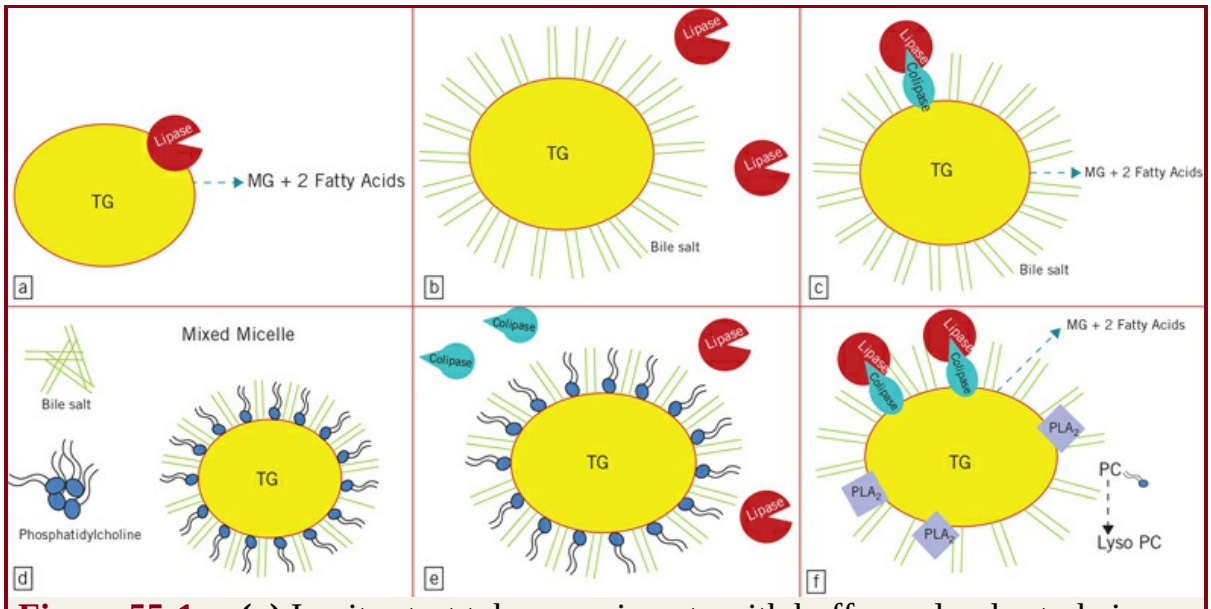


Figure 55-1 **(a)** In vitro test tube experiments with buffer and a short-chain triglyceride (SCT): pancreatic lipase comes into apposition with the lipid droplet and as hydrolysis of the triglyceride ensues, a monoglyceride and two fatty acids

are released into the medium. **(b)** In an in vitro experiment with buffer and the SCT, with a simple micelle with a bile acid concentration above the critical micellar concentration, the bile acids prevent adsorption of the lipase to the lipid surface, thus effectively completely inhibiting lipase activity. **(c)** This inhibition can be relieved by adding the smaller and less-charged molecule, colipase, which can interdigitate between the bile acids and then with its specific affinity for lipase bring that molecule into apposition with the surface of the lipid droplet and allow lipolysis to occur. **(d** and **e)** The mixed micelles containing bile acid and phosphatidylcholine (PC) actually prevent apposition of colipase with the lipid droplet, completely inhibiting lipolysis. **(f)** Following hydrolysis of PC by phospholipase A^2 (PLA^2) to lysoPC, which is water soluble and enters the aqueous phase of the medium, colipase can then gain access to the lipid droplet surface and then activate triglyceride hydrolysis by pancreatic lipase.

Normal Protein Digestion

Ingested protein consists of polypeptide chains composed of covalently bonded amino acid residues folded into quaternary structures with specific spatial arrangements and held together by noncovalent and disulfide bonds. Average recommended daily allowance (RDA) of protein vary according to age and weight from 13 g/d in toddlers 1–3 years of age up to 46–52 g/d in females and males of 14–18 year olds, to 46–56 g/d in adult females and males. In general, over 90% of the protein is absorbed through the small intestine and similar to fat, up to 5% of nitrogen is malabsorbed in normal human subjects.

Dietary protein is digested partially by pepsin in gastric juice, but mainly in the upper small intestine by pancreatic proteases, and enterocyte aminopeptidases, to amino acids and di- and tripeptides. Inactive pepsinogen is released from gastric chief cells and activated by hydrochloric acid, which allows the pepsinogen to unfold and undergo autocatalysis to form active pepsin which preferentially cleaves the aromatic amino acids, phenylalanine, tryptophan, and tyrosine. Pancreatic proteases in general are secreted as a mixture of proenzymes/zymogens, for example, trypsinogens, chymotrypsinogen, and procarboxypeptidase, avoiding activation in pancreatic tissue, which could lead to pancreatitis. Enteropeptidase, a brush border enzyme, is itself formed as a proenzyme and requires cleavage by duodenase to form the active enzyme.[44,45] The activated enterokinase, in turn, cleaves trypsinogens to trypsin which, in turn, cleaves chymotrypsinogen and procarboxypeptidase to the active chymotrypsin and carboxypeptidase. Chymotrypsin hydrolyzes protein to small di- and tripeptides and carboxypeptidase specifically hydrolyzes

amino acids from the carboxyl end of the peptide chain to produce free amino acids for absorption. The process is enhanced by trypsin autocatalysis which culminates in the production of the "proteolytic cascade." Residual di- and tri-peptides which have avoided luminal proteolysis are hydrolyzed to amino acid residues by brush border– and transmembrane–located aminopeptidases.

Protein Digestion and Absorption in EPD

Primary intestinal enterokinase deficiency has provided evidence for the important step of pancreatic proenzymes/zymogen activation necessary for the commencement of intestinal proteolysis by pancreatic proteases.[44,45] Nitrogen balance studies (as a surrogate for protein absorption) of patients with EPDs have virtually been confined to the disorder CF and, specifically, those patients with PI. Patients demonstrate nitrogen malabsorption of 5% to over 50% of ingested nitrogen, that is, a similar range to that of fat malabsorption,[56] but considering the complexities of nitrogen balances they are not as well utilized as fat balance studies in the studies of exocrine pancreatic insufficiency.

NUTRITION IN EXOCRINE PANCREATIC DISORDERS

Each of the EPDs, particularly CF and SDS, and the isolated pancreatic colipase/lipase and enterokinase deficiency (although not a pancreatic disorder, it effectively is, as its deficiency prevents intestinal activation of pancreatic proteases), have helped to elucidate the underlying pathophysiology of pancreatic maldigestion and subsequent malabsorption in human pancreatic diseases. The consequences of pancreatic insufficiency are common to the inherited disorders, in terms of maldigestion and malabsorption, growth, and fat-soluble vitamin deficiencies. These consequences have been best described in the CF population considering the rarity of the other disorders, and the remaining part of the chapter will focus on the nutritional aspects of CF, with reference to the other disorders where relevant.

Consequences of Maldigestion/Malabsorption

A classical feature of pancreatic insufficiency in children is the passing of bulky, malodorous stools which contain oil variably described as melted butter, cheese, or bacon fat. Stool microscopy readily demonstrates the

presence of oil droplets, confirming the clinical suspicion of an EPD. Beyond infancy, fat maldigestion and malabsorption have been well documented in 85%–90% of CF patients,[18] averaging near 40% and up to 80% of fat intake in 3–5 day fat balance studies.[54] Nitrogen losses are less impressive, varying up to 50% of nitrogen intake.[56] Similar fecal fat excretions have been evident in SDS patients and in patients with isolated lipase and colipase deficiency. Exceptions to these findings are seen in CF patients at the time of neonatal diagnosis as some 25%–40% have normal fat excretion.[29,30] A variable 20%–50% of these PS patients will develop PI in the first 10 years of life, likely related to the presence of two Class I–III mutations, as opposed to those who remain PS with at least one Class IV or V mutation.[29] In contrast, in SDS, while most are PI when diagnosed in the first year of life, up to 50% of these improve their exocrine function and develop normal fat excretion by at least 5 years of age.[33]

Prediagnosis and postdiagnosis if noncompliant to PERT, all PI patients are at risk of the complications outlined in Table 55-5. These include the obvious effects of poor weight gain with wasting and hypoproteinemia and edema akin to primary protein energy malnutrition or Kwashiorkor. Prolonged malnutrition, in turn, will impair linear growth as also seen in non-CF populations. Patients with PI are at ongoing risk of fat-soluble vitamin deficiencies prediagnosis and postdiagnosis if noncompliant to oral vitamin therapy and/or PERT. The classical clinical features of these deficiencies, as per Table 55-5,[57–66] are now rarely seen in infants diagnosed by neonatal screening. However, subclinical biochemical deficiencies are apparent and if untreated will eventually result in clinical deficiencies, for instance vitamin D deficient rickets, particularly in northern hemisphere winters.

Current treatment guidelines aim at keeping serum vitamin levels above the lower limit of normal, that is, vitamin A levels >0.7 μmol/L, 25-hydroxyvitamin D (25-OH-D) levels >50 nmol/L, vitamin E levels >7 μg/ml, and the maintenance of a normal international normalized ratio (INR) <1.4 as a surrogate marker for vitamin K deficiency. However, while intuitively a valid approach, these threshold levels have not been completely validated against functional effects of the deficiency.

A cross-sectional study of electroretinograms in CF PI patients showed that they remained normal in those with serum vitamin A levels above 0.6 μmol/L, indicating adequate function,[67] but whether those with persistent borderline levels suffer retinal degeneration in the long term is unknown. Subnormal levels of 25-OH-D of between 25 and 50 nmol/L are common

during Northern hemisphere winters with low sunlight exposure,[61] and rickets and osteomalacia have occurred. Recently we are seeing a similar phenomenon in the Southern hemisphere in spite of sunlight exposure throughout the year, but likely this relates to recent Public Health measures to reduce sun exposure to prevent skin cancer, which is prevalent in these populations. Clinical rickets/osteomalacia due to vitamin D deficiency are, in the main, associated with 25-OH-D levels <20 nmol/L, and those with vitamin D insufficiency without rickets are associated with 25-OH-D levels in the borderline range of 25–50 nmol/L. Maintenance of levels >50 nmol/L would appear appropriate. In regard to vitamin E levels, patients with abetalipoproteinemia or cholestasis develop peripheral neuropathy with persistently low or absent serum levels, but in CF populations a threshold of vitamin E, below which nerve conduction is impaired, has not been defined.[63,64] Vitamin K deficiency has traditionally been evaluated by measurement of prothrombin levels on INR, but they appear less sensitive than two other markers, PIVKA-II or osteocalcin, which are currently undergoing further evaluation.

TABLE 55-5 Consequences of Malabsorption

Gut
- Oily malodorous bowel movements
- Diarrhea
- Bile acid losses
- Risk of gallstones

Systemic
- Nutritional:
  - General:
    - Poor weight gain and subcutaneous fat and muscle wasting
    - Impaired linear growth with stunting
    - Hypoproteinemia with edema, ascites, hydrothorax
  - Fat-soluble vitamins:
    - Vitamin A deficiency:
      - → Raised intracranial pressure, with bulging fontanelle in infants
      - → Impaired dark adaptation with night blindness
      - → Xerophthalmia
      - → Bitot spots
    - Vitamin D deficiency:
      - → Rickets, osteomalacia
    - Vitamin E deficiencies:
      - → Loss of deep tendon reflexes with peripheral neuropathy
      - → Spinocerebellar tract degeneration

- → External ophthalmoplegia
- Vitamin K deficiency:
  - → Coagulopathy
- Essential fatty acid deficiency:
  - → Seborrheic dermatitis

In general, PS patients should not require routine fat-soluble vitamin supplementation. Considering vitamin A toxicity has occurred in PI patients with excessive use of vitamin A, theoretically this could be even more evident in PS patients who have normal absorption.[68] However, PS patients in northern winter climates could become vitamin D deficient, as occurs in normal absorbing non-CF normal populations. Ideally, all PS patients require annual monitoring of serum vitamin levels as, not only will this detect the occurrence of subclinical deficiency but also it has the potential to detect PS patients who become PI and develop subsequent fat-soluble vitamin deficiencies. Similarly, essential fatty acid deficiency, which occurs in PI patients, could be a marker for PS patients becoming PI. Of recent interest, an increased arachidonic acid to docosahexaenoic acid (DHA) ratio has been demonstrated in lung, pancreas, and intestine from a CF knockout mouse and supplementation with DHA has reversed the early pancreatic ductal dilatation.[15] Similar findings have occurred in human CF patients in circulating monocytes[69] and currently the outcome of a North American randomized controlled trial on the effect of DHA is awaited.

PI patients are at risk of vitamin B-12 malabsorption if noncompliant to PERT. The B-12 intrinsic factor complex is bound to R proteins in the stomach and prior to binding to the specific terminal ileal receptors, R proteins are cleaved from the complex by pancreatic proteases. Inadequate PERT theoretically could, therefore, be associated with poor uptake and subsequent cobalamine deficiency. Ileal resection associated with complex neonatal meconium ileus or other surgical complications, for example, intussusception, could also interfere with B-12 absorption mandating regular 3–6 monthly monitoring of serum B-12 levels.

Other Contributing Factors to Malnutrition and Growth Failure in CF

In addition to maldigestion/malabsorption, other factors, including inadequate nutritional intake and excessive energy expenditure, can contribute to the occurrence of malnutrition with concomitant wasting and stunting and CF-related bone disease (CFRBD), as summarized in Table

55-6.

Inadequate Intake

Up until 1980 poor growth was a recognized feature in CF clinics globally, with nearly 50% of patients in early adolescence being below the third percentile for height and weight.[70,71] These findings were attributed to the then undiscovered cause of the underlying disease process and the lack of efficacy of the nonmicrospheric PERT preparations used in that era. However, in 1980, Corey[72] published near normal height percentile distributions in both males and females from the Toronto, Canada, clinic and observed males also approached normal weight percentiles. Females over 8 years of age were more wasted although still only 16% were below the third percentile, as opposed to 42% of those from another similarly sized clinic.[73] The much improved average growth figures from the Toronto clinic suggested that neither the underlying disease nor the use of the less efficacious PERT were major factors to the globally observed growth failure in CF populations. Bell et al.[74] later published dietary intakes on patients from Toronto demonstrating that on a high-fat diet regimen they achieved an average energy intake of 113% of recommended dietary intake (RDI), well in excess of the average 80%–90% RDI reported from clinics utilizing low-fat diets.[73] "Low-fat diets" were used in virtually every clinic during the 1960s and 1970s to control steatorrhea in the absence of efficacious PERT preparations, but the Toronto clinic did not adhere to this policy. Instead, they advised a high-fat diet with LCT comprising 40%–50% of energy intake[74] on the basis that if one increased dietary fat intake both the fat absorbed and the fecal fat in absolute terms would increase concomitantly. In effect, the dietary fat intake increase was replacing fat malabsorbed. This difference in dietary policy was later shown to be the most important single factor of the era in improving nutritional status and growth, but also was a major factor impacting on survival. The latter was best demonstrated by a retrospective epidemiological study comparing growth and survival between the Toronto clinic and the low-fat diet clinic in Boston.[75] Growth was superior in the Toronto clinic, and although average FEV_1% at age 20 was similar, median survival was significantly better with a near 10-year survival advantage to the Toronto clinic (≈30 years vs. 20 years). Nutrition and maintenance of normal diet has now become a major focus for CF clinics given the increasing amount of supportive data and the recognized median survival improvement to the mid-40s in some North American and

Canadian clinics.[21]

TABLE 55-6 Contributing Factors to Malnutrition in Cystic Fibrosis

Inadequate Intake:

- Inappropriate therapy with low-fat diets
- Anorexia:
  - Lung infections
  - Lung disease
  - Antibiotics
  - Malnutrition
  - Esophagitis
  - Gut/biliary tract disease:
    - Celiac disease
    - Persistent DIOS
    - Giardia
    - Crohn's disease
    - Biliary tract stones or strictures
    - Multilobular biliary cirrhosis

Malabsorption (see Table 55-5)

Excessive Energy Expenditure:

- Lung disease
- Lung infections
- Gene defect
- Female gender

DIOS, distal intestinal obstruction syndrome.

Currently, most clinics globally adhere to relatively high-fat diets and the microspheric PERT preparations available from the early 1980s have helped in this process. However, 20% or more patients have difficulties in maintaining their growth and dietary intervention is required if intakes are suboptimal and/or if patients fail to respond to changes in PERT including improvement of compliance and adjuvant therapy to their PERT regimen. Some patients are clearly anorexic, related to their disease complications including lung disease, antibiotics used, esophagitis, gut (celiac disease) and biliary disease (gallstones, bile duct strictures), psychosocial factors, micronutrient deficiencies, and, in temperate climates, salt deficiency. Although investigation and treatment of these complications, including intensification of lung and psychosocial therapy, may improve appetite and growth in some patients, others may continue to deteriorate and more invasive feeding techniques may be required to correct the nutritional

deficits.

Increased Energy Expenditure in CF

This phenomenon has been considered since at least the early 1980s to explain the lack of weight gain in some CF patients regardless of their excellent intake and energy consumption often in excess of 120% of RDI, and appropriate control of fecal fat excretion to less than 20% of dietary fat intake.[76] Similar findings occurred in undernourished CF patients undergoing nutritional rehabilitation, demonstrating energy requirements could be in excess of 150% RDI to achieve weight gain.[77,78] These studies further postulated as most subjects had moderate to severe lung disease the latter was the major contributing factor to the increased resting energy expenditure (REE).

However, in vitro experiments assessing tissue oxygen consumption had in fact demonstrated this occurrence in both cultured CF fibroblasts with a doubling of oxygen consumption,[79] and 6 years later, to a lesser degree, in CF nasal epithelium.[80] These findings indicated that at least a component of the increased energy expenditure was related to the intrinsic disease and independent of lung disease.

Consistent with this hypothesis, Shepherd et al.[81] demonstrated total energy expenditure (TEE) measured by doubly labeled water in CF infants with assumed minimal lung disease averaged 123% of control values suggesting if lung disease was minimal the major factor contributing was the underlying disease. Questions remained regarding the undefined minimal lung disease and the accuracy of the doubly labeled water technique measuring TEE in these infants. Using another approach, O'Rawe et al.[82] measured REE in three genotypic groups: ΔF508 homozygotes, ΔF508 compound heterozygotes, and non-ΔF508 compound heterozygotes and demonstrated an average REE of 121%, 109%, and 104% predicted, respectively, further supporting the contention that REE was related to the severity of genotypic disease. However, average FEV_1% was 56% predictive in the ΔF508/ΔF508 group, 63% in ΔF508/other groups, and near normal in non-ΔF508 groups, demonstrating an effect of lung disease in these three genotypic groups. Fried et al.[83] later demonstrated, retrospectively, a curvilinear effect of lung disease exponentially increasing REE as FEV_1% dropped below 75% predicted, confirming a major effect of deteriorating lung disease escalating energy expenditure.

Having established the deleterious effect of deteriorating lung disease

on energy expenditure, the question still remained as to whether the underlying gene defect contributed to increasing EE, as suggested by the in vitro work above, and whether there was any gender effect given the female survival disadvantage in this disorder. In a large study[84] of 104 CF patients with near normal lung function (average FEV_1% predicted in males 87% and females 83%), PI patients (Class I–III mutations) had an average REE of 110% of that seen in the PS patients (mild Class IV and V mutations) adjusting for lean body mass, thus identifying a very significant effect of those with the severe PI mutations, in spite of the similarity of lung dysfunction in these patients and linking the effect to the intrinsic gene defect. Moreover, in comparison with age-matched controls CF females had very significantly elevated average REE adjusted for lean mass of 111% of controls, whereas CF males had only 104% of their controls. These differences persisted in a subsequent longitudinal study[85] identifying a female disadvantage, which others suggested may contribute to the female survival disadvantage.[86] Overall, although the increase in EE in CF females remains enigmatic, in combination with the EE related to the intrinsic disease and deterioration of lung function into the moderate and severe ranges, one could expect to observe energy requirements escalating over 120% of RDA in many of these patients.

CF-Related Bone Disease (CFRBD)

With improved survival in CF, an increasingly large number of publications[87–92] have reported high clinical fracture rates and decreased bone mineral density (BMD) in the patient population. Fracture rates were twice as high in CF females (16–34 years old) compared with CF males (25–45 years old) with vertebral and rib fractures 100× and 10×, respectively, more common than in control populations. High vertebral fracture rates of 41% historically and 19% radiologically in one adult study, and overall in 34% of adults in another study suggest this is an ever-increasing problem with advancing age and prolonged survival. BMD studies emphasize the extent of the problem, noting impaired BMD [below −1 standard deviation (SD)] in up to 85% of adult patients, and 20%–34% having scores below −2 SD. High fracture rates and BMD scores below −1 SD are observed in patients with low FEV_1% (30%) and BMD scores averaging −2 SD, and alarmingly high fractures rates of up to 42% have been reported in patients post-lung transplantation.[89]

Bone histomorphometric studies demonstrate severe osteopenia and decreased intratrabecular and cortical bone formation with reduced

osteoblastic and increased osteoclastic activity, but surprisingly minimal, if any, evidence for osteomalacia.[93] Clinical BMD studies indicate that the peak BMD accrual that occurs in normal adolescence is impaired in CF populations. The resultant osteopenia and osteoporosis is seemingly independent of country of origin and is in keeping with the concept above that CFRBD is not usually related to vitamin D deficiency and osteomalacia. Bone turnover studies demonstrate, similar to the CFTR-null mouse,[94] bone resorption is in excess of bone formation, as evident by high urinary hydroproline levels and the more recent isotopic calcium kinetic studies showing decreased calcium deposition.

Irrespective of the histological and turnover studies, vitamin D insufficiency (25-OH-D levels of >25–50 nmol/L) is common and deficiency occurs in 5%–10%. CF adults are often in this deficient range and in end stage lung disease may affect up to 33% of patients. The deficiency is likely due to malabsorption, as noted in studies where CF patients absorbed 50% less of vitamin D than controls and had an absence of rise of their serum 25-OH-D levels. Of considerable interest, 1,25-dihydroxyvitamin D levels are usually similar to controls and as it is the most active vitamin D metabolite this may explain the minimal effects of vitamin D deficiency on bone histology and turnover.

Other contributing factors to the development of CFRBD include vitamin K deficiency, CF-related diabetes, physical inactivity and lack of weight-bearing exercise, delayed puberty with gonadal failure, chronic infection with release of inflammatory cytokines [tumor necrosis factor-α, interleukin (IL)-1, IL-6], parathormone, vascular growth factors, and the use of glucocorticoids.

Considering the frequency and severity of CFRBD particularly in the adult population, it is essential that CF children achieve their peak BMD accrual in their adolescent years.[93] In this regard, a consensus committee has recommended a baseline BMD assessment [dual-energy X-ray absorptiometry (DXA)] for all patients ≥18 years of age. In those 8 years and older, a BMD should be performed if ideal weight is <90%, FEV_1% predicted <50% or glucocorticoids ≥5 mg/kg/d for ≥90 d/yr, delayed puberty, and/or fractures. If BMD is ≥−1.0 T or Z score, DXA should be repeated 5 yearly; if T or Z score is −1.0 to −2.0, 2–4 yearly; and ≤ −2.0, yearly, as listed in Table 55-7.

NUTRITIONAL ASSESSMENT

Most CF clinics worldwide would undertake a baseline nutritional assessment at diagnosis with at least an annual review of the same parameters. As most patients are reviewed 3–6 monthly in-between annual visits, more limited evaluation with anthropometry and physical examination can be assessed and, if anomalies occur, further investigations at that time may be required. Baseline and annual reviews require height, weight, ideal weight for height and BMI, and head circumference measurements, with percentiles or Z scores and examination for micronutrient deficiencies. Blood work should include hemoglobin, coagulation profile, serum albumin, liver function tests, U&E (urea and electrolytes), serum fat-soluble vitamins and 25-OH-D levels, and serum essential fatty acids. Iron and trace metals, zinc and selenium levels may be required depending on clinical findings or geographic location and serum vitamin B-12 if patients are noncompliant to PERT. Fasting blood glucose is recommended annually after 5 years of age or sooner if diabetic symptoms occur (polyuria, polydipsia). After neonatal screening diagnosis, a formal three-day fecal fat study is essential to determine if the infant is PI or PS as neither fecal elastase (FE-1) nor the mixed triglyceride breath test (MTBT) have been fully evaluated in this population. A three-day fecal fat result in full-term babies of ≥10% of fat intake is indicative of fat malabsorption in bottle-fed infants and ≥2 g/d in breast-fed babies.[30] In nonscreened populations with children presenting for diagnosis at over 1 year of age, either FE-1 or MTBT can be used reliably to evaluate the pancreatic phenotype (PI or PS). The latter tests can also be used in later childhood to determine if a PS patient has become PI because of symptoms suggesting PI, for example, weight loss, oily stools, night blindness, absence of deep tendon reflexes, rickets and coagulation problems, or low-serum fat-soluble vitamin levels. If fresh symptoms of malabsorption occur in spite of compliance to an adequate dose of PERT, a formal fecal fat may be required to assess the failure of the medication. Prior to doing so it is worth considering intercurrent illness, for example, giardiasis or cryptosporidiosis and to perform celiac serology to exclude celiac disease. Annual food diaries to assess macro- and micronutrient intakes are undertaken at many clinics, particularly in patients with evidence of growth or nutritional deficits. However, in patients maintaining normal growth who are asymptomatic with normal blood profiles, the use of food diaries is questionable.

TABLE 55-7 Guidelines for the Management of CF-

Related Bone Disease
Regular/Routine Therapy • Maintain serum 25-OH vitamin D levels >50 ng/mL • Vitamin K supplements 0.3–0.5 mg/d • Calcium intake up to 1500 mg/d • Weight-bearing exercise • Intensification of treatment of lung infections • Cessation of oral corticosteroids • Treat diabetes • Endocrine referral for the management of delayed puberty
For T or Z scores for bone mineral density of −1.0 to −2.0—oral bisphosphonates. For T or Z scores for bone mineral density below −2.0—oral bisphosphonates, and if not tolerated, intravenous bisphosphonates.

In the absence of symptoms of malabsorption, growth failure can signal a number of significant problems, particularly exacerbation of pulmonary disease. However, if adequate respiratory therapy is given and the growth problems continue one has to consider poor intake related to esophagitis, liver, and biliary tract disease, and celiac disease, all of which require further investigation. Moreover, if the growth failure continues despite negative investigations, they may have excessive energy expenditure as seen in pubertal PI females colonized with *Pseudomonas* either with or without significant lung disease. Such patients, if unable to increase their nutrient intake orally, should then be considered for enteral tube feeding.

Nutritional Management

Nutrient requirements for EPD patients with PI are summarized in Table 55-8, including a salt supplement regimen for CF patients. EPD patients with PS should grow normally on an average normal diet without macro- or micronutrient supplements with the exception that CF patients still require salt supplements. In terms of macronutrients, average protein intake in CF PI patients is well in excess of requirements when consuming a liberal relatively high-fat diet, where LCT constitutes over 40% of the energy intake. However, energy requirements vary considerably depending on residual fat malabsorption, excessive energy expenditure, and the degree of physical activity undertaken. Maintenance of normal height and weight percentiles with patients tracking on the best previously achieved percentiles is the appropriate evidence of an adequate energy intake. Deviation below the achieved percentile is indicative of a negative energy balance related to decreased appetite, increased malabsorption or excessive

utilization, or some combination of these factors, and requires appropriate investigation.

Micronutrient balance is best monitored as above measuring serum levels of fat-soluble vitamins and trace metals, and maintenance of these levels within a range of normal defined by a clinic's local laboratory. Subnormal levels may be related to noncompliance and if not, an increase in vitamin dose would be appropriate, but with the caution that vitamin A toxicity has occurred in CF PI individuals.

TABLE 55-8 Nutrient Requirements in PI Including CF

Nutrient	Requirement	
Protein	>120% of RDI	
Energy	Variable, depending on disease status, but in PI patient should average near 120% RDI, with 40% as LCT and 3%–5% as EFA	
Vitamins:		
A	5,000 IU/d[a]	1–3 years of age
	10,000 IU/d[a]	>3 years of age
D	400–800 IU/d[b]	
E	200–400 IU/d[a]	
K	5 mg twice weekly[a]	
Salt intake[c]		
Age:		
0–6 months	0.5 g/d	
6–12 months	1 g/d	
1–5 years	2 g/d	
>5 years	3–5 g/d	

CF, cystic fibrosis; EFA, essential fatty acid; LCT, long-chain triglyceride; PI, pancreatic insufficiency; RDI, recommended dietary intake.
Can use vitamins A, D, E, and K (Scandipharm) or vitamins A, B, D, E, and K (Technipro): 4–10 years, one cap daily; >10 years, two caps daily; 0–1 year, 1 mL daily; 1–3 years, 2 mL daily; or Micelle A&E, 1–2 mL daily, or Aquasol E (Rover Pharmaceutical, Fort Washington, PA), 1 mL daily.
[a]If serum levels and coagulation profile are normal, patients may not need supplement.
[b]Not required if exposed to sunlight throughout the year.
[c]Double salt dose in hot weather or during strenuous exercise.

CF patients living in warm to hot climates in subtropical/tropical zones are at constant risk of salt depletion. The average sweat chloride in CF is

in the region of 100 mmol/L and is the equivalent concentration of a solution of 2/3 N saline or 6 g/L. During prolonged exercise on a hot day, an older child could easily lose a liter or more of sweat and replacement electrolyte solution or salt tablets are required during the exercise. The proportionally large body surface area of small CF infants leads to a more rapid salt depletion in hot enclosed areas, and they may present in shock with a hyponatremic, hypokalemic, hypochloridemic metabolic alkalosis requiring emergency resuscitation with normal saline boluses 10–20 mL/kg intravenously or intraosseously if intravenous cannulation is not possible.

Malabsorption is currently managed with microspheric PERT. Many different regimes have been developed based on dosage per kilogram of body weight per day or dosage per gram of fat per meal. In the 1980s when microspheric preparations were first used, a number of studies were conducted in older children and adolescents (in general, over 30 kg in weight) using a standard capsule containing 5000 IU of lipase. In each study, 3-day fecal fat balance studies on and off PERT preparations were performed, but with each new study a higher number of capsules and therefore a higher dose of lipase were used.[76,95,96] Studies using 25–30 capsules/d showed average fecal fat as a percent of fat intake had decreased from near 40% off PERT to 12%–15% on PERT, suggesting there was a plateau effect on fat excretion when the dose averaged between 20 and 30 capsules/d. Using a dose of one capsule of 5000 U of lipase per kilogram per day to a maximum of 30 per day ensured normal growth of infants associated with a newborn screening program,[30] and in older children near 40% had normal fat absorption (<10% of fat intake) in fat balance studies.[76] However, a variable number of patients (<20%) maintained fat excretions in excess of 20% of fat intake. Others had also observed this phenomenon and suggested it was related to poor pancreatic HCO_3^- secretion and subsequent failure to elevate the duodenal pH above 6,[96] which was required for the microsphere dissolution. Initial studies using H_2 receptor antagonists to suppress gastric acid secretion in fact demonstrated marked improvement in fat excretion in these patients[96] and have led to the use of proton pump inhibitors in patients with excessive steatorrhea in spite of compliance to PERT.

During the late 1980s and early 1990s, manufacturers of PERT increased the lipase dosage per capsule so that a variety of capsules with lipase levels ranging from 5,000 IU and up to 25,000 IU became available, on the basis that the lesser number of capsules required would improve

compliance. However, many units worldwide permitted patients to self-modulate their doses and they ended up taking as many or more capsules per day of the high-dose preparations, giving them a lipase dose of 50,000 U/kg/d, that is, up to 10 times higher than their previous dose. Contemporaneously, the first descriptions of FC appeared suggesting a link between the excessive use of high-dose lipase capsules and the occurrence of FC.[97] Two subsequent epidemiological studies[98,99] confirmed this association leading to the recommendation that the lipase dose should not exceed more than 10,000 U/kg/d. Subsequently, FC has been virtually nonexistent.

In order to improve fat absorption in patients on conventional microspheric PERT, other investigators have trialed a recombinant breast milk lipase and a recombinant microbial lipase. Breast milk lipase is a BSSL and based on in vitro work has been shown to digest LCT similar to pancreatic lipase. However, at diagnosis in a newborn screening program, breast-fed PI CF infants pre-PERT have virtually identical ranges of fat excretion as those PI CF infants who were bottle fed pre-PERT indicating the BSSL had minimal, if any, effect.[29] Similarly, in older children, the microbial lipase had a minimal effect when studying fecal fats on and off the preparation.[53] As per the Figure, the pancreatic lipase/colipase complex can only gain access to the lipid droplet incorporated into the complex bile salt/phospholipid micelles if the phospholipid has been hydrolyzed by pancreatic PLA_2. The capsules trialed for either the recombinant BSSL or the microbial lipase did not have PLA_2 and one is left to assume the unhydrolyzed phospholipids which have remained in the micelle have prevented access of the lipase/colipase complex to the lipid droplet. It would be worth reevaluating the combined effect of these two new lipases in combination with a minimal dose of conventional microspheric PERT, which contains PLA_2.

Infant Nutrition

Prior to neonatal screening programs for early diagnosis of CF, CF infants often presented clinically with a history of fatty diarrhea and associated hypoproteinaemia, edema, ascites, and/or hydrothorax, with evidence of fat-soluble vitamin deficiencies. The hypoproteinemia could be attributed to the lack of proteolytic enzymes, but it was also more common in breast-fed CF infants, related to the lower protein content of human milk. Moreover, after diagnosis, the nonmicrospheric enzyme preparations used in the 1960s and 1970s were problematic as they could not be used in an

encapsulated form for infants. Mixing with milk or fruit gels and sauces provided a vehicle for ingestion, but when given orally infants often experienced mouth ulceration because of the premature activation of the enzymes by the more alkaline salivary secretions.

To overcome these problems in the early 1970s, the newly marketed hydrolyzed protein and MCT-containing semielemental formulae were trialed for use in CF infants. Fat balance studies showed near normal or normal fecal fat excretion, and the hydrolyzed protein overcame the problem of lack of pancreatic proteolytic enzymes.[100] However, long-term studies demonstrated poor growth of these infants so treated.[101] In addition, a further study[102] indicated that the Van de Kamer method for measuring the fecal fat in the original study was incorrect as it only measured LCT and not MCT. Thus, the fecal fat was seriously underestimated and no doubt the ongoing malabsorption contributed to the poor growth of this cohort.

An advantage of neonatal screening for diagnosis of CF has been the early recognition of infants with PI and commencement of PERT. This has enabled reevaluation of breast-feeding of CF infants[103] and demonstrated that exclusive breast-feeding, for at least 4 months and up to 12 months, is associated with average normal height and weight percentiles at 12 and 24 months, without the occurrence of hypoalbuminemia. These infants still require fat-soluble vitamin supplementation. Non-breast-fed PI infants also grew normally on regular cow's milk formulae with appropriate PERT supplements. These studies clearly indicate that specialized semielemental formulae as used in some clinics[101,104] are unnecessary for normal growth in CF infants with the exception of those with postsurgical short gut syndrome which these days is a rare occurrence.

Enteral Tube Feeding

Multiple studies[105–108] have been published on the efficacy of nocturnal gastrostomy feeding regimens for nutritional rehabilitation and restoration of normal growth in malnourished CF patients. Definitions for considering such therapy have varied, but center around the premise that if a patient has fallen below the third percentile for weight and/or height, and more recently below the third percentile for BMI, and the patient does not respond to oral supplements over a 3–6 months' duration, then tube feeding is an optional therapy. However, tube feeding is very demanding and may prove to be overburdening for families considering the multitude of other patient requirements. Some cope well, embracing the therapy and

ensuring its success. Those who express reluctance to have enteral feeding, who have psychosocial problems, and/or not coping with regular therapy are very unlikely to achieve nutritional rehabilitation. Thus, before embarking on this line of treatment, families require a thorough psychosocial evaluation and/or education program, including discussions with patients who have successful gastrostomies to ensure this type of therapy is appropriate for individual patients and their families.[109] Motivation and compliance are particularly important factors and it is imperative that teams guiding such programs bear this in mind both before commencing the therapy and throughout its duration, which may extend for many months or even years. Nutritional rehabilitation with enteral feeding prelung transplant intuitively is appropriate and in some units it is a prerequisite to lung transplant. However, such recommendations have yet to be completely evaluated.

CONCLUSION

The EPDs, except for isolated pancreatic enzyme deficiencies and enteropeptidase deficiency, are multisystem disorders and require a multidisciplinary team to achieve optimal care. These teams should include medical personnel with specific expertise in diagnosing and managing exocrine pancreatic disease, and expert nutritional support teams for managing the nutritional problems associated with pancreatic insufficiency. Optimal dietary management, ensuring adequate macro- and micronutrient intake and PERT control of malabsorption provides the best possible chance of achieving normal growth and ensuring normal bone development with adolescent accrual of peak BMD. Likewise, regular monitoring is necessary to detect deviations, particularly decline in growth or bone health, to ensure intensification of therapy and prevent further decline, and to restore growth and bone parameters to normal.

REFERENCES

1. Wilcken B, Brown AR, Urwin R, Brown DA. Cystic fibrosis screening by dried blood spot trypsin assay: results in 75,000 newborn infants. *J Pediatr.* 1983;102:383–387.
2. Shwachman H, Diamond LK, Oski FA, Khaw KT. The syndrome of pancreatic insufficiency and bone marrow dysfunction. *J Pediatr.* 1964;65:645–663.
3. Goobie S, Popovic M, Morrison J, et al. Shwachman-Diamond syndrome with exocrine pancreatic dysfunction and bone marrow failure maps to the centromeric region of chromosome 7. *Am J Hum Genet.* 2001;68:1048–1054.
4. Pearson HA, Lobel JS, Kocoshis SA, et al. A new syndrome of refractory anemia

with vacuolization of marrow precursors and exocrine pancreatic dysfunction. *J Pediatr*. 1979;95:976–984.
5. Johanson A, Blizzard R. A syndrome of congenital aplasia of the alae nasi, deafness, hypothyroidism, dwarfism, absent permanent teeth, and malabsorption. *J Pediatr*. 1971;79:982–987.
6. Howard CP, Go VLW, Infante AJ, et al. Long-term survival in a case of functional pancreatic agenesis. *J Pediatr*. 1980;97:786–789.
7. Riordan JR, Rommens JM, Kerem BS, et al. Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. *Science*. 1989;245:1066–1072.
8. Rommens JM, Iannuzzi MC, Kerem B, et al. Identification of the cystic fibrosis gene: chromosome walking and jumping. *Science.* 1989;245:1059–1065.
9. Bear CE, Li C, Kartner N, et al. Purification and functional reconstitution of the cystic fibrosis transmembrane conductance regulator (CFTR). *Cell.* 1992;68:809–818.
10. Quinton PM. Chloride impermeability in cystic fibrosis. *Nature.* 1983;301:421–422.
11. Ishiguro H, Steward C, Wilson RW, Case RM: Bicarbonate secretion in interlobular ducts from guinea pig pancreas. *J Physiol.* 1996;495:179–191.
12. Quinton PM. Role of epithelial HCO3– transport in mucin secretion: lessons from cystic fibrosis. *Am J Physiol Cell Physiol.* 2010;299:C1222–C1233.
13. Gaskin KJ, Durie PR, Corey M, et al. Evidence for a primary defect in pancreatic HCO3- secretion in cystic fibrosis. *Pediatr Res.* 1982;16:554–557.
14. Kopelman H, Corey M, Gaskin KJ, et al. Impaired chloride secretion, as well as bicarbonate secretion, underlies the fluid secretory defect in the cystic fibrosis pancreas. *Gastroenterology.* 1988;95:349–355.
15. Freedman SD, Katz MH, Parker EM, et al. A membrane lipid imbalance plays a role in the phenotypic expression of cystic fibrosis in cftr(-/-) mice. *Proc Natl Acad Sci USA.* 1999;96(24):13995–14000.
16. Wilschanski M, Zielenski J, Markiewicz D, et al. Correlation of sweat chloride concentration with classes of the cystic fibrosis transmembrane conductance regulator gene mutations. *J Pediatr.* 1995;127:705–710.
17. Ahmed M, Corey M, Forstner G, et al. Molecular consequences of cystic fibrosis transmembrane regulator (CFTR) gene mutations in the exocrine pancreas. *Gut.* 2003;52:1159–1164.
18. Gaskin KJ, Gurwitz D, Durie PR, et al. Improved respiratory prognosis in CF patients with normal fat absorption. *J Pediatr.* 1982;100:857–862.
19. Gaskin KJ, Wilcken. Long-term outcomes for patients with cystic fibrosis in Australia. *Med J Aust.* 2011;195:370–371.
20. Dodge JA, Lewis PA, Stanton M, Wilsher J. Cystic fibrosis mortality and survival in the UK: 1947-2003. *Eur Respir J.* 2007;29:522–526.
21. Cystic Fibrosis Canada. Canadian Cystic Fibrosis Patient Data Registry Report, 2009, http://www.cysticfibrosis.ca/assets/files/pdf/CPDR_ReportE.pdf. Updated 2010. Accessed August 2011.
22. Phelan P, Hey E. Cystic fibrosis mortality in England and Wales and in Victoria, Australia 1976-1980. *Arch Dis Child.* 1984;59:71–73.
23. Waters DL, Wilcken B, Irwig L, et al. Clinical outcomes of newborn screening for cystic fibrosis. *Arch Dis Child Fetal Neonatal.* 1999;80:F1–F7.
24. McKay KO, Waters DL, Gaskin KJ. The influence of newborn screening for cystic fibrosis on pulmonary outcomes in New South Wales. *J Pediatr.* 2004;147:S47–S50.
25. Dijk FN, McKay K, Barzi F, et al. Improved survival in cystic fibrosis patients diagnosed by newborn screening compared to a historical cohort from the same

centre. *Arch Dis Child.* 2011;96(12):1118–1123.
26a. Grosse SD, Rosenfeld M, Owen JD, et al. Potential impact of newborn screening for cystic fibrosis on child survival: a systematic review and analysis. *J Pediatr.* 2006;149:362–366.
26b. Yen EH, Quinton H, Borowitz D. Better nutritional status in early childhood is associated with improved clinical outcomes and survival in patients with cystic fibrosis. *J Pediatr.* 2013;162:530–535.
27. Kerem E, Corey M, Kerem B, et al. The relation between genotype and phenotype in cystic fibrosis—analysis of the most common mutation (DF508). *N Engl J Med.* 1990;323:1517–1522.
28. Novelli G, Sangiuolo F, Maceratesi P, et al. The up-to-date molecular genetics of cystic fibrosis. *Biomed Pharmacother.* 1994;48:455–463.
29. Cipolli M, Castellani C, Wilcken B, et al. Pancreatic phenotype in infants with cystic fibrosis identified by mutation screening. *Arch Dis Child.* 2007;92:842–846.
30. Waters DL, Dorney SFA, Gaskin KJ, et al. Pancreatic function in infants identified as having cystic fibrosis in a neonatal screening program. *N Engl J Med.* 1990;322:303–308.
31. Mack DR, Forstner GG, Wilschanski M, et al. Shwachman syndrome: exocrine pancreatic dysfunction and variable phenotypic expression. Gastroenterology. 1996;1593–1602.
32. Boocock GR, Morrison JA, Popovic M, et al. Mutations in SBDS are associated with Shwachman-Diamond syndrome. Nat Genet. 2003;33:97–101.
33. Hill RE, Durie PR, Gaskin KJ, et al. Steatorrhoea and pancreatic insufficiency in Shwachman's syndrome. Gastroenterology. 1982;83:22–27.
34. Dror Y, Freedman MH. Shwachman-Diamond syndrome. *Br J Haematol.* 2002;118:701–713.
35. Makitie O, Ellis L, Durie PR, et al. Skeletal phenotype in patients with Shwachman-Diamond syndrome and mutations in SBDS. *Clin Genet.* 2004;65:101–112.
36. Sano T, Ban K, Ichiki T, et al. Molecular and genetic analyses of two patients with Pearson's marrow-pancreas syndrome. *Pediatr Res.* 1993;34:105–110.
37. Zenker M, Mayerle J, Leach MM, et al. Deficiency of UBR1, a ubiquitin ligase of the N-end rule pathway, causes pancreatic dysfunction, malformations and mental retardation (Johanson-Blizzard syndrome). *Nat Genet.* 2005;37:1345–1350.
38. Whitcomb DC, Gorry MC, Preston RA, et al. Hereditary pancreatitis is caused by a mutation in the cationic trypsinogens gene. *Nat Genet.* 1996;14:141–145.
39. Szabo A, Sahin-toth M. Increased activation of hereditary pancreatitis-associated human cationic trypsinogens mutants in the presence of chymotrypsin C. *J Biol Chem.* 2012; 287:20701–20710.
40. Gorry MC, Gabbaizedeh D, Furey W, et al. Mutations in the cationic trypsinogen gene are associated with recurrent acute and chronic pancreatitis. *Gastroenterology.* 1997;113:1063–1068.
41. Stoffers DA, Zinkin NT, Stanojevic V, et al. Pancreatic agenesis attributable to a single nucleotide in the human IPF1 gene coding sequence. *Nat Genet.* 1997;15:106–110.
42. Tutak E, Satar M, Yapicioglu H, et al. A Turkish newborn infant with cerebellar agenesis/neonatal diabetes and PTF1A mutation. *Genet Counsel.* 2009;20:46–52.
43. Allen HL, Flannagan SE, Shaw-Smith C, et al. GATA6 haploinsufficiency causes pancreatic agenesis in humans. *Nat Genet.* 2012;44:20–22.
44. Zamolodchikova TS, Sokolova EA, Lu D, Sadler JE. Activation of recombinant proenteropeptidase by duodenase. *FEBS Lett.* 2000;466:295–299.
45. Holzinger A, Maier EM, Buck C, et al. Mutations in the proenteropeptidase gene are

the molecular cause of congenital enteropeptidase deficiency. *Am J Hum Genet.* 2002;70:20–25.
46. Ghishan FK, Moran JR, Durie PR, et al. Isolated congenital lipase-colipase deficiency. *Gastroenterology.* 1984;86:1580–1582.
47. Figarella C, De Caro A, Leupold D, Poley J. Congenital pancreatic lipase deficiency. *J Pediatr.* 1980;96:412–416.
48. Hildebrand H, Borgstrom B, Bekassy A, et al. Isolated colipase deficiency in two brothers. *Gut.* 1982;23:243–246.
49. Morgan R, Barrowman J, Borgstrom B. The effect of sodium taurodeoxycholate and pH on the gel filtration behaviour of rat pancreatic protein and lipases. *Biochim Biophys Acta.* 1969;175:65–75.
50. Morgan R, Hoffman N. The interaction of lipase, lipase cofactor and bile salts in triglyceride hydrolysis. *Biochim Biophys Acta*. 1971;248:143–148.
51. Gaskin KJ, Durie PR, Hill RE, et al. Colipase and maximally activated pancreatic lipase in normal subjects and patients with steatorrhoea. *J Clin Invest.* 1982;69:427–434.
52. Borgstrom B. Importance of phospholipids, pancreatic phospholipase A2 and fatty acids for the digestion of dietary fat: in vitro experiments with the porcine enzymes. *Gastroenterology.* 1980;78:954–962.
53. Borowitz D, Goss CH, Limauro S, et al. Study of a novel pancreatic enzyme replacement therapy in pancreatic insufficient subjects with cystic fibrosis. *Pancreas.* 2006;32:258–263.
54. Forstner GG, Gall G, Corey M, et al. Digestion and absorption of nutrients in cystic fibrosis. In: Sturgess JM, ed. *Proceedings of the 8th International Congress on CF*. Mississauga, ON: Imperial Press Ltd; 1980. pp. 137–148.
55. Gaskin KJ, Durie PR, Hill RE, et al. Colipase and lipase secretion in childhood onset pancreatic insufficiency: delineation of patients with steatorrhoea secondary to relative colipase deficiency. *Gastroenterology.* 1984;86:1–7.
56. Lapey A, Kirkpatrick J, di Sant'Agnese P, Laster L. Steatorrhoea and azotorrhoea and their relation to growth and nutrition in adolescents and young adults with cystic fibrosis. *J Pediatr.* 1974;84:328–334.
57. Abernathy RS. Bulging fontanelle as presenting sign in cystic fibrosis. *Am J Dis Child*. 1976;130:1360–1362.
58. Rayner RJ, Tyrell JC, Hiller EJ, et al. Night blindness and conjunctival xerosis due to vitamin A deficiency in cystic fibrosis. *Arch Dis Child*. 1989;64:1151–1156.
59. Scott J, Elias E, Moult PJA, et al. Rickets in adult cystic fibrosis with myopathy, pancreatic insufficiency and proximal renal tubular dysfunction. *Am J Med*. 1977;63:488–492.
60. Hubbard VS, Farrell PM, di Sant'Agnese PA. 25-Hydroxycholecalciferol levels in patients with cystic fibrosis. *J Pediatr*. 1979;94:84–86.
61. Reiter EO, Brugman SM, Pike JW, et al. Vitamin D metabolites in adolescents and young adults with cystic fibrosis: effects of sun and season. *J Pediatr*. 1985;106:21–25.
62. Farrell PM, Bieri JG, Fratantoni JF, et al. The occurrence and effects of human vitamin E deficiency. A study in patients with cystic fibrosis. *J Clin Invest.* 1977;60:233–241.
63. Elias E, Muller DPR, Scott J, et al. Association of spinocerebellar disorders with cystic fibrosis or chronic childhood cholestasis and low serum vitamin E. *Lancet.* 1981;ii:1319–1321.
64. Sitrin MD, Lieberman F, Jensen WE, et al. Vitamin E deficiency and neurologic disease in adults with cystic fibrosis. *Ann Intern Med*. 1987;107:51–54.

65. Walters TR, Koch HF. Hemorrhagic diathesis and cystic fibrosis in infancy. *Am J Dis Child*. 1972;124:641–647.
66. Komp DM, Selden RF. Coagulation abnormalities in cystic fibrosis. *Chest*. 1970;58:501–508.
67. Whatham A, Suttle C, Blumenthal C, et al. ERGs in children with pancreatic enzyme insufficient and pancreatic enzyme sufficient cystic fibrosis. *Doc Ophthalmol*. 2009;119:43–50.
68. Graham-Maar RC, Schall JI, Stettler N, et al. Elevated vitamin A intake and serum retinol in preadolescent children with cystic fibrosis. *Am J Clin Nutr*. 2006;84:174–182.
69. Freedman SD, O'Sullivan BP, Martinez-Clark P, et al. An abnormality in AA/DHA metabolism is present in CFTR-regulated tissues from CF patients. *Pediatr Pulmonol*. 2000;20:158–159.
70. Sproul A, Huang H. Growth patterns in children with cystic fibrosis. *J Pediatr*. 1964;65:664–676.
71. Berry HK, Kellogg FW, Hunt MM, et al. Dietary supplement and nutrition in children with cystic fibrosis. *Am J Dis Child*. 1975;129:165–171.
72. Corey ML. Longitudinal studies in cystic fibrosis. In: Sturgess JM, editor. *Perspectives in Cystic Fibrosis*. Toronto, ON: The Imperial Press Ltd; 1980. pp. 246–255.
73. Soutter VL, Kristidis P, Gruca MA, Gaskin KJ. Chronic undernutrition, growth retardation in cystic fibrosis. *Clin Gastroenterol*. 1986;15:137–155.
74. Bell L, Durie P, Forstner GG. What do children with cystic fibrosis eat? *J Pediatr Gastroenterol Nutr*. 1984;3(Suppl 1):137–146.
75. Corey M, McLaughlin FJ, Williams M, Levison H. A comparison of survival, growth and pulmonary function in patients with cystic fibrosis in Boston and Toronto. *J Clin Epidemiol*. 1988;41:588–591.
76. Gaskin KJ. Impact of nutrition in cystic fibrosis. A review. *J Pediatr Gastroenterol Nutr*. 1988;7(Suppl 1):12–17.
77. Pencharz P, Hill R, Archibald E, et al. Energy needs and nutritional rehabilitation in undernourished adolescents and adults with cystic fibrosis. *J Pediatr Gastroenterol Nutr*. 1984;3(Suppl 1):147–153.
78. Vaisman N, Pencharz PB, Corey M, et al. Energy expenditure of patients with cystic fibrosis. J Pediatr. 1987;111:496–500.
79. Feigel RJ, Shapiro BL. Mitochondrial calcium uptake and oxygen consumption in cystic fibrosis. *Nature*. 1979;178:276–277.
80. Stutts MJ, Knowles MR, Gatzy JT, Boucher RC. Oxygen consumption and ouabain binding-sites in cystic fibrosis nasal epithelium. *Pediatr Res*. 1986;20:1316–1320.
81. Shepherd RW, Holt TL, Vasques-Velasquez L, et al. Increased energy expenditure in children with cystic fibrosis. *Lancet.* 1988;1:1300–1303.
82. O'Rawe A, McIntosh I, Dodge JA, et al. Increased energy expenditure in cystic fibrosis is associated with specific mutations. *Clin Sci*. 1992;82:71–76.
83. Fried MD, Durie PR, Tsui LC, et al. The cystic fibrosis gene and resting energy expenditure. *J Pediatr*. 1991;119:913–916.
84. Allen JR, McCauley J, Selby A, et al. Differences in resting energy expenditure between male and female children with cystic fibrosis. *J Pediatr*. 2003;142:15–19 (editorial)
85. Magoffin A, Allen JR, McCauley J, et al. Longitudinal analysis of resting energy expenditure in patients with cystic fibrosis. *J Pediatr*. 2008;152:703–708.
86. Stallings VA. Gender, death and cystic fibrosis: is energy expenditure a component? *J Pediatr*. 2003;142:4–6. (Editorial)

87. Boyle, MP. Update on maintaining bone health in cystic fibrosis. *Curr Opin Pulm Med*. 2006;12:453–458.
88. Haworth CS, Selby PL, Webb AK, et al. Low bone mineral density in adults with cystic fibrosis. *Thorax*. 1999;54:961–967.
89. Aris RM, Neuringer IP, Weiner MA, et al. Severe osteoporosis before and after lung transplantation. *Chest*. 1996;109:1176–1183.
90. Aris RM, Renner JB, Winders AD, et al. Increased rate of fractures and severe kyphosis: sequelae of living into adulthood with cystic fibrosis. *Ann Intern Med*. 1998;128:186–193.
91. Shead EF, Haworth CS, Gunn E, et al. Osteoclastogenesis during infective exacerbations in patients with cystic fibrosis. *Am J Respir Crit Care Med*. 2006;174:306–311.
92. Javier RM, Jacquot J. Bone disease in cystic fibrosis: what's new? *Joint Bone Spine*. 2011;78:445–450.
93. Aris RM, Merkel PA, Bachrach LK, et al. Guide to bone health and disease in cystic fibrosis. *J Clin Endocrinol Metab*. 2005;90:1888–1896.
94. Dif F, Marty C, Baudoin C, et al. Severe osteopenia in CFTR-null mice. *Bone*. 2004;35:595–603.
95. Mitchell EA, Quested C, Marks RD, et al. Comparative trial of viokase, pancreatin and Pancrease® pancrelipase (enteric-coated beads) in the treatment of malabsorption in cystic fibrosis. *Aust Paediatr J*. 1982;18:114–117.
96. Gow R, Bradbear R, Francis P, Shepherd RW. Comparative study of varying regimens to improve steatorrhoea and creatorrhoea in cystic fibrosis. *Lancet*. 1981;i:1071–1074.
97. Smyth RL, van Velzen D, Smyth AR, et al: Strictures of ascending colon in cystic fibrosis and high-strength pancreatic enzymes. *Lancet*. 1994;343(8889):85–86.
98. FitzSimmons SC, Burkhart GA, Borowitz D, et al. High-dose pancreatic-enzyme supplements and fibrosing colonopathy in children with cystic fibrosis. *N Engl J Med*. 1997;336:1283–1289.
99. Smyth RL, Ashby D, O'Hea U, et al: Fibrosing colonopathy in cystic fibrosis: results of a case-control study. *Lancet*. 1985;346(8985):1247–1251.
100. Kuo PT, Huang NN. The effect of medium-chain triglycerides upon fat absorption and plasma lipids and deposition of fat of children with cystic fibrosis of the pancreas. *J Clin Invest*. 1965;44:1924–1929.
101. Duhamel DJ, Lenoir G, Brouard J, et al. Prise en charge nutritionelle des nouveau-nés et nourrissons atteints de fibrose kystique du pancréas et dépistés à la naissance. Proceedings of the International Conference on Neonatal Screening for Cystic Fibrosis; October 5– October 6, 1988,; Caen, France. pp. 183–199.
102. Durie PR, Newth CJ, Forstner GG, Gall DG. Malabsorption of medium-chain trigylcerides in infants with cystic fibrosis: correction with pancreatic enzyme supplements. *J Pediatr*. 1980;96:862–864.
103. Holliday KE, Allen JR, Waters DL, et al. Growth of human milk-fed and formula-fed infants with cystic fibrosis. *J Pediatr*. 1991;118:77–79.
104. Marcus MS, Sondel SA, Farrell PM, et al. Nutritional status of infants with cystic fibrosis associated with early diagnosis and intervention. *Am J Clin Nutr*. 1991;54:578–585.
105. Shepherd R, Cooksley EGE, Donvill Cooke WD. Improved growth and clinical, nutritional and respiratory changes in response to nutritional therapy in cystic fibrosis. *J Pediatr*. 1980;97:351–357.
106. Levy LD, Durie PR, Pencharz PB, Corey ML. Effects of long-term nutritional rehabilitation on body composition and clinical status in malnourished children and

adolescents with cystic fibrosis. *J Pediatr*. 1985;107:225–230.
107. Boland MP, Stocki DS, Macdonald NE, et al. Chronic jejunostomy feeding with a non-elemental formula in undernourished patients with cystic fibrosis. *Lancet*. 1986;i:232–234.
108. Shepherd RW, Holt TL, et al. Nutritional rehabilitation in cystic fibrosis: controlled studies of the effects on nutritional growth retardation, body protein turnover and cause of pulmonary disease. *J Pediatr*. 1986;109:788–794.
109. Gunnell S, Christensen NK, McDonald C, Jackson D. Attitudes towards percutaneous endoscopic gastrostomy placement in cystic fibrosis patients. *J Pediatr Gastroenterol Nutr*. 2005;40:334–338.

Acute and Chronic Liver Disease

56

Deirdre A. Kelly, MD, FRCP, FRCPI, FRCPCH, Susan Protheroe, MD, MRCP, FRCPCH, and Sara Clarke, BSc (Hons)

The liver has a central role in energy metabolism, nutritional homeostasis, and absorption of nutrients. Severe liver disease, whether acute or chronic, leads to multiorgan failure, which can have significant effects on growth and development in the long term. The pathophysiology of malnutrition in liver disease is complex and multifactorial and has extensive implications (Table 56-1). It is most severe in infants with chronic cholestatic liver disease, who are particularly vulnerable to the effects of malnutrition because of their high energy and growth requirements.[1] Insufficient dietary intake is often a crucial factor that can be rectified. This chapter reviews the pathophysiology of malnutrition in pediatric liver disease and current treatment strategies, including liver transplantation.

PATHOPHYSIOLOGY OF MALNUTRITION IN LIVER DISEASE

Prevalence of Malnutrition

Malnutrition is common in children with chronic liver disease. Estimates vary from 50% to 80% in studies from North America and Europe.[1–4] In general, children who present with acute liver disease such as viral hepatitis or fulminant hepatitis are not malnourished at presentation but

can become so as a result of their illness.[5]

TABLE 56-1 Pathophysiology of Malnutrition in Liver Disease

Reduced calorie intake
Anorexia
Fat malabsorption
Use of bile salt resins
Portal hypertension
Unpalatable feeds
Inappropriate substrate use
Abnormal nitrogen metabolism
Reduction in glycogen stores
Negative protein balance
Increased metabolic needs
Energy expenditure
Calorie requirements
Hormonal dysregulation GH/IGF-I
Insulin resistance

Protein energy malnutrition (PEM) used to be inevitable in chronic liver disease, particularly in the developing infant. Severe malnutrition (weight and/or height less than 2 standard deviations below the mean) with loss of fat stores and muscle wasting used to affect 60% of infants with liver disease[1]; but with modern management with early referral, diagnosis, and intensive nutrition, less children in the developed world develop this problem today. Both morbidity and mortality post liver transplantation are related to the degree of pretransplant malnutrition, and thus nutritional status is an important risk factor for survival.[1]

Reduced Energy Intake

Anorexia is a common symptom in children with either acute or chronic liver disease. Due to ill health or anorexia, children may take less than the recommended requirements or less than is appropriate for their increased energy requirements due to excess energy expenditure.[6] The consequent reduction in enteral intake may lead to considerable loss of body stores or lean body mass, which alters body composition as determined by anthropometry.[7]

In children with chronic liver disease, the complications of portal hypertension, such as ascites and hepatosplenomegaly, can lead to malabsorption of nutrients. The treatment of ascites, which includes fluid restriction, prescription of unpalatable feeds, or both, can exacerbate

anorexia and reduce energy intake further.[2]

Carbohydrate Metabolism

Under normal circumstances, the liver receives portal vein blood rich in absorbed glucose, which can be stored in the liver as glycogen or circulated to extrahepatic tissues, especially muscle, where lactate, pyruvate, and alanine are generated by glycolysis.[8,9] In children with liver disease, this substrate supply and use can be abnormal. The loss of glycogen stores in chronic liver disease leads to fasting hypoglycemia and an inability to meet energy demands. In addition, any significant hepatocyte loss, especially in acute liver failure,[10] has an immediate effect on glucose metabolism, leading to hypoglycemia.

In cirrhotic adults, the pancreas secretes increased amounts of insulin, which is not metabolized, either because of portosystemic shunting around the liver or because of reduced degradation owing to hepatic dysfunction[11] or reduced glucose uptake by hepatocytes.[12] Although there is increased circulation of insulin, glucose uptake in the peripheral tissues, especially muscles, is reduced as a result of insulin resistance. Adult studies suggest that this could be attributable to impaired glycogen synthesis in the muscle, because of either abnormal insulin receptor numbers or affinity.[13,14] These data have not been confirmed in children; but it is possible that similar mechanisms occur, although a few children (apart from those with cystic fibrosis) develop overt diabetes mellitus unlike adults.[13]

Protein Metabolism

Amino acids are absorbed by the intestine directly into the portal vein and transferred to the liver, where they are synthesized into protein or used for energy. The liver is responsible for approximately 10% of plasma protein synthesis; thus, amino acids are constantly recycled.[9,15] Nonessential amino acids are oxidized in both liver and muscle. The seven essential aromatic amino acids (AAAs) (arginine, histidine, lysine, methionine, phenylalanine, tryptophan, and threonine) are metabolized in the liver, whereas the three essential branched-chain amino acids (BCAAs) (leucine, isoleucine, and valine) are metabolized predominantly in muscle and pass unaltered through the liver to the periphery, where their uptake is regulated by insulin.[16] The liver detoxifies nitrogenous wastes via the urea cycle, leading to the production of ammonia—hence the rise of plasma ammonia in both acute and chronic liver failure.[17]

Reduced hepatic and muscle glycogen stores lead to early recruitment of fat and increased dependence on amino acids as an alternative fuel.[18] Abnormal protein use by the liver leads to a rise in AAAs and a reduction in BCAAs, which are metabolized in muscle in both children and adults.[19–21] An abnormal ratio of BCAAs to AAAs correlates with histologic damage and encephalopathy in adults[22] and children.[20,23]

Studies in adults demonstrating abnormalities in BCAA metabolism using studies of BCAA fluxes and protein turnover have given conflicting results. It is possible that the hyperinsulinemia detected in cirrhosis could cause decreased BCAA levels by increasing BCAA uptake in muscle or adipose tissue. Alternatively, it is possible that there is an increase in liver transamination of BCAAs in addition to abnormal protein turnover.[24] Three adult studies demonstrated that protein synthesis can be increased or decreased depending on clinical condition and that muscle wasting is attributable to a relative increase in protein breakdown.[25–27] Those patients who had more efficient protein retention were less malnourished.

A study of protein metabolism in infants with liver disease, which used a whole-body leucine turnover model, demonstrated that muscle protein degradation and protein oxidation were increased, possibly as a result of a reduction in carbohydrate metabolism and the use of protein as an energy supply.[27] In contrast to normal children, muscle protein degradation continued in these children even when they were fed, suggesting that this could be a factor in the muscle loss common in infants with chronic liver disease. A study in cholestatic children also documented increased leucine oxidation in the postabsorptive state but not in the fed state, but these children had less severe liver disease than in the aforementioned study.[23]

In addition to protein synthesis and degradation, the liver is the site of synthesis of all of the coagulation factors except factor VIII, von Willebrand's factor, which is synthesized by vascular endothelial cells.[28–30] Thus, coagulopathy is an early sign of significant acute or chronic liver failure.

In chronic liver disease, these metabolic changes result in muscle wasting, hyperammonemia, hypoalbuminemia, hypoglycemia, hypolipidemia, and reduced circulating triglyceride levels secondary to increased fat oxidation. Hypoalbuminemia is associated with a reduction in plasma oncotic pressure, exacerbating the development of ascites and peripheral edema. In acute liver failure, the main effects of deranged protein metabolism are hyperammonemia and abnormal coagulation.[30]

Fat Metabolism

Most dietary fat is in the form of long-chain triglycerides (LCTs), which are a high-energy source.[31] The first step in fat digestion is emulsification in the stomach; it is followed by hydrolysis of triglycerides by pancreatic lipase in the intestinal lumen and then micellar solubilization of di- and monoglycerides by bile acids, which are then transported into enterocytes. Once they are in the enterocytes, fatty acids are reesterified and chylomicrons are formed and removed via the lymphatics through the portal system to the liver and other tissues.[31]

In contrast, medium-chain triglycerides (MCTs) do not depend on micellar solubilization for absorption and can be transferred directly from the enterocytes to the portal circulation without reesterification.[31,32] In the liver, free fatty acids are metabolized into triglycerides or oxidized for energy. The very-low-density lipoproteins and high-density lipoproteins are synthesized in the liver, as is cholesterol, which is the precursor for many hormones, vitamins, and bile acids.

In all forms of chronic liver disease, there is reduction in the synthesis and secretion of bile salts, although this is more severe in cholestatic diseases such as biliary atresia and in children with progressive familial cholestasis who have a bile salt transport defect. Up to 50% of LCTs, fat-soluble vitamins, and essential polyunsaturated fatty acids (PUFAs) might not be absorbed because of reduced biliary secretion and reduction in intraluminal bile concentration.[33–35] In contrast, 95% of water-soluble lipids, such as MCTs, which do not depend on bile solubility, are absorbed even in cholestatic infants[36] and form the basis for nutritional replacement.[37]

Fat malabsorption can also be affected by portal hypertension, which leads to congested gastric and intestinal mucosa, and by small bowel overgrowth in the Roux-en-Y blind loop created by a Kasai portoenterostomy. In addition, therapies such as cholestyramine to reduce pruritus can exacerbate steatorrhea because it binds bile salts, decreasing micellar solubilization.

Pancreatic function is usually normal except in children with Alagille syndrome, in whom pancreatic lipase can be reduced.[36,38,39]

Long-Chain Polyunsaturated Fatty Acids

Long-chain polyunsaturated fatty acids (LCPs) such as arachidonic acid and docosahexaenoic acid (DHA) are essential nutrients in neonates and infants. LCPs, in particular DHA, play a major role in the development of

visual acuity and mental development in the first year of life, particularly in preterm infants.[40–42] The main source of LCPs is maternal, in the last trimester of pregnancy and through breast-feeding, because breast milk is a rich source of LCPs and contains both arachidonic acid and DHA in a combination of phospholipid and triglyceride forms.

Studies in adults have demonstrated that LCP deficiency can develop either from malabsorption of long-chain fat and essential fatty acids or from reduced synthesis or function of the A-6-desaturase enzyme required for the de novo synthesis of very-long-chain[20–22] carbon PUFAs from their essential fatty acid 18-carbon dietary precursors.[43,44]

A recent study that compared plasma and liver levels of very-long-chain PUFAs in adults undergoing liver transplantation with controls undergoing hepatic resection demonstrated low plasma but normal liver levels in cirrhotic adults, suggesting that tissue stores were not affected.[45] It is unclear how this would compare with infants who are growing rapidly, and certainly deficiencies have been described in pretransplant infants.[46]

Children with cholestatic liver disease have normal LCP and DHA levels at birth but can become deficient within 8–12 weeks[47] from malabsorption of LCTs, from prescription of formula feeds rich in MCTs, or as a result of inadequate liver desaturase enzyme activity.[43,48,49]

Fat-Soluble Vitamin Deficiency

Chronic liver disease affects vitamin absorption, metabolism, and storage. Reduction in bile salt secretion leads to malabsorption of the fat-soluble vitamins A, D, E, and K. Fat-soluble vitamin deficiency can develop within 6–12 weeks of birth, depending on body stores and availability of vitamin supplementation. Vitamin A deficiency can also develop secondary to reduction in protein synthesis or depletion of hepatic stores. Vitamin D deficiency can occur either from fat malabsorption or from reduction in hepatic 25-hydroxylation. Vitamin K deficiency arises partly from fat malabsorption and partly from a reduction in intake, particularly in breast-fed infants. Deficiency in all fat-soluble vitamins is as high as 30% despite supplementation in severely cholestatic children.[50,51]

Because the liver has a central role in lipoprotein metabolism and cholesterol synthesis, hypercholesterolemia and hypertriglyceridemia are common in chronic liver disease. There can be increased synthesis of cholesterol esters as a result of loss of hepatic lecithin cholesterol acyltransferase, which can alter lipoprotein fractions. In cholestatic liver

disease such as Alagille syndrome, unusual lipoproteins, such as lipoprotein X, are produced with cutaneous xanthomata and hemolysis from alterations in red cell and platelet membranes.[52]

In terminal liver disease, however, the reduction in cholesterol synthesis leads to hypocholesterolemia, which is a poor prognostic sign, now usually seen only in the developing world.[53]

Endocrine Dysfunction

Growth failure in chronic liver disease can be exacerbated by an impaired growth hormone (GH)/insulin-like growth factor (IGF-I) axis[54,55] because IGF-I and its major circulating binding protein, IGF-BP3, are synthesized in the liver. It seems that IGF-I is nutritionally regulated[56] because there is animal evidence that nutrients influence synthesis and action of IGF-I and its binding proteins at many levels, including defects in pre- and posttranslational expression. Thus, protein malnutrition not only decreases the IGF-I production rate but also can increase serum clearance and degradation. Evaluation of GH, GH-binding proteins, and IGF-I in children with protein-energy malnutrition before and after nutritional rehabilitation has confirmed the role of malnutrition in these hormones.[57]

Children with chronic liver disease have GH resistance due to downregulation of GH receptor expression resulting in low IGF-I levels[58–64], despite elevated levels of GH, which could be a result of diminished hepatic synthesis or malnutrition or related to end-organ insensitivity to IGF-I.[60,63,64] Additional reductions (50%-68%) in IGF binding protein 3 (IGF BP3) and acid-label subunit (ALS), as seen in adult cirrhosis,[65] are likely but have not been investigated in children. Reductions in these transport proteins lead to rapid clearance of IGF-I from the circulation resulting in growth failure.

Increased Energy Expenditure

Adult studies have produced conflicting data on energy expenditure, but the situation is different in a growing child. A number of studies have indicated an increase in energy requirements of up to 140% in children with both acute and chronic liver disease.[66–68] Mechanisms implicated include portosystemic shunting and ascites; abnormal intermediate metabolism; and energy demands of specific complications, such as sepsis and variceal hemorrhage. A study has confirmed that children with end-stage liver disease have a hypermetabolic state with increased metabolic activity in the body cell mass with excess lipid oxidation during fasting

and at rest, once again confirming the necessity for appropriate nutrition and the avoidance of prolonged fasting in these children.[69] In contrast, children with stable liver disease such as Alagille syndrome did not have increased energy expenditure, suggesting that abnormal energy expenditure is related to severity of liver disease.[70] Children with acute liver failure also have excess energy expenditure and requirements because of multiorgan failure and may also develop electrolyte loss such as hypomagnesmia.

Trace Elements and Metals

Biochemical deficiencies of water-soluble vitamins such as thiamin and pyridoxine can occur and lead to nutritional cardiomyopathy and peripheral neuropathy.[71] Trace metal deficiencies include iron deficiency secondary to gastrointestinal bleeding or diminished intake and zinc and selenium deficiencies caused by reduced enteral intake, malabsorption, or increased losses.[72]

Consequences of Malnutrition

Many different nutrient deficiencies occur in children with chronic liver disease (Table 56-2), and malnutrition per se can increase liver dysfunction because of the energy required for synthesis, storage, and detoxification. Children with progressive cholestasis, such as those with biliary atresia or Alagille syndrome, develop significant fat malabsorption, which leads to steatorrhea, fat-soluble vitamin deficiency, essential fatty acid deficiency, loss of fat stores, and a reduction in growth[51,73,74] (Tables 56-2 and 56-3). Essential fatty acid deficiency can present with a rash,[48] whereas DHA deficiency is associated with abnormalities in visual function, as demonstrated by electroretinograms in cholestatic infants.[47]

TABLE 56-2 Clinical Manifestations of Malnutrition in Liver Disease

Nutritional Deficit	Clinical Manifestation
Protein-energy malnutrition	Stunting, muscle wasting, motor, developmental delay
Fat malabsorption	Steatorrhea, loss of fat stores
Essential fatty acid deficiency	Peeling rash
Vitamin A deficiency	Conjunctival and corneal drying, night blindness

Vitamin E deficiency	Peripheral neuropathy, ophthalmoplegia, ataxia, hemolysis
Vitamin D deficiency	Osteopenia, rickets, fractures, reduced bone density
Vitamin K deficiency	Bruising, epistaxis, coagulopathy
Zinc deficiency	Acrodermatitis, anorexia, poor growth
Hypercholesterolemia	Xanthomata
Immunosuppression	Systemic infections secondary to reduced cell-mediated immunity

TABLE 56-3 Anthropometric Definitions in Children with Liver Disease

Acute malnutrition	Decreased weight for height Decreased triceps skinfold or midarm muscle area
Chronic malnutrition	Decreased height for age
Severe malnutrition	<2 SDSs below the mean <3rd percentile

Abbreviation: SDS, standard deviation score (z-score), 0 = 50th percentile.

Fat-soluble vitamin deficiency is more common and can be detected biochemically before clinical symptoms, or severe deficiencies.[51,73,74] Clinical signs and symptoms of vitamin A deficiency are rare but include night blindness, xerophthalmia, and keratomalacia.[75] Vitamin D deficiency leads to hypocalcemia, hypophosphatemia, rickets, and pathologic fractures, particularly in infants who are rarely exposed to sunlight or who have inherited metabolic liver disease and a renal tubular disorder.[63] Deficiency of vitamin E leads to hemolysis, peripheral neuropathy, and occasionally visual loss.[76,77] Vitamin K deficiency can present as hemorrhagic disease of the newborn, particularly in breast-fed babies who are given insufficient vitamin K at birth.[78] It leads to coagulopathy, which is exacerbated by decreased synthesis of liver-dependent clotting factors. In vitamin K deficiency secondary to fat malabsorption, parenteral vitamin K will improve the coagulation profile, whereas it will be ineffective in parenchymal liver disease.

Metabolic bone disease with reduced bone density is usually observed in end-stage liver disease[63] and has been related to fat absorption rather than dietary intake.[79] Although malabsorption of vitamin D is a factor, the etiology is more complex as normal levels of vitamin D do not prevent bone demineralization.[63] Children with liver disease have a higher incidence of dental caries and enamel hypoplastic defects, which could be

related to nutritional deficiency or to bone disease. They also have discolored teeth as a result of hyperbilirubinemia.[80]

Trace element and mineral deficiencies include iron deficiency with anemia and zinc deficiency, which leads to acrodermatitis, immunodeficiency, and altered protein metabolism; both zinc and selenium deficiencies can exacerbate growth failure and poor protein synthesis.[1,72] Recently, the development of scurvy was reported in a child awaiting combined liver and kidney transplantation.[81]

In the course of chronic liver failure, fat malnutrition develops first, as demonstrated by the loss of fat stores. Protein malnutrition is a late development and is associated with a reduction in muscle bulk, stunting, and significant motor developmental delay. In time, children with significant malnutrition will have impaired growth and psychosocial development[82–84]; thus, malnutrition is an important indication for liver transplantation Severe malnutrition has a significant effect on mortality and morbidity and correlates with prolonged hospital stay[85] and is one of the most important prognostic factors[83] for survival post liver transplantation.[4,83,86,87]

NUTRITIONAL ASSESSMENT

Accurate nutritional assessment is essential to the management of children with acute or chronic liver disease. It is important to start with a comprehensive clinical history, feeding history, and careful physical examination. Serial anthropometric examination is critical and can identify early malnutrition. Standard weight and height ratios are of little value in children with liver disease because of misinterpretation caused by fluid overload, ascites, and visceromegaly. Many researchers, using sophisticated methods such as whole-body potassium and dual-energy X-ray absorptiometry (DXA) scanning, have demonstrated that body weight underestimated the incidence of malnutrition by 50% in both adults and children[2] and, for this reason, simple methods of measuring body composition, such as body impedance analysis, are not reliable in children with liver disease.[87,88]

Thus, the assessment of malnutrition should be performed using a number of parameters, such as triceps or subscapular skinfolds, midarm circumference, and arm muscle measurements (midarm muscle area).[7,89] Triceps, skinfolds, and midarm circumference are useful indicators of body fat and protein and serial recordings demonstrate early loss of fat

stores before weight and height changes become obvious.[7,89] Although linear growth is a sensitive parameter, it is a late sign of growth failure in infancy; stunting (or negative height velocity) might not be apparent until 1 year of age (Table 56-3).[90]

A number of evidence-based tools are under evaluation to assess nutrition in seriously ill children and may be of future relevance for liver disease.[91]

Growth data are best expressed as standard deviation scores (or z-scores) related to the median value for the child's age and sex in which the z-score of 0 = 50th percentile. This is particularly useful for comparisons between centers and for evaluation of new feeds.

Biochemical evaluation of vitamin deficiency is a useful adjunct to nutritional assessment. Plasma vitamin A levels can be measured but might not reflect hepatic stores.[92] Vitamin E deficiency is monitored by serum vitamin E levels or the ratio of vitamin E to total lipids. Vitamin D deficiency is evaluated by measuring serum levels of vitamin D, parathyroid hormone, calcium, phosphate, and alkaline phosphatase, whereas the diagnosis of rickets is confirmed by radiographic examination of the wrist or knee.[63] Vitamin K deficiency is identified by measuring coagulation times and monitoring the response to parenteral vitamin K, but a more accurate assessment can be obtained by measuring plasma PIVKA-II (protein induced in vitamin K absence).[50]

Acute malnutrition is evidenced by loss of weight compared to height and loss of fat stores, whereas chronic malnutrition is documented by stunting and reduction in the height-for-age index (Table 56-3).

INDICATIONS FOR NUTRITIONAL THERAPY

The aim of nutritional therapy is to prevent or treat malnutrition by providing adequate calories for energy and sufficient nitrogen for protein synthesis, restore plasma amino acid imbalance, prevent vitamin and trace element deficiency, and achieve normal growth and activity. The need for nutritional support is often underestimated particularly in infants with liver disease, who might have an increased appetite in the first few months of life.

Children who are at particular risk for developing malnutrition include the following:

- Children under 2 years of age who have severe cholestasis (serum bilirubin > 70 mmol/L [4 mg/dL])

- Those with progressive liver disease such as biliary atresia or severe familial intrahepatic cholestasis
- Those with end-stage liver failure awaiting liver transplantation
- Children with recurrent hepatic complications such as ascites and bleeding varices

STRATEGIES FOR NUTRITIONAL SUPPORT

Increased energy intake: because the resting energy requirements of infants and children with liver disease are increased,[66] it is important to increase the energy intake to 140%–200% of estimated average requirements (EARs). Cholestatic infants in the early stages of disease will compensate for malabsorption and increased energy requirements by increasing their intake, often consuming 120%–200% more formula than the recommended intake for age. In ongoing cholestasis, energy intakes often fall due to associated symptoms of progressive liver disease and strategies to increase the nutrient density of feeds must be employed. This can be achieved by using recently available ready to feed nutrient-dense MCT-containing formula (Infatrini Peptisorb, Nutricia Advanced Medical Nutrition), which provides enhanced protein and energy intakes (100 kcal/100 mL, 10% energy from protein). Alternatively, where this formula is unavailable MCT-containing formula can be concentrated from 13% standard dilution to 15%–19% dilution. This increases the energy density from 67 to 80-100 kcal/100 mL and protein from 1.9 g/100 mL to 2.24–2.84 g/protein/100 mL. Concentrating infant formula feeds increases the intake of all nutrients and maintains the delicate balance between energy, protein, and micronutrients. The practice of concentrating formula should only be undertaken by an experienced pediatric dietitian who can ensure appropriate monitoring of intake and growth (Table 56-4). There are many high-calorie, nutrient-dense supplement drinks available (Fortini—Nutricia Advanced Medical Nutrition, Paediasure Plus, and Abbott Nutrition), some containing MCT fat (Paediasure Pepdite—Abbott Nutrition, Peptamen Vanilla Bottle—Nestle Healthcare Nutrition), which can be effective for older children to enhance nutritional intake; however, compliance in taking these supplements over the long term is poor. If there is no response to an increase in energy intake alone, enteral feeding by nasogastric tube as either daytime bolus feeds or continuous overnight feeds is beneficial (Figure 56-1).

MCTs: because fat is an important calorie source, with an energy value of 8 to 9 kcal/g, it is a major source of energy for infants. Because

absorption is deranged in liver disease, the fat content of the diet needs careful consideration. Low-fat diets are no longer considered appropriate, but an increase in MCT content will reduce steatorrhea.[34,37,48] For infants, a whole protein formula such as Caprilon (SHS Nutrition) containing 75% fat as MCT or hydrolyzed-protein infant formulas, such as Peptijunior (Cow & Gate), Pregestimil (Mead Johnson), or Infatrini Peptisorb, (Nutricia Advanced Medical Nutrition), which contain 50% MCTs, will maximize fat absorption and improve steatorrhea,[48] but formulas with more than 80% MCTs can lead to essential fatty acid deficiency.[48] Although MCTs are a useful energy source, there is no evidence that growth improves with formulas having a high MCT content,[93,94] so it is necessary to increase the total fat intake with both LCTs and MCTs. This will increase the overall amount of fat absorbed and improve growth despite increasing steatorrhea.[37] In older children, MCT oil or emulsions (Liquigen—SHS Nutrition) can be added to meals and should be balanced by fats with high LCP content.

LCPs: the malabsorption of long-chain fats in chronic liver disease means that it is important to ensure adequate intake of LCPs. The minimal intake of linoleic acid recommended for infants is 1% to 2% of energy in a ratio of linoleic to linolenic acid of 5:15.1.[41]

TABLE 56-4 Formulas for Children with Liver Disease[a]

Formula	kcal/kj	Protein (g)	Fat (% MCT)	Sodium (mmol)	Essential Fatty Acids (omega-6: omega-3)
Pregestimil[b]	68/285	1.9	55	1.26	10:1
Peptijunior[c]	67/280	1.8	50	0.9	7:1
Caprilon[d]	66/276	1.5	75	0.8	7.5:1
Generaid Plus[d]	102/428	2.4	32%	0.7	4:1
Infatrini Peptisorb	100/415	2.6	50%	1.1	418.1
Modular feed[d]	Flexible	Flexible	0–100	Flexible	None

[a] Per 100 mL.
[b] Mead Johnson.
[c] Cow & Gate.
[d] Scientific hospital supplies.

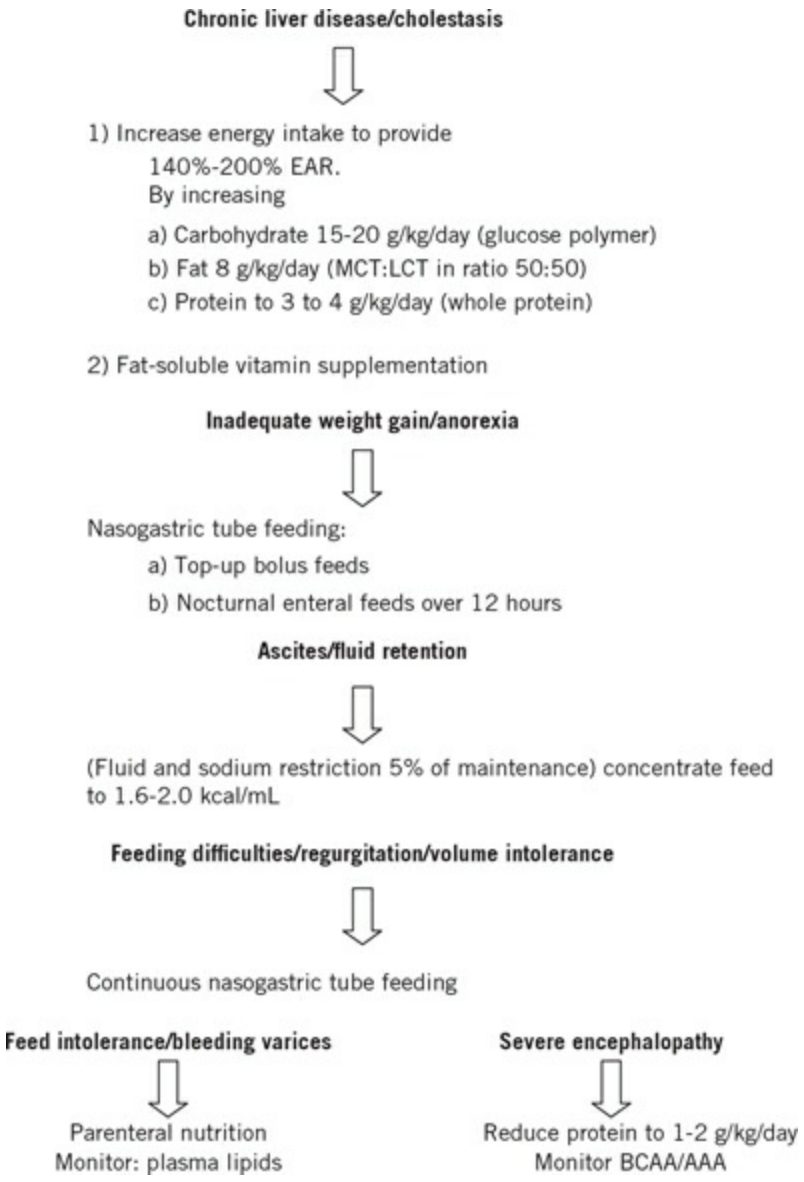


Figure 56-1 Nutritional support for chronic liver disease.

The LCPs, or PUFAs, can be supplemented by the addition of walnut oil to provide 1% to 2% of total energy, whereas LCP supplementation is provided in dietary products such as egg yolk (which is rich in arachidonic acid) or fish oil (which is rich in DHA). Alternatively, infants can be given conventional LCP-supplemented formulas, which are commercially available.[95]

Structured lipids: recently, chemically defined structured lipids have been developed to increase absorption of both medium-chain and long-

chain and essential fatty acids.[96] These lipids combine pure MCTs with LCTs, resulting in a triglyceride that contains combinations of short-, medium-, and long-chain fatty acids on a single glycerol backbone, which should be absorbed like MCTs. To date, clinical studies in adults have evaluated structured lipids in postoperative patients receiving parenteral nutrition and demonstrated that they are safe and effective compared to LCP emulsion.[97] Although clinical studies of these modified lipids in children are currently in the preliminary stages,[98] studies in rats have demonstrated improved fat absorption and reversal of essential fatty acid deficiency in vitro in caco-2 cells.[99,100] If successful, there might be considerable benefit for cholestatic infants.

Carbohydrate: carbohydrate is a major source of energy and is particularly useful for increasing calorie intake. It can be given as a monomer, a short-chain polymer, or starch, but complex carbohydrates such as maltodextrin or glucose polymer restrict the osmolality of feed while maintaining a high energy density of more than 1 kcal/mL. This allows fluid restriction if necessary while providing up to 20 g/kg/day of carbohydrate. In infants, glucose polymers are best added to milk feeds; in older children, they can be added to drinks, mixed in with foods, or given as a supplemental drink.

PROTEIN INTAKE

Historical advice, which restricted protein in advanced end-stage liver disease, is now considered inappropriate in both children and adults.[101] Children with end-stage liver disease require minimal protein intakes of approximately 2 to 3 g/kg/day but will tolerate up to 4 g/kg/day without developing encephalopathy or a significant increase in plasma amino abnormalities.[102] Severe protein restriction (less than 2 g/kg/day) might be required for acute, severe encephalopathy but should be avoided in the long term as it can lead to endogenous muscle protein consumption.[66] There is no necessity to use semielemental diets or protein hydrolysates because there is no evidence of protein malabsorption.

In view of the abnormal aromatic acid to BCAA ratio, there has been considerable interest in the use of a modified amino acid formulation designed to improve this imbalance. Numerous studies in adults have not demonstrated a clear benefit for BCAA nutritional supplementation, despite the evidence that BCAAs did improve nitrogen retention and protein synthesis; their effect on patient outcome was not proved.[103]

The necessity for BCAA supplementation may persist post transplantation, as demonstrated in a recent study in five clinically stable children, although the requirement was less than before transplantation.[104]

There is both animal and clinical evidence that suggests that BCAA-enriched formulas can have significant nutritional benefit in children. BCAAs increased weight gain, protein and muscle mass, body composition, and bone mineral density in an animal model of biliary obstruction.[105] A study that compared a feed containing 32% BCAAs with a standard feed demonstrated improved lean body mass in children awaiting liver transplantation but no improvement in amino acid levels.[106] In another study in infants with liver disease, the effect of a modified amino acid feed containing 50% BCAAs was compared with an isonitrogenous formula containing 22% BCAAs by measuring whole-body protein turnover.[67] The BCAA-supplemented feed improved protein retention compared with the standard formula by suppressing endogenous protein catabolism and normalizing the plasma amino acid profile. Formulas rich in BCAAs, complete with MCTs and vitamin and mineral supplements, have been developed for use in infants, and oral supplements rich in BCAAs are available for older children, but both are particularly unpalatable.

VITAMIN AND MINERAL SUPPLEMENTATION

Both water- and fat-soluble vitamins should be supplemented in children with chronic liver disease. Supplementation should be based on plasma levels of selenium, zinc, calcium, and magnesium. Iron supplementation might be required in children with chronic blood loss.

Fat-soluble vitamins are required in all children with prolonged or cholestatic liver disease (Table 56-5). Most children will be maintained adequately on oral fat-soluble vitamin administration, but monthly intramuscular administration is occasionally required for children with severe cholestasis.

MODE OF DELIVERY

Nutritional supplementation should be provided enterally for as long as possible because it has a number of advantages over parenteral feeding. It not only is cheaper and more physiologic but also maintains gastrointestinal tract immunity, reduces bacterial overgrowth, and maintains the integrity of the gut barrier to microorganisms. It is also

essential to maintain normal feeding behavior. Many children with progressive liver disease will not be able to consume the prescribed increase in nutritional intake necessary to prevent or reverse malnutrition orally; thus, early nasogastric tube feeding is indicated for children who have a poor response to an increase in nutritional intake or who become anorexic.

Despite anxiety about using nasogastric feeding in children with esophageal varices, there is no evidence that the use of modern soft Silastic™ nasogastric tubes is harmful. Nocturnal feeding is the method of choice because it allows nutritional supplementation while supporting normal oral feeding during the day. For infants with severe liver disease, nocturnal feeding can be beneficial because it will prevent fasting hypoglycemia and reduce protein catabolism. Continuous enteral infusion might be required in children with severe malabsorption or feed intolerance. Intensive enteral nasogastric feeding has effectively reversed malnutrition in infants and children with liver disease, and it can relieve parental anxiety and may induce a transformation in the child's affect and increase voluntary intake.[37,102,107] Under normal circumstances, it should be possible to provide nocturnal or continuous nasogastric feeding at home with appropriate support from a nutritional care team including a dietitian, a liaison nurse, pediatric community nurses, and clinicians.

Gastrostomy tube placements might be preferable in older children but should be avoided in children with severe portal hypertension because of the development of stomal varices. In children with stable compensated liver disease (e.g., cystic fibrosis), percutaneous endoscopic gastrostomy might be beneficial.

GROWTH HORMONE THERAPY

In view of the disruption of the GH/IGF-I axis, it is tempting to prescribe GH therapy for children with significant growth failure, but this has not been proved beneficial in children with end-stage liver disease.[60,63]

PARENTERAL NUTRITION

In view of the well-recognized hepatobiliary complications that occur in premature infants with short-gut syndrome or intestinal failure, parenteral nutrition has been used cautiously in children with liver disease. Particular concern has been focused on whether the potentially toxic components of parenteral nutrition could be exacerbated in children with established

chronic liver disease or cholestasis. There is no evidence that amino acids, carbohydrates, or lipid emulsions are any more toxic in children with established liver disease, although amino acid levels and plasma lipids need careful monitoring.

Although aluminum toxicity is well recognized in parenteral nutrition and can lead to bone disease, there is no evidence that it is implicated in parenteral nutrition liver disease or that it exacerbates the metabolic bone disease associated with chronic liver disease.[108] In contrast, chromium toxicity has been reported in animals on parenteral nutrition. Although both serum and urine chromium levels are higher in children on long-term parenteral nutrition, there is no correlation with liver disease.[109]

However, manganese toxicity in children on long-term parenteral nutrition has recently been reported.[110,111] Although 79% of children on long-term parenteral nutrition had whole-blood manganese concentrations above the reference range, children with impaired liver function had the highest manganese levels. Although manganese is toxic to both brain and liver, it is not clear whether the high manganese levels led to cholestasis or were secondary to it because manganese is excreted in the bile. Nevertheless, it is likely that manganese toxicity exacerbates cholestasis in children with established liver disease; thus, monitoring of manganese levels is particularly important in this group.

TABLE 56-5 Assessment and Management of Nutritional Deficiency in Pediatric Liver Disease

Deficit	Investigation	Management
Energy	Calorie intake, energy expenditure, anthropometry (body cell mass [TBK], DXA)	Increase calorie intake, achieve 130%–150% EAR, nocturnal enteral nutrition, continuous enteral nutrition
Protein	Plasma proteins (albumin), BCAA/AAA ratio, protein stores (muscle mass, TBN)	Provide adequate protein, (3 to 4 g/kg/d), BCAA-enriched protein (32%), albumin infusion (if serum albumin < 25 g/L)
Fat	Triceps skinfolds, body composition, EFA deficiency, plasma lipid profile	Improve fat absorption (MCT:LCT, 50:50), provide saturated fats high in EFA, ? supplement DHA
Fat-	Plasma 25-OH-D (skeletal	Light exposure, vitamin D-1-a (50

soluble Vitamins	X-rays, DXA), prothrombin time ([<138 c] INR = 1), plasma vitamins E and A	ng/kg), vitamin K (2.5–5 mg/d), vitamin E (50–400 IU/day as TPGS), vitamin A (5,000–10,000 IU/d)
Water-soluble count vitamins	Specific levels, blood	Supplement as requested
Minerals	Specific levels, cardiac evaluation	Supplement as requested

Abbreviations: EFA, essential fatty acid; INR, international normalized ratio; LCT, long-chain triglyceride; MCT, medium-chain triglyceride; TBK, total body potassium; TBN, total body nitrogen; TPGS, tocopherol polyethylene glycol-1000 succinate.

In general, the use of parenteral nutrition in children with established liver disease is for short-term purposes only; thus, they are unlikely to develop the complications seen in children on long-term parenteral nutrition,[112] although prescription of ursodeoxycholic acid (15–20 mg/kg) might be of value.[113]

Parenteral nutrition should be considered in children with chronic liver disease only if they cannot be enterally fed because of feed intolerance or secondary to complications such as recurrent variceal bleeding or abdominal sepsis. As stated earlier, there is no evidence that short-term parenteral nutrition is associated with hepatobiliary dysfunction or an increase in cholestasis; it can improve nutritional status in the short term. Standard amino acid and lipid solutions are well tolerated in stable patients, and lipids can be particularly beneficial in achieving adequate calorie intake. If encephalopathy develops, the amino acid content of the feed can be reduced to 1 to 2 g/kg/day; lipid administration requires careful monitoring in children with severe liver dysfunction, hepatic encephalopathy, or sepsis. Parenteral structural lipid emulsions have not yet been evaluated in children.

The use of parenteral nutrition is particularly important for children with acute fulminant hepatitis who are awaiting liver transplantation because they are hypercatabolic. Standard formulas should be used, although the volume needs to be restricted to 75% of maintenance and the concentration should be increased to maintain glucose levels (less than 4 mmol/L). There is no need to reduce protein content, particularly if the patient is electively ventilated. As indicated earlier, intravenous use of BCAAs for the treatment of hepatic encephalopathy or coma has not been evaluated in children, although they can be of value in adults.

BEHAVIORAL FEEDING PROBLEMS

Behavioral feeding problems are common in this group of children, particularly in infants who have had long-term nasogastric feeding. Feeding problems are compounded by the use of unpalatable feeds, medications, and excess parental anxiety about feeding and growth.

Strategies to prevent behavior problems include promoting daytime feeding to provide oral stimulation and encouraging children to experiment with flavors and textures appropriate to their age. The success of these techniques is dependent on a multidisciplinary approach involving a clinician, nurse specialist, dietitian, clinical psychologist, speech therapist, and play therapist.[37]

SPECIFIC DISEASE STATES

Acute Liver Disease

Most children with acute viral or autoimmune hepatitis require no nutritional supplementation, although recent studies indicate that their energy intake, body composition, and vitamin status may be altered.[68,73] Fulminant hepatic failure requires careful management of protein to prevent hepatic encephalopathy and reduce excess protein catabolism. Protein should be restricted only for short periods to treat severe encephalopathy (see earlier). Fluid volume can be reduced to prevent or treat cerebral edema; it is critical to maintain adequate glucose levels by infusing 6–8 mg/kg/min of glucose to prevent hypoglycemia. Enteral nutrition should be continued as long as the child is conscious and in the early stages of elective ventilation. Once the requirement for liver transplantation becomes obvious, parenteral nutrition may be required to provide sufficient calories to prevent additional protein catabolism should enteral feeding fail to be tolerated (see earlier).

Chronic Liver Disease

The majority of children with chronic liver disease will have cholestasis, from biliary atresia, Alagille syndrome, or familial intrahepatic cholestasis, and should be managed as indicated earlier.[3,114,115] Children who are not jaundiced require similar support, although their requirement for fat-soluble vitamin supplementation will be less.

Problems can arise with children who have ascites and fluid retention; fluid and sodium restriction might be required. In these circumstances when commercial feeds may not deliver appropriate nutrition, a modular

feeding system may be of benefit. The modular feeding system is extremely flexible in its composition and allows individual prescription of protein, sodium, and water to which complex carbohydrate polymers, fats, vitamins, and minerals are added to produce a highly energy-dense/nutrient-dense feed to suit the child's specific nutritional requirements.

Inborn Errors of Metabolism

A number of inborn errors of metabolism rely on dietary modification as part of therapy. These include galactosemia, hereditary fructose intolerance, and tyrosinemia type I.

Galactosemia is an autosomal recessive disorder in which there is a reduction in the enzyme activity of galactose-1-phosphate uridyl transferase, which prevents the conversion of galactose to glucose. Dietary treatment includes removing all products containing galactose (i.e., milk and milk products).[116]

Hereditary fructose intolerance is an autosomal recessive disorder caused by a deficiency or the absence of the enzyme aldolase B, which leads to cirrhosis and renal tubular disorders. Dietary management consists of elimination of fructose, sucrose, and sorbitol from the diet; this must be maintained for life. This means that all fruit and fruit juices are eliminated from the diet and only certain vegetables are permitted. Commercial products such as breads and cereals can also contain small amounts of sucrose, as can medications and toothpaste.[116]

Tyrosinemia type I is an autosomal recessive disorder caused by a defect in the enzyme fumarylacetoacetase, which leads to hypertrophic cardiomyopathy, cirrhosis, and renal tubular acidosis. For many years, dietary treatment was the only method of therapy and included restriction of phenylalanine, tyrosine, and methionine. Although restriction of these amino acids was effective in normalizing plasma amino acids, it did not prevent the progression of liver failure or the development of metabolic crises. The development of Nitosone (2-[2-nitro-4-trifluoromethylbenzoyl]-1,3-cyclohexenedione) in association with dietary treatment has altered the natural history and outcome of the disease.[117]

Glycogen Storage Disease

Hepatic glycogen storage disorders are a group of autosomal recessive inherited disorders that lead to defects in the metabolism of glycogen. Liver and skeletal muscle are usually affected, but heart, kidney, bones,

and brain can also be involved. The presentation and biochemical features depend on the precise enzyme deficiency. Nevertheless, the treatment is dietary: provide a continuous supply of exogenous glucose, either intravenously or enterally; maintain normal glucose; and suppress counterregulatory responses. In infants, this is best achieved by frequent daytime feedings or continuous nocturnal enteral glucose feeding. In older children, the oral feeding of uncooked cornstarch (1 to 2 g/kg every four hours), which is hydrolyzed in the gut to release glucose slowly, can reduce the need for frequent daytime feedings or continuous nocturnal feeding.[118] Glycogen storage disease type III requires similar treatment but with a higher protein intake (3 g/kg). In most instances, dietary management is successful and can prevent the long-term complications of these diseases.

PREPARATION FOR LIVER TRANSPLANTATION

Liver transplantation is indicated for both acute and chronic liver failure. Nutritional support is of particular importance for children with chronic liver failure because malnutrition is a significant risk factor for mortality and morbidity related to liver transplantation.[83,86,87]

Children awaiting liver transplantation require intensive nutritional support, depending on the etiology of their liver failure, as documented earlier, but the main principle is to provide a sufficient nutrient intake (Table 56-6).

TABLE 56-6 Nutritional Support in Children Before and After Liver Transplantation

Requirement	Preoperative	Postoperative
Carbohydrate	Glucose polymer 15–20	Glucose polymer 6–8 (g/kg/day)
Protein (g/kg/d)	Low-salt protein 3–4	Whole protein 2.5–3
Fat (g/kg/d)	50%–70% MCT 8	LCT 5–6
Energy intake	130%–150%	120% (EAR)

EFFECT OF LIVER TRANSPLANTATION ON NUTRITIONAL STATUS

Many recent studies have documented the nutritional rehabilitation achieved by successful liver transplantation.[4,90,119] Following liver

transplantation, surviving children experience a rapid return to normal midarm muscle area and midarm fat within three to six months.[92,119] Initial weight gain can be rapid because of the effects of corticosteroids on appetite and salt and water retention, but most children return to a normal weight once corticosteroids are reduced or discontinued. Linear growth is often delayed by 6–24 months depending on the rate of hepatic complications and the withdrawal or reduction of corticosteroids.[83]

Preoperative nutritional status has a strong influence on posttransplantation growth. Children who were less stunted at transplantation grew slowly initially but finally achieved normal growth velocity, whereas older children who were more stunted (height standard deviation score [SDS] > –2) grew more quickly initially post transplantation but did not achieve normal height.[120] There are no differences in catch-up growth between the sexes or with pretransplantation disease, apart from Alagille syndrome, which is associated with growth failure before and after transplantation in 50% of patients.[121,122]

Although successful liver transplantation reverses most aspects of malnutrition, it can take some months or years before bone density returns to normal. Persistent osteoporosis and bone fractures have been described at three to six months following transplantation[63] and were exacerbated by the effects of corticosteroid therapy.

Likewise, it may take some time before long-chain PUFA and BCAA metabolism return to normal[46,103] and these factors should be taken into consideration when planning posttransplant nutrition.

EFFECT OF GROWTH HORMONE AND IGF-I

As indicated earlier, both GH and IGF-I are necessary for normal growth, and it has been shown that plasma IGF-I levels are low in children with chronic liver disease despite elevated serum GH. Although posttransplantation IGF-I and IGF-BP3 rise significantly and IGF levels return to normal, abnormalities in the IGF-binding proteins BP1, BP2, and BP3 remain for some months.[55]

PREVENTION OF GROWTH FAILURE AFTER TRANSPLANTATION

The nutritional strategy for prevention of growth failure before and after

liver transplantation begins with intensive nutritional support preoperatively. Children who are malnourished before transplantation should be maintained with parenteral nutrition perioperatively until normal nutrition is resumed, whereas children with normal nutritional status should start enteral feeding with a rapid buildup of calorie intake 3–5 days following transplantation.

It is important to ensure sufficient energy intake (at least 120% of EAR) following the operation. This can be provided as a high-energy pediatric enteral feed, a high-energy infant formula, or a modular feed taken either orally or enterally via a nasogastric tube for infants and children who have poor oral intake. Nasogastric tube feeds alongside additional supplements of fat and carbohydrate may be required for some time to maintain growth while establishing normal oral intake. The energy intake should include 2.5 to 3 g protein/d and glucose polymer (6–8 g/kg/d), but fat can now be provided entirely as long-chain fat (5 to 6 g/kg/d) unless cholestasis secondary to chronic rejection is a complication (Table 56-6).[123]

Glucocorticoids, used to prevent organ rejection, interfere with GH excretion. As GH, IGF-I, IGF-BP1, IGF-BP2, and IGF-BP3 remain abnormal following transplantation, it is rational to consider whether GH therapy might be helpful. Recombinant human GH has been evaluated in eight growth-retarded children following liver transplantation.[124] The median growth rate increased from 3.2 to 7.1 cm/year (p = .025) and height SDSs increased from –2.9 to –3.1 (p = .036) during the first year of GH treatment. Both IGF-I and insulin IGF-BP3 levels increased significantly during treatment. There was no effect on graft function, and no child developed rejection or any other complication, but growth velocity was not maintained beyond the first year.

THE OBESITY EPIDEMIC

Survivors of liver transplantation are not immune from the effects of the obesity epidemic, and there are reports of obesity following all forms of solid organ transplantation.[125] Likewise, the development of nonalcoholic fatty liver disease is related to a number of inherited disorders, such as Bardet-Biedl, polycystic ovary disease, and obesity,[126] but a detailed description of this new emerging chronic disease in childhood is outside the scope of this chapter.

CONCLUSION

The essential role of liver in energy and growth has particular relevance for infants and children with end-stage liver failure. Considerable progress has been made in understanding the pathophysiology of malnutrition in liver disease, and this has improved our ability to support these children. Nutritional rehabilitation has been achieved by intensive nutritional support, and mortality following liver transplantation has been reduced. Its success depends on a multidisciplinary approach based on early intervention and prevention of malnutrition. Caution about the effects of overfeeding may be relevant to the development of obesity in the long term.

REFERENCES

1. Chin SE, Shepherd RW, Thomas BJ, et al. The nature of malnutrition in children with end stage liver disease. *Am J Clin Nutr*. 1992;56:164–168.
2. Trocki O, Wotton MJ, Cleghorn GJ, Shepherd RW. Value of total body potassium in assessing the nutritional status of children with end-stage liver disease. *Ann N Y Acad Sci*. May 2000;904:400–405.
3. Kaufman SS, Murray ND, Wood RP, et al. Nutritional support for the infant with extrahepatic biliary atresia. *J Pediatr*. May 1987;110(5):679–686.
4. Beath S PG, Kelly D, McMaster P, et al. Liver transplantation in babies and children with extrahepatic biliary atresia. *J Pediatr Surg*. 1993;28(8):1044–1047.
5. Novy MA, Schwarz KB. Nutritional considerations and management of the child with liver disease. *Nutrition*. Mar 1997;13(3):177–184.
6. Roggero P, Cataliotti E, Ulla L, et al. Factors influencing malnutrition in children waiting for liver transplants. *Am J Clin Nutr*. Jun 1997;65(6):1852–1857.
7. Sokal RJ, Stall C. Anthropometric evaluation of children with chronic liver disease. *Am J Clin Nutr*. 1990;52:203–208.
8. Romjin JA, Endert E, Suerwein HP. Glucose and fat metabolism during short-term starvation in cirrhosis. *Gastroenterology*. 1991;100:731–737.
9. Hellerstein MK, Munro HN. Interaction of liver, muscle and adipose tissue in the regulation of metabolism in response to nutritional and other factors. In: Arias IM, Boyer JL, Fausto N, et al. eds., *The Liver: Biology and Pathobiology*. 3rd ed. New York: Raven Press; 1994. pp. 1169–1191.
10. Felig P, Brown WV, Levine RA, Klatskin G. Glucose homeostasis in viral hepatitis. *N Engl J Med*. Dec 24 1970;283(26):1436–1440.
11. Petrides AS, DeFronzo RA. Glucose and insulin metabolism in cirrhosis. *J Hepatol*. Jan 1989;8(1):107–114.
12. Nielsen MF, Caumo A, Aagaard NK, et al. Contribution of defects in glucose uptake to carbohydrate intolerance in liver cirrhosis: assessment during physiological glucose and insulin concentrations. *Am J Physiol Gastrointest Liver Physiol*. Jun 2005;288(6):G1135–G1143.
13. Cleghorn GJ. The role of basic nutrional research in pediatric liver disease: an historical perspective. *Gastroneterol Hepatol*. 2009;24(3):S93–S96.
14. Petrides AS, Strohmeyer G, DeFronzo RA. Insulin resistance in liver disease and portal hypertension. *Prog Liver Dis*. 1992;10:311–328.

15. Yudkoff M NI, McNellis W, Polin R. Albumin synthesis in premature infants: determination of turnover with [15N]glycine. *Pediatr Res.*1987;21:49–53.
16. Miller L. The role of the liver and the non-hepatic tissues in the regulation of free amino acid levels in the blood. In: Holden JT, ed., *Amino Acid Pools A.* Amsterdam: Elsevier; 1962. pp. 708–709.
17. Powers Lee SG ML. Urea synthesis and ammonia metabolism. In: Arias IM, Boyer JL, Fausto N, et al. eds., *The Liver: Biology and Pahtobiology.* New York: Raven Press; 1988. pp: 317–330.
18. McCullough AJ MK, Smanik EJ, Tabaa M, Szauter K. Nutritional therapy and liver disease. *Gastroenterol Clin North Am.* 1989;18:619–643.
19. Weisdorf SA FD, Fath J, Tsai MY, Cerra FB. Amino acid abnormalities in infants with extrahepatic biliary atresia and cirrhosis. *Paed Gastroenterol Nutr.* 1987;6:860–864.
20. Protheroe S, Jones R, Kelly DA. Evaluation of the role of branched chain amino acids in the treatment of protein malnutrition in infants with liver disease. *Gut.* 1995;37:A30.
21. Kawaguchi T, Izumi N, Charlton MR, Sata M. Branched-chain amino acids as pharmacological nutrients in chronic liver disease. *Hepatology.* Sep 2 2011;54(3):1063–1070.
22. Cascino A, Cangiano C, Calcaterra V, et al. Plasma amino acids imbalance in patients with liver disease. *Am J Dig Dis.* Jul 1978;23(7):591–598.
23. Mager DR WL, Roberts EA, Ball RO, Pencharz PB. Mild-to-moderate chronic cholestatic liver disease increases leucine oxidation in children. *J Nutr.* 2006 Apr;136(4):965–970.
24. Blonde-Cynober F, Aussel C, Cynober L. Abnormalities in branched-chain amino acid metabolism in cirrhosis: influence of hormonal and nutritional factors and directions for future research. *Clin Nutr.* Feb 1999;18(1):5–13.
25. Tessari P, Zanetti M, Barazzoni R, et al. Response of phenylalanine and leucine kinetics to branched chain-enriched amino acids and insulin in patients with cirrhosis. *Gastroenterology.* Jul 1996;111(1):127–137.
26. Dichi I, Dichi JB, Papini-Berto SJ, et al. Protein-energy status and 15N-glycine kinetic study of child a cirrhotic patients fed low- to high-protein energy diets. *Nutrition.* Jul-Aug 1996;12(7–8):519–523.
27. Protheroe S, McNurlan M, Garlick P, et al. Failure to suppress protein breakdown contributes towards malnutrition in infants with liver disease. *Hepatology.* 1996;24:141A.
28. Kelly DA, Summerfield JA. Hemostasis in liver disease. *Semin Liver Dis.* Aug 1987;7(3):182–191.
29. Wion KL, Kelly D, Summerfield JA, et al. Distribution of factor VIII mRNA and antigen in human liver and other tissues. *Nature.* Oct 24-30 1985;317(6039):726–729.
30. Devictor D, Tissieres P, Afanetti M, Debray D. Acute liver failure in children. *Clin Res Hepatol Gastroenterol.* Jun 2011;35(6-7):430–437.
31. Van Dyke RW. Mechanisms of digestion and absorption of fiid. In: Sleisenger MH, Fordtran JD, eds., *Gastrointestinal Disease, Pathyophysiology, Diagnosis, Management.* 4th ed. Philadelphia: WB Saunders Co; 1989. pp. 1062–1088.
32. Carey M, Small DM, Bliss CM. Lipid digestion and absorption. *Annu Rev Physiol.* 1983;45:651–677.
33. Greenberger NJ, Skillman TG. Medium-chain triglycerides. *N Engl J Med.* May 8 1969;280(19):1045–1058.
34. Glasgow JF, Hamilton JR, Sass-Kortsak A. Fat absorption in congenital

obstructive liver disease. *Arch Dis Child.* Aug 1973;48(8):601–607.
35. Phan CT, Tso P. Intestinal lipid absorption and transport. *Front Biosci.* 2001;6:D299–D319.
36. Beath S, Hooley I, Willis K, et al. Long chain triacyglycerol malabsorption; pancreatic function in children with protein, malnutrition complicating severe liver disease. *Prot Nil Soc.* 1993b;52:252A.
37. Beath SV BI, Kelly DA. Nutritional support in liver disease. *Arch Dis Child.*1993;69:545–549.
38. Pawlowska J, Socha P, Socha J. Nutrition in pediatric patients before liver transplantation. *Rocz Panstw Zakl Hig.* 2007;58(1):111–118.
39. Chong SKF, Lindridge J, Moniz C, Mowat AP. Exocrine pancreatic insufficiency in syndromic paucity of interlobular bile ducts. *J Pediatr Gastroenterol Nutr.* 1989;9(4):445–449.
40. Makrides M, Neuman M, Simmer K, et al. Are long-chain polyunsaturated fatty acids essential nutrients in infancy? *Lancet.* 1995;345:1463–1468.
41. Koletzko B, Agosoni C, Carlson SE, et al. Long chain polyunsaturated fatty acids (LC-PUFA) and perinatal development. *Acta Paediatr.* 2001;90:460–464.
42. Hoffman DR BE, Birch DG, Uauy RD. Effects of supplementation with omega 3 long-chain polyunsaturated fatty acids on retinal and cortical development in premature infants. *Am J Clin Nutr.* 1993;57:807S–812S.
43. Burke PA, Ling PR, Forse RA, Bistrian BR. Conditionally essential fatty acid deficiencies in end-stage liver disease. *Nutrition.* Apr 1999;15(4):302–304.
44. Cabre E, Gassull MA. Polyunsaturated fatty acid deficiency in liver diseases: pathophysiological and clinical significance. *Nutrition.* Jul-Aug 1996;12(7-8):542–548.
45. Burke PA, Ling PR, Forse RA, et al. Sites of conditional essential fatty acid deficiency in end stage liver disease. *JPEN J Parenter Enteral Nutr.* Jul-Aug 2001;25(4):188–193.
46. Lapillonne A, Hakme C, Mamoux V, et al. Effects of liver transplantation on long-chain polyunsaturated fatty acid status in infants with biliary atresia. *J Pediatr Gastroenterol Nutr.* May 2000;30(5):528–532.
47. Spray CH BS, Willis KD, et al. Docosahexaenoic acid (DHA) and visual function in infants with fat malabsorption secondary to liver disease. *Proceed Nutr.* 1995;54:108A.
48. Kaufman SS, Scrivner DJ, Murray ND, et al. Influence of portagen and pregestimil on essential fatty acid status in infantile liver disease. *Pediatrics.* Jan 1992;89(1):151–154.
49. Larque E, Zamora S, Gil A. Dietary trans fatty acids in early life: a review. *Early Hum Dev.* Nov 2001;65 Suppl:S31–S41.
50. Mager DR, McGee PL, Furuya KN, Roberts EA. Prevalence of vitamin K deficiency in children with mild to moderate chronic liver disease. *J Pediatr Gastroenterol Nutr.* Jan 2006;42(1):71–76.
51. Shneider BL, Magee JC, Bezerra JA, et al. Efficacy of fat-soluble vitamin supplementation in infants with biliary atresia. *Pediatrics.* Sep 2012;130(3):e607–e614.
52. Powell EE, Kroon PA. Liver, lipoproteins and disease: II. Clinical relevance of disordered cholesterol metabolism in liver disease. *J Gastroenterol Hepatol.* Mar-Apr 1992;7(2):225–231.
53. Malatack JJ, Schaid DJ, Urbach AH, et al. Choosing a pediatric recipient for orthotopic liver transplantation. *J Pediatr.* Oct 1987;111(4):479–489.
54. Quirk P OP, Moyse K, Chin S, et al. Insulin-like growth factors I and II are

reduced in plasma from growth retarded children with chronic liver disease. *Growth Regulation.* 1994;4:35–38.
55. Holt RI, Jones JS, Stone NM, et al. Sequential changes in insulin-like growth factor I (IGF-I) and IGF-binding proteins in children with end-stage liver disease before and after successful orthotopic liver transplantation. *J Clin Endocrinol Metab.* Jan 1996;81(1):160–168.
56. Ketelslegers JM, Maiter D, Maes M, et al. Nutritional regulation of insulin-like growth factor-I. *Metabolism.* Oct 1995;44(10 Suppl 4):50–57.
57. Zamboni G, Dufillot D, Antoniazzi F, et al. Growth hormone-binding proteins and insulin-like growth factor-binding proteins in protein-energy malnutrition, before and after nutritional rehabilitation. *Pediatr Res.* Mar 1996;39(3):410–414.
58. Held MA, Cosme-Blanco W, Difedele LM, et al. Alterations in growth hormone receptor abundance regulate growth hormone signaling in murine obstructive cholestasis. *Am J Physiol Gastrointest Liver Physiol.* May 2005;288(5):G986–G993.
59. Maes M, Sokal E, Otte JB. Growth factors in children with end-stage liver disease before and after liver transplantation: a review. *Pediatr Transplant.* Nov 1997;1(2):171–175.
60. Bucuvalas JC, Horn JA, Chernausek SD. Resistance to growth hormone in children with chronic liver disease. *Pediatr Transplant.* Aug 1997;1(1):73–79.
61. Baruch Y, Assy N, Amit T, et al. Spontaneous pulsatility and pharmacokinetics of growth hormone in liver cirrhotic patients. *J Hepatol.* Oct 1998;29(4):559–564.
62. Assy N, Pruzansky Y, Gaitini D, et al. Growth hormone-stimulated IGF-1 generation in cirrhosis reflects hepatocellular dysfunction. *J Hepatol.* Jul 2008;49(1):34–42.
63. Hogler W, Baumann U, Kelly D. Endocrine and bone metabolic complications in chronic liver disease and after liver transplantation in children. *J Pediatr Gastroenterol Nutr.* Mar 2012;54(3):313–321.
64. Holt RI MJ, Jones JS, Mieli-Vergani G, Baker AJ. Nasogastric feeding enhances nutritional status in paediatric liver disease but does not alter circulating levels of IGF-I and IGF binding proteins. *Clin Endocrinol* 2000;52:217–224.
65. Moller S, Juul A, Becker U, Henriksen JH. The acid-labile subunit of the ternary insulin-like growth factor complex in cirrhosis: relation to liver dysfunction. *J Hepatol.* Mar 2000;32(3):441–446.
66. Pierro A KB, Carnielli V, Superina RA, et al. Resting energy expenditure is increased in infants and children with extrahepatic biliary atresia. *J Ped Surgery.* 1989;24:534–538.
67. Protheroe SM, McKiernan PJ, Kelly DA. Can measurement of dietary-induced thermogenesis (DIT) predict response to nutritional intervention in infants with liver disease? *Clin Nutr.* 1996;15:39.
68. Cortez AP, de Morais MB, Speridiao Pda G, et al. Food intake, growth and body composition of children and adolescents with autoimmune hepatitis. *J Clin Gastroenterol.* Mar 2010;44(3):200–207.
69. Greer R, Lehnert M, Lewindon P, et al. Body composition and components of energy expenditure in children with end-stage liver disease. *J Pediatr Gastroenterol Nutr.* Mar 2003;36(3):358–363.
70. Rovner AJ, Stallings VA, Piccoli DA, et al. Resting energy expenditure is not increased in prepubertal children with Alagille syndrome. *J Pediatr.* May 2006;148(5):680–682.
71. Rossouw JE, Labadarios D, Davis M, Williams R. Water-soluble vitamins in severe liver disease. *S Afr Med J.* Jul 29 1978;54(5):183–186.

72. Umusig-Quitain P, Gregorio GV. High incidence of zinc deficiency among Filipino children with compensated and decompensated liver disease. *J Gastroenterol Hepatol.* Feb 2010;25(2):387–390.
73. Strople J, Lovell G, Heubi J. Prevalence of subclinical vitamin K deficiency in cholestatic liver disease. *J Pediatr Gastroenterol Nutr.* Jul 2009;49(1):78–84.
74. Saron ML, Godoy HT, Hessel G. Nutritional status of patients with biliary atresia and autoimmune hepatitis related to serum levels of vitamins A, D and E. *Arq Gastroenterol.* Jan-Mar 2009;46(1):62–68.
75. Mourey MS, Siegenthaler G, Amedee-Manesme O. Regulation of metabolism of retinol-binding protein by vitamin A status in children with biliary atresia. *Am J Clin Nutr.* Apr 1990;51(4):638–643.
76. Guggenheim MA, Ringel SP, Silverman A, Grabert BE. Progressive neuromuscular disease in children with chronic cholestasis and vitamin E deficiency: diagnosis and treatment with alpha tocopherol. *J Pediatr.* Jan 1982;100(1):51–58.
77. Sokol RJ, Heubi JE, Iannaccone S, et al. Mechanism causing vitamin E deficiency during chronic childhood cholestasis. *Gastroenterology.* Nov 1983;85(5):1172–1182.
78. van Hasselt PM, Kok K, Vorselaars AD, et al. Vitamin K deficiency bleeding in cholestatic infants with alpha-1-antitrypsin deficiency. *Arch Dis Child Fetal Neonatal Ed.* Nov 2009;94(6):F456–F460.
79. Olsen IE, Ittenbach RF, Rovner AJ, et al. Deficits in size-adjusted bone mass in children with Alagille syndrome. *J Pediatr Gastroenterol Nutr.* Jan 2005;40(1):76–82.
80. Hosey MT. Dental care of children with liver disease. In: Kelly DA, editor. *Diseases of the Liver and Biliary System in Children.* 3rd ed. Malden, MA: Wiley Blackwell; 2008. pp. 420–432.
81. Samonte VA, Sherman PM, Taylor GP, et al. Scurvy diagnosed in a pediatric liver transplant patient awaiting combined kidney and liver retransplantation. *Pediatr Transplant.* May 2008;12(3):363–367.
82. Wayman KI, Cox KL, Esquivel CO. Neurodevelopmental outcome of young children with extrahepatic biliary atresia 1 year after liver transplantation. *J Pediatr.* Dec 1997;131(6):894–898.
83. Barshes NR, Chang IF, Karpen SJ, et al. Impact of pretransplant growth retardation in pediatric liver transplantation. *J Pediatr Gastroenterol Nutr.* Jul 2006;43(1):89–94.
84. Erlichman J, Hohlweg K, Haber BA. Biliary atresia: how medical complications and therapies impact outcome. *Expert Rev Gastroenterol Hepatol.* Aug 2009;3(4):425–434.
85. Alonso EM. Growth and developmental considerations in pediatric liver transplantation. *Liver Transpl.* May 2008;14(5):585–591.
86. Rodeck B, Melter M, Kardorff R, et al. Liver transplantation in children with chronic end stage liver disease: factors influencing survival after transplantation. *Transplantation.* Oct 27 1996;62(8):1071–1076.
87. Chin SE, Shepherd RW, Cleghorn GJ, et al. Survival, growth and quality of life in children after orthotopic liver transplantation: a 5 year experience. *J Paediatr Child Health.* Dec 1991;27(6):380–385.
88. Holt TL, Cui C, Thomas BJ, et al. Clinical applicability of bioelectric impedance to measure body composition in health and disease. *Nutrition.* May-Jun 1994;10(3):221–224.
89. Shepherd RW, Chin SE, Cleghorn GJ, et al. Malnutrition in children with chronic

liver disease accepted for liver transplantation: clinical profile and effect on outcome. *J Paediatr Child Health.* Oct 1991;27(5):295–299.
90. van Mourik ID, Beath SV, Brook GA, et al. Long-term nutritional and neurodevelopmental outcome of liver transplantation in infants aged less than 12 months. *J Pediatr Gastroenterol Nutr.* Mar 2000;30(3):269–275.
91. Hartman C, Shamir R, Hecht C, Koletzko B. Malnutrition screening tools for hospitalized children. *Curr Opin Clin Nutr Metab Care.* May 2012;15(3):303–309.
92. Amedee-Manesme O, Furr HC, Alvarez F, et al. Biochemical indicators of vitamin A depletion in children with cholestasis. *Hepatology.* Nov-Dec 1985;5(6):1143–1148.
93. Beath SV, Johnson S, Willis KD, et al. E. Growth and luminal fat solubilisation in cholestatic infants on medium chain triglyceride (MCT). *J Pediatr Gastroenterol Nutr.* 1996;22:443.
94. Pettei MJ, Daftary S, Levine JJ. Essential fatty acid deficiency associated with the use of a medium-chain triglyceride infant formula in pediatric hepatobiliary disease. *Am J Clin Nutr.* May 1991;53(5):1217–1221.
95. Socha P, Koletzko B, et al. LCP-supplementation in infants with cholestasis. *J Pediatr Gastroenterol Nutr.* 2000;30:528–532.
96. Stein J. Chemically defined structured lipids: current status and future directions in gastrointestinal diseases. *Int J Colorectal Dis.* Apr 1999;14(2):79–85.
97. Bellantone R, Bossola M, Carriero C, et al. Structured versus long-chain triglycerides: a safety, tolerance, and efficacy randomized study in colorectal surgical patients. *JPEN J Parenter Enteral Nutr.* May-Jun 1999;23(3):123–127.
98. Canielli VP LI, Van Goudoever J, Aulders EJ, et al. Structural position and amount of palmitic acid in infant formulas: effects on fat, fatty acid, and mineral balance. *JPGN.* Dec 1996;23(5):553–560.
99. Spalinger JH, Seidman EG, Lepage G, et al. Uptake and metabolism of structured triglyceride by Caco-2 cells: reversal of essential fatty acid deficiency. *Am J Physiol.* Oct 1998;275(4 Pt 1):G652–G659.
100. Straarup EM, Hoy CE. Structured lipids improve fat absorption in normal and malabsorbing rats. *J Nutr.* Nov 2000;130(11):2802–2808.
101. Protheroe SM. Feeding the child with chronic liver disease. *Nutrition.* Oct 1998;14(10):796–800.
102. Charlton CP, Buchanan E, Holden CE, et al. Intensive enteral feeding in advanced cirrhosis: reversal of malnutrition without precipitation of hepatic encephalopathy. *Arch Dis Child.* May 1992;67(5):603–607.
103. Platell C, Kong SE, McCauley R, Hall JC. Branched-chain amino acids. *J Gastroenterol Hepatol.* Jul 2000;15(7):706–717.
104. Mager DR, Wykes LJ, Roberts EA, et al. Effect of orthotopic liver transplantation (OLT) on branched-chain amino acid requirement. *Pediatr Res.* Jun 2006;59(6):829–834.
105. Sokal EM, Baudoux MC, Collette E, et al. Branched chain amino acids improve body composition and nitrogen balance in a rat model of extra hepatic biliary atresia. *Pediatr Res.* Jul 1996;40(1):66–71.
106. Chin SE, Shepherd RW, Thomas BJ, et al. Nutritional support in children with end-stage liver disease: a randomized crossover trial of a branched-chain amino acid supplement. *Am J Clin Nutr.* Jul 1992;56(1):158–163.
107. Moreno LA, Gottrand F, Hoden S, et al. Improvement of nutritional status in cholestatic children with supplemental nocturnal enteral nutrition. *J Pediatr Gastroenterol Nutr.* Feb 1991;12(2):213–216.

108. Moreno A, Dominguez C, Ballabriga A. Aluminum in the neonate related to parenteral nutrition. *Acta Paediatr.* Jan 1994;83(1):25–29.
109. Bougle D, Bureau F, Deschrevel G, et al. Chromium and parenteral nutrition in children. *J Pediatr Gastroenterol Nutr.* Jul 1993;17(1):72–74.
110. Fell JM, Reynolds AP, Meadows N, et al. Manganese toxicity in children receiving long-term parenteral nutrition. *Lancet.* May 4 1996;347(9010):1218–1221.
111. Huang CC, Chu NS, Lu CS, et al. Chronic manganese intoxication. *Arch Neurol.* Oct 1989;46(10):1104–1106.
112. Kelly D, Tong C. Neonatal and paediatric infection. In: *Viral Hepatitis.* 4th ed. Wylie; 2012.
113. Heubi JE, Wiechmann DA, Creutzinger V, et al. Tauroursodeoxycholic acid (TUDCA) in the prevention of total parenteral nutritional associated liver disease. *J Pediatr Gastroenterol Nutr.* 2003;36(3):422–423.
114. Kamath BM, Loomes KM, Piccoli DA. Medical management of Alagille syndrome. *J Pediatr Gastroenterol Nutr.* Jun 2010;50(6):580–586.
115. Smart KM, Alex G, Hardikar W. Feeding the child with liver disease: a review and practical clinical guide. *J Gastroenterol Hepatol.* May 2011;26(5):810–815.
116. Green A, Kelly DA. Metabolic liver disease in older children. In: Kelly DA, editor. *Diseases of the Liver and Biliary System in Children. Oxford, UK: Blackwell Science;* 1999. pp. 157–166.
117. Lindstedt S, Holme E, Lock EA, et al. Treatment of hereditary tyrosinaemia type I by inhibition of 4-hydroxyphenylpyruvate dioxygenase. *Lancet.* Oct 3 1992;340(8823):813–817.
118. Mayatepek E, Hoffmann B, Meissner T. Inborn errors of carbohydrate metabolism. *Best Pract Res Clin Gastroenterol.* Oct 2010;24(5):607–618.
119. Holt RI, Broide E, Buchanan CR, et al. Orthotopic liver transplantation reverses the adverse nutritional changes of end-stage liver disease in children. *Am J Clin Nutr.* Feb 1997;65(2):534–542.
120. Bartosh SM, Thomas SE, Sutton MM, et al. Linear growth after pediatric liver transplantation. *J Pediatr.* Nov 1999;135(5):624–631.
121. Cardona J, Houssin D, Gauthier F, et al. Liver transplantation in children with Alagille syndrome—a study of twelve cases. *Transplantation.* Aug 27 1995;60(4):339–342.
122. Kamath BM, Yin W, Miller H, et al. Outcomes of liver transplantation for patients with Alagille syndrome: the studies of pediatric liver transplantation experience. *Liver Transpl.* Aug 2012;18(8):940–948.
123. Kelly D. Posttransplant growth failure in children. *Liver Trans and Surgery.* Sep 1997;3(5 Suppl 1):S32–S39.
124. Sarna S, Sipila I, Ronnholm K, et al. Recombinant human growth hormone improves growth in children receiving glucocorticoid treatment after liver transplantation. *J Clin Endocrinol Metab.* Apr 1996;81(4):1476–1482.
125. Schutz T, Hudjetz H, Roske AE, et al. Weight gain in long-term survivors of kidney or liver transplantation—another paradigm of sarcopenic obesity? *Nutrition.* Apr 2012;28(4):378–383.
126. Mouzaki M, Allard JP. The role of nutrients in the development, progression, and treatment of nonalcoholic fatty liver disease. *J Clin Gastroenterol.* Jul 2012;46(6):457–467.

Cancer Prevention

57

A. Lindsay Frazier, MD, ScM

INTRODUCTION

Cancer is undeniably a disease with an established genetic basis, but the field of epidemiology has shown that the development of cancer is the result of an interaction between genes and the environment. A simple example are migrant studies that show an increase in cancer risk in the second generation after migration but not in the first generation, theoretically after sufficient exposure to the environment of the new country has taken place. These studies strongly suggest that it is the environment that modifies underlying genetic risk. Diet has long been postulated to be one of the most important environmental factors in the etiology of cancer.[1]

Pinpointing exactly which elements of diet are implicated in the pathogenesis of cancer is complicated; diet itself is inherently complex, even one single food can simultaneously contain both cancer-promoting and cancer-preventing substances. Moreover, diet does not happen in a vacuum but takes place in the context of other human behaviors, and these other risk factors must be simultaneously taken into account. Precise measurement of diet is challenging due to the day-to-day variability in consumption; measurement is also dependent on the accurate recall and estimation by the individual (see Chapter 17, Nutritional Epidemiology for details). Finally, the critical time of the exposure may vary by type of cancer. For instance, in the case of breast cancer, the theory has been advanced that there is a critical period for carcinogenic exposures between onset of menarche and first birth. The hormone-induced proliferation occurs with each menstrual cycle prior to the differentiation of the breast that occurs at the time of pregnancy and lactation. Diet during this

adolescent period likely has different implications for breast cancer than diet in later life. Despite this substantial list of caveats, the evidence has accumulated that diet throughout the life course, beginning with in-utero exposures and extending through adult life, has an impact on the risk of malignancy, both in childhood and adulthood.

The evidence for the relationship between nutritional intake and cancer comes in several forms. The simplest line of evidence is the relationship between growth and risk of malignancy. Although studies showing a relationship between birth weight, height, or growth velocity do not help to identify a causative agent, these studies do confirm that what one eats, and therefore what fuels growth, is related to cancer. The second line of evidence attempts to identify the causative agent through the use of dietary records, either in the setting of a case-control study or in a prospective cohort study. Animal studies in which diet can be more tightly controlled, can either suggest or support observations in humans.

In this chapter, we review the evidence that childhood nutrition can impact the risk of subsequent malignancy. Using a wide lens, we examine several different aspects of nutrition. The first principle will be to examine the impact of diet over the entire span of childhood, beginning with in utero exposures through adolescence. Second, we consider the impact of childhood diet on the incidence of both childhood and adult malignancies. To do so, we review the evidence linking childhood growth and cancer, making the argument that growth is a proxy for nutrition. In addition, using case-control studies, we review the specific associations between childhood diet and cancer. Finally, we review the evidence that for breast cancer at least, the period of childhood and adolescence may be the critical period for exposures that ultimately determines risk of disease as an adult (see Figure 57-1).

The Connection Between Growth and Cancer

Cancer, simply put, is uncontrolled growth. Where does the uncoupling occur between controlled and uncontrolled growth? In pediatrics, there are a set of clinical syndromes notable for “overgrowth,” or excessive growth in the pre- and postnatal periods that occur at a frequency of approximately 1/10,000 children. Over 20 pediatric overgrowth syndromes have been described, each with the shared feature of excessive growth, but also with unique abnormalities due to the specific underlying genetic cause. Most of these syndromes are also associated with an increased risk of cancer, principally embryonal tumors. These syndromes are provocative paradigms of the malignant hazards of excessive growth.

The most widely recognized of pre- and postnatal growth syndromes is Beckwith–Wiedemann syndrome (BWS). BWS is often associated with advanced bone age, macroglossia, hemihypertrophy, neonatal hypoglycemia, and umbilical hernia or omphalocele. Approximately 1%–8% of children with BWS will present with Wilms' tumor and less commonly, hepatoblastoma. Genetic abnormalities have been located primarily to the short arm of chromosome 11(11p15), known as the WT2 locus. This region is heavily imprinted; the paternal gene for insulin-like growth factor 2 (IGF-2) is normally constitutively activated. The presence of not just one but two active copies of the IGF-2 gene, due to paternal uniparental disomy, which is demonstrable in 10%–20% of BWS patients, is hypothesized to be one of the molecular causes of the syndrome.

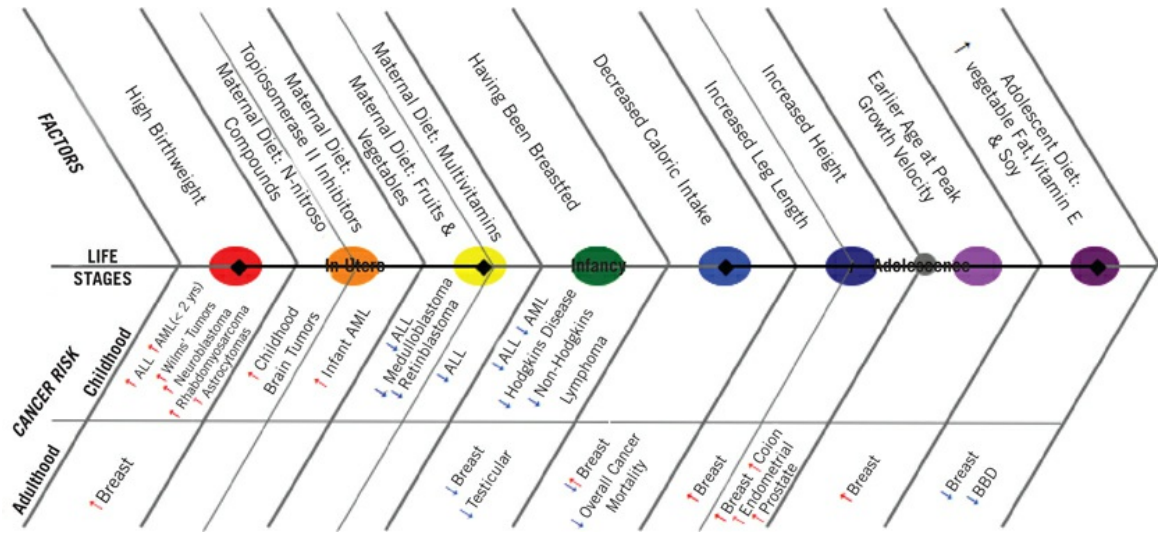


Figure 57-1 Nutritional factors in childhood and lifetime prevention of cancer.

Simpson–Golabi–Behmel syndrome (SGBS) is an X-linked disorder that has many of the same clinical features as BWS: pre- and postnatal overgrowth, macroglossia, cleft plate, advanced bone age, and umbilical hernia. Approximately 5%–20% of patients with SGBS will present with Wilms' tumor. SGBS patients often have other features including coarse facies, down-slanting palpebrae, polydactyly, syndactyly, intracardiac defects, and cardiac conduction defects. SBGS is caused by a deletion in the glypican 3 gene on the X chromosome. Glypican 3 is a heparan sulfate proteoglycan that is attached to the cell surface via a glycosyl-

phosphatidylinositol anchor. Its role in the cell is to regulate the growth and differentiation of predominately mesodermal cells through modulating the interaction of growth factors, particularly IGF-2, with their receptors. Glypican 3 is usually expressed in embryonic tissues and downregulated in adult tissues. Glypican 3 is found to be overexpressed in Wilms' tumor, hepatocellular carcinoma, and neuroblastoma. It is interesting that IGF-2 is implicated in two of the most common pediatric overgrowth syndromes: BWS and SBGS. Understanding the molecular basis for these overgrowth syndromes will likely highlight genes critical for the proper balance of growth during childhood and may help us to understand the link between the genetics of growth and the genetics of cancer and the environmental influences, such as diet, that can tip the balance in favor of malignant growth.

IN-UTERO EXPOSURES AND INFANT LEUKEMIAS AND OTHER CHILDHOOD CANCERS

When does cancer begin? The genetic evidence is now quite compelling that infant leukemias are initiated in utero. Infant leukemias have certain pathognomonic translocations such as t(12;21), t(4,11), or t(8:21). Among infants who developed leukemia in the first 2 years of life, scientists used neonatal blood spots collected to neonatal screening purposes at birth to get a picture of the chromosomal makeup of the child at birth. They have been able to identify the leukemia-causing translocation clone present at birth, even though the leukemia itself did not become manifest for several months or years.[2,3] Epidemiologic evidence supports the notion that in utero exposures are etiologic in cancer causation. For instance, there is strong association between birth weight and childhood cancer, especially leukemia. Other environmental factors, particularly maternal—or in utero—diet have also been shown to influence the risk of certain childhood cancers, particularly leukemia, and central nervous system tumors in children.

Birth Weight and Pediatric Malignancies

The most straightforward marker of intrauterine growth, birth weight, is in large part determined by levels of maternal insulin-like growth factor-1, which has both mitotic and anti-apoptotic functions. These same functions of IGF may be the link between larger birth weight and the observed

associations with several types of childhood cancers including acute lymphoblastic leukemia (ALL), acute myelogenous leukemia (AML), Wilms' tumor, neuroblastoma, rhabdomyosarcoma, and astrocytomas (but not other forms of childhood brain tumors).[4–8] The results of some studies have been inconsistent but some of the variability in results is likely due to the fact that the association between birth weight and cancer may operate for only for one gender and not the other, or only for certain histologic subtypes, or only for earlier age at diagnosis, depending on the underlying mechanism responsible for the association. Reviewing the adequacy of stratified analyses is imperative in interpreting the findings.

The data linking higher birth weight and childhood malignancy are most consistent for childhood leukemias. A meta-analysis of 18 studies of birth weight and childhood leukemia,[9] estimated that for each one kilogram (kg) increase in birth weight, risk of ALL increased by 14% (95% CI 1.08–1.20) and risk of AML increases by 29% (95% CI 0.73–2.20).[9] There was evidence of a dose-response relationship for both types of leukemia, although it was stronger for ALL (likely because ALL is more prevalent than AML and hence there is more power to detect an association). The association is strongest in children under the age of 2 years; in fact, for AML, the association appears to be limited to children diagnosed before age 2.[10] Indeed, for children under age 2, risk of leukemia may increase by as much as 100% for children who weigh > 4,000 g at birth.[11] In a pooled analysis of three case-controls studies from the United States, United Kingdom and Germany that comprised 4,075, the estimated excess risk per kg of bodyweight was similar, however, in this analysis, the authors showed that case children were on average heavier at every gestational age. The authors also noted a paucity of cases in the very low birth weight group (<1500 g) and speculate that if indeed ALL is initiated in utero but not manifest at birth, that it may still predispose these "occult" cases to increased perinatal mortality.[12] Interestingly, both the prevalence of children who are large for gestational age (birth weight > 90th percentile) as well as the incidence of childhood leukemia and brain tumors have been increasing in parallel in the United States over the past several decades.[4]

Several studies report no relationship between birth weight and soft tissue sarcoma, but soft tissue sarcoma encompasses a very wide range of histologies and subgroup analysis is important. For instance, in a case-control study that linked state cancer registry and birth registry data, the authors showed that larger birth weight was associated with risk of

rhabdomyosarcoma, but only the embryonal histology. This makes intuitive sense in that embryonal rhabdomyosarcoma is diagnosed at a much younger age than alveolar, usually within the first 5 years of life, suggesting that perinatal exposures may be more relevant.[13] Moreover, the authors showed that it was not just birth weight but increased fetal growth that was specifically associated with embryonal rhabdomyosarcoma because the association was replicated for babies large for gestational age, not just those who met a certain birth weight cut-off.

A case-control study of the risk of osteosarcoma was conducted in orthopedic surgery departments in 10 US medical centers between 1994 and 2000. Controls (n = 151) were from the same departments as cases (n = 158) and the study included both children and adults. The study found that a higher birth weight was associated with an increased risk of osteosarcoma and the odds were greater for females. However, these results were inconsistent with two prior studies, which found no association, though these results were primarily conducted in children.

High birth weight and Wilms' tumor has been examined in 12 studies to date, including three prospective birth cohorts. Results of a meta-analysis show that a high birth weight is associated with a significantly increased risk of Wilms' tumor (RR = 1.08 (95% CI: 1.04–1.12).[14] Additionally in the National Wilms Tumor Study, a high percentage of cases had birth weights higher than the national median.

The results may vary by gender as well. Two studies have reported that the association between birth weight and Wilms' tumor was only observed for girls but not for boys.[15] Dorak et al. reported that the association for birth weight and ALL was present only for boys but not for girls.[16]

Several biologic explanations for the association between birth weight and childhood cancer have been suggested. One hypothesis has focused on the possibility that higher birth weight is a simple proxy for a greater number of cells proliferating at any one time and therefore greater absolute number of cells that are at potential risk for malignant transformation. Another possible mechanism may be a surfeit of IGF-1, which is known to be associated with higher birth weight. Others have suggested that excess of iron, which has also been linked to both increased birth weight as well as enhanced cancer cell growth, may explain the relationship between birth weight and childhood cancer. The major hemochromatosis allele, HFE C282Y, which is associated with increased serum iron levels in 15%–25% of heterozygotes, is also present in higher frequency among boys diagnosed with childhood ALL than controls.[17]

It should be noted that a variety of studies have also reported an

association between birth weight and cardiovascular disease, but in the opposite direction as that observed for cancer. Low birth weight has been associated with an increased risk of ischemic heart disease, as well as many of the intermediate markers of cardiovascular disease, such as blood pressure, body mass index, waist/hip ratio, serum lipids, plasma clotting factors, and impaired glucose tolerance.[18] Adverse conditions in utero, whether it is a surfeit of growth (linked to cancer) or a paucity of growth (linked to cardiovascular disease), appear to have long lasting implications on the human for risk of subsequent disease (see Chapter 26, Human Growth and Disease in Later Life for details).

IN-UTERO DIET (MATERNAL DIET) AND CHILDHOOD CANCER

Topoisomerase II Inhibitors and Infant Leukemias

Infant leukemias, although rare, clearly represent a unique subset of childhood leukemia because of their high resistance to standard chemotherapy (20% survival), in contrast to leukemias that occur later in childhood (80% survival). The majority of infant leukemias display a gene rearrangement at 11q23 in the MLL gene. This gene has been shown to play a major role in normal hematopoiesis.

The same gene, MLL, is implicated in secondary leukemias that result from treatment with a class of drugs—epipodophyllotoxins—that inhibit the function of DNA topoisomerase II (DNAt2). DNAt2 first relaxes the supercoiled DNA double helix by cleaving the bonds between the strands and then religates the two strands. Epipodophyllotoxins act by inhibiting religation by the enzyme, thereby triggering apoptosis. Topoisomerase inhibitors have wide activity against a number of different types of malignancies, but a small percent of treated patients will develop secondary leukemia, usually AML, with a characteristic cytogenetic marker of an 11q23 abnormality.

The de novo presence of the MLL abnormalities in infant leukemias led Ross et al. to propose in 1994 that there might be a link between in-utero exposure to DNAt2 inhibitors and infant leukemias.[19] The principal source of DNAt2 inhibitors in the environment is diet, primarily flavonoids, genistein, and catechins. Flavonoids, such as quercetin, are found in citrus fruits, apples, onions, parsley, tea, red wine, and dark berries such as blueberries and blackberries; genistein is found in soy products and catechins are found in green and black tea, cocoa, and red

wine. In the largest case-control study to date (n = 240), an increase in the odds of infant AML but not infant ALL was observed.[20] The effect was limited to in infants who were MLL+. Mothers in the top quartile of consumption of DNAt2-containing foods during pregnancy had 3.2 times (95% CI: 0.9–11.9) the odds of having an infant with AML (MLL+); however, the P value for the test for trend was .10 likely due to the fact that the study was based only on 29 patients in this subgroup.

N-Nitroso Compounds and Brain Tumors

Animal experiments have definitively shown that feeding pregnant animals nitrosamines causes nervous system tumors in the offspring. In the human diet, cured meats are the major source of nitrites and vegetables a major source of nitrates. Cured meats are treated with nitrites to prevent spoilage and unappealing changes in the color of the meat. The most common types of cured meats consumed include hot dogs, bacon, sausage, cured ham, and packaged lunch meats. In the acid environment of the stomach, nitrates and nitrites are converted to *N*-nitroso compounds, which can cause the formation of DNA adducts. Vitamins C and E block the nitrosation reactions in the intestines and have been shown in animals to protect against the carcinogenic effect of *N*-nitroso compounds.

Numerous case-control studies have examined the relationship between maternal consumption of nitrate and nitrite containing foods and childhood brain tumors; a consistent relationship has been seen for cured meats but not for vegetables.[21,22] In a meta-analysis of maternal cured meat consumption during pregnancy and the risk of childhood brain tumors that included seven observational studies, the pooled RR was 1.68.[23] As part of the US West Coast Childhood Brain Tumor Study, Preston-Martin et al. examined the mitigating effects of vitamin supplementation on risk of childhood brain tumors among women who consumed cured meat.[24] Risk of diagnosis of a brain tumor in her offspring was greater among women who did not take supplemental prenatal vitamins than among women who did take vitamins (OR = 2.4 vs. 1.3). In the group overall, daily use of prenatal vitamins decreased risk of childhood brain tumors by nearly 50% (OR = 0.54).

Multivitamins Supplementation During Pregnancy and Childhood Cancer

Numerous studies have examined how the use of multivitamins periconceptually and during pregnancy affect cancer risk in offspring.[25] In

a meta-analysis,[26] prenatal vitamin use was associated with a reduced risk of brain tumors (OR = 0.73; 95% CI 0.60–0.88), ALL (OR = 0.61; 95% CI 0.50–0.74), neuroblastoma (OR = 0.53; 95% CI 0.42–0.68). One study of retinoblastoma has also shown a reverse association. Studies have consistently have shown a decreased risk of childhood ALL, but not AML, among the offspring of women who took vitamins at time of pregnancy. In a case-control study of children with Down syndrome, chosen as the study population because of their 20-fold increase in risk of leukemia, vitamin use by the mother in the periconceptual period was associated with a reduced risk of ALL (OR = 0.63; 95% CI 0.39–1.00) whereas vitamin use later in pregnancy was associated with an increase in risk.[27]

Because of the evidence that adequate preconceptual levels of folate prevent neural tube defects and other congenital anomalies, both the US and Canada mandated universal fortification of flour products with folate in the mid 1990s. Two ecological studies of the effect of flour fortification with folic acid on rates of childhood malignancy have been reported. The first study from Canada showed that the incidence rate ratios of both neuroblastoma and Wilms' tumor decreased after universal flour fortification with folic acid.[28] The second study examined the impact of universal fortification in the United States.[29] Using SEER data, the incidence of both Wilms tumor and possibly primitive neuroectodermal tumors sharply declined after fortification. There was no observed effect on the rates of neuroblastoma in the United States postfortification.

Fruit and Vegetable Intake During Pregnancy and Childhood Cancers

Two studies have examined the relationship between fruit and vegetable consumption of the mother during pregnancy and risk of childhood ALL and both have shown an inverse association.[30] Another study has shown a moderate association between maternal consumption of fruits and vitamin C and a decreased risk of medulloblastoma/PNET in the offspring.[31] An additional case-control study from Mexico City has shown that incidence of sporadic retinoblastoma in the offspring is associated with lower maternal intake of fruits and vegetables, in particular folate and lutein/zeaxanthin.[32]

Birth Weight and Adult Malignancies

The association between birth weight and breast cancer is the most studied (Figure 57-2); evidence for other adult malignancies is limited. The

hypothesis that the roots of breast cancer could originate in utero was proposed by Trichopoulos in 1990.[33] He suggested that higher levels of maternal estrogens and other intrauterine growth factors could produce higher birth weight[34] and simultaneously affect the developing fetal breast tissue.[35,36] both of which might contribute to a higher risk of breast cancer.

There is a strong relationship between high birth weight and increased risk of premenopausal cancer. Evidence is remarkably consistent across study types (i.e., case-control versus cohort), exposure ascertainment (birth records versus self-reported birth weight), and even countries including populations in Scandinavia, United Kingdom, and the United States.[35] To date, 12 of the 15 studies provide supportive evidence of a relation between premenopausal breast cancer and high birthweight.[35] In meta-analyses of birth weight and premenopausal breast cancer, cohort studies showed a 20% (95% CI: 1.07–1.35I) increase whereas case-control studies showed a 36% (95% CI: 1.15–1.61) increase in risk with each 1,000 g increase in birth weight.[35] In an analysis that combined data from the Nurses' Health Study (NHS) and NHS II (*n* = 152,608 women), women with low birth weights (<5.5 lbs) had a 34% reduction in risk of premenopausal breast cancer incidence (95% CI: 0.47–0.93) compared with high birth weight (>8.5 lbs).[37] The association of birth weight was stronger in estrogen-receptor positive (ER+) and progesterone-receptor positive (PR+) tumors, indicating steroid hormones as a possible mediator between birth weight and breast cancer.[37] Finally, the association of birth weight and future risk of premenopausal breast cancer was independent of other factors later in life (i.e., age of menarche, family history of breast cancer), with the exception of adult height,[37] another risk factor for breast cancer that is established during childhood (see Figure 57-2).[35]

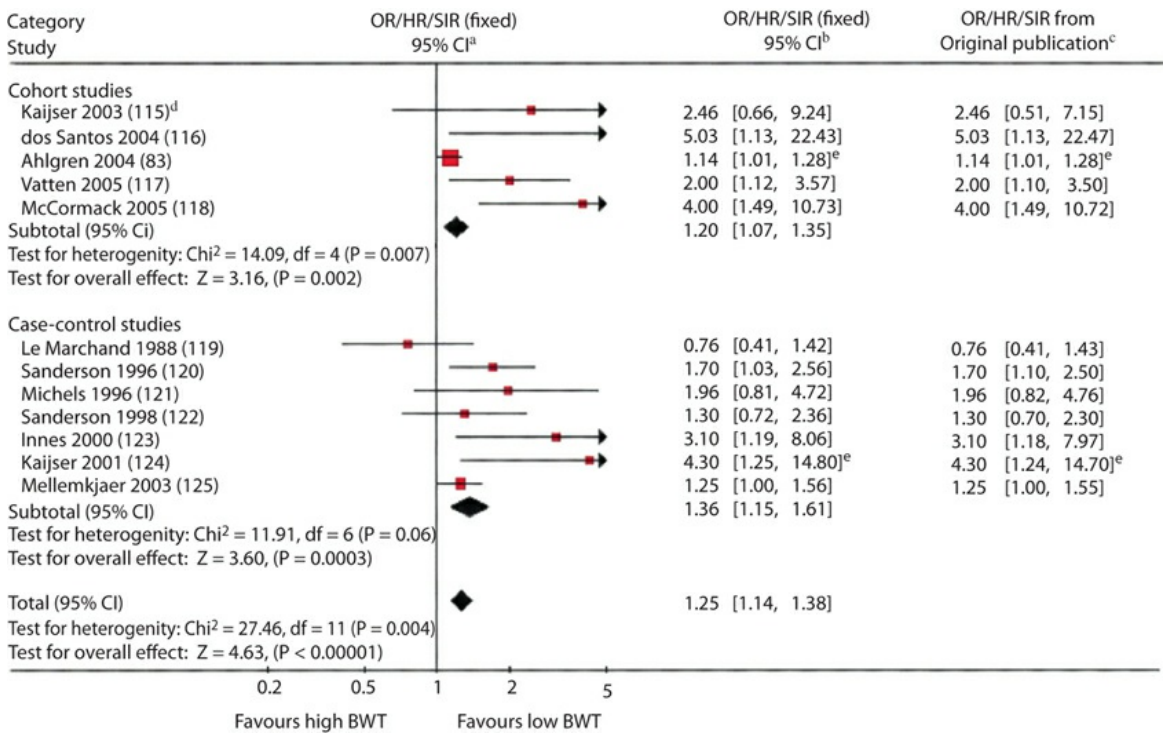


Figure 57-2 Studies on birthweight and the risk of breast cancer among premenopausal women. [a]OR, odds ratio; HR, hazard ratio; SIR, standardized incidence ratio; CI, confidence interval. The square indicates the odds ratio, hazard ratio, or standardized incidence ratio; the size of the square indicates the statistical weight each study contributes to the overall estimate; the line indicates the 95% confidence intervals. [b]Odds ratios, hazard ratios, standardized incidence rate ratios, and 95% confidence intervals generated by Revman through the generic inverse variance method and used in the analysis. [c]Odds ratios, hazard ratios, standardized incidence rate ratios, and 95% confidence intervals from original publication. [d]The number following the last name of the first author indicates the year of publication; the number in parenthesis indicates the reference number of the publication. [e]Effect estimate associated with each 1,000 g increase in birthweight.

In contrast, all but one study to date report no association between birth weight and postmenopausal cancer.[35] The pooled analysis of birth weight and postmenopausal breast cancer reported a relative risk of 1.04 (95% CI: 0.91–1.19) in cohort studies and a RR of 1.04 in (95% CI: 0.66–1.64) case-control studies.[35] The data from the combined NHS cohorts also support this meta-analysis, reporting a RR of 0.97 (CI: 0.08–1.16) of

postmenopausal breast cancer comparing women with a birth weight >8.5 lbs. compared with those <5.5 lbs.[37]

The relationship between birth weight and other cancers have been investigated, although the studies are sparse. Several studies have investigated the relationship between prostate cancer and birth weight but results are inconclusive. In a large Norwegian cohort (n = 19,681), a weak positive association was found between birth weight and risk of prostate cancer comparing the highest quartile of birth weight (>3,925 g) and the lowest quartile (<3,260 g), (RR 1.3; 95% CI: 0.8–2.0).[38] However, the study also reported a significant positive association between birth length comparing the highest (>53 cm) to lowest (<51 cm) quartile and risk for metastatic prostate cancer (n = 33). Also, men in the highest quartiles for both birth weight and birth length had nearly a 4-fold increase in metastatic prostate cancer (RR: 3.8; 95% CI: 1.2–2.0). These findings are consistent with a small Swedish cohort study[39] and the health professionals follow-up study which reported a weak positive association between birth weight and risk of advanced prostate cancer.[40] However, to date, the health and professionals follow-up study and two other studies reported no association of birth weight and risk of total prostate cancer.[40–42]

The association of birth weight and testicular cancer is also unclear. A large cohort study of Swedish men (n = 337,249) found no association between birth weight and total testicular cancer (n = 144) nor in a subset of nonseminomatous testicular cancer (n = 120),[43] although three previous case-control studies had reported a positive association.[44–46]

Two case-control studies have been conducted regarding birth weight and colorectal cancer.[47] Of note, in one case-control study, lower birth weight was associated with increased risk, though the trend across categories was not consistent.[47] Another prior study reported a J-shaped relationship between birth weight and colorectal cancer; the study was based only a few cases and included both men and women in analyses.[48]

Breastfeeding and Childhood Malignancy

Breastfeeding already has many proven benefits for the mother and child, including potentially decreasing the mothers' own risk for ovarian and breast cancer.[49,50] Breastfeeding likely also reduces risk of hematologic malignancy in the offspring.[51,52] Children who have been breastfed for at least six months have approximately a 30% lower likelihood of developing ALL and AML.[53] The effect appears restricted to pre-B cell ALL and M0,

M1, and M2 subtypes of AML. Other studies have suggested a reduction in risk for non-Hodgkin's lymphoma, although not all studies have reached statistical significance due to small number of cases.[51,54,55] The risk of Hodgkin's Lymphoma also appears reduced in two studies[54,56] but inference is limited in other studies due to small sample size.[53,55] The mechanism by which breastfeeding protects against hematologic malignancies in children is postulated to be immunomodulatory, protecting against an inappropriate lymphoproliferative response to an infection or other antigen challenge, rather than due to the nutritive components of breast milk.

Having Been Breastfed and Adult Malignancies

The anticarcinogenic effects of having been breastfed during infancy may persist into adulthood. The most studied relationship is the association of having been breastfed and risk of breast cancer, although the relationship between having been breastfed and site-specific cancers including gastric, colorectal, prostate, and testicular, has also been investigated.[57]

Several hypotheses have been advanced to explain the link between having been breastfed and subsequent risk of breast cancer.

- Early research (1930s) focused on breast milk as a deleterious exposure. The theory was that an "oncogenic" virus could be transmitted from the mother to infant via breast milk, increasing risk of breast cancer later in life for offspring. This hypothesis was first supported in animal models where particular strains of mice could be infected with a tumor virus via breast milk inducing mammary carcinogenesis.[58] Additionally, there is evidence of viral DNA, RNA, and antigen in human breast tissue and milk, and thus there has been continued interest in the possible viral etiology of breast cancer.[59–62]
- Organochlorines, such as polychlorinated biphenyls (PCBs) and Dichloro-Diphenyl-Trichloroethane (DDT), are carcinogenic substances preferentially secreted into breast milk.[63] It has been postulated that breastfeeding is a method used by the body to reduce the mother's burden of these chemicals; and women with higher levels of these chemicals are associated with decreased lactation,[64] thereby possibly reducing breast cancer risk for the mother.[65] However, the infant is exposed to these carcinogenic substances which are implicated in increasing risk of cancer

(including breast) in adulthood. This physiologic situation raises the question as to the optimal duration of breastfeeding, balancing the positive attributes of breast milk against possible extended exposure to carcinogens secreted into the breast milk.[66] At present, the World Health Organization recommends exclusive breastfeeding for the first 6 months of life because of well-documented benefits to both the mother and child.[67]

- Breast milk contains many bioactive components not present in formula such as prostaglandins, hormones, and cytokines, present at especially high levels in colostrum[68] that may affect the offspring's normal breast development but in addition reduce overall child morbidity.[67] Breastfed children may have altered patterns of growth which may impact subsequent risk of breast cancer risk later in life, as reviewed later in this chapter.[60]
- A protein fraction of breast milk, MAL (multimeric alpha-lactalbumin), is a potent agent inducing apoptosis in tumor and immature cells, while sparing mature cells. This unique property of MAL has been proposed as possibly guiding the breast epithelium of the infant toward healthy maturation and away from neoplasm.[69]

The Boyd Orr cohort is one of the few cohort studies with prospective data (rather than recalled data) on breastfeeding and subsequent risk of cancer. The Boyd Orr cohort is comprised of 4,999 children who were ages 0–19 when enrolled as part of Carnegie survey of family diet and health in prewar Britain (1937–1939). In 1988, the children who had been part of the study were recontacted by a team of researchers who were then able to link their childhood experiences with adult disease outcomes. Using data from the National Health Services Central Registry, cancer outcomes from 1948 to 2003 were available on 88% of the original cohort. Martin et al. reported the association of having been breastfed with risk of total cancer, smoking-related and nonrelated cancers, as well as site-specific cancers including breast, prostate, testicular, colorectal, and gastric cancers. For each cancer site, Martin et al. also conducted a meta-analysis of data on breastfeeding and cancer using data available from other cohort and case-control studies.[57]

Analysis of cases from the Boyd Orr cohort itself showed no association between having been breastfed and total cancer incidence nor any of the individual cancers (prostate, breast, colorectal, gastric). In the meta-analyses using data pooled from other studies, there was an 18%

reduction in risk for testicular cancer and having been breastfed, though the association failed to reach statistical significance (95% CI = 0.62–1.10).[57] This result was consistent with one of the individual case-control studies included in the meta-analysis which found that men who had been breastfed greater than 6 months compared with those never breastfed had a reduction in risk of 55% of borderline statistical significance in a dose-responsive manner (95% CI: 0.41–1.04, p-trend = 0.05).[70]

Having been breastfed is associated with a lower risk of premenopausal cancer. The Boyd Orr meta-analysis of breastfeeding and premenopausal breast cancer which included nine studies and a total of 3,346 premenopausal breast cancer incident cases, showed a 12% reduction in risk for premenopausal breast cancer (95% CI: 0.79–0.98).[57] This is consistent with the majority of case-control studies, although the prospective NHS and NHS II cohorts reported a nonsignificant reduction in risk of having been breastfed and premenopausal breast cancer.[57,60,71] In contrast, the Boyd Orr meta-analysis that focused solely on postmenopausal breast cancer showed reported no association (RR: 1.0; 95% CI: 0.86–1.16) (n = 5,069)[57] and is consistent with individual studies.

There are several limitations to the research of having been breastfeeding and later risk of cancer in adulthood. First, in meta-analyses described above, subjects were born in 1874–1972 when alternatives to breastfeeding were primarily unmodified cow's milk. Preparations of infant formula have changed dramatically over time. Rates of breastfeeding have decreased as depicted in NHS cohort, in which 82% of women were breastfed as infants as compared with the younger cohort of women in NHS II, among whom only 31% report having been breastfed as an infant; there was also a decrease in exclusive breastfeeding, likely resulting in mixed feeding.[60] This raises a concern of generalizability of past research to modern day populations worldwide. Second, ascertainment of breastfeeding exposure was generally a simple yes/no question, a rather crude estimate and breastfeeding duration was ascertained in only three studies.[34] Third, timing of breastfeeding initiation is a possible important question since colostrum is dense with immunological factors, a possible mechanism of protection in childhood cancers with possible implications for disease in adulthood. Lastly, residual confounding is possible since infant feeding practices are associated with social, educational, and economical factors.

Adolescence and Breast Cancer: Critical Time Period for

Breast Carcinogenesis

Data from both animal and human studies suggest strongly that the breast is particularly sensitive to carcinogenic exposures that occur after menarche and prior to first pregnancy. In the rat, Russo and Russo have elegantly studied the underlying biology of mammary gland development that explains the phenomenon of this time-sensitive window for exposure.[72,73] The mammary gland, a specialized accessory organ of the skin, is unusual in that, unlike most other organs in the body, it develops entirely postnatally. It is the terminal end of the duct, farthest from the nipple, or terminal end bud (TEB) that undergoes growth and differentiation at the time of puberty. With the advent of menarche and the monthly surges in progesterone and estrogen, the TEB undergoes further cleavage into alveolar buds (AB). At the time of pregnancy, these AB differentiate into lobules that are capable of lactation. Pregnancy permanently alters the architecture of the mammary gland; the multiparous rat's mammary gland has virtually no TEBs and the number of lobules is four times that seen in an age-matched virgin rat.[72,73]

This change in structure is important in terms of carcinogenesis because of the concomitant change that has occurred within the epithelial cells that line these three developmentally distinct structures. The epithelial cells that line the TEB likely represent a pluripotent stem cell that can differentiate into either a myoepithelial cell or a ductal cell. Cell kinetics of TEB are quite different than their more differentiated counterparts. Compared with cells that line the AB, cells in the TEB have much higher DNA labeling index, as measured by incorporation of ^{3}H thymidine, and fewer cells in the G_0 portion of the cell cycle.[72,73] Higher rates of cell proliferation, and less time in the G_0 phase of the cell cycle allow less time for DNA repair, which may be one reason that TEB are more susceptible to carcinogenic exposures than the more differentiated structure of the AB or lobule.

In animals, one of the most reproducible ways to decrease cancer incidence is to restrict calories. The effect of caloric restriction is most pronounced for hormone-sensitive cancers. The effect of reducing calories during different age periods of the rat has been tested and restricting the peripubertal diet of the rat, as compared with the adult diet, is the most potent inhibitor of future cancer risk.[72,73] If the caloric intake of the rats is restricted early in life, the architecture of the mammary gland is permanently altered, with more terminal end buds and lower rates of proliferation in the gland overall.[74] These structural changes remain

evident into the adulthood of the rat, and persist even after pregnancy-induced differentiation. Some insight into the mechanism by which a reduction in calories also reduces risk of cancer is evident from experiments in the worm *Caenorhabditis elegans.* In *C. elegans,* reduced food induces a decrease in an IGF-1–like compound, *daf,* that results in the induction of "diapause," a hibernation-like state of the larva between the juvenile state and adulthood. The breast may also be undergoing a similar developmental arrest.[75]

Few human studies exist on the effect childhood caloric intake and risk of cancer as it is rare to have a study that has data on both childhood diet and adult cancer risk. Analysis of the data from the Boyd Orr cohort did show that for each one megajoule increase in total calories during childhood, there was a 20% increase in nonsmoking related cancers observed as an adult.[76]

Sadly, the Second World War has provided another mechanism to study the effect of war-induced famine on human health. The residents of Norway experienced a chronic reduction in calories due to rationing over the entire timeperiod of the war. Females who were adolescents during the war subsequently had a 13% reduction in risk of breast cancer.[77] A famine in the Netherlands at the end of the war, known as "the Hunger Winter," was more abrupt and severe.[78] The Dutch government in exile ordered a railroad strike to try to cripple the German army; the Nazis retaliated with a transport embargo that severely limited the transport of all food and fuel. Average caloric intake dropped to 600 kcal/d. Women exposed to the famine as children had, as expected, a later age at menarche, reduced height and earlier age at menopause, three factors shown to reduce risk of breast cancer. The effects were strongest for prepubertal children who were 2–9 years old during the Hunger Winter. However, Dutch scientists found that paradoxically the risk of breast cancer was increased, not decreased, among childhood survivors of the famine. Consistent with this increased risk of breast cancer, scientists showed that hormone levels of estrone sulfate were higher in the women exposed to the famine as children, measured 60 years postexposure. Likewise, IGF-1 was also higher among the women who had been exposed to the famine. The authors propose the immediate effects of the famine were as predicted, but that the effect of severe deprivation followed by refeeding reset hypothalamic set points, resulting in lifelong elevations in IGF-1 and steroid hormones. This resetting overrode the short-term results of famine that should have reduced risk of breast cancer and resulted in higher observed rates of breast cancer among the famine-exposed. These two

experiences of famine point out the complexity of the human organism's response to environmental exposures and how the response can modulated by dose (severity of the famine) and by susceptibility of the organism (age at exposure).

Other animal experiments, and human experiences, have highlighted how the age at which exposure occurs can influence the risk of subsequent breast cancer. For instance, chemical induction of tumors in the rat is most potent when the carcinogen is administered *after* 30 weeks of age and *before* 60 weeks of age, the time period analogous to the window between menarche and first pregnancy.[79] In humans, women who survived the atomic bombs in Japan have an elevated risk of breast cancer, but increase in risk is inversely proportional to age at the time of the bombing. Interestingly, no increase in risk at all was observed among women 35 years and older at the time of the bombing. Effects of the treatment for Hodgkin's disease is another example of the importance of age of exposure to radiation in determining subsequent risk of breast cancer. When the mediastinal lymph nodes are involved with lymphoma, Hodgkin's disease patients receive what is known as "mantle irradiation," a radiation field that includes the breast. Women who survive Hodgkin's disease have a markedly increased risk of developing breast cancer, but again, risk is dependent on age at exposure to the ionizing radiation.[80] Similar to the experience of women who were exposed in Japan to the radiation fall out of the atomic bomb, no increase in risk of breast cancer is observed among women who were over age 30 at the time of treatment, but risk rises exponentially the younger the woman was at the time of her mantle irradiation for Hodgkin's disease.

Migrant studies are another indicator of the importance of age at exposure. Migrant studies examine how risk of disease changes when an individual migrates from a country with a low incidence of disease to a country with a high incidence of disease. In the case of breast cancer, studies of Japanese immigrants (low incidence) to Hawaii (higher incidence), rates of breast cancer did not approximate those of US citizens for the immigrants themselves but only for their children and grandchildren.[81] The interpretation of this data is that the immigrant must grow up in the adopted country to "inherit" the cancer risk of the country, pinpointing again the importance of early life and adolescent exposures in breast cancer risk induction.

Childhood and Adolescent Growth and Breast Cancer

Adult height has been correlated with several adult-onset cancers, including colon, prostate, and endometrial cancer, but the association between tall stature and cancer has been most consistently observed in terms of risk of breast cancer, primarily postmenopausal breast cancer. A review of 24 prospective studies[82] found a positive relationship in all but one and since the publication of that review several more studies have confirmed the relationship.[83,84] On average, risk of breast cancer increases by 11% for each 5 cm increase in height.[83,85]

Height is the composite of leg length plus trunk length. Interestingly, leg length and trunk length, though both strongly correlated with total height, are poorly correlated with one another (r = 0.1–0.2)[84,86] and appear to be determined by different factors during childhood and at different times during childhood growth.[87] Growth immediately after birth is due in larger part to leg length than trunk growth. This is demonstrated by the changing ratio of trunk length: height over time. At birth, the ratio is around 0.66 but by puberty, this ratio has declined to 0.50.[87] After puberty, linear growth occurs proportionately in the trunk and the leg. It has been suggested therefore that separate analysis of these two components of height may help elucidate the biologic connections between growth and breast cancer.

Insight on the determinants of leg and trunk height has been gleaned from the 1946 National Birth Cohort study[86] (n = 5,362), a social-class stratified sample of all legitimate births in England, Scotland, and Wales from March 3–9, 1946 who have been followed up on 21 separate occasions between birth and age 53. Adult leg and trunk height were measured by a research nurse at home visit when the subjects were 43 years old. The most important predictor of leg length was early childhood nutrition: having been breastfed (reported at age 2) and total energy intake (reported at age 4).[86] Not living in an “overcrowded” home, a proxy for socioeconomic conditions, was also associated with longer leg length. The determinants of trunk length are less clear than those of leg length. Prolonged childhood illness (> three months or requiring hospital admission) were associated with shorter trunk length as was parental separation, whereas childhood nutrition was not associated with trunk length.

Using data from the Boyd Orr cohort, Gunnell et al. argue that the effect of stature on cancer risk is more strongly correlated with leg length; in fact, the authors report no association was observed between trunk length and risk.[88] Overall, Gunnell reports an 80% increase in risk of non-

smoking related cancers with each one z-score increase (approximately 3–4 mm) in leg length; this effect was even more pronounced in sex-hormone–dependent cancers, such as breast cancer. The association between longer leg length and breast cancer was confirmed in the French E3N cohort.[89] Further elucidation of the separate contributions of trunk length and leg length to the overall association of height and breast cancer may provide insight into the key developmental periods for induction of breast cancer risk.

Earlier age at onset of peak growth velocity and more total growth during puberty have also been associated with increased risk of breast cancer, independent of total stature.[83] A study from Denmark linked school health records in Copenhagen from 1930 to 1975 to the Danish Cancer Registry, resulting in a cohort of 117,415 women, among whom 3340 cases of breast cancer had been diagnosed. Age at peak growth was divided into quintiles. Girls whose peak growth occurred in the oldest quintile (median age 13.5 years) had a decreased risk of breast cancer (RR 0.84) compared with those whose peak growth occurred in the youngest quintile (median age: 10.4 years). Earlier age at peak growth velocity is associated with earlier onset of menarche and therefore more prolonged exposure of the never-pregnant and thus undifferentiated breast to the higher levels of estrogens postpuberty. In the Danish study, age at menarche was a significant predictor of subsequent risk of breast cancer in univariate analysis but not when age at peak growth velocity was also included in the model, indicating that although both are likely a measure of onset of puberty, it is rapid early growth that appears to confer risk. Larger increases in height during puberty (between ages 8 and 14) conferred an increased risk of breast cancer; whereas total height accrued prior to age 8 was not significantly associated with risk of breast cancer.

Adolescent Diet and Breast Cancer

Concern that the period of adolescence is a key and critical period for the induction of breast cancer has lead investigators to also study adolescent diet and breast cancer. The study of adolescent diet and breast cancer is challenging in that it is usually based on recall of diet from the past, which undermines its accuracy due to measurement error.[90] In addition, in the situation where the recall of adolescent diet is collected after the diagnosis of breast cancer, there is the potential for recall bias.

In the NHS and the NHS II, the reproducibility of the recall of adolescent diet has been assessed and effort made to estimate validity of

diet recalled from 15 to 30 years previously.[91,92] In NHS, a short 24-item food frequency questionnaire (FFQ) was added to the main questionnaire in 1986. In NHS II, participants were asked on the main questionnaire if they would be willing to fill out a supplemental questionnaire on adolescent diet. The 124-item supplemental adolescent diet FFQ was completed by 45,947 NHS II participants in 1998. In both cohorts, a subset of the nurse participants who had completed the initial FFQ on adolescent diet were asked to complete a second survey 4–8 years later. In both cohorts, the reproducibility of recall of both foods and nutrients was reasonable (r = 0.5–0.6 [NHS] and r = 0.6–0.7 [NHS II]). The mothers of the participants from the NHS II were also asked to recall their daughters' diets as adolescents. The correlation between the report of the mother versus the daughter was moderate (r = 0.4) but implies some modicum of validity especially when one acknowledges that the mother of an adolescent no longer has the control over diet that a mother would have had at an earlier age, and therefore she may have been unaware of what the daughter was eating outside the home. As an additional approach to assessing the validity of the report, the basal metabolic rate of the NHS II participant as an adolescent was calculated on the basis of her reported weight at age 18. This predicted basal metabolic rate was then used to calculate expected total energy intake. The expected energy intake was compared with the reported energy intake. Participants were categorized as under-, over-, or acceptable reporters. Correlation between the two reports of adolescent diet did not differ by reporter status. In addition, in both cohorts, current adult diet was minimally correlated (r = 0.2) with report of adolescent diet which implies minimal confounding by the content of the current diet. Finally, foods and nutrients were ranked into quintiles. 80% of the reports on the two surveys of adolescent diet were within one quintile of each other. These data in sum show that recall of adolescent diet using a FFQ provides a reasonable estimate, given the many limitations of the method.

In the NHS II, vegetable fat was also observed to be protective to the development of breast cancer (RR 0.58; 95% CI 0.38–0.86) as was Vitamin E (RR 0.61; 95% CI 0.42–0.89).[91] Vitamin E is a fat-soluble vitamin, and vegetable fat (principally peanut butter) is the major source of vitamin E in the adolescent diet of the NHS II participant. In addition, in the analysis of the adolescent diet of the NHS II participant, foods with a higher glycemic index, which are foods that invoke a steeper rise in blood sugar and therefore a compensatory rise in insulin and insulin-like growth factors, were associated with an increase in risk of breast cancer (RR 1.47;

95% CI 1.04–2.08). In the NHS, a significant protective effect was also observed for higher dietary intake of vegetable fat (RR 0.85) and risk of breast cancer.[93]

Red Meat Consumption in Adolescence and Risk of Breast Cancer.

Several case-control studies have examined the issue of adolescent red meat consumption and breast cancer. In a case-control study using cases identified through the SEER registry in three sites, Potischman et al. found a borderline effect for high-fat meats.[94] Hislop et al. conducted a case-control study in British Columbia among 846 incident cases of breast cancer between 1980 and 1982.[95] Women were asked to recall dietary intake of 31 foods during early childhood (up to age 13). Among premenopausal women, consumption of meat with visible fat during childhood was associated with increased risk of breast cancer (RR for "three or more times per week" compared with "less than once per month" = 2.69; 95% CI 1.26–5.80). A more recent analysis of premenopausal women in NHS II showed that increased levels of red meat consumed during adolescence was associated with a significant linear trend for premenopausal breast cancer risk; this risk was most pronounced for estrogen-positive tumors.[96] Supporting the possible role of red meat in breast cancer etiology, a case-control study of Chinese immigrant women reported that high consumption of red meat during adolescence, but not during adulthood, was associated with a significantly higher adult mammographic breast density.[97]

In another case-control study that used a limited version of the Bloch FFQ to assess only foods high in fat and fiber, Pryor et al. reported that higher intake of fat from dairy sources was associated with decreased risk of breast cancer for both pre- and postmenopausal women.[98] In the same study, higher intake of fiber decreased risk among premenopausal women, whereas higher intake of fiber was associated with an increased risk of breast cancer among postmenopausal women. Consumption of vegetable oils was associated with reduced risk among premenopausal women (RR = 0.48; 95% CI 0.25–0.89), analogous to the findings from the NHS and NHS II.

Adolescent Soy Intake and Breast Cancer

Several recent studies have focused on adolescent soy intake and breast cancer. Phytoestrogens have been shown in animal models to promote

differentiation of the mammary gland by enhancing the transition from terminal end buds to AB or lobules.[99] All four case-control studies to date have shown a protective effect for adolescent soy intake,[100–103] which was especially pronounced in the two studies that compared adolescent versus adult intake. In one study those individuals who indicated high intake in both time periods had 50% reduction in risk of breast cancer,[101] whereas in the other study, the greatest reduction in risk was associated with consumption during childhood (OR = 0.42; 95% CI 0.20–0.90).[102]

Adolescent Diet and Benign Breast Disease

Benign breast disease (BBD) is catch-all phrase that includes several different types of pathologic lesions, some with no relation to breast cancer and some that dramatically heighten risk. Non-proliferative BBD has no bearing on subsequent risk of breast cancer, whereas women with proliferative lesions without atypia have 1.5- to 2-fold great risk; women who have proliferative lesions with atypia have a 3.5- to 5-fold greater risk of breast cancer.[104] A mathematical model of breast cancer also has shown that BBD confers an increased risk of breast cancer, even when adult risk factors are included in the model, suggesting that it is early life events that determine risk. Therefore, proliferative BBD is considered an intermediate marker of risk for breast cancer. That reflects adolescent exposures in particular.

In an analysis from the NHS, a retrospective analysis of diet during adolescence and subsequent risk of proliferative BBD showed that higher intakes of vegetable fat, vitamin E and fiber during adolescence were inversely related to risk. In addition, it was shown that whereas recent alcohol did not affect risk of BBD, remote consumption of alcohol between ages 18 and 22 increased risk for both proliferative and nonproliferative BBD.[105]

Childhood Diet and Age at Menarche

Earlier age at menarche is a strong risk factor for breast cancer. In fact, it has been estimated that risk of breast cancer increases by 4% for each yearly decrease in age at menarche.[106] Earlier age at menarche is also a risk factor for endometrial cancer. Examinations of the dietary predictors of age at menarche have been sparse. Most dietary hypotheses have had null results.[107–109] A study in Germany showed that energy-adjusted fat intake was associated with accelerated menarche,[110] but this finding was not replicated in a study in Canada.[110] In fact, the Canadian study reported

that higher intake of monounsaturated fat was associated with a delay in menarche. Several studies have shown an association between higher intakes of dietary fiber and later age at menarche.[111–114]

SUMMARY

Data from a variety of sources support the notion that diet during childhood, beginning in utero and extending through adolescence, has an impact on the incidence of childhood as well as adult malignancies. Some of the evidence is based on the relationship between childhood growth and cancer; both abnormal growth patterns as in the Beckwith–Wiedemann syndrome, and "normal" growth, for instance, the relationship between birth weight and childhood ALL or height and breast cancer. Other studies have been able to pinpoint foods or nutrients that may impact risk of cancer, such as nitrate-containing meats and pediatric brain tumors, periconceptual folate and Wilms' tumor, or topoisomerase II inhibitors and infant AML. Timing of the exposure is a key variable; for instance, topoisomerase II inhibitors are not implicated in the genesis of AML that develops later in childhood and vegetable oil has not been found to have an effect on rates of breast cancer when consumed in the adult diet, only in the adolescent diet. The availability of validated tools such as the FFQ that allow diet to be measured efficiently and accurately in prospective large-scale, population-based studies will allow for the ascertainment of further relationships between diet and cancer, avoiding the inherent recall bias of case-control studies. Finally, the interaction between nutrition, genes, and cancer will become more interesting as the inter-relationships are elucidated and we understand more about how diet interacts with genetic susceptibility.

REFERENCES

1. Doll R, Peto R. *The Causes of Cancer*. New York, NY: Oxford University Press; 1981.
2. Wiemels JL, Cazzaniga G, Daniotti M, et al. Prenatal origin of acute lymphoblastic leukaemia in children. *Lancet*. 1999;354:1499–1503.
3. Gale KB, Ford AM, Repp R, et al. Backtracking leukemia to birth: identification of clonotypic gene fusion sequences in neonatal blood spots. *Proc Natl Acad Sci U S A*. 1997;94:13950–13954.
4. Ross JA. High birthweight and cancer: evidence and implications. *Cancer Epidemiol Biomarkers Prev*. 2006;15:1–2.
5. Heuch JM, Heuch I, Akslen LA, Kvale G. Risk of primary childhood brain tumors related to birth characteristics: a Norwegian prospective study. *Int J Cancer*.

1998;77:498–503.
6. Emerson JC, Malone KE, Daling JR, Starzyk P. Childhood brain tumor risk in relation to birth characteristics. *J Clin Epidemiol*. 1991;44:1159–1166.
7. Von Behren J, Reynolds P. Birth characteristics and brain cancers in young children. *Int J Epidemiol*. 2003;32:248–256.
8. Mogren I, Malmer B, Tavelin B, Damber L. Reproductive factors have low impact on the risk of different primary brain tumours in offspring. *Neuroepidemiology*. 2003;22:249–254.
9. Hjalgrim LL, Westergaard T, Rostgaard K, et al. Birth weight as a risk factor for childhood leukemia: a meta-analysis of 18 epidemiologic studies. *Am J Epidemiol*. 2003; 158:724–735.
10. Yeazel MW, Ross JA, Buckley JD, et al. High birth weight and risk of specific childhood cancers: a report from the Children's Cancer Group. *J Pediatr*. 1997;131:671–677.
11. Hjalgrim LL, Rostgaard K, Hjalgrim H, et al. Birth weight and risk for childhood leukemia in Denmark, Sweden, Norway, and Iceland. *J Natl Cancer Inst*. 2004;96:1549–1556.
12. Roman E, Lightfoot T, Smith AG, et al. Childhood acute lymphoblastic leukaemia and birthweight: insights from a pooled analysis of case-control data from Germany, the United Kingdom and the United States. *Eur J Cancer*. 2013;49(6):1437–1447. doi: 10.1016/j.ejca.2012.11.017.
13. Ognjanovic S, Carozza SE, Chow EJ, et al. Birth characteristics and the risk of childhood rhabdomyosarcoma based on histological subtype. *Br J Cancer*. 2010;102(1):227–231.
14. Chu A, Heck JE, Ribeiro KB, et al. Wilms' tumour: a systematic review of risk factors and meta-analysis. Paediatr Perinat Epidemiol. 2010; 24(5):449–469.
15. Schüz J, Schmidt LS, Kogner P, et al. Birth characteristics and Wilms tumors in children in the Nordic countries: a register-based case-control study. *Int J Cancer*. 2011;128(9):2166–2173.
16. Dorak MT, Pearce MS, Hammal DM, et al. Examination of gender effect in birth weight and miscarriage associations with childhood cancer (United Kingdom). *Cancer Causes Control*. 2007;18:219–228.
17. Dorak MT, Burnett AK, Worwood M, et al. The C282Y mutation of HFE is another male-specific risk factor for childhood acute lymphoblastic leukemia. *Blood*. 1999;94:3957.
18. Barker DJ. Growth in utero and coronary heart disease. *Nutr Rev*. 1996;54:S1–S7.
19. Ross JA, Potter JD, Robison LL. Infant leukemia, topoisomerase II inhibitors, and the MLL gene. *J Natl Cancer Inst*. 1994;86:1678–1680.
20. Spector LG, Xie Y, Robison LL, et al. Maternal diet and infant leukemia: the DNA topoisomerase II inhibitor hypothesis: a report from the children's oncology group. *Cancer Epidemiol Biomarkers Prev*. 2005;14:651–655.
21. Preston-Martin S, Yu MC, Benton B, Henderson BE. N-Nitroso compounds and childhood brain tumors: a case-control study. *Cancer Res*. 1982;42:5240–5245.
22. Kuijten RR, Bunin GR, Nass CC, Meadows AT. Gestational and familial risk factors for childhood astrocytoma: results of a case-control study. *Cancer Res*. 1990;50:2608–2612.
23. Huncharek M, Kupelnick B. A meta-analysis of maternal cured meat consumption during pregnancy and the risk of childhood brain tumors. *Neuroepidemiology*. 2004;23:78–84.
24. Preston-Martin S, Pogoda JM, Mueller BA, et al. Maternal consumption of cured meats and vitamins in relation to pediatric brain tumors. *Cancer Epidemiol*

Biomarkers Prev. 1996;5:599–605.
25. Goh YI, Koren G. Prenatal supplementation with multivitamins and the incidence of pediatric cancers: clinical and methodological considerations. *Pediatr Blood Cancer*. 2008;50(2 suppl):487–489; discussion 498.
26. Goh YI, Bollano E, Einarson TR, Koren G. Prenatal multivitamin supplementation and rates of congenital anomalies: a meta-analysis. *J Obstet Gynaecol Can*. 2006;28(8):680–689.
27. Ross JA, Blair CK, Olshan AF, et al. Periconceptional vitamin use and leukemia risk in children with Down syndrome: a Children's Oncology Group study. *Cancer*. 2005;104:405–410.
28. French AE, Grant R, Weitzman S, et al. Folic acid food fortification is associated with a decline in neuroblastoma. *Clin Pharmacol Ther*. 2003;74:288–294.
29. Linabery AM, Johnson KJ, Ross JA. Childhood cancer incidence trends in association with US folic acid fortification (1986-2008). *Pediatrics*. 2012;129(6):1125–1133.
30. Petridou E, Ntouvelis E, Dessypris N, et al. Maternal diet and acute lymphoblastic leukemia in young children. *Cancer Epidemiol Biomarkers Prev*. 2005;14:1935–1939.
31. Bunin GR, Kushi LH, Gallagher PR, et al. Maternal diet during pregnancy and its association with medulloblastoma in children: a children's oncology group study (United States). *Cancer Causes Control*. 2005;16:877–891.
32. Orjuela MA, Titievsky L, Liu X, et al. Fruit and vegetable intake during pregnancy and risk for development of sporadic retinoblastoma. *Cancer Epidemiol Biomarkers Prev*. 2005;14:1433–1440.
33. Trichopoulos D. Hypothesis: does breast cancer originate in utero? *Lancet*. 1990;335:939–940.
34. Potischman N, Troisi R. In-utero and early life exposures in relation to risk of breast cancer. *Cancer Causes Control*. 1999;10:561–573.
35. Michels KB, Xue F. Role of birthweight in the etiology of breast cancer. *Int J Cancer*. 2006;119:2007–2025.
36. de Assis S, Khan G, Hilakivi-Clarke L. High birth weight increases mammary tumorigenesis in rats. *Int J Cancer*. 2006;119:1537–1546.
37. Michels KB, Xue F, Terry KL, Willett WC. Longitudinal study of birthweight and the incidence of breast cancer in adulthood. *Carcinogenesis*. 2006;27:2464–2468.
38. Nilsen TI, Romundstad PR, Troisi R, Vatten LJ. Birth size and subsequent risk for prostate cancer: a prospective population-based study in Norway. *Int J Cancer*. 2005;113:1002–1004.
39. Tibblin G, Eriksson M, Cnattingius S, Ekbom A. High birthweight as a predictor of prostate cancer risk. *Epidemiology*. 1995;6:423–424.
40. Platz EA, Giovannucci E, Rimm EB, et al. Retrospective analysis of birth weight and prostate cancer in the Health Professionals Follow-up Study. *Am J Epidemiol*. 1998;147:1140–1144.
41. Ekbom A, Hsieh CC, Lipworth L, et al. Perinatal characteristics in relation to incidence of and mortality from prostate cancer. *BMJ*. 1996;313:337–341.
42. Ekbom A, Wuu J, Adami HO, et al. Duration of gestation and prostate cancer risk in offspring. *Cancer Epidemiol Biomarkers Prev*. 2000;9:221–223.
43. Rasmussen F, Gunnell D, Ekbom A, et al. Birth weight, adult height, and testicular cancer: cohort study of 337,249 Swedish young men. *Cancer Causes Control*. 2003;14:595–598.
44. Moller H, Skakkebaek NE. Testicular cancer and cryptorchidism in relation to prenatal factors: case-control studies in Denmark. *Cancer Causes Control*.

1997;8:904–912.
45. Akre O, Ekbom A, Hsieh CC, et al. Testicular nonseminoma and seminoma in relation to perinatal characteristics. *J Natl Cancer Inst*. 1996;88:883–889.
46. Brown LM, Pottern LM, Hoover RN. Prenatal and perinatal risk factors for testicular cancer. *Cancer Res*. 1986;46:4812–4816.
47. Nilsen TI, Romundstad PR, Troisi R, et al. Birth size and colorectal cancer risk: a prospective population based study. *Gut*. 2005;54:1728–1732.
48. Sandhu MS, Luben R, Day NE, Khaw KT. Self-reported birth weight and subsequent risk of colorectal cancer. *Cancer Epidemiol Biomarkers Prev*. 2002;11:935–938.
49. Siskind V, Green A, Bain C, Purdie D. Breastfeeding, menopause, and epithelial ovarian cancer. *Epidemiology*. 1997;8:188–191.
50. Lipworth L, Bailey LR, Trichopoulos D. History of breast-feeding in relation to breast cancer risk: a review of the epidemiologic literature. *J Natl Cancer Inst*. 2000;92:302–312.
51. Davis MK. Review of the evidence for an association between infant feeding and childhood cancer. *Int J Cancer*. 1998;78:29–33.
52. Parker L. Breast-feeding and cancer prevention. *Eur J Cancer*. 2001;37:155–158.
53. Shu XO, Linet MS, Steinbuch M, et al. Breast-feeding and risk of childhood acute leukemia. *J Natl Cancer Inst*. 1999;91:1765–1772.
54. Davis MK, Savitz DA, Graubard BI. Infant feeding and childhood cancer. *Lancet*. 1988;2:365–368.
55. Bener A, Denic S, Galadari S. Longer breast-feeding and protection against childhood leukaemia and lymphomas. *Eur J Cancer*. 2001;37:234–238.
56. Schwartzbaum JA, George SL, Pratt CB, Davis B. An exploratory study of environmental and medical factors potentially related to childhood cancer. *Med Pediatr Oncol*. 1991;19:115–121.
57. Martin RM, Middleton N, Gunnell D, et al. Breast-feeding and cancer: the Boyd Orr cohort and a systematic review with meta-analysis. *J Natl Cancer Inst*. 2005;97:1446–1457.
58. Bittner J. Some possible effects of nursing on the mammary gland tumor incidence in mice. *Science*. 1936;84:162.
59. Titus-Ernstoff L, Egan KM, Newcomb PA, et al. Exposure to breast milk in infancy and adult breast cancer risk. *J Natl Cancer Inst*. 1998;90:921–924.
60. Michels KB, Trichopoulos D, Rosner BA, et al. Being breastfed in infancy and breast cancer incidence in adult life: results from the two nurses' health studies. *Am J Epidemiol*. 2001;153:275–283.
61. Wang Y, Holland JF, Bleiweiss IJ, et al. Detection of mammary tumor virus env gene-like sequences in human breast cancer. *Cancer Res*. 1995;55:5173–5179.
62. Levine PH, Mesa-Tejada R, Keydar I, et al. Increased incidence of mouse mammary tumor virus-related antigen in Tunisian patients with breast cancer. *Int J Cancer*. 1984;33:305–308.
63. Jensen A, Slorach SA, eds. *Chemical Contaminants in Human Milk*. Boca Roton, FL: CRC Press; 1991.
64. Nickerson K. Environmental contaminants in breast milk. *J Midwifery Womens Health*. 2006;51:26–34.
65. Dewailly E, Yotte PA, Brisson J. Protective effect of breast feeding on breast cancer and body burden. *J Natl Cancer Inst*. 1994;86:603.
66. Rogan W, Ragan N. Chemical contaminants, pharmacokinetics, and lactating mother. *Environ Health Perspect*. 1994;102 (suppl 11):89–95.
67. Chantry CJ, Howard CR, Auinger P. Full breastfeeding duration and associated

decrease in respiratory tract infection in US children. *Pediatrics*. 2006;117:425–432.
68. Freudenheim JL, Marshall JR, Graham S, et al. Exposure to breastmilk in infancy and the risk of breast cancer. *Epidemiology*. 1994;5:324–331.
69. Hakansson A, Zhivotovsky B, Orrenius S, et al. Apoptosis induced by a human milk protein. *Proc Natl Acad Sci U S A*. 1995;92:8064–8068.
70. Coupland CA, Forman D, Chilvers CE, et al. Maternal risk factors for testicular cancer: a population-based case-control study (UK). *Cancer Causes Control*. 2004;15:277–283.
71. Barba M, McCann SE, Nie J, et al. Perinatal exposures and breast cancer risk in the Western New York Exposures and Breast Cancer (WEB) Study. *Cancer Causes Control*. 2006;17:395–401.
72. Russo IH, Russo J. Mammary gland neoplasia in long-term rodent studies. *Environ Health Perspect*. 1996;104:938–967.
73. Russo J, Russo IH. Toward a physiological approach to breast cancer prevention. *Cancer Epidemiol Biomarkers Prev*. 1994;3:353–364.
74. Engelman RW, Day NK, Good RA. Calorie intake during mammary development influences cancer risk: lasting inhibition of C3H/HeOu mammary tumorigenesis by peripubertal calorie restriction. *Cancer Res*. 1994;54:5724–5730.
75. Dillin A, Crawford DK, Kenyon C. Timing requirements for insulin/IGF-1 signaling in C. elegans. *Science.* 2002;298:830–834.
76. Frankel S, Gunnell DJ, Peters TJ, et al. Childhood energy intake and adult mortality from cancer: the Boyd Orr Cohort Study. *BMJ*. 1998;316:499–504.
77. Tretli S, Gaard M. Lifestyle changes during adolescence and risk of breast cancer: an ecologic study of the effect of World War II in Norway. *Cancer Causes Control*. 1996;7:507–512.
78. van Noord PAH. Breast cancer and the brain: a neurodevelopmental hypothesis to explain the opposing effects of caloric deprivation during the Dutch famine of 1944-1945 on breast cancer and its risk factors. *J Nutr*. 2004;134:3399S–3406S.
79. Ariazi JL, Haag JD, Lindstrom MJ, Gould MN. Mammary glands of sexually immature rats are more susceptible than those of mature rats to the carcinogenic, lethal, and mutagenic effects of N-nitroso-N-methylurea. *Mol Carcinog*. 2005;43:155–164.
80. Hancock SL, Tucker MA, Hoppe RT. Breast cancer after treatment of Hodgkin's disease. *J Natl Cancer Inst*. 1993;85:25–31.
81. Ziegler RG, Hoover RN, Pike MC, et al. Migration patterns and breast cancer risk in Asian-American women. *J Natl Cancer Inst*. 1993;85:1819–1827.
82. Gunnell D, Okasha M, Smith GD, et al. Height, leg length, and cancer risk: a systematic review. *Epidemiol Rev*. 2001;23:313–342.
83. Ahlgren M, Melbye M, Wohlfahrt J, Sorensen TI. Growth patterns and the risk of breast cancer in women. *N Engl J Med*. 2004;351:1619–1626.
84. Lawlor DA, Okasha M, Gunnell D, et al. Associations of adult measures of childhood growth with breast cancer: findings from the British Women's Heart and Health Study. *Br J Cancer*. 2003;89:81–87.
85. Tretli S. Height and weight in relation to breast cancer morbidity and mortality. A prospective study of 570,000 women in Norway. *Int J Cancer*. 1989;44:23–30.
86. Wadsworth ME, Hardy RJ, Paul AA, et al. Leg and trunk length at 43 years in relation to childhood health, diet and family circumstances; evidence from the 1946 national birth cohort. *Int J Epidemiol*. 2002;31:383–390.
87. Gunnell D. Can adult anthropometry be used as a 'biomarker' for prenatal and childhood exposures? *Int J Epidemiol*. 2002;31:390–394.

88. Gunnell DJ, Smith GD, Holly JM, Frankel S. Leg length and risk of cancer in the Boyd Orr cohort. *BMJ*. 1998;317:1350–1351.
89. Fagherazzi G, Vilier A, Boutron-Ruault M, et al. Height, sitting height, and leg length in relation with breast cancer risk in the E3N cohort. *Cancer Epidemiol Biomarkers Prev*. 2012;21:1171–1175.
90. Friedenreich CM, Orenstein MR. Physical activity and cancer prevention: etiologic evidence and biological mechanisms. *J Nutr*. 2002;132:3456S–3464S.
91. Frazier AL, Willett WC, Colditz GA. Reproducibility of recall of adolescent diet: Nurses' Health Study (United States). *Cancer Causes Control*. 1995;6:499–506.
92. Maruti SS, Feskanich D, Colditz GA, et al. Adult recall of adolescent diet: reproducibility and comparison with maternal reporting. *Am J Epidemiol*. 2005;161:89–97.
93. Frazier AL, Ryan CT, Rockett H, et al. Adolescent diet and risk of breast cancer. *Breast Cancer Res*. 2003;5:R59–R64.
94. Potischman N, Weiss HA, Swanson CA, et al. Diet during adolescence and risk of breast cancer among young women. *J Natl Cancer Inst*. 1998;90:226–233.
95. Hislop TG, Coldman AJ, Elwood JM, et al. Childhood and recent eating patterns and risk of breast cancer. *Cancer Detect Prev*. 1986;9:47–58.
96. Linos E, Willett WC, Cho E, Frazier L. Adolescent diet in relation to breast cancer risk among premenopausal women. *Cancer Epidemiol Biomarkers Prev*. 2010;19:689–696.
97. Tseng M, Olufade TO, Evers KA, Byrne C. Adolescent lifestyle factors and adult breast density in U.S. Chinese immigrant women. *Nutr Cancer*. 2011;63(3):342–349.
98. Pryor M, Slattery ML, Robison LM, Egger M. Adolescent diet and breast cancer in Utah. *Cancer Res*. 1989;49:2161–2167.
99. Hilakivi-Clarke L, Onojafe I, Raygada M, et al. Prepubertal exposure to zearalenone or genistein reduces mammary tumorigenesis. *Br J Cancer*. 1999;80:1682–1688.
100. Shu XO, Jin F, Dai Q, et al. Soyfood intake during adolescence and subsequent risk of breast cancer among Chinese women. *Cancer Epidemiol Biomarkers Prev*. 2001;10:483–488.
101. Wu AH, Wan P, Hankin J, et al. Adolescent and adult soy intake and risk of breast cancer in Asian-Americans. *Carcinogenesis*. 2002;23:1491–1496.
102. Korde LA, Wu AH, Fears T, et al. Childhood soy intake and breast cancer risk in Asian American women. *Cancer Epidemiol Biomarkers Prev*. 2009; 18(4):1050–1059.
103. Thanos J, Cotterchio M, Boucher BA, et al. Adolescent dietary phytoestrogen intake and breast cancer risk (Canada). *Cancer Causes Control*. 2006;17:1253–1261.
104. Schnitt SJ, Connolly JL. *Pathology of Benign Breast Disorders. Diseases of the Breast*. 2nd ed. In: Harris JR, Lippmann ME, Osborne CK, eds. Philadelphia, PA: Lippincott Williams & Wilkins; 2000.
105. Baer HJ, Schnitt SJ, Connolly JL, et al. Early life factors and incidence of proliferative benign breast disease. *Cancer Epidemiol Biomarkers Prev*. 2005;14:2889–2897.
106. Kvale G, Heuch I. Menstrual factors and breast cancer risk. *Cancer*. 1988;62:1625–1631.
107. Maclure M, Travis LB, Willett W, MacMahon B. A prospective cohort study of nutrient intake and age at menarche. *Am J Clin Nutr*. 1991;54:649–656.
108. Koprowski C, Ross RK, Mack WJ, et al. Diet, body size and menarche in a

multiethnic cohort. *Br J Cancer*. 1999;79:1907–1911.
109. Meyer F, Moisan J, Marcoux D, Bouchard C. Dietary and physical determinants of menarche. *Epidemiology*. 1990;1:377–381.
110. Merzenich H, Boeing H, Wahrendorf J. Dietary fat and sports activity as determinants for age at menarche. *Am J Epidemiol*. 1993;138:217–224.
111. Koo MM, Rohan TE, Jain M, et al. A cohort study of dietary fibre intake and menarche. *Public Health Nutr*. 2002;5:353–360.
112. Soriguer FJ, Gonzalez-Romero S, Esteva I, et al. Does the intake of nuts and seeds alter the appearance of menarche? *Acta Obstet Gynecol Scand*. 1995;74:455–461.
113. Hughes RE, Jones E. Intake of dietary fibre and the age of menarche. *Ann Hum Biol*. 1985;12:325–332.
114. de Ridder CM, Thijssen JH, Van't Veer P, et al. Dietary habits, sexual maturation, and plasma hormones in pubertal girls: a longitudinal study. *Am J Clin Nutr*. 1991;54:805–813.
115. Kaijser M, Akre O, Cnattingius S, Ekbom A. Preterm birth, birth weight, and subsequent risk of female breast cancer. *Br J Cancer*. 2003;89:1664–6.
116. De Stavola BL, dos Santos Silva I, McCormack V, et al. Childhood growth and breast cancer. *Am J Epidemiol*. 2004;159:671–82.
117. Vatten LJ, Nilsen TI, Tretli S, et al. Size at birth and risk of breast cancer: prospective population-based study. *Int J Cancer*. 2005 Apr 10;114:461–4
118. McCormack VA, dos Santos Silva I, Koupil I, et al. Birth characteristics and adult cancer incidence: Swedish cohort of over 11,000 men and women. *Int J Cancer*. 2005;115:611–7
119. Le Marchand L, Kolonel LN, Earle ME, Mi MP. Body size at different periods of life and breast cancer risk. *Am J Epidemiol*.1988;128:137–52
120. Sanderson M, Williams MA, Malone KE, et al. Perinatal factors and risk of breast cancer. *Epidemiology*. 1996;7:34–7
121. Troy LM, Michels KB, Hunter DJ, et al. Self-reported birthweight and history of having been breastfed among younger women: an assessment of validity. *Int J Epidemiol*. 1996;25:122–7
122. Sanderson M, Williams MA, Daling JR, et al. Maternal factors and breast cancer risk among young women. *Paediatr Perinat Epidemiol*. 1998;12:397–407.
123. Innes K, Byers T, Schymura M. Birth characteristics and subsequent risk for breast cancer in very young women. *Am J Epidemiol*. 2000;152:1121–8.
124. Kaijser M, Lichtenstein P, Granath F, et al. In utero exposures and breast cancer: a study of opposite-sexed twins. *J Natl Cancer Inst*. 2001;93:60–2.
125. Mellemkjaer L, Olsen ML, Sørensen HT, Thulstrup AM, et al. Birth weight and risk of early-onset breast cancer (Denmark). *Cancer Causes Control*. 2003;14:61-4

Cancer Treatment

58

Lori J. Bechard, PhD, MEd, RD, LDN and Christopher Duggan, MD, MPH

INTRODUCTION

Nutritional care for children with cancer is an essential component of treatment. Changes in nutrient metabolism, dietary intake, and body composition[1] occur in pediatric cancer patients as a result of disease and subsequent treatment. These changes can obscure nutritional deficits for growing children. Nutritional assessment of the child with cancer is important for identifying and treating malnutrition, and for ensuring the appropriate provision of nutrients to promote recovery and long-term health. Deviations in normal growth patterns may occur as a result of the disease, treatment, or other underlying causes. Being overweight or underweight has been associated with greater treatment-related mortality.[2]

Chronic health conditions, many of which have a relationship with nutritional intake, are common among survivors of pediatric cancer.[3] Obesity, hyperlipidemia, and diabetes combined with risk for subsequent cancers reveal the importance of nutrition beyond the acute stages of treatment. Families of children with cancer may be especially concerned with weight[4] and nutritional therapy, for both the important psychosocial aspects of eating and the potential for synergistic intervention with immediate and long-term medical therapies. We review the metabolic effects of pediatric cancers and their treatment, highlighting the nutritional conditions and interventions encountered by children from diagnosis to long-term survival. The role of nutrition in the prevention of cancer is described in Chapter 57.

PREVALENCE OF MALNUTRITION IN

PEDIATRIC CANCER

Among children from infancy to age 14, cancer is the leading cause of death by disease in the United States.[5] The incidence of childhood cancer in the United States during 2007-2009 was 17.2 cases per 100,000 children under the age of 20 years; the most common pediatric malignancy was acute lymphocytic leukemia (ALL), followed by brain and other central nervous system (CNS) tumors.[5] Although US pediatric cancer incidence has increased slightly over the past 30 years, mortality rates have declined by more than 50% for many childhood cancers, and the five-year survival rate for all pediatric cancers has increased from 63% in the 1970s to 83% in 2012.[5]

Malnutrition prevalence at presentation and during treatment varies with underlying history, host susceptibility, tumor type and location, and treatment regimen.[1] Relapsed malignant disease, hematopoietic stem cell transplantation (HSCT); tumors of the abdomen, head, or neck; and other advanced-stage solid tumors are clinically associated with poorer nutritional status than lower risk diseases. In general, children with solid tumors have a greater incidence of malnutrition.[6] Malnutrition is more prevalent among children with cancer in developing countries, which may impact survival rates in these areas.[7] High rates of obesity have been observed during and following treatment for acute leukemia.[8] A variety of pediatric cancer diagnoses can be grouped according to their nutritional risk (Table 58-1).

Weight loss may be considered the final step in a long process of nutritional perturbation in patients with cancer, and as such it may not be a useful indicator of malnutrition. The use of body mass index (BMI) as a marker of nutritional status may be inaccurate due to its inability to distinguish changes in lean body mass. Children undergoing treatment for cancer have reduced body cell mass and increased fat mass compared to control subjects, and these changes do not correlate well with BMI.[9] Overt signs of malnutrition do not typically present at diagnosis with leukemia, although elevations in cytokine antagonists and reductions in plasma proteins, compared to controls, have been observed, suggestive of an early malnourished state.[10] Although measures of body weight are commonly used to identify nutritional problems, the weight of tumor masses may obscure changes in lean body mass. A study of Canadian children diagnosed with cancer found that measurements of midarm circumference correlated better with lean body mass, as assessed by dual-energy X-ray absorptiometry (DXA), than BMI.[11] Lean body mass declines substantially

with the use of chemotherapy.[12] Assessment of the nutritional status of children with cancer should include consideration of changes in body composition that may result from disease, treatment, or both.

TABLE 58-1 Nutritional Risk Groupings of Pediatric Malignancies and Associated Conditions

- High risk
  - Relapsed disease
  - High-risk ALL
  - Acute myelogenous leukemia
  - Advanced-stage solid tumors
    - Neuroblastoma
    - Wilm's tumor
    - Lymphoma (Hodgkin's and non-Hodgkin's)
    - Hepatoblastoma
    - Medulloblastoma
    - Ewing's sarcoma
    - Osteosarcoma
  - Tumors involving the head, neck, or abdomen
  - Hemophagocytic lymphohistiocytosis
  - HSCT
- Low risk
  - Standard-risk leukemia
  - Early-stage solid tumors
- Obesity risk
  - ALL
  - Brain tumors with hypothalamic damage
  - Treatment including extended courses of corticosteroids

Cancer Cachexia

Wasting is defined as involuntary weight loss and is found in many patients with chronic disease, including cancer, kidney disease, cardiac disease, and advanced HIV infection. Overall loss of body weight, including fat and fat-free mass, occurs as a result of the energy imbalance caused by disease. Cachexia, in contrast with wasting, is defined as involuntary loss of lean body mass with or without overall weight loss. In this scenario, patients of normal or even excessive weight can still be malnourished due to diminished lean body mass. This loss of muscle is known to negatively influence recovery and reduce survival in patients with cardiac disease and cancer.[13] Because this decline in lean body mass has much functional and prognostic significance, this type of malnutrition is important to identify.

Differential loss of body fat versus fat-free mass implies a different

etiology and prognosis of malnutrition than simple starvation. Prolonged fasting typically leads to a predictable decrement first in body fat and then in body protein stores. In this case, energy repletion is usually successful with the careful provision of adequate energy, protein, and micronutrients.

In contrast, children with cancer experience reductions in fat-free mass both at diagnosis and during treatment, and some have concomitant increases in fat mass.[1] These changes are significant, because loss of lean body mass is associated with important functional changes including loss of strength, decreased immune function, decreased pulmonary function, increased disability, and death.

METABOLIC EFFECTS OF CANCER

The presence of malignancy results in several important changes in macronutrient metabolism, and these metabolic alterations can in turn affect energy balance. Metabolic effects of cancer are variable and depend in part on tumor types and sizes, chemotherapies used in treatment, as well as several host factors, including nutritional status. However, a general understanding of these aberrations is useful in anticipating potential metabolic complications as well as planning nutritional therapy.

Carbohydrate

Multiple metabolic shifts in glucose homeostasis tend to increase blood concentrations of glucose in cancer. These include increased gluconeogenesis (conversion of amino acid carbon skeletons to glucose) and increased Cori cycling (hepatic conversion of lactate to glucose). Increased conversion of lactate to glucose may be due in part to increased lactate production by selected tumors. Combined with insulin resistance, the enhanced production of glucose provides the tumor with ample substrate that is readily metabolized under both aerobic and anaerobic conditions.

Lipid

Multiple changes in lipid metabolism in cancer include increased free fatty acid turnover, increased free fatty acid oxidation, increased glycerol turnover, greater lipolysis, and lower rates of lipogenesis. A lipid mobilizing factor has been isolated from the urine of cachectic cancer patients and shows bioactivity with isolated murine adipocytes.[14] In addition, there can be a marked loss of body fat that can occur early in the

evolution of malignancy. Hypertriglyceridemia can be observed among children with acute lymphoblastic leukemia, those with widespread solid tumors, and patients treated with L-asparaginase.[15] The provision of glucose does not seem to resolve the high rates of fatty acid and glycerol turnover evident in weight-losing cancer patients.[16] Treatment with chemotherapy is associated with decreased fat utilization in children newly diagnosed with acute lymphoblastic leukemia.[17]

Protein

Cancer is often associated with an acute-phase response blood profile, with low synthesis rates of albumin, prealbumin, and transferrin and increased synthesis rates of C-reactive protein, fibrinogen, and ferritin.[18] Some tumors such as hepatocellular cancer exhibit very high rates of protein turnover and increased protein degradation.[19] A net loss of skeletal muscle protein is a common finding with malignancy and is particularly problematic in the growing child. An increase in muscle protein breakdown mobilizes amino acids that may assist tumor growth as well as fuel gluconeogenesis. This increased protein breakdown in pediatric cancer patients may be related to falling levels of insulin-like growth factor (IGF-1) and insulin-like growth factor binding proteins[20] and decreased skeletal muscle protein synthesis.

Cytokines

Proinflammatory cytokines commonly associated with cancer cachexia include tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6), interleukin-1 (IL-1), and interferon-γ (INF-γ). These cytokines are secreted by macrophages and lymphocytes and may represent the host response to cancer. Administering TNF-α to humans produces effects associated with cancer cachexia including increased fatty acid turnover, elevated glycerol turnover, and increased whole-body protein turnover.[21] Not all patients with cancer cachexia have detectable levels of TNF-α; hence, a paracrine or autocrine mechanism of action may be responsible. Other factors also seem to be involved with the catabolic response to cancer. Circulating IL-6 levels are generally elevated in cancer patients, supporting its role in causing cachexia. As with TNF-α, the precise mechanism of action of IL-6 is unknown. The metabolic roles of IL-1 and INF-γ are less well characterized in tumor models; however, like TNF-α and IL-6, they appear to inhibit lipoprotein lipase and hence facilitate lipolysis.

Energy Balance

Energy expenditure measurements of children with cancer have been observed in a variety of clinical scenarios. Stallings and others measured resting energy expenditure (REE) in nine patients with ALL and found that patients with a higher tumor burden (elevated white blood cell count, organomegaly) had an increased REE.[17] On the contrary, a study of 26 patients with ALL or solid tumors in remission showed no elevations in REE compared to age- and sex-matched healthy controls.[22] Reilly and others[23] studied 20 preadolescent children with ALL with doubly labeled water. Compared to healthy controls matched for age, sex, and body composition, children with ALL had significantly lower total energy expenditures than controls by an average of 282 kcal/d. Most of this reduction was due to reduced physical activity. Any deviation in energy expenditure can result in energy imbalance and potentially undesirable weight loss or gain; thus, a careful look at changes in expenditure is worthwhile.

Lean body mass is the primary predictor of energy expenditure, so it follows that reductions in lean body mass related to cancer treatment could affect energy expenditure. Patients with head and neck cancer experienced changes in REE, as measured by indirect calorimetry, when prospectively evaluated prior to treatment; at weeks 2, 4, and 6 during treatment; at the end of treatment; and at two weeks following completion of treatment. Whereas REE was higher than that predicted by standardized equations in this small group of adults, REE declined significantly over time and followed a U-shaped curve from pretreatment to posttreatment.[24] Studies of children undergoing HSCT have implicated a relationship between changes in REE and reductions in lean body mass during this medically intense treatment. Ringwald-Smith and others compared weekly measured REE in 34 children undergoing autologous or allogeneic stem cell transplantation to standardized equations and found that REE was significantly less than predicted except at 14 days after transplant.[25] A prospective study of 37 children undergoing allogeneic transplantation described a significant decline in REE from baseline to week 3 after HSCT.[26] Moreover, a second study of children undergoing HSCT demonstrated significant reductions in REE over the course of transplant, which correlated with reductions in lean body mass.[27] These changes appeared to be unaffected by the level of energy provision.[28] Studies suggest that treatment-related changes in REE occur, which may be related to reductions in lean body mass. Further explorations of REE as it relates

to changes in body composition in children with cancer are needed.

METABOLIC EFFECTS OF TREATMENT

Treatments used in pediatric cancer cause systemic side effects that exacerbate the symptoms of the tumor itself. A combination of agents, including radiotherapy, chemotherapeutic drugs, and surgery, is commonly used to treat pediatric cancers, particularly high-risk diseases. Radiation therapy, widely used in the treatment of leukemia and brain tumors, is known to cause localized end-organ damage. Diarrhea and malabsorption with fluid and electrolyte losses may result when the abdomen or pelvis is radiated. Radiation to the head and neck areas may similarly cause damage to taste, chewing, and swallowing functions. Nutritional status may be affected by surgical interventions depending on the site of the tumor and the extent of the resection. The efficient digestion, ingestion, absorption, and utilization of food are complex processes. Surgical disruption with impaired swallowing, decreased gastric reserve, or reduction in intestinal length can result in decreased oral intake as well as absorption of nutrients.

Many chemotherapeutic agents are known to cause anorexia, with or without nausea and vomiting; enteritis with malabsorption and diarrhea; mucositis; and constipation. Taste perception is often altered in cancer patients receiving chemotherapy; this phenomenon further contributes to anorexia and reduced food intake. Mucosal damage is generally dose related, with increased risk of mucosal toxicity with high-dose induction therapy, escalating dosage patterns, continuous infusion (versus bolus doses), and combination chemotherapy treatments. Oral and gastrointestinal mucositis represent major complications of cytotoxic chemotherapy and radiotherapy, which lead to significant morbidity, pain, odynophagia, and poor intake as well as nausea and vomiting. Examples of commonly used chemotherapies and applicable nutrition-related toxicities are presented in Table 58-2.

Myeloablative HSCT is typically reserved for those cases having no other reasonable options, because it is known to be a high-risk, highly toxic intervention (Table 58-3). High-dose chemotherapy and total-body irradiation as conditioning for HSCT frequently cause prolonged oral and gastrointestinal mucositis that can reduce nutritional intake for days to weeks. Another adverse effect of HSCT is hepatic veno-occlusive disease (VOD), a syndrome of hepatomegaly, jaundice, and fluid retention, which in its severe form, may cause renal and respiratory failure. The cholestasis and fluid management of hepatic VOD has nutritional implications in that

it will almost certainly impact available volume for nutrient and fluid provision. Graft-versus-host disease (GVHD), a condition involving the attack of newly engrafted cells on the host recipient, can occur following allogeneic HSCT. Acute gastrointestinal GVHD can cause symptoms ranging from mild nausea and anorexia to profuse, bloody diarrhea. Acute skin GVHD can similarly manifest as a mild rash or progress to severe epithelial sloughing, akin to a burn injury. Other organ systems may also be affected. Most children with severe acute GVHD will require intensive, individualized nutrition support while high-dose immunosuppression is used to treat the disease process.

Chronic GVHD typically occurs more than 100 days following transplant and while its initial presentation tends to be less severe than acute GVHD, it is associated with significant long-term morbidity and mortality.[31] Skin, liver, mouth, and eyes are the most commonly affected sites.[32] Multiorgan disease often affects the skin and oral cavity, as well as the lung and liver, while single-organ disease is more frequently found in the liver or skin.[33] The incidence of chronic GVHD is lower in children than in adults; however, its manifestations are uniquely problematic to children. Malnutrition and accompanying growth failure is common, but poorly understood.[33] Other complications include bone loss, steroid-induced weight gain, dysphagia, and depression. Treatment of chronic GVHD typically employs therapy beyond first-line immunosuppression with steroids and calcineurin inhibitors.

TABLE 58-2 Nutrition-Related Side Effects of Commonly Used Chemotherapies in Childhood Cancer

Chemotherapeutic Drug	Diseases Targeted	Toxicities
Alkylating agents		
Mechlorethamine (nitrogen mustard)	Hodgkin's disease	N&V, phlebitis, mucositis
Cyclosphosphamide	Lymphomas, leukemias, sarcomas, neuroblastoma	N&V, cystitis, water retention, cardiac (HD)
Ifosfamide	Sarcomas, germ cell	N&V, cystitis, renal, cardiac (HD)
Melphalan	Rhabdomyosarcoma, sarcomas, neuroblastoma,	N&V, mucositis, diarrhea (HD)

	leukemias (HD)	
Lomustine, carmustine (CCNU, BCNU)	Brain tumors, lymphoma, Hodgkin's disease	N&V, renal, pulmonary
Busulfan	CML, leukemias (HSCT)	Pulmonary, N&V, mucositis, hepatic (HD)
Cisplatin	Testicular, other germ cell, brain tumors, osteosarcoma, neuroblastoma	N&V, renal
Carboplatin	Brain tumors, germ cell, neuroblastoma, sarcomas	N&V, hepatic
Dacarbazine	Neuroblastoma, sarcomas, Hodgkin's disease	N&V, flu-like syndrome, hepatic
Temozolomide	Brain tumors	N&V
Procarbazine	Hodgkin's disease, brain tumors	N&V, rash, mucositis
Antimetabolites		
Methotrexate	Leukemia, lymphoma, osteosarcoma	Mucositis, rash, hepatic, renal
Mercaptopurine (6MP)	Leukemia (ALL, CML)	Hepatic, mucositis
Thioguanine	Leukemia (ALL, AML)	N&V, mucositis, hepatic (VOD)
Fludarabine, cladribine	Leukemia, lymphoma	Opportunistic infections, renal
Clofarabine	Leukemia	Hepatic, hypokalemia, inflammation
Nelarabine	T-cell leukemia	Somnolence, peripheral neuropathy
Gemcitabine	Hodgkin's, sarcomas	N&V, hepatic, mucositis, flu-like syndrome, edema, rash
Cytarabine (Ara-C)	Leukemia, lymphoma	N&V, mucositis, GI, flu-like syndrome, ocular, skin (HD)
Fluorouracil (5FU)	Carcinomas, hepatic tumors	Mucositis, N&V, diarrhea, skin, ocular, cardiac
Antitumor antibiotics		
Doxorubicin (adriamycin), daunomycin, idarubicin	Leukemia (ALL, ANLL), lymphomas, solid tumors	Mucositis, N&V, diarrhea, cardiac
Mitoxantrone	Leukemia (ALL, ANLL), lymphomas	Mucositis, N&V, bluish color to urine, veins, sclerae, nails

Bleomycin	Lymphoma, testicular, other germ cell	Lung, skin, fever, mucositis, Raynaud's, N&V
Dactinomycin (actinomycin-D)	Wilm's, sarcomas	N&V, mucositis, hepatic (VOD)
Plant products		
Vincristine	Leukemia (ALL), lymphomas, solid tumors	SIADH, hypotension, constipation
Vinblastine	Histiocytosis, Hodgkin's disease, testicular	Mucositis
Etoposide	Leukemias (ALL, ANLL), lymphomas, neuroblastoma, sarcomas, brain tumors	N&V, mucositis, hypotension, secondary leukemia, diarrhea (PO dosing)
Teniposide	Leukemia (ALL)	N&V, mucositis, hypotension
Topotecan	Neuroblastoma, rhabdomyosarcoma	Diarrhea, mucositis, N&V, rash, hepatic
Chemotherapeutic Drug	Diseases Targeted	Toxicities
Irinotecan	Rhabdomyosarcoma	Diarrhea, N&V, hepatic, dehydration, ileus
Kinase inhibitors		
Imatinib mesylate (Gleevec®)	Ph chromosome + CML	N&V, fatigue, GI, hepatic
Dasatinib	Ph chromosome + ALL, CML	Fluid retention, rash, nausea, bleeding, diarrhea
Sorafenib	Renal cell carcinoma, hepatocellular carcinoma	Rash, hypertension, diarrhea, N&V, bleeding
Sunitinib	GI stromal tumors, renal cell carcinoma	Cardiac, hypertension, diarrhea, N&V, GI, mucositis, bleeding, rash
Bevacizumab	Carcinomas, glioblastoma	Infusional reaction, rash, proteinuria
Erlotinib, gefitinib,	Carcinomas	Rash, diarrhea
Other agents		
Corticosteroids (prednisone, methylprednisolone, dexamethasone, decadron)	Leukemia, lymphomas, brain tumors	Increased appetite, centripetal obesity, myopathy, osteoporosis, pancreatitis, hypertension, diabetes, growth failure, impaired wound healing

Asparaginase (L-ASP, PEG-ASP)	Leukemia (ALL), lymphoma	Pancreatitis, hepatic
All-trans-retinoic acid (ATRA, tretinoin)	Acute promyelocytic leukemia	Retinoic acid syndrome (weight gain, respiratory distress, cardiac & renal failure), cheilitis, dry skin, η-triglycerides
13-*cis*-Retinoic acid (isotretinoin, accutane)	Minimal residual disease neuroblastoma	Cheilitis, dry mouth, bone & joint pain, η-triglycerides, η-Ca^{++}

Abbreviations: N&V, nausea and vomiting; HD, high dose; CML, chronic myelocytic leukemia; AML, acute myelogenous leukemia; CLL, chronic lymphocytic leukemia; GI, gastrointestinal; ANLL, acute nonlymphoblastic leukemia; SIADH, syndrome of inappropriate antidiuretic drug hormone secretion; ↑, increased.

Source: Adapted from Adamson PC, Bagatell R, Balis FM, Blaney SM. General principles of chemotherapy. In: Pizzo PA, Poplack DG, editors. *Principles and Practice of Pediatric Oncology*. 6th ed. Philadelphia: Lippincott Williams & Wilkins; 2011. pp. 279–355.[29]

TABLE 58-3 Diseases Commonly Treated with HSCT

Autologous Transplantation	Allogeneic Transplantation	
Non-Hodgkin's lymphoma	Cancers	Other diseases
Hodgkin's disease	Acute myeloid leukemia	Aplastic anemia
Acute myeloid leukemia	Acute lymphoblastic leukemia	Fanconi's anemia
Neuroblastoma	Chronic myeloid leukemia	Blackfan–Diamond anemia
Germ-cell tumors	Myelodysplastic syndrome	Thalassemia major
	Non-Hodgkin's lymphoma	Sickle cell anemia
	Hodgkin's disease	Severe combined immunodeficiencies
	Juvenile chronic myeloid leukemia	Wiskott–Aldrich syndrome

Source: Adapted from Copelan EA. Hematopoietic stem-cell transplantation. *N Engl J Med*. Apr 27 2006;354(17):1813-1826.[30]

ETIOLOGY OF ANOREXIA IN CANCER TREATMENT

As indicated in earlier sections, malignancies and their treatment can have a considerable effect on a child's nutritional status. Frequently occurring nutritional problems with cancer treatment are depicted in Figure 58-1.

Anorexia can be a side effect of any disease. Food aversion and anorexia may be linked with nausea and vomiting. While the pathophysiology of cancer and its treatment undoubtedly cause nutritional risk, eating problems are multifactorial in nature. Psychological factors are also important to consider in evaluating the reasons for inadequate dietary intake. Depression-related anorexia is probably underappreciated as a cause.

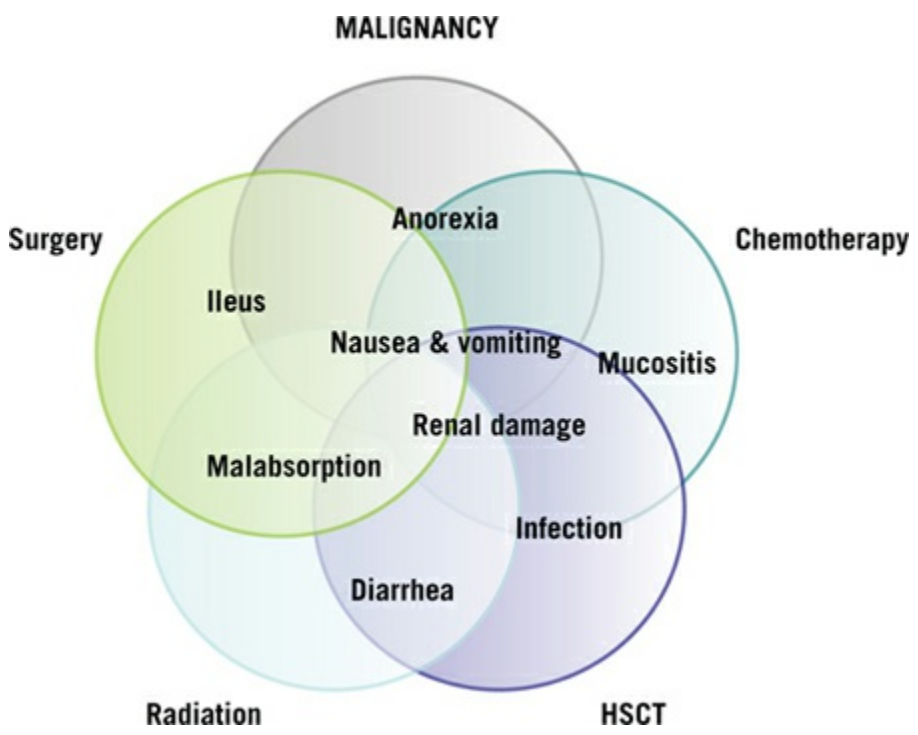


Figure 58-1 Nutrition-related problems in pediatric cancer treatment

The ability of children to eat at their appropriate developmental level may affect the nutritional adequacy of their diet. Anorexia, mucositis, and other effects of cancer treatment may interrupt the normal progression of feeding skills in infants and young children. Once arrested, the development of these skills may be difficult to restore. The resumption of eating can be a lengthy and difficult process and may extend the time frame of cancer recovery. Poor swallowing and chewing abilities will lengthen the time required to complete a meal and thus may lead to inadequate consumption of many required nutrients. Children may refuse to eat their preferred foods due to adverse associations or other impairments and thus may self-restrict their intake.

The impact of a cancer diagnosis on family dynamics and related behaviors should not be overlooked. Relationships between patients and caregivers influence nutritional intake, both positively and negatively. Appetite and feeding behaviors are inherently complicated activities of all children, and this behavior can obviously be affected by the onset of illness, its treatment, as well as the psychosocial impact that cancer has on the child and family. The nature of cancer medical care is such that parents often feel that they are relinquishing much of their usual caregiving behavior to the medical and nursing staff. Some parents understandably cling to the provision of food and nutrients as a critical part of parenting. Occasionally, such strong feelings concerning food and dietary intake can cause conflict between parent and child and be counterproductive to the goal of optimal nutrition. Discord among family members can lead to further negative consequences to the feeding relationship.

NUTRITIONAL ASSESSMENT OF THE CHILD WITH CANCER

The assessment of a child with cancer includes the standard elements of a nutritional evaluation: history of past and present illness, review of dietary intake and nutritional history, physical examination, and anthropometric measurements. Inquiry for suspected or measureable body composition changes should also be considered, as well as a review of pertinent laboratory measures.

History

A detailed medical history is essential to the nutritional evaluation of a patient. Important oncologic history includes the type and stage of the tumor, intensity of the antitumor therapy, presence or absence of remission, and timing and type of future treatment. Nutritional history entails (1) current symptoms of cancer and its therapy and their effect on nutrient intake, absorption, and retention; (2) growth history, previous antitumor therapy and its effects on nutritional status; (3) developmental status, with special attention to milestones of feeding skills and swallowing function; (4) known or perceived food allergies or intolerances; (5) medications, with special attention to those with gastrointestinal and metabolic side effects; (6) family history, parental heights, and sibling growth patterns; (7) social history, food preferences/beliefs, and food availability; and (8) a thorough diet history

obtained from the patient and/or caregivers.

Physical Examination

The physical characteristics of a child with cancer may suggest the presence of nutritional problems. Indeed, the predisposition of chemotherapy to affect tissues of rapid turnover (e.g., hair and gastrointestinal mucosa) parallels the symptoms of deficiency of several nutrients (e.g., stomatitis with some B vitamins and alopecia with biotin deficiency). Careful inspection and palpation of subcutaneous fat and muscle stores are particularly important in the physical examination, because deficits of these two components of body stores are common in malnutrition.

The general physical examination may show edema as a result of low concentrations of circulating albumin. This can be a subtle finding with only mild hypoalbuminemia. Edema may actually mask progressive muscle wasting and loss of lean body mass. Hypoalbuminemia may be due to poor liver function related to tumor or treatment, inadequate protein intake, or nutrient losses in excess of intake. Painful mouth or esophageal sores caused by mucositis can reduce a child's ability to eat and thus be a reason to consider nutritional support. Children who have limited mobility due to impairment by disease or treatment side effects may experience changes in appetite or satiety. Recognition of these aspects of the physical examination is a valuable component of the overall nutritional assessment of the child with cancer.

Anthropometry

Weight and height measured over time are the foundation of nutritional monitoring and evaluation. These objective measures of nutritional status provide the most information with the least inconvenience and cost. Weight should be measured daily in the hospital setting and at each office visit for outpatients. Height/length measurements should be recorded monthly. Accurate technique is critical to the validity of the measures; detailed methodologies are reviewed in Chapter 2.

The incidence of weight and height deficits in children with cancer has been reported to be quite high, depending on the severity and type of cancer and its treatment.[1,34] However, with the increased incidence of obesity in children, and the predisposition of some pediatric cancer patients to become obese,[8] it is also important to screen for overweight. For children under the age of 2 years, this can be done by examining the

weight for height z scores using the World Health Organization (WHO) 2006 growth curves. For older children, BMI is a more suitable measure of body fatness. Children with BMIs greater than the 85th percentile for age and sex are termed overweight, and those with BMIs greater than the 95th percentile are obese.[35] Growth curves for BMIs are included with the National Center for Health Statistics 2000 growth curves as well as the WHO 2007 growth reference. See Appendix I.

Arm Anthropometrics

Arm anthropometrics are an inexpensive, noninvasive, and widely accepted technique for estimating body composition against established normal values (see Chapter 2). Triceps skinfold, mid-upper arm circumference, and computed values of midarm muscle area and midarm fat area correlate well with more accurate and sensitive measures of body composition.[36] Percentiles and standards for comparisons for triceps skinfolds and midarm muscle area are available[37] and may indicate the presence of malnutrition when weight or height is unavailable or unreliable, as in the case of a growing tumor mass.

In a convenience sample of children undergoing treatment for cancer in Australia, skinfold measurements were more closely correlated with percentage body fat measured by air displacement plethysmography than weight, height or BMI z scores.[38] Midarm circumference was significantly correlated with lean body mass as measured by DXA in a study of children newly diagnosed with cancer.[11] These studies suggest that arm anthropometrics may be reasonable surrogates for precise measures of body composition and likely better indicators of nutritional status in children with malignancy than weight and height alone.

Other Measures of Body Composition

Anthropometric measurements may be confounded by technique, inaccuracy of equipment, lack of patient cooperation, and poor interindividual reliability. Because lean body mass is considered the most metabolically active component of the body, it would be preferable, therefore, to characterize nutritional status by an accurate and dependable measure of lean body mass. Patients with solid tumors prior to reduction or excision and patients with fluid imbalances are particularly susceptible to inaccurate nutrition assessment based on anthropometric measurements.

DXA is becoming increasingly available for clinical use. A whole-body DXA assessment measures total-body bone mineral content, nonbone

lean tissue, and fat, as well as bone mineral density, thereby providing body composition information using a three-compartment model. Elevations in body fat mass measured by DXA without a concomitant rise in BMI (kilogram per square meter) have been reported in several studies of cancer survivors.[39,40] Others have shown similar trends in body fat mass measurements. Warner and others found that a group of children treated for ALL with chemotherapy and cranial radiation had a significantly greater percent of body fat, as measured by DXA, than age-matched groups with other malignancies or healthy controls.[41] When combining the DXA measurements of fat-free mass with measures of total-body water and total-body potassium, children with ALL in this cohort had a significantly lower hydration and potassium state of their fat-free mass than those with other malignancies and controls. Conversely, a cross-sectional study of children in remission from ALL showed a significantly higher hydration fraction of fat-free mass compared to healthy age- and sex-matched controls, using DXA and a four-component model of body composition assessment.[42] Another study of 25 children who had undergone allogeneic HSCT at least four years previously showed that their mean BMI was 0.45 z scores lower ($p < .05$) but that percent body fat as measured by DXA was 1.1 z scores higher than controls ($p < .001$).[43] Furthermore, deficits in lean mass exceed overall BMI declines in survivors of childhood malignancy and HSCT.[44] These studies confirm the utility of assessing body composition in children with cancer to reveal clinically important changes relevant to nutrient provision. While the ultimate applicability of DXA scanning to nutritional assessment is still evolving, it is now widely used as a reasonable method to evaluate body composition in both clinical and research settings.

Laboratory Evaluation

Although laboratory evaluation of the pediatric cancer patient is commonly performed to assess the hematologic and metabolic response to cancer and its treatment, nutritional status should also be monitored with select biochemical parameters. Serum electrolytes and mineral concentrations are commonly deranged during treatment of cancer, especially when nephrotoxic agents are employed. A specific metabolic emergency termed “tumor lysis syndrome” occurs in the early stages of tumor diagnosis and treatment as a result of tumor cell breakdown. Massive cell lysis can lead to life-threatening electrolyte abnormalities, including hyperphosphatemia, hyperkalemia, hyperuricemia, and resultant

renal failure.

Visceral proteins synthesized by the liver are often used to assess nutritional status, because decreased levels presumably reflect a reduced supply of amino acid precursors and/or decreased hepatic and other visceral protein mass. Blood concentrations of these proteins, however, are dependent on their rates of synthesis, degradation, and escape from the circulatory system. Serum proteins are also affected by infectious or catabolic processes. The concentrations of positive acute-phase proteins (e.g., C-reactive protein, ferritin, and ceruloplasmin) are increased in infectious or other catabolic illnesses, whereas negative acute-phase proteins (e.g., albumin, prealbumin, transferrin, and retinol-binding protein) are decreased in these circumstances.

Albumin is a relatively insensitive marker of nutritional status owing to its long half-life of 20 days and its ability to be mobilized from extravascular stores even in the presence of prolonged energy and protein deficiency. Hypoalbuminemia can occur in situations of decreased synthesis (e.g., liver disease, age over 70 years, and malignancy), increased losses (e.g., nephrosis, protein-losing enteropathy, and burn injuries), or displacement to extravascular spaces (e.g., acute catabolic stress with capillary leak syndrome) and is not necessarily related to the presence of malnutrition. Fluid overload, a condition common to acutely ill cancer patients, can also dilute albumin concentrations.

Prealbumin is a more sensitive measure of visceral protein status than albumin because of its short half-life (two days) and high ratio of essential to nonessential amino acids. A small study of children with solid tumors correlated disease status and treatment intensity with changes in prealbumin levels, suggesting that it is likely the best available serum marker of nutritional status.[45] Like albumin, its concentration falls with an acute-phase protein response or liver disease. Levels increase with renal failure.

In a study of 170 children with an assortment of cancers, albumin and prealbumin levels were significantly lower on presentation in patients than in controls, whereas the acute-phase indicator C-reactive protein was elevated. After six months of chemotherapy, however, the C-reactive protein levels were comparable in the groups but the visceral protein levels were still lower in those with cancer.[46] This suggests that even though acute-phase response is responsible for some alterations in visceral protein levels cancer patients still have poorer visceral protein nutrition than controls.

Assessment of vitamin and mineral nutriture, particularly in the patient

on total enteral or parenteral nutritional support, may also be helpful. At the time of diagnosis, the effect of tumor burden on metabolic processes may be profound, as demonstrated by laboratory analysis. Vitamin D insufficiency in survivors of childhood cancer is common[47] and may predispose them to low bone mineral density and fractures.[48] Whether vitamin D status might be associated with cancer-related outcomes remains unknown.

Antioxidant nutrients, including vitamins A and E, β-carotene, zinc, and selenium have also been studied. Children newly diagnosed with a variety of cancers had lower concentrations of retinol, β-carotene, vitamin E, and zinc prior to treatment compared to healthy controls in a multicenter French study. After six months of treatment, most of these parameters were largely unchanged; however, zinc concentrations in patients with CNS tumors and malignant bone tumors showed significant decline, suggesting the possibility of losses related to therapy, or a reduction in intake.[46] Zinc deficiency was also found to be common in children following stem cell transplantation. Nineteen of 28 children developed biochemical zinc deficiency at a median of seven days post transplantation. Zinc blood levels were positively correlated with serum concentrations of alkaline phosphatase, a zinc-dependent enzyme.[49] Another study of 103 children with ALL examined plasma antioxidant levels and chemotherapy side effects at diagnosis and three months and six months after diagnosis. After adjustment for multiple comparisons, there was a significant relationship between shorter length of hospital stay and higher total carotenoid to total cholesterol ratio at diagnosis. Additionally, subjects were more likely to experience a treatment-related toxicity if they had a low plasma total carotenoid level.[50] More research is needed to further elucidate the relationship of nutrient levels with clinical outcomes in cancer.

NUTRITION INTERVENTION

Food Safety

Because cancer and its treatment inherently cause immunosuppression to varying degrees, low microbial diets have historically been used in an effort to minimize risk of illness from foodborne pathogens. Although there is little evidence to support the efficacy of these regimens in preventing clinical infections,[51] it is intuitive to rule out possible causes when little potential harm might be involved. Versions of the neutropenic

or low microbial diet continue to be used widely in pediatric cancer treatment centers, although the impact of diet adherence and quality of life must be considered when comparing this more regimented approach to general food safety guidelines.[52] Table 58.4 summarizes the prudent principles recommended for HSCT recipients.

TABLE 58-4 Food Safety Recommendations for HSCT recipients[a]

- Avoid raw or undercooked meat, including beef, poultry, pork, lamb, venison, sweetbreads, other game meat
- Avoid any foods containing undercooked meat, including sausages and casseroles
- Avoid raw or undercooked eggs or foods that contain them, such as hollandaise sauce, homemade eggnog, homemade mayonnaise or salad dressing
- Avoid raw or undercooked seafood, such as clams, oysters, sushi
- Avoid raw fruits and vegetables from developing countries
- Drink only tap water that has been treated and tested, or bottled water
- Avoid unpasteurized milk or other dairy products
- Avoid unpasteurized fruit juices
- Avoid food and drinks from street vendors

[a]In addition to careful attention to food safety guidelines recommended for all children, those who have undergone HSCT should adhere to the recommendations in this table for three months, or until immunosuppresive medications have been discontinued.

Source: Adapted from Guidelines for preventing opportunistic infections among hematopoietic stem cell transplant recipients: recommendations of CDC, the Infectious Disease Society of America, and the American Society of Blood and Marrow Transplantation. MMWR. Vol 49;RR-10: Centers for Disease Control and Prevention; 2000:50–52.[53]

Indications for Feeding Modalities

The method of nutrition support selected for the child with cancer is determined by a timely clinical assessment and estimation of the child's nutrient requirements.[54] Some children may thrive well on volitional oral intake, whereas others may require specialized enteral or parenteral support. Available evidence suggests that enteral nutrition is a less expensive, safer, and more effective way of nourishing the child with cancer than parenteral nutrition.[54] However, there is no standardized approach to nutrition intervention in pediatric oncology.[55] It is axiomatic that enteral feeding should be considered prior to the initiation of parenteral nutrition and that oral supplements should be offered before

considering tube feeding. The most appropriate feeding method for the child with cancer is that which provides complete nutrient requirements with least risk. Patients and their family members should of course be included in the discussion of nutrition options, with discussion of the risks and benefits of appropriate and available methods for the patient's current situation.

In the case of malnutrition, it is imperative to consider the manifestations of refeeding the body in a starved or cachectic state. A profound hypophosphatemia, in particular, can characterize the rapid cellular influx of nutrients during the anabolic phase following starvation. The risk of this complication is especially present with parenteral nutrition. Severe metabolic complications may be avoided by cautious advancement of macronutrients over a period of days to weeks combined with close laboratory monitoring of electrolytes, glucose, and minerals.

Oral Diet

Because painful oral mucositis is a common side effect of chemotherapy with few effective treatments, a soft diet is often better tolerated. A diet high in magnesium or potassium may be useful for the child with excessive urinary losses of minerals due to chemotherapies or antibiotics. Other diet alterations may be indicated for a variety of comorbidities, for example, a reduced sodium diet for hypertension and a bland, low-residue diet for gastrointestinal GVHD.[56] Most commonly, however, calorie enhancement is the primary manipulation in an effort to promote weight gain or weight maintenance during treatment-associated anorexia. This can be accomplished with usual foods, or in combination with commercial supplemental drinks, bars, or calorie additives. See Chapter 70 for further specifications of the high-calorie diet. The use of appetite-enhancing medications such as cyproheptadine hydrochloride and megestrol acetate has been associated with weight gain and increases in leptin levels in children with cancer cachexia,[57] although megestrol acetate has also been linked to adrenal suppression and is therefore not recommended for routine use.[58] Some children, including those with standard-risk ALL and long-term survivors of cancer with activity limitations, may be aided by calorie moderation when excessive weight gain is a concern.

Enteral Feeding

If children are unable to meet their nutrient needs orally, tube feedings should be considered to preserve or achieve optimal nutritional status.

Nasogastric tubes have been used with success in many pediatric oncology patients. In 21 children newly diagnosed with cancer, nasogastric tube feedings were given in addition to volitional oral intake. After 16 weeks of feedings, weights increased significantly.[59] Several studies have reported the use of nasogastric feedings in children after stem cell transplantation with varying results. One group demonstrated improved weight gain in 21 patients, compared to 8 children receiving dietary counseling alone. There was a positive correlation between increases in weight and midarm circumference and the duration of enteral feedings. However, 8 of the 21 patients stopped feedings after 10 days due to vomiting or diarrhea. Six of these patients were then switched to parenteral nutrition support.[60] Langdana and others retrospectively analyzed their intensive enteral feeding protocol for children undergoing HSCT, which included routine insertion of nasogastric tubes during conditioning or when oral intake had decreased. Of their 49 evaluable patients, 86% (42) were maintained on enteral nutrition alone, including 8% who took adequate oral intake.[61] Another study compared tube feeding to parenteral nutrition in a group of children undergoing allogeneic HSCT, with just 3 of 34 patients supported exclusively by tube feeding; the remainder required partial or total parenteral nutrition. Cholestasis occurred significantly less often in children who received at least some tube feeding compared to children who were exclusively parenterally fed; nutritional costs were also less in children receiving tube feeding.[62]

These studies suggest the feasibility of nasogastric feeding as a method of nutrition support for pediatric cancer and HSCT patients, although clearly some patients will be unable to tolerate the adequate volumes to support their nutritional needs. Gastrostomy tube feedings are generally considered to be more cosmetically acceptable and comfortable than nasogastric feedings while still providing the same advantages over parenteral nutrition. In a number of studies, significant weight gain in pediatric cancer patients has been demonstrated with the use of surgically placed or percutaneous endoscopic gastrostomy (PEG) feedings.[63–66] Although some patients may ultimately require parenteral support due to gastrointestinal intolerance, the use of enteral nutrition substantially reduces the amount and duration of parenteral nutrition required, thus reducing the risks and costs with which it is associated.[62–65] Further studies assessing the effect of proactive enteral feeding in this population are needed.

Nutritionally complete formulas for oral supplementation or tube

feeding are available for a variety of ages and conditions (see Appendix III). Most pediatric cancer patients will tolerate intact protein, 1 or 1.5 kcal/mL, formulas either orally or via tube feeding. On rare occasions, a specialized formula for tube feeding may be indicated. Formulas with elemental (free amino acids) or semielemental (small peptides) protein are available, primarily for the purpose of allergy or intolerance. Medium-chain triglycerides are also used in many formulas intended for patients with fat malabsorption. An abundance of defined formula diets are available for varying conditions and ages.

Parenteral Feeding

When the gastrointestinal tract is nonfunctional or unavailable, nutrients may be infused via central venous catheters. Peripheral venous lines may also be used temporarily to provide a nutrient solution with less than 900 mOsm/L, usually 10% dextrose and 1.5%-2% amino acids. Parenteral nutrition has been widely used in the oncology population due to the cytotoxic effects of many treatment regimens. Most chemotherapies commonly cause some degree of nausea and vomiting. Radiation therapy, when directed to gastrointestinal organs, also causes cell damage that may impair digestive or absorptive function. High-dose chemotherapeutic regimens and total-body irradiation, used in preparation for stem cell transplantation, may cause severe mucositis and enteritis, making significant oral or enteral intake difficult for many patients to achieve for several weeks. Gastrointestinal GVHD, a complication of allogeneic stem cell transplantation, typically causes impaired absorption of nutrients, either due to anorexia and diminished oral intake in its mildest cases or as profuse, bloody diarrhea in its more severe cases.

The value of parenteral nutrition in cancer patients has been questioned; in fact, its routine use in cancer treatment has been discouraged due to the risks of infectious complications.[67] A small study of pediatric cancer patients with severe chemotherapy-induced mucositis failed to demonstrate a significant benefit of parenteral nutrition over standard intravenous fluid. This prospective, randomized trial compared body composition measurements (bioelectrical impedance analysis), laboratory parameters, and other clinical outcomes after 10 days of therapy. Only total weight gain was greater in the parenteral nutrition group, with similar measures of fat-free mass, thus implying exclusive accumulation of body fat in the parenteral nutrition group.[68] HSCT, however, is one of the few clinical situations where parenteral nutrition has

historically demonstrated benefit.[69] A few well-designed trials have further advanced the science of nutrition support in children with cancer. A study of 20 children receiving parenteral nutrition during HSCT attempted to clarify a relationship between changes in parenteral energy intake and clinical outcomes. A greater difference between expected and actual parenteral energy intakes was associated with longer time to engraftment in this retrospective analysis. Actual energy expenditure and oral intake were not reported.[70] A randomized controlled trial designed to compare parenteral energy intake titrated to measured REE with standard energy intake, typically 130%-150% of REE, found significant declines in lean body mass in both groups, with a trend toward higher body fat in the standard energy group.[28] These results raise concern for overfeeding when parenteral nutrition energy intake exceeds REE. More research examining the specific regimens and components of nutrition support is warranted for children with cancer.

When clearly indicated, parenteral nutrition should be initiated according to standard recommendations (see Chapter 68). Goals for parenteral nutrition infusions will be based on estimated nutrient requirements, fluid requirements, and available venous access. Electrolytes are added to promote metabolic homeostasis; pediatric multivitamin and trace element preparations are generally used as well. Attention should also be directed to concomitant and expected medication use, particularly those with known nutritional interactions.

The risks of parenteral nutrition can be significant and include infections, hepatotoxicity, suppression of oral intake, and metabolic abnormalities. Most children undergoing aggressive cancer treatment will require indwelling central venous catheters for chemotherapy, but the use of parenteral nutrition has been associated with a 2.4-fold increase in the risk of infection in children receiving chemotherapy who have central access devices in place.[71] A specific role for intravenous fats in increasing infectious complications is not supported in the literature.[72]

Many of the medicines used in oncologic treatment can cause liver injury. The use of parenteral nutrition, singly or in combination with these medicines, may cause biliary dysfunction or steatosis.[73] Another potential risk of parenteral nutrition administration is the generation of peroxide compounds, which may have harmful effects in neonates and others with immune dysfunction and/or reduced antioxidant defenses.[74] Covering parenteral nutrition bags and intravenous tubing with opaque plastic coloring may reduce this effect.[75]

Parenteral nutrition may also cause early satiety and decreased oral intake. In a randomized controlled trial of children and young adults following marrow transplantation, outpatient parenteral nutrition delayed resumption of oral intake by six days compared to 5% dextrose hydration. The clinical outcome of both groups was not significantly different.[76] Metabolic derangements are also associated with parenteral nutrition. Over- or underhydration, electrolyte imbalance, hyperglycemia, and hypoglycemia are among the most common concerns. While there remains a role for parenteral nutrition in selected pediatric cancer patients, close monitoring of laboratory values, clinical condition, and ability to enterally or orally feed are important to minimize risks associated with long-term parenteral nutrition.

The significant metabolic and nutritional ramifications of cancer and its therapy have led investigators to evaluate whether the addition of various nutrients may improve clinical outcomes. Glutamine, an important amino acid for rapidly turning over cells, has a documented role in ameliorating the mucosal atrophy seen in prolonged states of parenteral nutrition,[77] the healing of gastrointestinal mucosa after damage from radio- or chemotherapy,[78,79] and reducing episodes of clinical sepsis.[80] Evidence supporting its use in cancer treatment has been conflicting, however, and pediatric data are minimal. ω-3 Fatty acids, derived from the essential fatty acid α-linolenic acid, have also been under consideration, owing to the antiangiogenic and antitumoral properties observed in animal models, and the anticachectic effect seen in a small number of clinical studies.[81] One small study suggested that supplementation with a formula containing ω-3 fatty acids may reduce weight loss associated with pediatric cancer treatment, although the effect may have been a result of additional energy rather than a specific effect of the fatty acids.[82] Further research is needed to advance this science as it pertains to the nutritional therapy of pediatric cancer.

NUTRITIONAL CONCERNS OF CHILDHOOD CANCER SURVIVORS

Many nutritional issues during childhood cancer treatment are associated with weight loss or poor growth, but more recently the risk of obesity and excess body fat accumulation has been elucidated. The Childhood Cancer Survivor Study, a multicenter, retrospective cohort study of adults who survived pediatric cancer greater than five years, described a greater

prevalence of overweight and obesity in cancer survivors who received high-dose cranial radiation (>20 Gy), compared to their healthy siblings.[83] Survivors of ALL were more likely to be obese, whereas survivors of other cancer types were more likely to be underweight, suggesting an overall abnormal growth pattern resulting from pediatric cancer treatment.[84] A study using a combination of indirect calorimetry and ambulatory heart rate monitoring to measure basal metabolic rate and total energy expenditure concluded that ALL survivors have lower levels of energy expenditure for activity.[85] Percent body fat, as measured by DXA, was significantly higher among ALL survivors, whereas their BMI remained similar to controls,[86] suggesting the need for body composition measurements to elucidate changes in body fat. These changes in growth and body composition may be subsequent risk factors for cardiovascular disease and other chronic illnesses as this population ages.[84]

The impact of cancer treatment on linear growth has also been examined. A cohort of HSCT survivors, engrafted before or at the onset of puberty, had a significant decrease in final height compared to pretransplant height standard deviation scores. However, most of the patients achieved a height considered to be normal.[87] Similarly, another study of HSCT survivors, who were treated with total-body irradiation, found a reduction in peak height velocity following transplantation.[88] Again, radiation therapy seems to be the common element. A reduction in linear growth has also been described in survivors of ALL and brain tumors.[84] Nutritional intervention may play a key role in limiting the extent of growth faltering, though the primary cause is likely a deficiency of growth hormone caused by radiation damage. Although the clinical significance of this growth stunting is as yet unclear, the relationship of height to nutrient intake, body weight, and body composition is certainly worthy of consideration when planning nutritional therapy.

Conflicting reports of a greater prevalence of osteopenia in childhood cancer survivors have been published,[89–92] perhaps related to differences in treatment, time of follow-up, or variability in measurements. Bone loss has been described in adult HSCT survivors,[93] likely related to steroid treatment post transplantation. Declines in bone mineral density z scores have also been observed in children following HSCT. One study demonstrated an increasing incidence of osteopenia (defined as a z score between 1 and 2 SD below the mean) and osteoporosis (defined as a z score >2 SD below the mean) in a prospective evaluation of bone mineral density at the time of HSCT and 100 days, six months, and one year after

HSCT.[94] These changes in bone mass have yet to be correlated with clinical findings, such as fractures; however, their relationship to the overall health of the growing group of childhood cancer survivors may be substantial.

A significant incidence of insulin resistance and dyslipidemia among survivors of HSCT has been reported.[95] Likewise, features of the metabolic syndrome have been found in ALL survivors[96]; however, the findings appear to be most pronounced in those receiving HSCT. Unquestionably, complications of oncologic treatment, including obesity and insulin resistance, need further evaluation and attention. Dietary intake among long-term survivors may fall short of recommendations for optimal health and chronic disease prevention.[97] Children undergoing cancer treatment should be taught healthy diet principles and acceptable activity options for weight maintenance.

CONCLUSION

From the time of diagnosis, nutrition is an important aspect of the care and recovery of the pediatric cancer patient. The effects of cancer and its treatment have lifelong implications. Age-appropriate growth and development should be the goal during extended treatment courses as well as following the completion of therapy. Serial nutritional assessments should be performed regularly with appropriate interventions and follow-up. Malnutrition should not be accepted as an inevitable consequence of cancer and/or its treatment. Similarly, effort should be made to prevent obesity and other nutrition-related chronic diseases in long-term cancer survivors. Effective supportive care of the pediatric cancer patient involves nutritional intervention and counseling with a multidisciplinary approach, emphasizing the quality of life of each child.

REFERENCES

1. Brinksma A, Huizinga G, Sulkers E, et al. Malnutrition in childhood cancer patients: a review on its prevalence and possible causes. *Crit Rev Oncol Hematol.* Aug 2012;83(2):249–275.
2. Lange BJ, Gerbing RB, Feusner J, et al. Mortality in overweight and underweight children with acute myeloid leukemia. *JAMA.* Jan 12 2005;293(2):203–211.
3. Oeffinger KC, Mertens AC, Sklar CA, et al. Chronic health conditions in adult survivors of childhood cancer. *N Engl J Med.* Oct 12 2006;355(15):1572–1582.
4. Gerhardt CA, Baughcum AE, Johnston A, et al. Parent perceptions of nutritional issues during their child's treatment for cancer. *J Pediatr Hematol Oncol.* Jul 2006;28(7):454–460.

5. Surveillance, Epidemiology, and End Results (SEER) Program Populations (1969-2009). National Cancer Institute, DCCPS, Surveillance Research Program, Surveillance Systems Branch, released January 2011. http://www.seer.cancer.gov/. Accessed January 2, 2013.
6. Garofolo A, Lopez FA, Petrilli AS. High prevalence of malnutrition among patients with solid non-hematological tumors as found by using skinfold and circumference measurements. *Sao Paulo Med J.* Dec 2005;123(6):277–281.
7. Sala A, Rossi E, Antillon F, et al. Nutritional status at diagnosis is related to clinical outcomes in children and adolescents with cancer: a perspective from Central America. *Eur J Cancer.* Jan 2012;48(2):243–252.
8. Withycombe JS, Post-White JE, Meza JL, et al. Weight patterns in children with higher risk ALL: a report from the Children's Oncology Group (COG) for CCG 1961. *Pediatr Blood Cancer.* Dec 15 2009;53(7):1249–1254.
9. Murphy AJ, White M, Davies PS. Body composition of children with cancer. *Am J Clin Nutr.* Jul 2010;92(1):55–60.
10. Schmid I, Schmitt M, Streiter M, et al. Effects of soluble TNF receptor II (sTNF-RII), IL-1 receptor antagonist (IL-1ra), tumor load and hypermetabolism on malnutrition in children with acute leukemia. *Eur J Med Res.* Nov 16 2005;10(11):457–461.
11. Barr R, Collins L, Nayiager T, et al. Nutritional status at diagnosis in children with cancer. 2. An assessment by arm anthropometry. *J Pediatr Hematol Oncol.* Apr 2011;33(3):e101–e104.
12. Delbecque-Boussard L, Gottrand F, Ategbo S, et al. Nutritional status of children with acute lymphoblastic leukemia: a longitudinal study. *Am J Clin Nutr.* 1997;65(1):95–100.
13. Wolfe RR. The underappreciated role of muscle in health and disease. *Am J Clin Nutr.* Sep 2006;84(3):475–482.
14. Todorov PT, McDevitt TM, Meyer DJ, et al. Purification and characterization of a tumor lipid-mobilizing factor. *Cancer Res.* 1998;58(11):2353–2358.
15. Halton JM, Nazir DJ, McQueen MJ, Barr RD. Blood lipid profiles in children with acute lymphoblastic leukemia. *Cancer.* 1998;83(2):379–384.
16. Shaw JH, Wolfe RR. Fatty acid and glycerol kinetics in septic patients and in patients with gastrointestinal cancer. The response to glucose infusion and parenteral feeding. *Ann Surg.* 1987;205(4):368–376.
17. Stallings VA, Vaisman N, Chan HS, et al. Energy metabolism in children with newly diagnosed acute lymphoblastic leukemia. *Pediatr Res.* Aug 1989;26(2):154–157.
18. Fearon KC, McMillan DC, Preston T, et al. Elevated circulating interleukin-6 is associated with an acute-phase response but reduced fixed hepatic protein synthesis in patients with cancer. *Ann Surg.* 1991;213(1):26–31.
19. O'Keefe SJ, Ogden J, Ramjee G, Rund J. Contribution of elevated protein turnover and anorexia to cachexia in patients with hepatocellular carcinoma. *Cancer Res.* 1990;50(4):1226–1230.
20. Attard-Montalto SP, Camacho-Hubner C, Cotterill AM, et al. Changes in protein turnover, IGF-I and IGF binding proteins in children with cancer. *Acta Paediatr.* 1998;87(1):54–60.
21. Starnes HF, Jr., Warren RS, Jeevanandam M, et al. Tumor necrosis factor and the acute metabolic response to tissue injury in man. *J Clin Invest.* 1988;82(4):1321–1325.
22. Bond SA, Han AM, Wootton SA, Kohler JA. Energy intake and basal metabolic rate during maintenance chemotherapy. *Arch Dis Child.* 1992;67(2):229–232.

23. Reilly JJ, Ventham JC, Ralston JM, et al. Reduced energy expenditure in preobese children treated for acute lymphoblastic leukemia. *Pediatr Res.* 1998;44(4):557–562.
24. Garcia-Peris P, Lozano MA, Velasco C, et al. Prospective study of resting energy expenditure changes in head and neck cancer patients treated with chemoradiotherapy measured by indirect calorimetry. *Nutrition.* Nov–Dec 2005;21(11-12):1107–1112.
25. Ringwald-Smith KA, Heslop HE, Krance RA, et al. Energy expenditure in children undergoing hematopoietic stem cell transplantation. *Bone Marrow Transplant.* 2002;30(2):125–130.
26. Duggan C, Bechard L, Donovan K, et al. Changes in resting energy expenditure among children undergoing allogeneic stem cell transplantation. *Am J Clin Nutr.* 2003;78(1):104–109.
27. Bechard LJ, Feldman HA, Venick R, et al. Attenuation of resting energy expenditure following hematopoietic SCT in children. *Bone Marrow Transplant.* Oct 2012;47(10):1301–1306.
28. Sharma TS, Bechard LJ, Feldman HA, et al. Effect of titrated parenteral nutrition on body composition after allogeneic hematopoietic stem cell transplantation in children: a double-blind, randomized, multicenter trial. *Am J Clin Nutr.* Feb 2012;95(2):342–351.
29. Adamson PC, Bagatell R, Balis FM, Blaney SM. General principles of chemotherapy. In: Pizzo PA, Poplack DG, editors. *Principles and Practice of Pediatric Oncology*. 6th ed. Philadelphia: Lippincott Williams & Wilkins; 2011. pp. 279–355.
30. Copelan EA. Hematopoietic stem-cell transplantation. *N Engl J Med.* Apr 27 2006;354(17):1813–1826.
31. Wingard JR, Piantadosi S, Vogelsang GB, et al. Predictors of death from chronic graft-versus-host disease after bone marrow transplantation. *Blood.* Sep 1989;74(4):1428–1435.
32. Bhushan V, Collins RH, Jr. Chronic graft-vs-host disease. *JAMA.* Nov 19 2003;290(19):2599–2603.
33. Browning B, Thormann K, Seshadri R, et al. Weight loss and reduced body mass index: a critical issue in children with multiorgan chronic graft-versus-host disease. *Bone Marrow Transplant.* Mar 2006;37(5):527–533.
34. Tenardi RD, Fruhwald MC, Jurgens H, et al. Nutritional status of children and young adults with Ewing sarcoma or osteosarcoma at diagnosis and during multimodality therapy. *Pediatr Blood Cancer.* Oct 2012;59(4):621–626.
35. Barlow SE. Expert committee recommendations regarding the prevention, assessment, and treatment of child and adolescent overweight and obesity: summary report. *Pediatrics.* Dec 2007;120(Suppl 4):S164-S192.
36. Chomtho S, Fewtrell MS, Jaffe A, et al. Evaluation of arm anthropometry for assessing pediatric body composition: evidence from healthy and sick children. *Pediatr Res.* Jun 2006;59(6):860–865.
37. Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr.* 1981;34:2540–2545.
38. White M, Davies P, Murphy A. Correlation between nutrition assessment data and percent body fat via plethysmography in pediatric oncology patients. *J Parenter Enteral Nutr.* Nov 2011;35(6):715–722.
39. Jarfelt M, Lannering B, Bosaeus I, et al. Body composition in young adult survivors of childhood acute lymphoblastic leukaemia. *Eur J Endocrinol.* Jul 2005;153(1):81–89.

40. Kyle UG, Chalandon Y, Miralbell R, et al. Longitudinal follow-up of body composition in hematopoietic stem cell transplant patients. *Bone Marrow Transplant.* Jun 2005;35(12):1171–1177.
41. Warner JT, Evans WD, Webb DK, Gregory JW. Pitfalls in the assessment of body composition in survivors of acute lymphoblastic leukaemia. *Arch Dis Child.* Jan 2004;89(1):64–68.
42. Murphy AJ, Wells JC, Williams JE, et al. Body composition in children in remission from acute lymphoblastic leukemia. *Am J Clin Nutr.* Jan 2006;83(1):70–74.
43. Nysom K, Holm K, Michaelsen KF, et al. Degree of fatness after allogeneic BMT for childhood leukaemia or lymphoma. *Bone Marrow Transplant.* 2001;27(8):817–820.
44. Inaba H, Yang J, Kaste SC, et al. Longitudinal changes in body mass and composition in survivors of childhood hematologic malignancies after allogeneic hematopoietic stem-cell transplantation. *J Clin Oncol.* Nov 10 2012;30(32):3991–3997.
45. Elhasid R, Laor A, Lischinsky S, et al. Nutritional status of children with solid tumors. *Cancer.* Jul 1 1999;86(1):119–125.
46. Malvy DJM, Arnaud J, Burtschy B, et al. Antioxidant micronutrients and childhood malignancy during oncological treatment. *Med Pediatr Oncol.* 1997;29:213–217.
47. Modan-Moses D, Pinhas-Hamiel O, Munitz-Shenkar D, et al. Vitamin D status in pediatric patients with a history of malignancy. *Pediatr Res.* Dec 2012;72(6):620–624.
48. Bilariki K, Anagnostou E, Masse V, et al. Low bone mineral density and high incidences of fractures and vitamin D deficiency in 52 pediatric cancer survivors. *Horm Res Paediatr.* 2010;74(5):319–327.
49. Papadopoulou A, Nathavitharana K, Williams MD, et al. Diagnosis and clinical associations of zinc depletion following bone marrow transplantation. *Arch Dis Child.* 1996;74(4):328–331.
50. Kennedy DD, Ladas EJ, Rheingold SR, et al. Antioxidant status decreases in children with acute lymphoblastic leukemia during the first six months of chemotherapy treatment. *Pediatr Blood Cancer.* Apr 2005;44(4):378–385.
51. Trifilio S, Helenowski I, Giel M, et al. Questioning the role of a neutropenic diet following hematopoetic stem cell transplantation. *Biol Blood Marrow Transplant.* Sep 2012;18(9):1385–1390.
52. Moody K, Finlay J, Mancuso C, Charlson M. Feasibility and safety of a pilot randomized trial of infection rate: neutropenic diet versus standard food safety guidelines. *J Pediatr Hematol Oncol.* Mar 2006;28(3):126–133.
53. Guidelines for preventing opportunistic infections among hematopoietic stem cell transplant recipients: recommendations of CDC, the Infectious Disease Society of America, and the American Society of Blood and Marrow Transplantation. *MMWR.* Vol 49;RR-10: Centers for Disease Control and Prevention; 2000:50–52.
54. Bauer J, Jurgens H, Fruhwald MC. Important aspects of nutrition in children with cancer. *Adv Nutr.* Mar 2011;2(2):67–77.
55. Ladas EJ, Sacks N, Brophy P, Rogers PC. Standards of nutritional care in pediatric oncology: results from a nationwide survey on the standards of practice in pediatric oncology. A Children's Oncology Group study. *Pediatr Blood Cancer.* Mar 2006;46(3):339–344.
56. Imataki O, Nakatani S, Hasegawa T, et al. Nutritional support for patients suffering from intestinal graft-versus-host disease after allogeneic hematopoietic

stem cell transplantation. *Am J Hematol.* Oct 2006;81(10):747–752.
57. Couluris M, Mayer JL, Freyer DR, et al. The effect of cyproheptadine hydrochloride (periactin) and megestrol acetate (megace) on weight in children with cancer/treatment-related cachexia. *J Pediatr Hematol Oncol.* Nov 2008;30(11):791–797.
58. Azcona C, Sierrasesumaga L. Adrenal suppression in children with malignant solid tumors treated with megestrol acetate. *J Pediatr.* Jul 2000;137(1):141–142.
59. den Broeder E, Lippens RJ, van't Hof MA, et al. Effects of naso-gastric tube feeding on the nutritional status of children with cancer. *Eur J Clin Nutr.* 1998;52(7):494–500.
60. Papadopoulou A, MacDonald A, Williams MD, et al. Enteral nutrition after bone marrow transplantation. *Arch Dis Child.* 1997;77(2):131–136.
61. Langdana A, Tully N, Molloy E, et al. Intensive enteral nutrition support in paediatric bone marrow transplantation. *Bone Marrow Transplant.* 2001;27(7):741–746.
62. Hopman GD, Pena EG, Le Cessie S, et al. Tube feeding and bone marrow transplantation. *Med Pediatr Oncol.* Jun 2003;40(6):375–379.
63. Aquino VM, Smyrl CB, Hagg R, et al. Enteral nutritional support by gastrostomy tube in children with cancer. *J Pediatr.* 1995;127:58–62.
64. Mathew P, Bowman L, Williams R, et al. Complications and effectiveness of gastrostomy feedings in pediatric cancer patients. *J Pediatr Hematol Oncol.* 1996;18(1):81–85.
65. Pedersen AM, Kok K, Petersen G, et al. Percutaneous endoscopic gastrostomy in children with cancer. *Acta Paediatr.* Aug 1999;88(8):849–852.
66. Skolin I, Hernell O, Larsson MV, et al. Percutaneous endoscopic gastrostomy in children with malignant disease. *J Pediatr Oncol Nurs.* Sep-Oct 2002;19(5):154–163.
67. Parenteral nutrition in patients receiving cancer chemotherapy. American College of Physicians. *Ann Intern Med.* May 1 1989;110(9):734–736.
68. Schmid I, Schmitt M, Streiter M, et al. Parenteral nutrition is not superior to replacement fluid therapy for the supportive treatment of chemotherapy induced oral mucositis in children. *Eur J Cancer.* Jan 2006;42(2):205–211.
69. Weisdorf S, Lysne J, Wind D, et al. Positive effect of prophylactic total parenteral nutrition on long-term outcome of bone marrow transplantation. *Transplantation.* 1987;43(6):833–838.
70. Forchielli ML, Azzi N, Cadranel S, Paolucci G. Total parenteral nutrition in bone marrow transplant: what is the appropriate energy level? *Oncology.* 2003;64(1):7–13.
71. Christensen ML, Hancock ML, Gattuso J, et al. Parenteral nutrition associated with increased infection rate in children with cancer. *Cancer.* 1993;72(9):2732–2738.
72. Lenssen P, Bruemmer BA, Bowden RA, et al. Intravenous lipid dose and incidence of bacteremia and fungemia in patients undergoing bone marrow transplantation. *Am J Clin Nutr.* 1998;67(5):927–933.
73. Copeman MC. Use of total parenteral nutrition in children with cancer: a review and some recommendations. *Pediatr Hematol Oncol.* 1994;11:463–470.
74. Laborie S, Lavoie JC, Chessex P. Paradoxical role of ascorbic acid and riboflavin in solutions of total parenteral nutrition: implication in photoinduced peroxide generation. *Pediatr Res.* 1998;43(5):601–606.
75. Laborie S, Lavoie JC, Pineault M, Chessex P. Protecting solutions of parenteral nutrition from peroxidation. *J Parenter Enteral Nutr.* 1999;23(2):104–108.

76. Charuhas PM, Fosberg KL, Bruemmer B, et al. A double-blind randomized trial comparing outpatient parenteral nutrition with intravenous hydration: effect on resumption of oral intake after marrow transplantation. *J Parenter Enteral Nutr.* 1997;21(3):157–161.
77. Van der Hulst R, van Kreel B, von Meyenfeldt M, et al. Glutamine and the preservation of gut integrity. *Lancet.* 1993;334:1363–1365.
78. Klimberg V, Souba W, Dolson D, et al. Prophylactic glutamine protects the intestinal mucosa from radiation injury. *Cancer.* 1990;66:62–68.
79. Fox A, Kripke S, DePaula J, et al. Effect of glutamine-supplemented enteral diet on methotrexate-induced enterocolitis. *J Parenter Enteral Nutr.* 1988;12:325–331.
80. Ziegler TR, Young LS, Benfell K, et al. Clinical and metabolic efficacy of glutamine-supplemented parenteral nutrition after bone marrow transplantation. A randomized, double-blind, controlled study. *Ann Intern Med.* 1992;116(10):821–828.
81. Hardman WE. (n-3) Fatty acids and cancer therapy. *J Nutr.* Dec 2004;134(12 Suppl):3427S-3430S.
82. Bayram I, Erbey F, Celik N, et al. The use of a protein and energy dense eicosapentaenoic acid containing supplement for malignancy-related weight loss in children. *Pediatr Blood Cancer.* May 2009;52(5):571–574.
83. Oeffinger KC, Mertens AC, Sklar CA, et al. Obesity in adult survivors of childhood acute lymphoblastic leukemia: a report from the Childhood Cancer Survivor Study. *J Clin Oncol.* Apr 1 2003;21(7):1359–1365.
84. Diller L, Chow EJ, Gurney JG, et al. Chronic disease in the Childhood Cancer Survivor Study cohort: a review of published findings. *J Clin Oncol.* May 10 2009;27(14):2339–2355.
85. Warner JT, Bell W, Webb DK, Gregory JW. Daily energy expenditure and physical activity in survivors of childhood malignancy. *Pediatr Res.* 1998;43(5):607–613.
86. Nysom K, Holm K, Michaelsen KF, et al. Degree of fatness after treatment for acute lymphoblastic leukemia in childhood.[erratum appears in *J Clin Endocrinol Metab*. 2001 Apr;86(4):1758]. *J. Clin. Endocrinol. Metab.* 1999;84(12):4591–4596.
87. Cohen A, Rovelli A, Van-Lint M, et al. Final height of patients who underwent bone marrow transplantation during childhood. *Arch Dis Child.* 1996;74:437–440.
88. Bakker B, Oostdijk W, Geskus RB, et al. Patterns of growth and body proportions after total-body irradiation and hematopoietic stem cell transplantation during childhood. *Pediatr Res.* Feb 2006;59(2):2z9–264.
89. Kadan-Lottick N, Marshall JA, Baron AE, et al. Normal bone mineral density after treatment for childhood acute lymphoblastic leukemia diagnosed between 1991 and 1998. *J Pediatr.* Jun 2001;138(6):898–904.
90. Arikoski P, Komulainen J, Riikonen P, et al. Reduced bone density at completion of chemotherapy for a malignancy. *Arch Dis Child.* Feb 1999;80(2):143–148.
91. Marinovic D, Dorgeret S, Lescoeur B, et al. Improvement in bone mineral density and body composition in survivors of childhood acute lymphoblastic leukemia: a 1-year prospective study. *Pediatrics.* Jul 2005;116(1):e102–e108.
92. Ruza E, Sierrasesumaga L, Azcona C, Patino-Garcia A. Bone mineral density and bone metabolism in children treated for bone sarcomas. *Pediatr Res.* Jun 2006;59(6):866–871.
93. Schulte CM, Beelen DW. Bone loss following hematopoietic stem cell transplantation: a long-term follow-up. *Blood.* May 15 2004;103(10):3635–3643.
94. Petryk A, Bergemann TL, Polga KM, et al. Prospective study of changes in bone

mineral density and turnover in children after hematopoietic cell transplantation. *J Clin Endocrinol. Metab.* Mar 2006;91(3):899–905.
95. Shalitin S, Phillip M, Stein J, et al. Endocrine dysfunction and parameters of the metabolic syndrome after bone marrow transplantation during childhood and adolescence. *Bone Marrow Transplant.* 2006;37(12):1109–1117.
96. Gurney JG, Ness KK, Sibley SD, et al. Metabolic syndrome and growth hormone deficiency in adult survivors of childhood acute lymphoblastic leukemia. *Cancer.* Sep 15 2006;107(6):1303–1312.
97. Robien K, Ness KK, Klesges LM, et al. Poor adherence to dietary guidelines among adult survivors of childhood acute lymphoblastic leukemia. *J Pediatr Hematol Oncol.* Nov 2008;30(11):815–822.

Diabetes Mellitus

59

Joseph I. Wolfsdorf, MB, BCh, Sanjeev Mehta, MD, MPH, Maryanne Quinn, MD, and Karen Y. Warman, MS, RD

MEDICAL NUTRITION THERAPY

Meal planning is a cornerstone of diabetes management, and nutrition education is an essential component of a comprehensive program of diabetes education for patients and their families.[1] Recommendations for medical nutrition therapy (MNT) of diabetes mellitus have continued to evolve since the advent of insulin therapy in 1922. In this chapter, we briefly review the history of MNT, the scientific basis on which the general principles are founded, and recent investigations that implicate dietary factors in the etiology of immune-mediated or type 1a diabetes mellitus. Thereafter, we discuss the goals of treatment, principles of meal planning, and formulation of an individualized meal plan based on current evidence as recommended by the Food and Nutrition Committee of the American Diabetes Association (ADA).[2]

PREVALENCE, DEFINITION, AND CLINICAL DESCRIPTION OF DIABETES MELLITUS

Diabetes mellitus is one of the most common chronic diseases in children and adolescents. Diabetes is not a single entity but a group of metabolic diseases characterized by hyperglycemia that is caused by defects in insulin secretion, defects in insulin action, or both.[3] Type 1 diabetes, caused by absolute deficiency of insulin secretion, is the most common form in children and adolescents. Its incidence widely varies among geographic regions, from 0.1/100,000 per year in China and Venezuela to

64/100,000 per year in Finland. In the United States in 2010, an estimated 215,000 people below 20 years had diabetes (type 1 or type 2), which represents 0.26% of all people in this age group.[4] The estimated crude prevalence rate was 1.82 cases per 1000 youth, being lower for children 0–9 years of age (0.79 cases per 1000 youth) than for those 10–19 years of age (2.80 cases per 1000 youth).

Type 2 diabetes occurs when insulin secretion is inadequate to meet the increased demand caused by insulin resistance. Until recently, immune-mediated type 1 diabetes was by far the most prevalent form of diabetes in children and adolescents in the United States; only 1% to 2% of children had type 2 or other, rare, forms of diabetes. In the past two decades, the prevalence of type 2 diabetes in youth has increased at an alarming rate throughout the developed world, and recent reports indicate that 8%–45% of children with newly diagnosed diabetes have nonimmune-mediated diabetes.[5] Among younger children type 1 diabetes accounts for at least 80% of diabetes, whereas among older youth the proportion of type 2 diabetes ranges from 6% for non-Hispanic white youth to 76% for American Indian youth.[6] The variation in incidence depends on age, race/ethnicity, and sampling strategy. Most children and adolescents who do not have immune-mediated diabetes have type 2 diabetes; however, other types, especially monogenic diabetes (formerly referred to as maturity-onset diabetes of youth), are being increasingly identified in adolescents and young adults and account for 1% to 2% of youth with diabetes.[7]

Type 1a diabetes occurs in genetically predisposed individuals as a consequence of immune-mediated destruction of the insulin-secreting β-cells of the pancreatic islets.[8] The onset of clinically overt diabetes represents a late stage in the process of chronic progressive immune-mediated β-cell destruction. The resulting absolute deficiency of insulin leads to hyperglycemia (the result of decreased uptake and utilization of glucose by liver, muscle, and adipose tissue and uncontrolled hepatic gluconeogenesis), unregulated release of free fatty acids from adipose tissue, and increased ketogenesis. The disease typically presents with polyuria, polydipsia, and weight loss.

Patients with type 1 diabetes depend on exogenous insulin to survive. Insulin is normally secreted in an oscillatory fashion at a low basal rate with periodic increases in secretion in response to ingestion of food. In contrast, when insulin is injected subcutaneously it is absorbed from the injection site in a somewhat predictable fashion depending on the specific types of insulin used. Each type of insulin has a characteristic time of onset

of action and maximum effect, duration of peak effect, and total duration of action (Table 59-1). When short- or intermediate-acting insulins are used, meals and snacks have to be timed to match the pharmacodynamics of insulin. Rapid-acting insulin analogs, delivered by injection or via continuous subcutaneous insulin infusion (CSII) (insulin pump), significantly reduce the variation in absorption of insulin and permit greater flexibility and accuracy with respect to matching insulin delivery to glucose absorption after meals and snacks.

Children with idiopathic or nonimmune-mediated type 1 diabetes may be difficult to distinguish from those with immune-mediated diabetes. In the United States, most patients with idiopathic type 1 diabetes are African-American and have so-called atypical diabetes mellitus or Flatbush diabetes.[9] They typically have a positive family history of early-onset diabetes in several relatives and in multiple generations. Although insulin may not be required for survival after the episode of acute metabolic deterioration resolves, without insulin therapy blood glucose control is usually poor and ketoacidosis may recur.

TABLE 59-1 Insulin Preparations Classified According to Their Pharmacodynamic Profiles

	Onset of Action	Peak Action	Duration of Action
Rapid acting[a]		30–90 min	3–5 h
Insulin lispro (Humalog®)[b]	5–15 min	30–90 min	3–5 h
Insulin aspart (Novolog®)[b]	5–15 min	30–90 min	3–5 h
Insulin glulisine (Apidra®)[b]	5–15 min		
Short acting[a] Regular (soluble)	30–60 min	2 to 3 h	5–8 h
Intermediate acting[a] NPH (isophane)	2–4 h	4–10 h	10–16 h
Long acting	2–4 h	Peakless	20–24 h
Insulin glargine (Lantus®)[b]	2–4 h	6–14 h	16–20 h
Insulin detemir (Levemir®)[b]			

[a]Premixed combinations of intermediate-acting and either rapid- or short-acting insulins are available whose pharmacodynamic profiles have a bimodal pattern reflecting the two insulin components.

[b]Insulin analog developed by modifying the amino acid sequence of the human insulin molecule.

Type 2 diabetes is characterized by decreased insulin sensitivity and inadequate insulin secretion and is commonly associated with other features of the insulin resistance syndrome (hyperlipidemia, hypertension, acanthosis nigricans, ovarian hyperandrogenism, and nonalcoholic fatty liver disease).[10] Changes in energy intake and expenditure have resulted in an epidemic of obesity in children and adolescents in the United States, and in the past two decades there has been an alarming increase in the prevalence of type 2 diabetes in children and adolescents, especially in minority populations.[10] Almost 95% of youth with type 2 diabetes are either overweight or frankly obese.[11] Body composition is a major determinant of insulin sensitivity in children. Adiposity accounts for 55% of the variance in insulin sensitivity in white children,[12] and an inverse relationship between increasing adiposity and insulin sensitivity has also been shown in black children. As adiposity and body mass index (BMI) increase, insulin sensitivity decreases and serum insulin levels increase.[13]

Monogenic forms of diabetes are caused by defects in β-cell function and are inherited in an autosomal dominant pattern.[7] Monogenic diabetes occurs in all racial and ethnic groups and has a broad clinical spectrum ranging from asymptomatic hyperglycemia to a severe acute presentation. Genetic testing is required to confirm specific diagnoses.[7]

Individuals with nonimmune-mediated diabetes may have clinical presentations indistinguishable from those of patients with immune-mediated type 1 diabetes. This is especially relevant because as the number of children with type 2 diabetes increases it is important to determine correctly the cause of a child's diabetes to be able to institute appropriate therapy. When the type of diabetes is uncertain, initial stabilization and management with insulin therapy should be the default approach to management.

The initial classification is usually based on the clinical and laboratory assessment at presentation. Children with immune-mediated type 1a diabetes are typically not overweight, have a short duration of symptoms, and frequently have ketosis, and approximately 15%–70% of newly diagnosed patients in North America and Europe present with ketoacidosis. However, as the childhood population in the United States has become increasingly overweight the percentage of children with immune-mediated type 1 diabetes who are coincidentally overweight, or obese, has increased. After metabolic stabilization, they may have an initial period of reduced insulin requirement (the honeymoon period); thereafter, they require daily insulin replacement for survival and are

always at risk for ketoacidosis. In contrast, approximately 95% of children with type 2 diabetes are either overweight or obese at the time of diagnosis and typically present with glucosuria, absent or mild polyuria and polydipsia, and little or no weight loss. In contrast to new-onset type 1 diabetes, only one-third of children diagnosed with type 2 diabetes have ketonuria at diagnosis, and 5%–25% present with ketoacidosis. Children with type 2 diabetes are usually more than 10 years old and are in middle to late puberty; 45%–80% have at least one parent with type 2 diabetes, and 74%–100% have a first- or second-degree relative with type 2 diabetes. Children and adolescents of African-American, Hispanic, Asian, American Indian, and Pacific Islander descent are disproportionately affected.[6]

Acute, life-threatening consequences of diabetes are hyperglycemia with ketoacidosis or the hyperglycemic hyperosmolar state syndrome from inadequate insulin, and severe hypoglycemia from excess insulin administration or, in the case of type 2 diabetes, oral antihyperglycemic medications. Long-term complications of chronic hyperglycemia include retinopathy with potential loss of vision; nephropathy leading to hypertension and renal failure; and peripheral and autonomic neuropathy with associated risk of foot ulcers and limb amputation and gastrointestinal, genitourinary, and cardiovascular symptoms.[14] Glycation of tissue proteins and other macromolecules and excess production of polyol compounds are among the mechanisms whereby chronic hyperglycemia produces tissue damage.[15]

HISTORY OF MEDICAL NUTRITION THERAPY FOR TYPE 1 DIABETES MELLITUS

Until the advent of insulin therapy in 1922, the only available treatment was diet and exercise. Meal plans severely restricted total energy intake and were low in carbohydrate and high in fat. Nutrition recommendations were often based on dogma and a limited understanding of pathophysiology. After the introduction of insulin, nutrition recommendations changed but generally involved restriction of carbohydrate to a maximum of 40% of total energy because this was thought to reduce the severity of hyperglycemia. Several studies early in the 20th century showed that a high-carbohydrate meal plan, provided it was restricted in total energy, had beneficial metabolic effects. Nonetheless, change was slow and, despite evidence to the contrary,

patients continued to be indoctrinated to use a low-carbohydrate, high-fat meal plan until 1971 when the Food and Nutrition Committee of the ADA formally recommended liberalizing the consumption of carbohydrate. Sucrose was not permitted in the meal plans of people with diabetes mellitus. Considerable scientific evidence since then has shown that sucrose as part of the meal plan does not impair blood glucose control in people with either type 1 or type 2 diabetes (see the section Nutritive Sweeteners).

In 1950, the ADA, American Dietetic Association, and US Public Health Service introduced the "exchange system" for meal planning to assist patients in choosing foods consistent with current nutrition recommendations.[16] The exchange system established portion sizes and grouped foods with similar macronutrient contents. The goal of the system was to enable individuals with diabetes to have greater flexibility in food choices by allowing them to exchange foods within a group and still consume comparable amounts of macro- and micronutrients. This was an important advance because it antedated the current widespread availability of standardized food composition analysis. Legislation mandating food labeling was not introduced in the United States until 1974. The exchange system has undergone four revisions. The 1976 revision improved the accuracy of the calorie content of foods within exchange lists and stratified foods by their fat content. The 1986 revision developed a database for nutrient composition and increased the carbohydrate content of fruits from 10 to 15 g, which is closer to their actual carbohydrate content. The 1995 revision grouped carbohydrate-containing foods into a single category to allow greater flexibility in meal planning. This revision also added many new food items (e.g., fat-modified, vegetarian, and fast foods) to reflect the US population's changing dietary habits and food choices. The most recent revision, in 2003, liberalized the use of sugar in the diet by establishing a food list entitled "Sweets, Desserts and Other Carbohydrates that could be exchanged for Grains, Fruits and Dairy." Another new category, "Combination foods," was added in response to the increased availability of processed foods.

Changes in food manufacturing and societal dietary habits, including increases in the consumption of prepared foods, access to international cuisines, eating meals outside the home, and modification of natural foods to lower their fat and carbohydrate content, have resulted in added complexity when teaching the exchange diet. For example, historically, bread contained 15 g of carbohydrate per ounce and presliced bread was generally sold in 1-oz. portions. Currently, some bread recipes are

formulated to provide 4 g of carbohydrate per ounce; thus, it has become important to understand the "evolution" of carbohydrate content in bread in order to explain why multiple bread slices may need to be consumed in order to satisfy the historical "15 gram" expectation within the exchange diet. Therefore, teaching patients and families how to perform carbohydrate counting will enable them to avoid problems with the exchange diet that are the result of altered manufacturing and consumption practices. It is also important to recognize that the portion size on food labels may differ from the portion size and carbohydrate content listed in the exchange system. Most food manufacturers do not list diabetes exchanges on their products. Since the introduction of mandatory food labeling, the general public is becoming more accustomed to reading nutrition facts on food labels. It is no longer essential, therefore, for people with diabetes to rely on the exchange system as the sole source of information regarding macronutrients for dietary planning.

In 1993, the landmark Diabetes Control and Complications Trial (DCCT) showed that a system of intensive diabetes management aimed at near-normal glycemic control dramatically reduces the risk of microvascular complications and favorably affects the risk of macrovascular complications.[17] Intensive insulin therapy using multiple daily injections (basal-bolus or flexible insulin regimen) or an insulin pump has become the standard of care for type 1 diabetes. Flexible insulin regimens in conjunction with carbohydrate counting (estimating the carbohydrate content of foods based on information available from food labels, reference books, and software) allow patients with type 1 diabetes to have a wide variety of dietary choices and to live more normal lifestyles without compromising glycemic control. Carbohydrate counting is discussed in more detail later in this chapter.

DIETARY FACTORS IN THE PATHOGENESIS OF TYPE 1 DIABETES MELLITUS

The increasing incidence of type 1 diabetes in recent decades[18] and the concordance rate of less than 50% in monozygotic twins suggest a role for environmental factors in the development of type 1 diabetes. The relatively greater increase in incidence in children below 5 years suggests that environmental exposures early in life could play an important pathogenetic role.[19] However, the contributions of genetic, environmental, and immunologic factors and their interactions in the pathogenesis of type 1a

diabetes are poorly understood.

Early nutrition is one of the first environmental determinants to which an infant is exposed, and evidence suggests that components of the diet may predispose to or protect against type 1 diabetes. Short duration of breast-feeding and early exposure to complex foreign proteins have been implicated as risk candidates. Other possible diet-related factors that have been implicated include fruits, berries, root vegetables, lack of vitamin D supplementation, heavy birth weight, and rapid weight gain in infancy.[20] It has been suggested that a dietary antigen may play a role as the factor driving the disease process toward clinical type 1 diabetes.

Changes in food, particularly its protein content, influence the onset of diabetes in rodent models of autoimmune diabetes, and these observations have been invoked as evidence that complex natural ingredients in human and rodent diets may be involved in the immune-mediated destruction of β-cells. Antibodies reactive against a peptide sequence contained within bovine serum albumin occur in humans, BB rats, and NOD mice, which cross-react with an islet antigen designated p69. Together with suggestive epidemiological data, these observations have given rise to the controversial theory that dietary cow's milk may contribute to the pathogenesis of type 1a diabetes in susceptible individuals. Removal of bovine proteins from the diet prevents diabetes in NOD mice. Some investigators have also shown that diets containing cow's milk are diabetogenic in the BB rat, but other investigators have not confirmed this. Although diet clearly has a major diabetogenic effect in BB rats and NOD mice, the exact chemical identity of the food diabetogens is still unclear.

Insulin is an important autoantigen in type 1a diabetes and is the only β-cell-specific autoantigen in postnatal life.[8] Insulin autoantibodies are the first or among the first autoantibodies to appear when the disease process leading to type 1a diabetes is initiated in young children. Because cow's milk formulas are generally the first and most common dietary source of foreign complex proteins to which infants are exposed in developed countries, the question has been asked whether early exposure to cow's milk formula results in an immune response to bovine insulin (which differs from human insulin in three amino acids) present in infant formulas. Bovine insulin is thought to be an attractive candidate because an immune response, initially induced by bovine insulin, cross-reacts with and may target human insulin in the β-cell.[21] Infants exclusively breast-fed up to the age of 3 months had substantially lower IgG class antibodies to bovine insulin than those exposed to formula before 3 months of age.[22] The IgG class insulin antibodies started to decrease slowly after the age of

3 months as a sign of the development of oral tolerance in those infants who were given formula before the age of 3 months. In contrast, antibody levels increased up to the age of 12 months before titers leveled off in infants exclusively breast-fed to the age of at least 3 months. It has been suggested that young children who present early signs of β-cell autoimmunity lack the capacity to develop oral tolerance to bovine insulin.[21] In these individuals, the initial immune response to bovine insulin appears to be diverted to a response targeting human insulin.

Data from three population-based case-control studies on cow's milk intake before the diagnosis of type 1 diabetes have yielded conflicting results. In New South Wales, Australia, cow's milk intake was higher in children who subsequently developed diabetes than in control children,[23] and in Finland a high consumption of cow's milk in childhood was associated with more frequent appearance of diabetes-associated autoantibodies and type 1 diabetes in a prospective cohort of initially unaffected siblings of children with type 1 diabetes.[24,25] In contrast, a Swedish retrospective survey showed that the frequency of cow's milk consumption was modestly lower among children who presented with type 1 diabetes than among unaffected children.[26] If confirmed, the increased risk of diabetes associated with formula feeding could reflect early exposure to a variety of cow's milk proteins, modification of the gut microflora,[27] and increased weight gain early in life.[28] A recent pooled analysis of data from 43 observational studies (9874 participants) suggests that exclusive breast-feeding in the early weeks of life (for more than two or three weeks) can reduce the risk of childhood-onset type 1 diabetes by 15%. There is little evidence, however, that longer exclusive or nonexclusive breast-feeding has a protective effect.[19] Knip et al.[27] have speculated that breast-feeding could protect against type 1 diabetes risk by causing a more rapid decrease in gut permeability or by reducing the risk of enteroviral infections, which have been associated with increased type 1 diabetes risk.[29]

A pilot intervention study sought to determine whether weaning to hydrolyzed formula over the first six to eight months of life decreased the cumulative incidence of diabetes-associated autoantibodies. Preliminary evidence has shown a decreased frequency of at least one diabetes-associated autoantibody including, specifically, islet cell antibody by the age of 6 years. In 2002, a randomized double-blind trial (Trial to Reduce IDDM in the Genetically at Risk) was initiated to determine whether it is possible to reduce the frequency of diabetes-associated autoantibodies

and/or clinical type 1 diabetes by the age of 6 years and the cumulative incidence of type 1 diabetes by the age of 10 years by weaning to a highly hydrolyzed formula over the first six to eight months of life.[27] During a median observation period of 10 years, weaning to a casein hydrolysate formula during infancy was associated with a lower risk of developing β-cell autoimmunity markers compared to weaning to cow's milk formula.[30]

Gluten or other cereal-derived proteins have also been implicated as potential driving antigens in type 1 diabetes. Dietary gliadin, a component of gluten found in wheat and other grains, may trigger intestinal inflammation in subjects with type 1 diabetes, independent of the presence of the human leukocyte antigen (HLA) DQ2/DQ8 haplotype known to confer risk for celiac disease. It has been suggested that gliadin could affect the immune activation stage of the gut immune system in individuals at genetic risk for type 1 diabetes and modify the autoimmune process involved in β-cell destruction.[21] Two prospective studies have reported an association between the introduction of cereals in infancy and the emergence of early β-cell autoimmunity. A study conducted in the United States suggested that both early (<4 months) and late (≥7 months) exposures to cereals, either gluten- or non-gluten-containing cereals, were associated with an increased risk of β-cell autoimmunity.[31] A similar study in Germany suggested that an increased risk of β-cell autoimmunity was related to exposure to cereals before age 3 months.[32] In contrast, a prospective analysis of data from the DIPP study conducted in Finland showed no relation between early or late introduction of cereals and emergence of advanced β-cell autoimmunity.[33] In small intervention studies performed in family members already positive for pancreatic autoantibodies, gluten elimination from the diet did not significantly affect autoantibody titers nor did titers change when gluten was reintroduced into the diet. A gluten-free diet does not appear to influence β-cell autoimmunity in first-degree relatives of affected patients.

There is seasonal variation in the initial appearance of pancreatic autoantibodies in young children with increased HLA-conferred susceptibility to type 1 diabetes. Seasonal viral infections have been implicated but not proved. There is marked seasonal variation in the amount of daylight and hours of sunshine, especially in Northern Europe, which has the highest incidence of type 1 diabetes in the world. Sun exposure may affect the immune system, possibly mediated by vitamin D. In 2003, Stene and Joner reported that the use of cod liver oil (containing high concentrations of vitamin D and ω-3-polyunsaturated fatty acids)

during the first year of life was related to a decreased risk of type 1 diabetes,[34] but the observation was not confirmed in a later study.[35] A lack of oral vitamin D supplementation in infancy may increase the subsequent risk of type 1 diabetes, and a recent meta-analysis based on five studies concluded that vitamin D supplementation in early childhood may provide protection against the development of type 1 diabetes.[36] There is countervailing evidence, however, arguing against a role of vitamin D deficiency as a trigger of β-cell autoimmunity. About 80% of all young children in Northern Europe receive a daily vitamin D supplement at least up to the age of 1 year. Furthermore, there were no significant differences in the circulating vitamin D concentrations in pregnant women and schoolchildren between the Karelian Republic of Russia, where the incidence is approximately one-sixth that of neighboring Finland, and Finland despite similar HLA-conferred genetic susceptibility.[37]

Increased weight gain in infancy is also a risk factor for developing type 1 diabetes.[38] Children with type 1 diabetes are both heavier and taller in infancy (despite no differences in birth weight or length) compared to unaffected children.[39] Individuals who develop type 1 diabetes between 15 and 39 years of age have a faster gain in BMI during infancy than unaffected individuals.[40] Accelerated linear growth and weight gain result in increased β-cell load and insulin resistance, and rapid growth and weight gain may induce β-cell stress. Indeed, compared to resting cells active β-cells are more prone to cytokine-induced damage.[41] According to Wilkin's accelerator hypothesis, rapid weight gain is an important factor accounting for the rising incidence of both type 1 and type 2 diabetes, but mediation through insulin resistance has yet to be established.[42,43]

Although no specific dietary factor or nutrient in infancy has, unequivocally, been shown to play a role in the development of type 1 diabetes, dietary components may predispose to or protect against type 1 diabetes.[27] Therefore, at the present time no definitive nutrition recommendations can be made for preventing type 1 diabetes. However, increasing BMI is associated with a younger age at diagnosis of type 1 diabetes in youth with reduced β-cell function,[44] and excessive weight gain during childhood should be avoided.

GOALS OF MEDICAL NUTRITION THERAPY

Medical nutrition therapy (MNT) is the process by which the nutrition prescription is tailored for the individual with diabetes based on medical,

lifestyle, and personal factors and is an integral component of diabetes management and diabetes self-management education.[45] The goals of MNT for children and adolescents are shown in Table 59-2.

TABLE 59-2 Goals of MNT for Diabetes Mellitus

Goals that apply to all persons with diabetes	Attain and maintain optimal metabolic outcomes including:
	Blood glucose levels in the normal range or as close to normal as is safely possible
	A lipid and lipoprotein profile that reduces the risk for vascular disease
	Blood pressure levels in the normal range or as close to normal as is safely possible
	To prevent, or at least slow, the rate of development of the chronic complications of diabetes by modifying nutrient intake and lifestyle.
	To address individual nutrition needs, taking into account personal and cultural preferences and willingness to change
	To maintain the pleasure of eating by only limiting food choices when indicated by scientific evidence
Goals specific to type 1 diabetes	To provide adequate energy to ensure normal growth and development and integrate insulin regimens into usual eating and physical activity habits
Goals specific to type 2 diabetes	To facilitate changes in eating and physical activity habits that reduce weight and insulin resistance, and improve metabolic control
Goals specific to patients treated with insulin or insulin secretagogues	To provide self-management education for prevention and treatment of hypoglycemia, acute illnesses, and exercise-related blood glucose problems.

Source: Adapted from Bantle JP, Wylie-Rosett J, Albright AL, et al. Nutrition recommendations and interventions for diabetes—2006: a position statement of the American Diabetes Association. *Diabetes Care*. Sep 2006;29(9):2140–2157[74]; Franz MJ, Bantle JP, Beebe CA, et al. Nutrition principles and recommendations in diabetes. *Diabetes Care*. Jan 2004;27(Suppl 1):S36–S46.[119]

TABLE 59-3 ADA-Recommended Glycemic Targets for Type 1 Diabetes Across all Pediatric Age Groups

Blood glucose goal range		
Before meals	**Bedtime/overnight**	**A1c**
90–130 mg/dL	90–150 mg/dL	<7.5%*

*A lower goal (<7%) is reasonable if it can be achieved without excessive hypoglycemia.

Key concepts in setting glycemic goals:

- Goals should be individualized, and lower goals may be reasonable based on benefit-risk assessment.
- Blood glucose goals should be modified in children with frequent hypoglycemia or hypoglycemia unawareness.
- Postprandial blood glucose values should be measured when there is a discrepancy between preprandial blood glucose values and A1c levels and to help assess glycemia in those on basal-bolus regimens.

Source: American Diabetes Association. Children and adolescents. Sec. 11. In Standards of Medical Care in Diabetes–2015. Diabetes Care 2015;38 (Suppl. 1): S70–S76.

The DCCT and the observational follow-up study of the DCCT cohort, the Epidemiology of Diabetes Interventions and Complications (EDIC) study, confirmed that long-term maintenance of blood glucose at levels near to normal delays or prevents the microvascular complications of type 1 diabetes[17] and the benefits of intensive treatment extend well beyond the period of its most vigorous implementation.[46] The DCCT also demonstrated that intensive management of type 1 diabetes is associated with an increased risk of severe hypoglycemia and a tendency to gain weight. Weight gain in intensively treated patients with type 1 diabetes has been primarily attributed to the elimination of glucosuria (70% of the weight gain) and a 5% reduction in daily energy expenditure (30% of the weight gain). Recurrent symptomatic hypoglycemia requiring oral carbohydrate to restore normal blood glucose levels also contributes to the tendency to gain weight in some intensively treated patients.

Our ability to help most patients achieve optimal glycemic control is limited by the imperfection of insulin replacement, which does not mimic normal physiology; inability to continuously monitor blood glucose concentrations; and risk of severe hypoglycemia when glucose levels are near to normal. In the future, insulin delivery regulated by a closed-loop feedback system will likely improve our ability to tightly control blood glucose and hemoglobin A1c levels for all individuals with diabetes.

The child with diabetes and his or her family have to assume primary responsibility for daily management, whereas the role of the health-care professional is that of educator, advisor, and motivator. Even modest

objectives are not achievable in all patients because educational, psychosocial, and economic factors make it impossible for some children and their families to perform daily all the tasks that comprise a comprehensive treatment program. Nonetheless, the goals of therapy can be accomplished in a substantial number of patients by using flexible insulin regimens and insulin dosage algorithms in conjunction with frequent self-monitoring of blood glucose (supplemented by continuous glucose monitoring), individualized meal planning, and educating and motivating older children and adolescents. The ultimate goal is to empower patients and enable them to independently make appropriate decisions about their day-to-day diabetes management.

GENERAL PRINCIPLES OF NUTRITION MANAGEMENT

There is little or no research on the unique nutrient requirements of children and adolescents with either type 1 or type 2 diabetes nor is there evidence that their nutritional needs differ from those of children without diabetes. Therefore, nutrient recommendations are based on requirements of the general pediatric population. Meal plans for children with diabetes should begin with the key recommendations outlined in Dietary Guidelines for Healthy Americans 2010.[47] This document provides guidelines that should serve as the foundation for nutrition management in each of the following areas: balancing calories to manage weight, foods and food components to optimize food safety, and the rationale and process for developing healthy eating patterns. The guidelines address issues specific to children and may require further definition as they relate to diabetes management. For example, the guidelines suggest that carbohydrate choices should include fiber-rich fruits, vegetables, and whole grains, and added sugar should be minimized.[47] People with diabetes treated with insulin or insulin secretagogues must be aware of the total amount of carbohydrate eaten at a meal and have a form of oral glucose available at all times to treat hypoglycemia.

ENERGY

It is important to assess a child’s usual dietary intake before the onset of diabetes to assist with the formulation of the initial meal plan. Clinical dietitians frequently use equations such as the Schofield, World Health

Organization (WHO), Harris-Benedict, or estimated energy requirements for age to calculate energy requirements (see Appendix II). Because energy requirements change with age, physical activity, and growth rate, an assessment of height, weight, and energy intake is recommended every three to six months.[48]

Growth is an excellent indicator of the adequacy of energy intake, which should be evaluated by regularly plotting weight and stature on WHO growth charts for children less than 3 years of age and Centers for Disease Control and Prevention (CDC) pediatric growth charts (http://www.cdc.gov/growthcharts) for older children. In addition to adequate daily caloric intake, good metabolic control is essential for normal growth and development. If growth is not optimal, the diet should be reviewed and, in patients with type 1 diabetes, one should screen for evidence of inadequate insulin delivery, hypo- or hyperthyroidism, celiac disease (see later), and adrenal insufficiency. For overweight and obese youth with either type 1 or type 2 diabetes, energy consumption must be reduced to arrest weight gain while supporting normal linear growth and physical development.

DISTRIBUTION OF MACRONUTRIENTS

Many studies have attempted to determine the optimal macronutrient combination that should comprise total energy intake. The general consensus is that a single prescription does not exist. The dietary reference intake (DRI) is cited as a guideline that should be used to formulate the initial prescription. DRI reports suggest a mix of 45%–65% of calories from carbohydrate, 20%–35% from fat, and 10%–35% from protein. Once the nutrition prescription is determined, the proportion of macronutrients can be modified, as necessary, according to blood glucose and plasma lipid goals and requirements for growth and development.

CARBOHYDRATE

Control of blood glucose in an effort to maintain normal or near to normal levels is the primary goal of diabetes management. Because dietary carbohydrate is the major determinant of postprandial glucose levels, food and nutrition interventions that reduce postprandial blood glucose excursions are extremely important.[49] Blood glucose concentrations after a meal are primarily determined by the rate of appearance of glucose in the blood (digestion and absorption) and its clearance from the circulation.

Normal insulin secretory response maintains blood glucose concentrations in a narrow range. In patients with diabetes, however, defects in insulin secretion, defects in insulin action, or both impair normal regulation of postprandial blood glucose concentrations in response to dietary carbohydrate. The amount, type, and source of carbohydrate in foods, as well as the dose and time interval between administration of rapid- or short-acting insulin and consumption of the meal, influence postprandial glucose levels.

The diet should include carbohydrates from fruits, vegetables, whole grains, legumes, and low-fat dairy products. Until the 1980s, dietary dogma had been that people with diabetes should avoid simple sugars and replace them with complex carbohydrates. This belief was based on the assumption that simple sugars are more rapidly digested and absorbed than starches and, therefore, would lead to more pronounced postprandial hyperglycemia. The blood glucose responses to various carbohydrate-containing foods have been extensively investigated and there is little scientific evidence to support this notion. It is now clear that there is a wide spectrum of biologic responses to different complex and simple carbohydrates with so much overlap that they cannot be simply classified into two distinct groups.

Numerous factors influence the glycemic response to food, including the amount of carbohydrate, type of sugar (glucose, fructose, sucrose, and lactose), nature of the starch (amylose, amylopectin, and resistant starch), cooking and food processing (degree of starch gelatinization, particle size, and cellular form), and food structure, as well as other food components (fat and natural substances that slow digestion—lectins, phytates, tannins, fiber, and starch-protein and starch-lipid combinations). Furthermore, the glycemic response to a particular carbohydrate food is dependent on whether it is eaten alone or as part of a mixed meal. Fasting and preprandial blood glucose concentrations, severity of glucose intolerance, and second meal effect (the ability of a meal to diminish the glucose response to carbohydrates in the subsequent meal) are other factors that can affect the glycemic response to foods.

The glycemic index (GI), proposed in 1981 as an alternative system for classifying carbohydrate-containing food, measures the glycemic response after ingestion of carbohydrate. Carbohydrates were previously classified only on the basis of their chemical structure: sugars, starches, or fiber. GI is defined as the incremental area under the plasma glucose response curve over two hours after consumption of a standard amount (usually a 50-g portion) of carbohydrate from a test food relative to that of a control food,

either white bread or glucose.[50] In individuals without diabetes, the glycemic and hormonal responses to a large number of ingested carbohydrates have been systematically examined and their GIs defined. A low-GI food releases glucose more slowly, leading to a flatter postprandial blood glucose curve, whereas a high-GI food causes a more rapid rise in blood glucose levels.

As a result of these studies, the validity of the dogma that ingestion of simple sugars causes a rapid and pronounced rise in blood glucose whereas complex carbohydrates produce only modest increments in blood glucose concentration began to be disputed. Even a single food produces a substantially different glycemic response when it is prepared in different ways. For example, a pureed apple causes a greater increment in blood glucose than does an apple eaten whole. Similarly, ground rice or rice flour causes a greater glycemic and insulinemic response than whole cooked rice and cooking increases the glycemic response to starch. Blood glucose levels after ingesting wheat flour in the form of pasta are lower than those after an equal amount of wheat flour in bread. Thus, the form of a carbohydrate-containing food, in addition to its chemical composition, influences its glycemic effect. The effect of food form alters the rate of digestion and absorption and, therefore, affects postprandial glycemia. Fruits (e.g., apples GI 40, oranges GI 48, peaches GI 42, and pears GI 38 vs. watermelon GI 72) and milk (e.g., whole milk GI 27 and skim milk GI 32)[51,52] cause a lower glycemic response than most starches, and sucrose causes a glycemic response similar to that of bread, rice, and potatoes. In general, most refined starchy foods eaten in the United States have a high GI, whereas nonstarchy vegetables, fruits, and legumes tend to have a low GI. Although insulin- and glucose-lowering medications for type 2 diabetes can adequately control the glycemic impact of added sucrose and other sugars, their impact on total caloric intake suggests that moderation in the use of added sugars is still warranted.

Use of the GI of a food alone ignores the glycemic response if the portion is other than 50 g. The concept of glycemic load was introduced to more accurately describe the postprandial glycemic response to a food when eaten in a typical portion size. The glycemic load is determined by multiplying the GI of a food by the amount (in grams) of carbohydrate in the portion size consumed. For example, 12 medium carrots contain 50 g of carbohydrate. Although carrots have a high GI, a typical portion would not significantly increase the glycemic load of the meal. The glycemic load of a meal reflects the sum of the contributions of each food that is eaten.

The practical usefulness of low-GI diets in type 1 diabetes is beginning

to be embraced as a therapeutic option that potentially enhances overall glycemic control. In 2006, the ADA rated the evidence for use of low-GI diets as an E (evidence based on expert opinion). By 2008, the ADA increased its support to a B (evidence from controlled cohort studies). A meta-analysis of randomized controlled trials (virtually all adult studies) shows that choosing low-GI foods in preference to conventional or high-GI foods has a small (0.4% decrement in hemoglobin A1c) but clinically beneficial effect on medium-term glycemic control in patients with diabetes.[53] It should be noted that no negative effects of the low-GI diet have been documented; however, if GI and load were used as the only criteria for meal planning the results could be a diet high in fat and low in vitamins and minerals. ChooseMyPlate.gov guidelines unveiled in 2011 by the US Department of Agriculture depicts a standard plate with recommended serving sizes from grains, fruits, vegetables, and protein with dairy and fats to the side. When the concepts of GI and load are taught within the context of these nutritionally based guidelines, the result is a healthy diet. This is a useful approach to improving glycemic control, especially in patients with pronounced postprandial hyperglycemia.

Concerns about the acceptance of lower GI foods have been expressed as a reason not to introduce a low-GI diet. At a diabetes summer camp for 7- to 16-year-old children, satisfaction ratings were equal when dinner and snacks containing lower GI foods replaced standard food choices; however, they were lower (although within the acceptable range) at breakfast and lunch.[54] In a different study, 8- to 13-year-old children who received low-GI dietary advice did not report more limited food choices or compromise their macronutrient intake compared to children who planned their meals using a traditional exchange system.[55] Once patients have mastered the concept of matching carbohydrate intake to insulin (discussed later), we recommend providing additional guidance on how to lower the GI of the diet.

FIBER

Fiber refers to the indigestible portion of a plant, and previously it was thought to have no nutritive value in humans. It is now known that fiber markedly influences the digestion, absorption, and metabolism of many nutrients. The fibers (cellulose, lignins, and certain hemicelluloses) in vegetables and grains are usually insoluble in water, whereas many fruits and certain legumes are rich in water-soluble fibers such as pectins, guar, and storage polysaccharides. When carbohydrate is ingested together with

soluble fiber, the blood glucose concentration increases less than when carbohydrate is ingested alone. A large test meal containing carbohydrate, protein, and fat causes significantly less postprandial glycemia when the meal is supplemented with guar due to delayed rather than incomplete absorption of carbohydrate.

Small, short-term studies using large amounts of fiber (>30 g/d) in suboptimally controlled individuals with type 1 diabetes suggest a positive effect of fiber on glycemia; however, in participants using intensive insulin therapy 56 g of fiber had no beneficial effect on glycemic control.[56] Subjects with baseline hemoglobin A1c levels of 7%–10% receiving more than or equal to two injections per day were randomized to either a high-fiber (50 g/d), low-GI diet or a low-fiber (15 g/d), high-GI diet for 24 weeks. The high-fiber diet significantly decreased mean daily blood glucose concentrations, number of hypoglycemic events, and hemoglobin A1c levels in the subgroup of study participants who were adherent to their diet. There were no beneficial effects, however, on total cholesterol, high-density lipoprotein (HDL) cholesterol, or triglyceride concentrations.[57] A cross-sectional analysis of dietary fiber in type 1 diabetes patients in EURODIAB IDDM showed that a higher intake of soluble fiber (from fruit, vegetables, and legumes) was independently associated with higher levels of HDL cholesterol in both men and women and lower low-density lipoprotein (LDL) cholesterol levels in men but not women.[58] No substantial differences were observed between soluble and insoluble fiber intakes. These observations suggest that inclusion of plant fiber in the diet may benefit patients with diabetes by decreasing postprandial hyperglycemia.

People with diabetes are encouraged to choose a variety of fiber-containing foods, including legumes, fiber-rich cereals (≥5 g of fiber per serving), fruits, vegetables, and whole grain products and should be encouraged to achieve the fiber intake goal set for the general population: 14 g per 1000 kcal. Alternatively, children more than 2 years old should consume an amount of dietary fiber greater than or equal to their age (in years) plus 5 g daily.[59]

PROTEIN

Protein requirements are not increased when diabetes is well controlled; therefore, children with diabetes should follow the recommended daily allowance (RDA) of protein for children without diabetes. The physiologic requirement is determined by the amount of protein necessary to sustain

normal growth; is based on ideal weight for height; and varies with age, being highest in infancy and early childhood. The protein requirement (per kilogram body weight) of infants, children, and adolescents is higher than that of adults. Protein intake decreases from approximately 2 g/kg/d in early infancy to 1 g/kg/d for a 10-year-old and is 0.8–0.9 g/kg/d in later adolescence. Children and adolescents with diabetes who have normal renal function should consume the same amount of protein as unaffected children. The 2010 DRIs for children 1–3 years old is 5%–20% and for children 4–18 years of age 10%–30%, respectively, of total energy intake. The recommendation for adults with diabetes is to maintain protein intake at less than 20% of energy needs. Any ingested protein that exceeds the body's requirement for new protein synthesis enters the carbon pool and is converted into either glucose or fat.

Dietary protein can be derived from both animal and vegetable sources; however, the source of protein is not especially important provided that all the essential amino acids are available in adequate amounts. Sources of good-quality protein include meat, poultry, fish, eggs, milk, cheese, and soy. To lower the consumption of saturated fat, less protein should be derived from red meat, whole milk, and high-fat dairy foods than is customary in the United States. With the onset of nephropathy, as evidenced by persistent microalbuminuria, excessive protein intake may be detrimental and it is prudent to advise a protein intake similar to the adult RDA (0.8 g/kg body weight per day), approximately 10% of daily energy intake. With a protein intake of 0.6 g/kg/d, evidence of protein undernutrition has been reported.

FAT

The American Heart Association (AHA) categorizes children with type 1 diabetes in the highest tier for cardiovascular risk.[60] People diagnosed with type 1 diabetes in childhood have a high risk of early subclinical[61] and clinical[62] cardiovascular disease (CVD). The prevalence of CVD is increased in both type 1 and type 2 diabetes. Type 1 diabetes is associated with a greater than or equal to 10-fold increase in CVD compared with an age-matched population without diabetes.[63] Atherosclerotic vascular disease affecting the coronary, cerebral, and lower extremity large arteries is the major cause of mortality and morbidity in patients with diabetes in Westernized populations, and about 75% of patients with diabetes in Europe and North America die of coronary heart disease. This terrible toll is not inevitable. In other parts of the world, individuals with diabetes who

escape atherosclerosis have low serum cholesterol levels and live on diets that are usually high in starch and have a low content of saturated fat.

The DCCT/EDIC report on the effect of intensive diabetes therapy on CVD in type 1 diabetes shows that higher total and LDL cholesterol levels at baseline are significantly associated with the occurrence of CVD independent of treatment assignment.[64] Saturated fatty acids and *trans*–fatty acids are the principal dietary determinants of plasma LDL cholesterol concentrations. In individuals without diabetes, reducing the consumption of saturated fatty acids and *trans*–fatty acids and cholesterol decreases plasma total and LDL cholesterol levels. Reducing the consumption of saturated fatty acids may also reduce HDL cholesterol concentrations; importantly, the ratio of LDL to HDL cholesterol is not adversely affected (Table 59-4). Therefore, to reduce CVD risk consumption of saturated fat, *trans*–fatty acids, and cholesterol must be limited while increasing the intake of ω-3 fatty acids and unsaturated fat. *Trans*–fatty acids, formed when vegetable oils are processed and solidified (hydrogenation), are found in margarine, deep-frying fat, cooking fat, and some processed foods such as cookies and cakes.

Children and adolescents with well-controlled type 1 diabetes are not at high risk for developing lipid abnormalities, but they should be screened and monitored according to specific guidelines.[65] For normally growing children and adolescents with normal plasma lipid levels, less than 10% of energy from saturated fat is recommended, the daily intake of cholesterol should be less than 300 mg/d, and consumption of *trans*–fatty acids should be minimal. In the overweight or obese child, total fat should be reduced to reduce total energy consumption. Drinking less whole milk; eating less red meat and high-fat dairy foods; and eating more poultry, fish, and vegetable proteins can reduce consumption of saturated fat.

For individuals with diabetes and increased LDL cholesterol concentrations, initial therapy is with a step 2 AHA diet, which restricts saturated fat to 7% of total calories and dietary cholesterol to 200 mg/d.[60] A recent survey of a representative sample of US youth with diabetes (89% with type 1 and 11% with type 2) aged 10–19 years shows that fat represents 37% to 38% of their total energy consumption. Only 6.5% of the cohort met the ADA recommendation of less than 10% of energy from saturated fat, and less than 50% met the recommendations for total fat, vitamin E, fiber, fruits, vegetables, and grains. It is evident that there is a critical need to improve the diet of youth with diabetes.[66]

TABLE 59-4 Practical Recommendations for Implementing the AHA Dietary Guidelines

- Reduce added sugars, including sugar-sweetened drinks and juices.
- Use canola, soybean, corn, safflower oil, or other unsaturated oil in place of solid fats during food preparation.
- Use fresh, frozen, and canned vegetables and fruits and serve at every meal; be careful with added sauces and sugar.
- Introduce and regularly serve fish as an entrée.
- Remove the skin from poultry before eating.
- Use only lean cuts of meat and reduced-fat meat products.
- Limit high-calorie sauces such as Alfredo, cream sauces, cheese sauces and hollandaise
- Eat whole-grain breads and cereals rather than refined products; read labels and ensure that “whole grain” is the first ingredient on the food label of these products.
- Eat more legumes (beans) and tofu in place of meat for some entrées.

Breads, breakfast cereals, and prepared foods, including soups, may be high in salt and sodium and/or sugar; read food labels for content and choose high-fiber, low-salt/low-sugar alternatives.

SODIUM

The average consumption of salt in populations correlates with the prevalence of hypertension, and a high intake of sodium causes high blood pressure in susceptible individuals. Most processed foods contain considerably more sodium than similar foods prepared from raw ingredients. Sodium is usually present in the form of sodium chloride; but it may also be present as sodium glutamate, added to enhance flavor, or added as sodium nitrite included as a preservative. The leaders in sodium content are takeout or fast foods, which account for an ever-increasing proportion of the US diet. It has been estimated that 70% of the sodium in the US diet is derived from prepared foods.

Sodium consumption in persons with diabetes should be the same as that for the general population, between 2400 and 3000 mg daily. A reduction in salt intake lowers blood pressure in both normotensive and hypertensive individuals. In adults who do not have diabetes, a reduction

in salt consumption may lower systolic blood pressure by 4 to 5 mmHg. A reduction in salt intake to 2400 mg daily is recommended for patients with diabetes and hypertension (in children and adolescents defined as the average of three separate systolic and/or diastolic blood pressures >95th percentile for age, sex, and height). To achieve these goals, individuals with diabetes should be advised to reduce their use of table salt and decrease their consumption of convenience foods.

SWEETENERS

The preference for a sweet taste is present at birth, and for most individuals sweetness adds significant enjoyment to eating. Sweeteners are available in two varieties, nutritive and nonnutritive.[67] If a patient on a fixed carbohydrate meal plan has exceeded his carbohydrate allotment and has a preference for sweeter products, nonnutritive sweeteners (NNSs) can be used. Current recommendations do not exclude the use of nutritive sweeteners, although nutritive sweeteners should be limited to 10% of total calories. It is important to note that the Nutrition Facts label does not differentiate between carbohydrate supplied by naturally occurring sugar and added sugar. This distinction is often a source of confusion to the general public.

NUTRITIVE SWEETENERS

Nutritive sweeteners contain carbohydrate and provide energy. Whereas monosaccharides or disaccharides provide 4 kcal/g, sugar alcohols (polyols) provide 2 kcal/g. These include sucrose; fructose; sorbitol; xylitol; and corn sweeteners such as corn syrup, fruit juice or fruit juice concentrate, honey, molasses, dextrose, and maltose.

Sucrose

Several studies have shown that sucrose as part of the meal plan does not adversely affect blood glucose control in individuals with type 1 or type 2 diabetes. In children, a moderate amount of sucrose does not increase glycemia more than an isocaloric amount of starch.[68] If a patient is on a fixed meal plan, sucrose and sucrose-containing foods may be substituted for other carbohydrates but may not simply be added to the meal plan. When making such substitutions, one must consider the nutrient content of sucrose-containing foods as well as the presence of other nutrients,

especially fat, frequently ingested with sucrose. Data are conflicting, and additional research will be necessary to determine whether dietary sucrose adversely affects serum lipids in people with diabetes. Sucrose-sweetened beverage consumption has been linked to excessive weight gain, and in an intervention that reduced consumption of sugar-sweetened beverages (replacement of sucrose-sweetened beverages by the provision of noncaloric beverages at home) among overweight and obese adolescents the increase in BMI was smaller in the experimental group than in the control group after one year.[69] Consumption of sugar- sweetened beverages should be strictly curtailed in youth with type 2 diabetes.

Fructose

Fructose is a constituent of sucrose and accounts for approximately 9% of energy intake in the United States. About one-third of dietary fructose is from fruits, vegetables, and other natural dietary sources, and about two-thirds is from the addition of fructose to food and beverages. Compared to sucrose, fructose is similar in taste but 1.2–1.8 times sweeter. It is absorbed more slowly from the intestinal tract than glucose, sucrose, or maltose and is converted to glucose and glycogen in the liver. Several studies have shown reduced postprandial plasma glucose levels when isocaloric amounts of fructose replaced sucrose or starch in the diets of people with diabetes.

Any potential benefit is tempered by the concern that excessive consumption of fructose may have adverse effects on serum lipid, especially LDL cholesterol, levels.[70] Consumption of large amounts of fructose (15%–20% of daily energy intake [90th percentile of usual intake]) increases fasting total and LDL cholesterol in people with and without diabetes.[70] Because of concern about the potential adverse effect of large amounts of fructose on serum lipids, fructose may have no advantage over other nutritive sweeteners and is not recommended. There is no reason, however, to avoid fruits, vegetables, and honey, in which fructose occurs naturally. People with dyslipidemia should be advised not to consume large amounts of fructose.

Sugar Alcohols (Polyols)

Sugar alcohols are classified as hydrogenated monosaccharides. These include erythritol; sorbitol; mannitol; xylitol; hydrogenated disaccharides (e.g., isomalt, maltitol, and lactitol); and mixtures of hydrogenated mono-, di-, and oligosaccharides (e.g., hydrogenated starch hydrolysates).[71] Sugar

alcohols contain, on average, about 2 cal/g (one-half the calories of other sweeteners such as sucrose). When calculating carbohydrate content of foods containing sugar alcohols, it is appropriate to subtract half the sugar alcohol grams from total carbohydrate grams. Studies of subjects with and without diabetes have shown that sugar alcohols produce a lower postprandial glucose response than sucrose or glucose and have a lower available energy.

Sorbitol

Sorbitol is about half as sweet as sucrose and is slowly and only partially absorbed from the gut and then metabolized to fructose in the liver by sorbitol dehydrogenase. Sorbitol has been assigned an energy value of 2.6 kcal/g; however, its precise caloric value is uncertain. Its slow absorption causes blood glucose levels to rise less rapidly than after the absorption of sucrose and other carbohydrates. When the daily intake exceeds 30–50 g, sorbitol causes flatulence, intestinal cramps, and osmotic diarrhea. Isocaloric substitution of sorbitol in the diet does not appear to alter metabolic control.

Xylitol

Xylitol, a pentahydric alcohol, is as sweet as sucrose and widely distributed in fruits and vegetables. It is metabolized to glucose, pyruvate, and lactate, mainly by the liver and, to a lesser extent, by the kidneys. Xylitol is slowly converted to glucose; however, ingestion of a single 30- to 40-g dose causes diarrhea, which limits its usefulness as an isocaloric carbohydrate substitute. Scant information is available on the effect of long-term oral administration on blood glucose control of diabetes. Many so-called dietetic foods marketed for people with diabetes contain sorbitol or xylitol instead of sucrose. In general, the sugar alcohols offer no nutritional advantage over other nutritive sweeteners and are usually more expensive.

Resistant Starch

Nondigestible oligosaccharides and the starch amylose are not digested and, therefore, not absorbed as glucose in the small intestine, but they are almost completely fermented in the colon and produce about 2 kcal/g of energy. It is estimated that resistant starch and unabsorbed starch represents approximately 2%–5% of the total starch ingested in the average Western diet. Legumes are the major source of resistant starch (2

to 3 g per 100 g of cooked legumes). Uncooked cornstarch contains about 6 g resistant starch per 100 g dry weight. Single meal studies have found that meals high in resistant starch caused some reduction in postprandial glucose and insulin responses compared to meals with equivalent amounts of digestible starch. Uncooked cornstarch as part of a bedtime snack has resulted in less overnight hypoglycemia in children and adults with type 1 diabetes; however, a study in intensively treated adults with type 1 diabetes showed that a bedtime snack bar (194 kcal, 39 g carbohydrate, 4 g fat, and 4 g protein) containing 6.25 g uncooked cornstarch did not prevent nocturnal hypoglycemia.[72]

Other Nutritive Sweeteners

These include corn sweeteners such as corn syrup, fruit juice or fruit juice concentrate, honey, molasses, dextrose, and maltose. These sweeteners have the same calorie value as sucrose, and there is no evidence that they cause a less pronounced increase in blood glucose concentrations than sucrose. All nutritive sweeteners contribute calories and, like sucrose and fructose, must be accounted for in meal planning.

NONNUTRITIVE SWEETENERS (LOW-CALORIE, OR ALTERNATIVE, SWEETENERS)

NNSs are widely used in beverages and other food products such as diet soft drinks, yogurts, desserts, and gum.[67] The Food and Drug Administration (FDA) has approved six NNSs for use in the United States: saccharin, aspartame, acesulfame potassium, neotame, sucralose, and stevia. When used judiciously, NNSs can facilitate reductions in intake of added sugars, thereby resulting in decreased total energy and weight loss/weight control.

Saccharin was discovered in 1897 and until the advent of aspartame was the most commonly used artificial sweetener. It leaves a slightly bitter, metallic aftertaste. Administration of large amounts of saccharin to rats was associated with an increased occurrence of bladder tumors, which raised concerns about its carcinogenic potential and safety for use in human beings, but it is no longer on the FDA list of cancer-causing chemicals.

Aspartame has been available since the early 1980s. It is a dipeptide consisting of phenylalanine and aspartic acid and is 200 times sweeter than sugar. At one-tenth of a calorie, it has the sweetness of a teaspoon of

sugar. It rapidly gained widespread popularity as an alternative to saccharin.

Acesulfame K is a white, colorless, and crystalline sweetener approximately 200 times sweeter than sucrose. It is a derivative of acetoacetic acid and has a structure similar to that of saccharin. It was approved for use by the FDA in 1988 and is marketed in the United States under the brand name Sunette® when used as an ingredient of foods and as Sweet One® when sold as a tabletop sweetener. It is stable both in liquids and during baking and cooking.

Sucralose is made from sucrose through a process in which three hydrogen-oxygen groups are replaced with chlorine atoms. Sucralose is 600 times sweeter than sucrose. It is very stable and can be used for baking and cooking and has no effect on glucose homeostasis in diabetic subjects.

Neotame was approved by the FDA in 2002 as a sweetener for general use in foods and beverages. It is also a derivative of the dipeptide composed of aspartic acid and phenylalanine and has a clean sweet taste like sugar. It is about 8000 times sweeter than sugar on a weight basis; therefore, only very small amounts are needed to sweeten foods and beverages.[73]

Stevia: steviol glycosides, rebaudioside A and stevioside, are extracted from leaves of the plant *Stevia rebaudiana* Bertoni. In 2008, the FDA granted Generally Recognized as Safe status for purified rebaudioside A and stevioside. Steviol glycosides have a sweet, clean taste at usual amounts. They are stable in dry form and more stable than aspartame or acesulfame K in liquid form.

MEDICAL NUTRITIONAL THERAPY

A registered dietitian skilled in MNT is the diabetes team member who plays the leading role in providing nutrition care; however, it is important that all members of the diabetes team, including physicians and diabetes nurse educators, be knowledgeable about MNT and help to support its implementation.[74]

MNT begins with a nutrition assessment, taking heed of the ethnic, religious, and economic factors that pertain to the individual with diabetes and his or her family. Family eating patterns, meal preparation methods, and nutrition beliefs must be identified. The initial evaluation of eating styles and nutrition beliefs within the family assists the dietitian to determine how best to begin educating the patient and family to achieve nutrition guidelines for optimal management of the disease. The first

priority is to teach families how diet and insulin (or other antihyperglycemic medications) affect blood glucose levels and overall metabolic control. Thereafter, it is important to teach dietary principles that will help patients reduce their risk of diabetes-related complications, including CVD.

The way meals are planned should be understandable to the patient and family. A vast lay literature about diet is available to the general public; some accurately describes evidence-based research, but much does not. Families that have received and applied advice consistent with the recommendations of the AHA, American Academy of Pediatrics, and Dietary Guidelines for Americans 2010 and information available at ChooseMyPlate.gov will benefit by an approach that builds on their current practices.

Changing how we think about our food and beverage selections on a day-to-day basis is, perhaps, the most difficult aspect of diabetes management. Before discovering that a member of the family has diabetes, families may give significant thought to their food purchases, but planning when and the precise amount to be eaten at a meal is not common in the general population. Patients with both type 1 and type 2 diabetes have the same goals, namely, to achieve and maintain optimal growth and metabolic control, which includes target blood glucose and glycated hemoglobin levels (Table 59-2). The initial focus of MNT, however, differs between the two major types of diabetes.

CONSIDERATIONS WHEN FORMULATING A MEAL PLAN FOR TYPE 1 DIABETES

In the child with type 1 diabetes, the primary goal is to match insulin delivery and carbohydrate consumption while optimizing growth and physical development. Newly diagnosed children with type 1 diabetes usually present with weight loss and are hungry for the first few days to weeks after starting insulin therapy. Therefore, the child's initial dietary allowance is determined by appetite. The meal plan should be formulated to restore and then maintain an appropriate body weight and allow for normal growth and development. Energy requirements vary with age, sex, height, and weight, as well as with physical activity, season of the year, and stage of puberty. If the child's usual food intake cannot be adequately determined, the nutrition prescription can be based on a formula for estimating energy and protein allowances (see the section "Energy").

The daily eating patterns of young children generally include three

meals and two or three snacks, depending on the length of time between meals, age of the child, usual bedtime, and level of physical activity. Although their daily energy intake is relatively constant over time, young children adjust their energy intake at successive meals. Because the energy needs of children continuously change, food intake should be reevaluated at three- to six-month intervals depending on the age of the child. If a pattern of premeal hyperglycemia is identified, it should be treated by adjusting the insulin dose and not by withholding food. Trends in hunger and satiety should be tracked. The meal plan should be adjusted when an abnormal trend in growth is observed, when the child frequently complains of being too full to eat the prescribed meal or frequently complains of being hungry after meals. Occasional hunger between meals need not lead to a major dietary change because calorie-free foods can be offered. Additional servings of protein and fat-rich foods should not be routinely offered to satisfy hunger, and force-feeding carbohydrate to achieve meal plan goals is discouraged.

Dietary strategies are dependent on the patient's insulin replacement regimen (Table 59-5). The concept of total carbohydrate intake determining the premeal insulin dosage is supported by data from the DCCT. Individuals who adjusted their premeal insulin dosages based on the carbohydrate content of their meals had a 0.5% lower hemoglobin A1c level than those who did not adjust premeal insulin doses.[75]

TABLE 59-5 General Approach to Meal Management

Type 1 diabetes	Basal-bolus or flexible insulin regimen (includes insulin pump therapy)	Carbohydrate content can vary
		Must accurately estimate carbohydrate and match insulin dose using predetermined insulin to carbohydrate ratio
		Should eat at least 3 meals daily
		Should not eat less than a predetermined amount of carbohydrate per day
		Continued education and assessment of readiness to change lifestyle to achieve heart-healthy diet
	Sliding scale or fixed dose insulin regimen	Meal times consistent from day to day
		Three meals and 2 or 3 snacks daily
		Meals and snacks spaced 2 to 3 hours apart
		Consistent carbohydrate intake
		Snack before bed to decrease risk of overnight

	hypoglycemia Continued education and assessment of readiness to change lifestyle to achieve heart-healthy diet
Type 2 diabetes	Meal plan to assist with evenly spaced carbohydrate intake and increased emphasis on reducing calories to promote weight loss
Age-related issues: children's activities often differ on weekdays compared to weekends and holidays, and appropriate allowance must be made for these differences. The meal plan must take into account the child's school schedule, early or late lunches, gym classes, and after-school physical activity.	

Rapid-acting insulin combined with intermediate-acting (neutral protamine Hagedorn [NPH]) insulin may be used at the outset because glycemia can often be controlled with two or three daily injections: typically at breakfast, dinner, and/or bedtime. Such regimens may be preferred, for example, when a young child attends a school that does not employ a nurse. With this regimen, rapid- and intermediate-acting insulins are released from the subcutaneous injection site into the circulation, reaching peak effects 1 to 2 hours and 4–10 hours after injection, respectively (Table 59-1). A meal plan is formulated to provide a constant carbohydrate intake because the breakfast dose of NPH will have its peak effect between lunch and dinner, and the dose is based on the glycemic load provided by lunch and a midafternoon snack. Therefore, consuming more or less than the recommended amounts of carbohydrate at meals will result in hyper- or hypoglycemia, respectively. Whenever the quantity of carbohydrate in the meal plan is changed, the insulin dose must be reviewed. The successful use of fixed doses of rapid-acting together with intermediate-acting insulin requires consistency in the timing and carbohydrate content of meals. These fixed insulin regimens are not ideal for very young children whose appetites are highly variable and for families whose schedules are too busy or unpredictable to allow a high level of consistency with meal times and amounts.

In general, more physiologic and flexible (basal-bolus) insulin replacement strategies are preferred.[76] These employ multiple daily doses of rapid-acting insulin together with a long-acting (basal) insulin, or CSII of rapid-acting insulin using an insulin pump. Meal plans are flexible with respect to the time at which meals are eaten and the amount of carbohydrate consumed at each meal. Patients adjust the premeal rapid-acting insulin dose to match the carbohydrate content of the meal. There is a strong relationship between the premeal insulin dose and the postprandial

response; therefore, patients who use these regimens are encouraged to perform pre- and postprandial blood glucose monitoring to optimize prandial insulin doses. The patient's blood glucose level prior to the meal and the carbohydrate content of the meal are used to determine the total mealtime insulin dose. Variation in the carbohydrate content of meals does not modify the patient's basal insulin requirement. In contrast to patients receiving intermediate-acting insulin, patients who use basal-bolus insulin therapy are not obliged to eat snacks to offset excessive insulin action between meals unless they engage in strenuous or prolonged physical activity. They must, however, administer rapid-acting insulin each time they consume carbohydrate. For young children with variable appetites and unpredictable eating patterns, administering rapid-acting insulin after the meal, although not ideal, relieves the stress parents experience at meal times and provides a practical solution to the uncertainty of knowing how much insulin to administer.

CONSIDERATIONS WHEN FORMULATING A MEAL PLAN FOR TYPE 2 DIABETES

Children with type 2 diabetes are overweight or obese at presentation and there is great emphasis on weight loss, limiting caloric intake, and distributing meals evenly throughout the day. Even modest weight reduction alone increases sensitivity to insulin and improves fasting and postprandial plasma glucose levels. Studies in adults show that structured, intensive lifestyle programs involving participant education, individualized counseling, reduced energy and fat intake (~30% of total energy), regular physical activity, and frequent participant contact are necessary to produce long-term weight loss of 5%–7% of starting weight. Accordingly, lifestyle changes that lead to weight loss are the cornerstone of therapy in type 2 diabetes. It is noteworthy, however, that in the TODAY study of treatments to maintain glycemic control in children and adolescents with recent-onset type 2 diabetes the effect of an intensive lifestyle intervention program focusing on weight loss through healthful eating and activity behaviors together with metformin was not superior to metformin alone in achieving durable glycemic control.[77]

Aims of treatment for type 2 diabetes are to lose weight, normalize fasting and postprandial blood glucose and hemoglobin A1c values, identify and treat associated comorbidities such as hypertension and hyperlipidemia,[78] and minimize the risk of acute and chronic

complications associated with diabetes. The early age of onset of type 2 diabetes in children can be expected to increase their risk of microvascular complications, which are directly related to the duration of diabetes and severity of hyperglycemia. Indeed, early-onset type 2 diabetes is associated with a high incidence of diabetic nephropathy[79] and vascular disease may already be present at diagnosis.

MNT for patients with type 2 diabetes emphasizes healthy eating to optimize metabolic control while achieving sustained weight loss. The individual's usual dietary and physical exercise habits can assist in determining the best approach to formulating a meal plan. Patients and their family members should receive guidance on behavior modification strategies to change their lifestyle, decrease their consumption of high-energy/high-fat foods,[80] and incorporate daily aerobic physical activity in their lives. To increase children's physical activity, the amount of time devoted to sedentary activities and "screen time" (viewing television, playing video games, surfing the Internet, etc.) must be strictly limited.[81,82] Recommendations outlined by the AHA (Table 59-4) that include but are not limited to simple suggestions such as eliminating sugar-containing sodas and juices, and decreasing the consumption of calorie-dense fast foods and high-fat snacks, can improve blood glucose levels and achieve weight loss. Patients who do not have regularly scheduled meals and eat large amounts of food toward the end of the day can improve their glycemic control when carbohydrate intake is more evenly distributed throughout the day. If weight loss is not achieved by revising eating habits and food selections, a more structured approach to define appropriate portions and calories is warranted. These diets are based on energy requirements calculated from resting energy expenditure equations and modified based on weight change. Younger children can achieve weight loss and better glycemic control when weight is maintained or the rate of weight gain is slowed so that with linear growth their weight for height gradually returns toward normal over time. A child whose BMI exceeds the 95th percentile, however, should be encouraged to lose weight. When the diet is implemented, linear growth should be monitored to assure that the decrease in calories does not adversely affect growth velocity.

Individuals with type 2 diabetes who do not achieve satisfactory glycemic control with a combination of weight loss, portion control, and optimal meal spacing require drug therapy. The meal plan then needs to be formulated based on the prescribed medical regimen. At the time of diagnosis, patients may require glucose-lowering therapies, including

insulin, to stabilize metabolic control while lifestyle changes are implemented.

TEACHING INDIVIDUAL NUTRITION AND MEAL PLANS

The dietitian has the important task of evaluating the patient's and family's understanding of basic principles of nutrition and then providing sufficient knowledge to help clients reach their nutrition goals. The aim is to lay a foundation for lifelong, healthy eating habits for the child and family. Even the most intensive regimens of insulin administration are not successful without careful attention to meal planning and the family's eating style. Nutrition education, like all aspects of diabetes education, is an ongoing process, requiring periodic review of the meal plan and to teach the complex interactions of insulin, carbohydrates, meal composition, and exercise on glycemic response in the context of a healthy diet.

Health-care professionals must communicate their recommendations (supported by scientific evidence) in practical terms and teach individuals with diabetes how to minimize the daily burden of food calculations, maintain the pleasure of eating, and enjoy a wide variety of food choices. When implementing a nutrition plan, the distribution of educational materials and approaches will help other involved health-care providers assist in evaluating their patient's understanding of and adherence to the plan. Appreciation of a patient's or family's learning style, math and reading skills, and attention to detail are important when devising and implementing the plan.

Several different meal planning strategies are used to manage individuals with diabetes.[83,84] The primary strategies include exchange lists, carbohydrate choices, and carbohydrate counting. Additional strategies include low-GI diets and healthy food choices. Despite the great importance of dietary management, there is little evidence to support the superiority of one dietary strategy over another nor are there universally agreed on techniques for providing dietary education. Combining several meal planning approaches and teaching methods is usually necessary to ensure that patients receive all the nutrients necessary to attain normal growth and development while optimizing glycemic control.[84]

MEAL PLANNING STRATEGIES

Exchange Diet

For decades, the exchange system approach for meal planning was the most widely used strategy. This approach defines carbohydrate intake within the context of food groups, thus controlling both carbohydrate intake and by recommending sources of carbohydrate ensuring that nutritional needs are being met. It was originally developed in 1950 by a joint committee of the ADA, American Dietetic Association (now the Academy of Nutrition and Dietetics), and Diabetes-Arthritis Program of the US Public Health Service. The Exchange List for Meal Planning was designed as an education tool that placed foods into groups that could be "exchanged" or traded with another group based on macro- and micronutrients. Revisions occurred in 1986, 1995, 2003, and 2008 to meet changing nutritional recommendations and in response to the variety of foods available in the market. The 2008 revision included a name change to Choose Your Foods: Exchange Lists for Diabetes to stress self-management while alerting health-care providers that it was a revision rather than a new approach. This continues to be an excellent tool for patients on oral antihyperglycemic medications or a fixed insulin regimen and for whom a defined amount of carbohydrate is recommended for each meal and snack.

Carbohydrate choices are a simplified version of the Choose Your Foods: Exchange lists for carbohydrate counting. The approach defines carbohydrates and which foods contain carbohydrates and then identifies a Carbohydrate Choice as the amount of the food that provides about 15 g of carbohydrate.

Carbohydrate counting in total grams is taught when rapid-acting insulin is used before the upcoming meal. The dose of insulin is calculated based on the amount of carbohydrate contained in the meal using an individualized insulin to carbohydrate ratio (e.g., 1 unit rapid-acting insulin per 10 g carbohydrate). Accurate measurement of portion sizes has to be learned, and weighing and measuring foods help to achieve familiarity. Active learning is required to acquire this skill. If blood glucose control inexplicably appears to deteriorate, effort should be renewed to improve the accuracy of carbohydrate counting. A recent study showed that adults with an average diabetes duration of 21 years underestimated the carbohydrate content of 62% of their meals, and this predicted higher glycemic variability. Smart et al. examined the accuracy of carbohydrate counting in children, adolescents, and their caregivers:

larger meals were more likely to be underestimated and subjects who had been carbohydrate counting the longest made the most errors.[85] These observations suggest that maintaining visual estimation of carbohydrate counting requires ongoing evaluation to ensure that desired glycemic control is maintained. Spiegel et al. used an increased-intensity interactive approach to teach carbohydrate counting to adolescents identified as having poor carbohydrate counting skills[86]; however, the intervention did not lead to a significant improvement in glycemic control. In contrast, a similar approach significantly improved glycemic control in adults with type 1 diabetes.[87] We have yet to identify the best approach to teaching carbohydrate counting or the frequency of weighing and measuring food required to learn and maintain the visual skills for accurate carbohydrate counting.

The advantage of carbohydrate counting is the availability of food labels and ready access to reference books and electronic resources. Patients may find it easier to count grams of carbohydrate than learn exchanges or carbohydrate choices, and this allows for a wider selection of foods. Most food labels do not provide exchange values, but all provide information on their carbohydrate content. Patients must be taught how to read and interpret food labels, and it is important to review sample food labels and ensure that patients demonstrate proficiency utilizing Nutrition Facts to determine portions per container and the amount of carbohydrate in the portion that they actually consume.

Advanced Carbohydrate Counting

Advanced carbohydrate counting is encouraged for patients using flexible, basal-bolus insulin regimens, either multiple daily injections or an insulin pump. This approach attempts to more accurately count carbohydrates, and insulin doses are adjusted to match anticipated or consumed carbohydrate intake. Portion sizes should first be determined by weighing or measuring foods to help develop excellent visual estimation skills. The Nutrition Facts on food labels and nutrition composition reference guides are used to determine the carbohydrate content of the portion eaten. The total amount of carbohydrate should be adjusted for fiber and sugar alcohol content to account for their decreased impact on glycemic response. General guidelines are to subtract all of the grams of fiber if it accounts for more than 5 g of the total carbohydrate in the meal and subtract half of the grams from sugar alcohols. For practical purposes, it may be easier and provide a clearer message to subtract the entire amount of fiber, especially

if a patient usually consumes a high-fiber diet. Insulin doses are not adjusted for GI or glycemic load.

If the premeal insulin dose is correctly adjusted for the carbohydrate content of the meal and premeal blood glucose level, the glycemic response two to four hours after the meal should be within target range.[88] Although fat intake does not contribute to glycemic load, a high-fat diet can prolong hyperglycemia due to delayed digestion and absorption of the meal, and insulin pump users are encouraged to use a dual-wave or square-wave (extended) insulin bolus when consuming a meal with high fat content.[89]

Celiac Disease and Type 1 Diabetes Mellitus

Celiac disease occurs in approximately 5%–7% of patients with type 1a diabetes mellitus. Weight loss or failure to gain weight, erratic blood glucose control, and unexpected hypoglycemic episodes should alert the clinician to obtain a tissue transglutaminase determination, along with measurement of serum IgA, to screen for celiac disease. Approximately 25% of patients are asymptomatic at the time of diagnosis.[90] Definitive diagnosis requires endoscopy and a duodenal biopsy before initiating treatment with a gluten-free diet. Meal planning requires identifying and eliminating all gluten-containing foods from the diet as well as counting carbohydrates, which presents a challenge for two reasons. First, gluten-free breads and cereals are denser than their gluten-containing counterparts. Therefore, a gluten-free product that appears the same size as its gluten-containing counterpart has more carbohydrate. Affected patients must relearn portion sizes when gluten-free foods are introduced. Second, most commercially available gluten-free breads and cereals are made from highly refined, high-GI grains. Consequently, consuming gluten-free products can cause more pronounced excursions in postprandial blood glucose levels. One strategy for minimizing postprandial glucose excursions on a gluten-free diet is to incorporate naturally occurring gluten-free whole grains, such as quinoa, buckwheat, and brown rice, in the diet.

Patients should also be alerted to the probability that they will need frequent changes in their prandial insulin to carbohydrate ratios as the gastrointestinal tract heals and is able to absorb more glucose. Asymptomatic patients are more likely to be noncompliant with gluten restriction, and less than absolute compliance with gluten restriction will prevent gut healing.

OUTPATIENT NUTRITION MANAGEMENT

A single instructional session is never adequate. To be effective, nutrition education must be an ongoing process with periodic review of the meal plan and assessment of accuracy of carbohydrate estimation. The parents and patient with newly diagnosed diabetes should meet with a dietitian several times during the first few weeks after diagnosis. Because children's energy needs change with growth and variations in physical activity, food intake and overall dietary quality, ideally, should be reassessed every three to six months.

Patients should be encouraged to keep records of food, medication, blood sugar levels, and physical activity to facilitate the recognition of glycemic patterns. By measuring pre- and postprandial glucose levels with various foods, many individuals use their empirical experience to optimize insulin dosing and achieve postprandial glucose goals. Reviewing these data with their dietitian can assist patients to consume preferred foods and optimize their glycemic control.

The use of imaginative, individualized teaching methods and materials will engage the attention and cooperation of children and parents more effectively than abstract concepts of nutritional principles and detailed diet sheets that list the composition of individual foodstuffs. Incorporating visual demonstrations with exercises such as sorting pictures for their carbohydrate content and then healthier carbohydrate content will keep the patient and family involved and has the advantage of offering insight into their understanding of important nutrition concepts.

Once the family is able to accurately apply the principles of nutrition management directed toward glycemic control, focus should turn to identifying food choices and cooking methods that will be consistent with guidelines aimed at lowering the risk of CVD. If the patient's glycemic control is poor, if growth is failing, if weight gain is excessive, or if the insulin regimen is changed, the dietitian should be reconsulted to review the meal plan and to make any necessary dietary recommendations.

MEAL PLANNING AND GLYCEMIC OUTCOMES

During the DCCT, adherence to prescribed meal plans, including appropriate treatment for hypoglycemia, was associated with a significant 0.25%–1.0% absolute reduction in hemoglobin A1c among intensively treated individuals.[75] Following the DCCT, the introduction of novel insulins and greater use of insulin pumps made greater mealtime flexibility while maintaining better glycemic control possible. Over the last five

years, there has been a renewed interest in the association between meal planning strategies, particularly carbohydrate counting, and glycemic outcomes.[85, 91–93] Empirical evidence suggests that absolute accuracy is unnecessary because carbohydrate estimates within 10–15 g of the actual carbohydrate content of a meal do not adversely affect postprandial glycemic excursions[85,92,93] or hemoglobin A1c levels[91] in youth with type 1 diabetes. Conversely, inconsistent carbohydrate estimation has been associated with higher hemoglobin A1c,[91] and estimation greater and less than 20 g of the actual meal carbohydrate content[92] is associated with postprandial hypoglycemia and hyperglycemia, respectively. Despite the current focus on carbohydrate counting, there is no evidence to suggest the superiority of one meal planning strategy over another. Several, small interventions aimed at improving carbohydrate counting proficiency have failed to improve glycemic outcomes in adolescents and adults with type 1 diabetes,[86] but meal planning strategies need to be evaluated in a larger, adequately controlled population. It has been suggested that the effectiveness of carbohydrate estimates within 10–15 g is consistent with exchange-based systems[85,94] and that the emphasis should remain on formulating a healthful meal plan consistent with the preferences and abilities of the individual patient with diabetes. Indeed, the greater focus on carbohydrate estimation may negatively impact overall diet quality for youth with type 1 diabetes,[95,96] highlighting the critical importance of educating patients with diabetes about both diet quantity and diet quality.

More recently, greater attention has been given to the impact of fat and protein on postprandial glycemic responses in youth with diabetes. In nondiabetic individuals, the ingestion of protein together with carbohydrate increases postprandial glucose levels due to gluconeogenesis and changes in insulin sensitivity. The inclusion of more fat in isocaloric mixed meals is associated with attenuated peaks but prolonged, postprandial glycemic responses owing to the incretin effect of fat and delayed gastric emptying.[97] Evidence suggests that additional insulin to account for protein and fat in a meal is superior to dosing insulin based solely on carbohydrate intake.[98,99] Pankowska et al. have introduced the notion of a “fat-protein unit” in which one additional unit of rapid-acting insulin can be administered for every 100 cal derived from protein or fat in a mixed meal.[89,100] In addition to selecting a dose of insulin for anticipated carbohydrate intake, the caloric values of fat and protein are used to calculate an additional amount of insulin to be delivered over several hours as a square- or dual-wave bolus. As the relative caloric contribution to the

meal from fat and protein increases, the length of the extended bolus is increased. This investigational methodology can effectively only be used by insulin pump users. Youth who employ this insulin dosing methodology are warned not to use the bolus calculators in their insulin pumps. There are insufficient data to support the superiority of this approach in terms of safety, ease of use, and effectiveness compared to extended insulin boluses for mixed meals based on carbohydrate quantity alone. Given the complexity of advanced carbohydrate counting, additional studies are required to confirm the translation and acceptance of this novel approach in routine clinical practice.

EXERCISE

A recent report concerning the effects of regular physical activity on health and behavioral outcomes in 6- to 18-year-old youths recommends that school-age children should participate daily in at least 60 minutes of moderate to vigorous physical activity that is developmentally appropriate, enjoyable, and involves a variety of activities.[101] Regular exercise improves blood glucose control, reduces cardiovascular risk factors, and assists in reaching and maintaining healthy weight.

Patients with type 1 diabetes should check blood glucose levels before exercise to determine whether additional carbohydrate is needed to avoid hypoglycemia. The amount of supplemental carbohydrate depends on the intensity and duration of exercise. If blood glucose is less than 100 mg/dL before exercise, added carbohydrate is required. If exercise is planned and the individual is familiar with his own glycemic response to exercise, the insulin dose given at the meal preceding exercise can be reduced rather than increasing carbohydrate intake. This approach is particularly applicable to patients who are attempting to lose weight and do not want to consume additional calories to offset the energy expenditure from exercise.

HYPOGLYCEMIA

Hypoglycemia is the principal barrier to optimizing glycemic control in youth with type 1 diabetes.[102] The risk of hypoglycemia is increased for several reasons, including pharmacologic insulin therapy and impaired glucagon and epinephrine responses to hypoglycemia.[103] Mild hypoglycemia, defined as an episode of hypoglycemia that is easily recognized and managed by the affected individual, occurs several times each week in well-controlled individuals with type 1 diabetes. An

important treatment goal is to avoid severe hypoglycemia, defined as an episode requiring assistance from another individual, use of parenteral therapy (e.g., glucagon or dextrose), and/or loss of consciousness or seizure. Patients are taught to take additional precautions in situations that may increase the risk of hypoglycemia: exercise, fasting, alcohol use, and when using certain medications. In these situations, more frequent blood glucose monitoring, reducing insulin doses, and/or ingesting additional "uncovered" carbohydrate are recommended.[65]

Given the frequent occurrence of mild hypoglycemia in youth with diabetes, MNT also focuses on the prevention and appropriate treatment of hypoglycemia.[104] Treatment of hypoglycemia entails immediate ingestion of 5–15 g of rapidly available carbohydrate, depending on the child's age and current blood glucose level (Table 59-6). The patient's blood glucose level should be remeasured in 15 minutes and additional rapid-acting carbohydrate can be administered if the blood glucose level remains less than 80 mg/dL. Fat-containing foods, such as chocolate or ice cream, are not recommended as fat delays the absorption of glucose, increasing the risk of prolonged or severe hypoglycemia. Rapidly available carbohydrate derived from glucose or sucrose is more effective than fructose-based sources.[105] Following resolution of an acute, mild episode of hypoglycemia, an additional 10–20 g of complex carbohydrate with protein decreases the risk of recurrent hypoglycemia if the next meal or snack is more than one hour away. Patients should be advised not to treat based on symptoms of hypoglycemia without confirmation by measuring a fingerstick blood glucose level. However, if a patient is unable to perform a timely blood glucose measurement and/or is about to drive (or is involved in another activity that exposes the individual to risk), it is better to assume the symptoms are due to hypoglycemia and treat accordingly until a blood glucose determination can be performed. Finally, if the child with diabetes is not alert and able to swallow safely, treatment must be with glucagon or intravenous dextrose (when this is a practical option).[65,104]

TABLE 59-6 Hypoglycemia Treatment: Sources and Quantities of Rapid-Acting Carbohydrate Based on Age

Age	≤5 Years	6–9 Years	≥10 Years
Amount	4–10 g	10–15 g	15–20 g
Glucose tablets[a]	1–2 Tablets	2–3 Tablets	3–4 Tablets

Glucose gel[b]	1/3–1/2 Tube	1/2–2/3 Tube	2/3–1 Tube
Regular soda[c]	2–3 oz.	4–6 oz.	6–8 oz.
100% Fruit juice (apple, orange)[d]	2–3 oz.	4–6 oz.	6–8 oz.

Products specifically designed to treat hypoglycemia contain pure glucose. Foods and beverages contain a variety of sugars, protein, and fat, which may slow absorption of glucose. Using pure glucose instead of other foods may help to prevent consumption of more calories than are needed to restore blood glucose to normal.

[a]1 Tablet = 4 g.

[b]Amount depends on product: Dex4 Glucose Gel® and Glutose 15® 1 tube = 15 g; Insta-Glucose® 1 tube = 24 g.

[c]Carbohydrate content (grams) of 8 oz.: Pepsi 28, Coca-Cola 27, Mountain Dew 31, Sprite 26, Schweppes Ginger Ale 22, and Canada Dry Ginger Ale 24.

[d]1 oz. Contains approximately 3 g of sugar.

ADHERENCE

Adherence to a meal plan is one of the behaviors required for optimal management of type 1 diabetes and correlates with blood glucose control in children and adults.[75,106,107] In a recent study of youth primarily using a flexible insulin regimen, greater dietary adherence was associated with lower hemoglobin A1c values.[107] Achieving optimal dietary adherence, however, is challenging and requires knowledge about healthy nutrition and meal planning, proficiency in counting carbohydrates, and motivation.[91,108] Furthermore, success requires cooperation between parent and child and appropriate parent-child mealtime behaviors.[109,110] Adherence to eating behaviors in youth with type 1 diabetes varies from 21% to 95%, depending on the specific behaviors studied and whether a fixed or flexible carbohydrate regimen is used.[111]

In the United States, a self-report survey of 144 adolescents who attended a tertiary care diabetes program showed that 15% took extra insulin to cover inappropriate food intake, 73% ate inappropriate foods, and 50% missed meals and snacks.[112] Many youth with type 1 diabetes do not adhere to recommendations for intake of fruits, vegetables, and whole grains.[66,113] A recent survey of usual dietary intake among a large ethnically diverse cohort of youth with diabetes aged 10 years or older (89% type 1) showed that only 6.5% of the cohort met the ADA recommendation of less than 7% of energy from saturated fat and 40% of youth with diabetes had a BMI greater than or equal to 85th percentile. Less than 50% met recommendations for total fat, vitamin E, fiber, fruits, vegetables, and grains.[66] Many youth with type 1 diabetes in the United States struggle with dietary adherence and do not meet nutrition

guidelines.[111]

Management of diabetes is difficult for children and adolescents and, as children progress through adolescence and into young adulthood, satisfactory adherence to therapy and optimal glycemic control become increasingly difficult to achieve and maintain. To achieve better diabetes outcomes, families must remain actively involved in their child's diabetes management, regardless of the child's age, even as the primary responsibility for diabetes care shifts from parent to adolescent.[114]

Disordered eating and eating disorders are a significant health problem among adolescents and the combination of an eating disorder and type 1 diabetes has major adverse health consequences.[115] Adolescent girls and young women with type 1 diabetes frequently decrease or omit insulin doses to maintain or lose weight.[116] Family meals are an important defense against disordered eating in adolescents with diabetes. However, in persistent or extreme cases the involvement of a mental health professional is essential.

An important role of health-care professionals involved in the care of children with diabetes is to motivate their patients to overcome the barriers to adherence so that they will be able to enjoy the benefits of good diabetes control.[17,117] Empirical research shows that adherence to the meal plan can be improved by psychosocial factors such as promoting a positive attitude toward the illness and its treatment as well as supportive relationships with the patient's family, peers, and health-care team. Frequent follow-up and ongoing education is essential to individualize the meal plan, set attainable goals, and teach the patient and his or her family how to match insulin to exercise and food.[118]

SUMMARY

Nutrition recommendations for treatment of diabetes have evolved since the pre-insulin era when a starvation diet with negligible carbohydrate and 70% of calories from fat was the recommended treatment. Today, there is no diabetic diet. The recommended diet can only be defined as a meal plan based on a nutrition assessment and mutually accepted treatment goals and desired outcomes. Nutrition therapy should be individualized. Consideration must be given to usual eating habits and other lifestyle factors. Monitoring clinical and metabolic parameters including blood glucose, glycated hemoglobin (hemoglobin A1c), lipids, blood pressure, and body weight, as well as quality of life, is crucial to ensure successful

outcomes. Modern diabetes management, combining frequent self-monitoring of blood glucose (supplemented with continuous glucose monitoring) with multiple dose flexible insulin regimens, or insulin pump therapy, and mastery of carbohydrate estimation, has made it possible for children and adolescents with diabetes to have considerable dietary flexibility and achieve better glycemic control with less frequent severe hypoglycemia.

Although the introduction of insulin analogs, flexible insulin regimens, and insulin pump therapy has permitted greater flexibility in carbohydrate intake, timing of meals, variety of food choices, and overall quality of life, these therapeutic innovations may also encourage unhealthy food choices.[95] Education must continue to emphasize dietary recommendations that maintain normal body weight and improve cardiovascular risk factors, including reduction of total fat, and especially saturated and *trans*-fats, and promote consumption of cardioprotective foods such as those containing monounsaturated fats and antioxidants.

REFERENCES

1. Smart C, Aslander-van Vliet E, Waldron S. Nutritional management in children and adolescents with diabetes. *Pediatr Diabetes*. Sep 2009;10 (Suppl 12):100–117.
2. Bantle JP, Wylie-Rosett J, Albright AL, et al. Nutrition recommendations and interventions for diabetes: a position statement of the American Diabetes Association. *Diabetes Care*. Jan 2008;31(Suppl 1):S61–S78.
3. American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care*. Jan 2012;35(Suppl 1):S64–S71.
4. Centers for Disease Control and Prevention. National diabetes fact sheet: national estimates and general information on diabetes and prediabetes in the United States, 2011. Atlanta, GA2011.
5. American Diabetes Association. Type 2 diabetes in children and adolescents. (Consensus statement). *Diabetes Care*. Mar 2000;23(3):381–389.
6. Liese AD, D'Agostino RB, Jr., Hamman RF, et al. The burden of diabetes mellitus among US youth: prevalence estimates from the SEARCH for Diabetes in Youth Study. *Pediatrics*. Oct 2006;118(4):1510–1518.
7. Hattersley A, Bruining J, Shield J, et al. The diagnosis and management of monogenic diabetes in children and adolescents. *Pediatr Diabetes*. Sep 2009;10(Suppl 12):33–42.
8. Eisenbarth GS. Banting Lecture 2009: an unfinished journey: molecular pathogenesis to prevention of type 1A diabetes. *Diabetes*. Apr 2010;59(4):759–774.
9. Smiley D, Chandra P, Umpierrez GE. Update on diagnosis, pathogenesis and management of ketosis-prone type 2 diabetes mellitus. *Diabetes Manag (Lond)*. Nov 1 2011;1(6):589–600.
10. Rosenbloom AL, Silverstein JH, Amemiya S, et al. Type 2 diabetes in children and adolescents. *Pediatr Diabetes*. Sep 2009;10(Suppl 12):17–32.

11. Dabelea D, Pettitt DJ, Jones KL, Arslanian SA. Type 2 diabetes mellitus in minority children and adolescents. An emerging problem. *Endocrinol Metab Clin North Am*. 1999;28(4):709–729, viii.
12. Arslanian S, Suprasongsin C. Insulin sensitivity, lipids, and body composition in childhood: is "syndrome X" present? *J Clin Endocrinol Metab*. Mar 1996;81(3):1058–1062.
13. Arslanian S, Danadian K. Insulin secretion, insulin sensitivity and diabetes in black children. *Trends Endocrinol Metab*. 1998;9:194–199.
14. Glastras SJ, Mohsin F, Donaghue KC. Complications of diabetes mellitus in childhood. *Pediatr Clin North Am*. Dec 2005;52(6):1735–1753.
15. Brownlee M. The pathobiology of diabetic complications: a unifying mechanism. *Diabetes*. Jun 2005;54(6):1615–1625.
16. Caso EK. Calculation of diabetic diets. *J Am Diet Assoc*. Aug 1950;26(8):575–583.
17. Diabetes Control and Complications Trial Research Group. The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. [see comments]. *N Engl J Med*. 1993;329(14):977–986.
18. Patterson CC, Dahlquist GG, Gyurus E, et al. Incidence trends for childhood type 1 diabetes in Europe during 1989–2003 and predicted new cases 2005–20: a multicentre prospective registration study. *Lancet*. Jun 13 2009;373(9680):2027–2033.
19. Cardwell CR, Stene LC, Ludvigsson J, et al. Breast-feeding and childhood-onset type 1 diabetes: a pooled analysis of individual participant data from 43 observational studies. *Diabetes Care*. Nov 2012;35(11):2215–2225.
20. Knip M, Virtanen SM, Becker D, et al. Early feeding and risk of type 1 diabetes: experiences from the Trial to Reduce Insulin-dependent diabetes mellitus in the Genetically at Risk (TRIGR). *Am J Clin Nutr*. Dec 2011;94(6 Suppl):1814S–1820S.
21. Knip M, Veijola R, Virtanen SM, et al. Environmental triggers and determinants of type 1 diabetes. *Diabetes*. Dec 2005;54(Suppl 2):S125–S136.
22. Vaarala O, Knip M, Paronen J, et al. Cow's milk formula feeding induces primary immunization to insulin in infants at genetic risk for type 1 diabetes. *Diabetes*. Jul 1999;48(7):1389–1394.
23. Verge CF, Howard NJ, Irwig L, et al. Environmental factors in childhood IDDM. A population-based, case-control study. *Diabetes Care*. Dec 1994;17(12):1381–1389.
24. Virtanen SM, Hypponen E, Laara E, et al. Cow's milk consumption, disease-associated autoantibodies and type 1 diabetes mellitus: a follow-up study in siblings of diabetic children. Childhood Diabetes in Finland Study Group. *Diabet Med*. Sep 1998;15(9):730–738.
25. Virtanen SM, Laara E, Hypponen E, et al. Cow's milk consumption, HLA-DQB1 genotype, and type 1 diabetes: a nested case-control study of siblings of children with diabetes. Childhood Diabetes in Finland Study Group. *Diabetes*. Jun 2000;49(6):912–917.
26. Dahlquist GG, Blom LG, Persson L-A, et al. Dietary factors in the risk of developing insulin-dependent diabetes in childhood. *BMJ*. 1990;300:302–306.
27. Knip M, Virtanen SM, Akerblom HK. Infant feeding and the risk of type 1 diabetes. *Am J Clin Nutr*. May 2010;91(5):1506S–1513S.
28. Hypponen E, Kenward MG, Virtanen SM, et al. Infant feeding, early weight gain, and risk of type 1 diabetes. Childhood Diabetes in Finland (DiMe) Study Group.

Diabetes Care. Dec 1999;22(12):1961–1965.
29. Yeung WC, Rawlinson WD, Craig ME. Enterovirus infection and type 1 diabetes mellitus: systematic review and meta-analysis of observational molecular studies. *BMJ*. 2011;342:d35.
30. Knip M, Virtanen SM, Seppa K, et al. Dietary intervention in infancy and later signs of beta-cell autoimmunity. *N Engl J Med*. Nov 11 2010;363(20):1900–1908.
31. Norris JM, Barriga K, Klingensmith G, et al. Timing of initial cereal exposure in infancy and risk of islet autoimmunity. *JAMA*. Oct 1 2003;290(13):1713–1720.
32. Ziegler AG, Schmid S, Huber D, et al. Early infant feeding and risk of developing type 1 diabetes-associated autoantibodies. *JAMA*. Oct 1 2003;290(13):1721–1728.
33. Virtanen SM, Kenward MG, Erkkola M, et al. Age at introduction of new foods and advanced beta cell autoimmunity in young children with HLA-conferred susceptibility to type 1 diabetes. *Diabetologia*. Jul 2006;49(7):1512–1521.
34. Stene LC, Joner G. Use of cod liver oil during the first year of life is associated with lower risk of childhood-onset type 1 diabetes: a large, population-based, case-control study. *Am J Clin Nutr*. Dec 2003;78(6):1128–1134.
35. Stene LC, Thorsby PM, Berg JP, et al. Peroxisome proliferator-activated receptor-gamma2 Pro12Ala polymorphism, cod liver oil and risk of type 1 diabetes. *Pediatr Diabetes*. Feb 2008;9(1):40–45.
36. Zipitis CS, Akobeng AK. Vitamin D supplementation in early childhood and risk of type 1 diabetes: a systematic review and meta-analysis. *Arch Dis Child*. Jun 2008;93(6):512–517.
37. Viskari H, Kondrashova A, Koskela P, et al. Circulating vitamin D concentrations in two neighboring populations with markedly different incidence of type 1 diabetes. *Diabetes Care*. Jun 2006;29(6):1458–1459.
38. Harder T, Roepke K, Diller N, et al. Birth weight, early weight gain, and subsequent risk of type 1 diabetes: systematic review and meta-analysis. *Am J Epidemiol*. Jun 15 2009;169(12):1428–1436.
39. Hypponen E, Virtanen SM, Kenward MG, et al. Obesity, increased linear growth, and risk of type 1 diabetes in children. *Diabetes Care*. Dec 2000;23(12):1755–1760.
40. Lammi N, Moltchanova E, Blomstedt PA, et al. Childhood BMI trajectories and the risk of developing young adult-onset diabetes. *Diabetologia*. Mar 2009;52(3):408–414.
41. Palmer JP, Helqvist S, Spinas GA, et al. Interaction of beta-cell activity and IL-1 concentration and exposure time in isolated rat islets of Langerhans. *Diabetes*. Oct 1989;38(10):1211–1216.
42. Wilkin TJ. The convergence of type 1 and type 2 diabetes in childhood: the accelerator hypothesis. *Pediatr Diabetes*. Jun 2012;13(4):334–339.
43. Rewers M. The fallacy of reduction. *Pediatr Diabetes*. Jun 2012; 13(4):340–343.
44. Dabelea D, D'Agostino RB, Jr., Mayer-Davis EJ, et al. Testing the accelerator hypothesis: body size, beta-cell function, and age at onset of type 1 (autoimmune) diabetes. *Diabetes Care*. Feb 2006;29(2):290–294.
45. Nutrition Recommendations and Interventions for Diabetes: A position statement of the American Diabetes Association. *Diabetes Care*. Jan 2007;30(Suppl 1):S48–S65.
46. Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications Research Group. Effect of intensive therapy on the microvascular complications of type 1 diabetes mellitus. *JAMA*. May 15 2002;287(19):2563–2569.
47. U.S. Department of Agriculture and U.S. Department of Health and Human

Services. Dietary Guidelines for Americans, 2010. 2010; 7th:http://www.health.gov/DietaryGuidelines/.
48. Kulkarni K, Castle G, Gregory R, et al. Nutrition Practice Guidelines for Type 1 Diabetes Mellitus positively affect dietitian practices and patient outcomes. Diabetes Care and Education Dietetic Practice Group. *J Am Diet Assoc*. Jan 1998;98(1):62–70; quiz 71–62.
49. Sheard NF, Clark NG, Brand-Miller JC, et al. Dietary carbohydrate (amount and type) in the prevention and management of diabetes: a statement by the American Diabetes Association. *Diabetes Care*. Sep 2004;27(9):2266–2271.
50. Jenkins DJA, Wolever TMS, Taylor RH, et al. Glycemic index of foods: a physiological basis for carbohydrate exchange. *Am J Clin Nutr*. 1981;34:362–366.
51. Brand-Miller J, Foster-Powell K, Gilbertson H. *The Glucose Revolution Pocket Guide to Childhood Diabetes*. New York: Marlowe and Company; 2004.
52. Foster-Powell K, Holt SH, Brand-Miller JC. International table of glycemic index and glycemic load values: 2002. *Am J Clin Nutr*. Jul 2002;76(1):5–56.
53. Brand-Miller J, Hayne S, Petocz P, Colagiuri S. Low-glycemic index diets in the management of diabetes: a meta-analysis of randomized controlled trials. *Diabetes Care*. Aug 2003;26(8):2261–2267.
54. Nansel TR, Gellar L, Zeitzoff L. Acceptability of lower glycemic index foods in the diabetes camp setting. *J Nutr Educ Behav*. May–Jun 2006;38(3):143–150.
55. Gilbertson HR, Thorburn AW, Brand-Miller JC, et al. Effect of low-glycemic-index dietary advice on dietary quality and food choice in children with type 1 diabetes. *Am J Clin Nutr*. 2003;77(1):83–90.
56. Lafrance L, Rabasa-Lhoret R, Poisson D, et al. Effects of different glycaemic index foods and dietary fibre intake on glycaemic control in type 1 diabetic patients on intensive insulin therapy. *Diabet Med*. Nov 1998;15(11):972–978.
57. Giacco R, Parillo M, Rivellese AA, et al. Long-term dietary treatment with increased amounts of fiber-rich low-glycemic index natural foods improves blood glucose control and reduces the number of hypoglycemic events in type 1 diabetic patients. *Diabetes Care*. Oct 2000;23(10):1461–1466.
58. Toeller M, Buyken AE, Heitkamp G, et al. Fiber intake, serum cholesterol levels, and cardiovascular disease in European individuals with type 1 diabetes. EURODIAB IDDM Complications Study Group. *Diabetes Care*. Mar 1999;22(Suppl 2):B21–B28.
59. Williams CL, Bollella M, Wynder EL. A new recommendation for dietary fiber in childhood. *Pediatrics*. Nov 1995;96(5 Pt 2):985–988.
60. Kavey RE, Allada V, Daniels SR, et al. Cardiovascular risk reduction in high-risk pediatric patients: a scientific statement from the American Heart Association Expert Panel on Population and Prevention Science; the Councils on Cardiovascular Disease in the Young, Epidemiology and Prevention, Nutrition, Physical Activity and Metabolism, High Blood Pressure Research, Cardiovascular Nursing, and the Kidney in Heart Disease; and the Interdisciplinary Working Group on Quality of Care and Outcomes Research: endorsed by the American Academy of Pediatrics. *Circulation*. Dec 12 2006;114(24):2710–2738.
61. Krantz JS, Mack WJ, Hodis HN, et al. Early onset of subclinical atherosclerosis in young persons with type 1 diabetes. *J Pediatr*. Oct 2004;145(4):452–457.
62. Orchard TJ, Forrest KY, Kuller LH, Becker DJ. Lipid and blood pressure treatment goals for type 1 diabetes: 10-year incidence data from the Pittsburgh Epidemiology of Diabetes Complications Study. *Diabetes Care*. Jun 2001;24(6):1053–1059.
63. Laing SP, Swerdlow AJ, Slater SD, et al. Mortality from heart disease in a cohort

of 23,000 patients with insulin-treated diabetes. *Diabetologia*. Jun 2003;46(6):760–765.
64. Nathan DM, Cleary PA, Backlund JY, et al. Intensive diabetes treatment and cardiovascular disease in patients with type 1 diabetes. *N Engl J Med*. Dec 22 2005;353(25):2643–2653.
65. Silverstein J, Klingensmith G, Copeland K, et al. Care of children and adolescents with type 1 diabetes: a statement of the American Diabetes Association. *Diabetes Care*. Jan 2005;28(1):186–212.
66. Mayer-Davis EJ, Nichols M, Liese AD, et al. Dietary intake among youth with diabetes: the SEARCH for Diabetes in Youth Study. *J Am Diet Assoc*. May 2006;106(5):689–697.
67. Fitch C, Keim KS. Position of the Academy of Nutrition and Dietetics: use of nutritive and nonnutritive sweeteners. *J Acad Nutr Diet*. May 2012;112(5):739–758.
68. Rickard KA, Loghmani ES, Cleveland JL, et al. Lower glycemic response to sucrose in the diets of children with type 1 diabetes. *J Pediatr*. Sep 1998;133(3):429–434.
69. Ebbeling CB, Feldman HA, Chomitz VR, et al. A randomized trial of sugar-sweetened beverages and adolescent body weight. *N Engl J Med*. Oct 11 2012;367(15):1407–1416.
70. Bantle JP, Swanson JE, Thomas W, Laine DC. Metabolic effects of dietary fructose in diabetic subjects. *Diabetes Care*. 1992;15(11):1468–1476.
71. *Report of a Joint FAO/WHO Expert Consultation. Carbohydrates in Human Nutrition*. Rome, Italy: Food and Agriculture Organization of the United Nations and World Health Organization; 1998.
72. Raju B, Arbelaez AM, Breckenridge SM, Cryer PE. Nocturnal hypoglycemia in type 1 diabetes: an assessment of preventive bedtime treatments. *J Clin Endocrinol Metab*. Jun 2006;91(6):2087–2092.
73. http://www.neotame.com. Accessed February 11, 2015.
74. Bantle JP, Wylie-Rosett J, Albright AL, et al. Nutrition recommendations and interventions for diabetes—2006: a position statement of the American Diabetes Association. *Diabetes Care*. Sep 2006;29(9):2140–2157.
75. Delahanty LM, Halford BN. The role of diet behaviors in achieving improved glycemic control in intensively treated patients in the Diabetes Control and Complications Trial. *Diabetes Care*. 1993; 16(11):1453–1458.
76. Mehta SN, Wolfsdorf JI. Contemporary management of patients with type 1 diabetes. *Endocrinol Metab Clin North Am*. Sep 2010; 39(3):573–593.
77. Zeitler P, Hirst K, Pyle L, et al. A clinical trial to maintain glycemic control in youth with type 2 diabetes. *N Engl J Med*. Jun 14 2012;366(24):2247–2256.
78. American Diabetes Association. Standards of medical care in diabetes—2006. *Diabetes Care*. Jan 2006;29(Suppl 1):S4–S42.
79. Eppens MC, Craig ME, Cusumano J, et al. Prevalence of diabetes complications in adolescents with type 2 compared with type 1 diabetes. *Diabetes Care*. Jun 2006;29(6):1300–1306.
80. Willett WC, Dietz WH, Colditz GA. Guidelines for healthy weight. *N Engl J Med*. Aug 5 1999;341(6):427–434.
81. Gortmaker SL, Must A, Sobol AM, et al. Television viewing as a cause of increasing obesity among children in the United States, 1986–1990. *Arch Pediatr Adolesc Med*. Apr 1996;150(4):356–362.
82. Epstein LH, Paluch RA, Gordy CC, Dorn J. Decreasing sedentary behaviors in treating pediatric obesity. *Arch Pediatr Adolesc Med*. Mar 2000;154(3):220–226.

83. Ross TA, O'Connell BS, Boucher JL. *ADA Guide to Diabetes Medical Nutrition Therapy and Education. Chicago, IL: American Dietetic Association; 2005.*
84. Wylie-Rosett J, Aebersold K, Conlon B, Ostrovsky NW. Medical nutrition therapy for youth with type 1 diabetes mellitus: more than carbohydrate counting. *J Acad Nutr Diet*. Nov 2012;112(11):1724–1727.
85. Smart CE, Ross K, Edge JA, et al. Can children with type 1 diabetes and their caregivers estimate the carbohydrate content of meals and snacks? *Diabet Med.* Mar 2010;27(3):348–353.
86. Spiegel G, Bortsov A, Bishop FK, et al. Randomized nutrition education intervention to improve carbohydrate counting in adolescents with type 1 diabetes study: is more intensive education needed? *J Acad Nutr Diet*. Nov 2012;112(11):1736–1746.
87. Hegar K, Heiber S, Brandle M, et al. Carbohydrate counting of food. *Swiss Med Wkly*. 2011;141:w13224.
88. Cobry E, McFann K, Messer L, et al. Timing of meal insulin boluses to achieve optimal postprandial glycemic control in patients with type 1 diabetes. *Diabetes Technol Ther*. Mar 2010;12(3):173–177.
89. Pankowska E, Szypowska A, Lipka M, et al. Application of novel dual wave meal bolus and its impact on glycated hemoglobin A1c level in children with type 1 diabetes. *Pediatr Diabetes*. Aug 2009;10(5):298–303.
90. Marchese A, Lovati E, Biagi F, Corazza GR. Coeliac disease and type 1 diabetes mellitus: epidemiology, clinical implications and effects of gluten-free diet. *Endocrine*. Feb 2013;43(1):1–2.
91. Mehta SN, Quinn N, Volkening LK, Laffel LM. Impact of carbohydrate counting on glycemic control in children with type 1 diabetes. *Diabetes Care*. Jun 2009;32(6):1014–1016.
92. Smart CE, King BR, McElduff P, Collins CE. In children using intensive insulin therapy, a 20-g variation in carbohydrate amount significantly impacts on postprandial glycaemia. *Diabet Med*. Jul 2012;29(7):e21–e24.
93. Smart CE, Ross K, Edge JA, et al. Children and adolescents on intensive insulin therapy maintain postprandial glycaemic control without precise carbohydrate counting. *Diabet Med*. Mar 2009;26(3):279–285.
94. Maahs DM, Higgins J. Is carbohydrate counting enough? Towards perfection or unwanted complexity? *Diabetes Technol Ther*. Jan 2012;14(1):3–5.
95. Mehta SN, Haynie DL, Higgins LA, et al. Emphasis on carbohydrates may negatively influence dietary patterns in youth with type 1 diabetes. *Diabetes Care*. Dec 2009;32(12):2174–2176.
96. Lawton J, Rankin D, Cooke DD, et al. Dose adjustment for normal eating: a qualitative longitudinal exploration of the food and eating practices of type 1 diabetes patients converted to flexible intensive insulin therapy in the UK. *Diabetes Res Clin Pract*. Jan 2011;91(1):87–93.
97. Carrel G, Egli L, Tran C, et al. Contributions of fat and protein to the incretin effect of a mixed meal. *Am J Clin Nutr*. Oct 2011;94(4):997–1003.
98. Kordonouri O, Hartmann R, Remus K, et al. Benefit of supplementary fat plus protein counting as compared with conventional carbohydrate counting for insulin bolus calculation in children with pump therapy. *Pediatr Diabetes*. Nov 2012;13(7):540–544.
99. Pankowska E, Blazik M, Groele L. Does the fat-protein meal increase postprandial glucose level in type 1 diabetes patients on insulin pump: the conclusion of a randomized study. *Diabetes Technol Ther*. Jan 2012;14(1):16–22.
100. Pankowska E, Blazik M. Bolus calculator with nutrition database software, a new

concept of prandial insulin programming for pump users. *J Diabetes Sci Technol*. May 2010;4(3):571–576.
101. Strong WB, Malina RM, Blimkie CJ, et al. Evidence based physical activity for school-age youth. *J Pediatr*. Jun 2005;146(6):732–737.
102. Cryer PE, Davis SN, Shamoon H. Hypoglycemia in diabetes. *Diabetes Care*. Jun 2003;26(6):1902–1912.
103. Cryer PE. The barrier of hypoglycemia in diabetes. *Diabetes*. Dec 2008;57(12):3169–3176.
104. Clarke W, Jones T, Rewers A, et al. Assessment and management of hypoglycemia in children and adolescents with diabetes. *Pediatr Diabetes*. Apr 2008;9(2):165–174.
105. Husband AC, Crawford S, McCoy LA, Pacaud D. The effectiveness of glucose, sucrose, and fructose in treating hypoglycemia in children with type 1 diabetes. *Pediatr Diabetes*. May 2010;11(3):154–158.
106. Patton SR, Dolan LM, Powers SW. Dietary adherence and associated glycemic control in families of young children with type 1 diabetes. *J Am Diet Assoc*. Jan 2007;107(1):46–52.
107. Mehta SN, Volkening LK, Anderson BJ, et al. Dietary behaviors predict glycemic control in youth with type 1 diabetes. *Diabetes Care*. Jul 2008;31(7):1318–1320.
108. Koontz MB, Cuttler L, Palmert MR, et al. Development and validation of a questionnaire to assess carbohydrate and insulin-dosing knowledge in youth with type 1 diabetes. *Diabetes Care*. Mar 2010;33(3):457–462.
109. Powers SW, Byars KC, Mitchell MJ, et al. Parent report of mealtime behavior and parenting stress in young children with type 1 diabetes and in healthy control subjects. *Diabetes Care*. Feb 2002;25(2):313–318.
110. Patton SR, Dolan LM, Powers SW. Mealtime interactions relate to dietary adherence and glycemic control in young children with type 1 diabetes. *Diabetes Care*. May 2006;29(5):1002–1006.
111. Patton SR. Adherence to diet in youth with type 1 diabetes. *J Am Diet Assoc*. Apr 2011;111(4):550–555.
112. Weissberg-Benchell J, Glasgow AM, Tynan WD, et al. Adolescent diabetes management and mismanagement. *Diabetes Care*. 1995;18(1):77–82.
113. Overby NC, Flaaten V, Veierod MB, et al. Children and adolescents with type 1 diabetes eat a more atherosclerosis-prone diet than healthy control subjects. *Diabetologia*. Feb 2007;50(2):307–316.
114. Anderson B, Ho J, Brackett J, et al. Parental involvement in diabetes management tasks: relationships to blood glucose monitoring adherence and metabolic control in young adolescents with insulin-dependent diabetes mellitus. *J Pediatr*. 1997;130(2):257–265.
115. Peveler RC, Bryden KS, Neil HA, et al. The relationship of disordered eating habits and attitudes to clinical outcomes in young adult females with type 1 diabetes. *Diabetes Care*. Jan 2005;28(1):84–88.
116. Polonsky WH, Anderson BJ, Lohrer PA, et al. Insulin omission in women with IDDM. *Diabetes Care*. 1994;17(10):1178–1185.
117. Diabetes Control and Complications Trial Research Group. Effect of intensive diabetes treatment on the development and progression of long-term complications in adolescents with insulin-dependent diabetes mellitus: Diabetes Control and Complications Trial. [see comments]. *J Pediatr*. 1994;125(2):177–188.
118. Williamson AR, Hunt AE, Pope JF, Tolman NM. Recommendations of dietitians for overcoming barriers to dietary adherence in individuals with diabetes.

Diabetes Educ. Mar-Apr 2000;26(2):272–279.
119. Franz MJ, Bantle JP, Beebe CA, et al. Nutrition principles and recommendations in diabetes. *Diabetes Care*. Jan 2004;27(Suppl 1):S36–S46.

Diarrheal Diseases

60

Kerri Gosselin, MD and Christopher Duggan, MD, MPH

There exists a strong association between diarrhea and malnutrition, whereby episodes of childhood diarrhea lead to worse nutritional status, and malnutrition predisposes children to episodes of diarrhea. Diarrheal diseases are a major cause of pediatric morbidity and mortality, causing an estimated 800,000 deaths of children under 5 years of age worldwide.[1] Over the past several decades, substantial progress has been made in reducing childhood deaths due to diarrhea, with the estimated mortality dropping below one million children per year for the first time in 2012. Comparing this value to those of previous years reveals significant progress in reducing mortality due to diarrhea. For example, 2.5 million child deaths from diarrhea were estimated in 2003,[2] 3.3 million annual deaths were estimated in 1992,[3] and 4.6 million deaths from diarrhea were estimated in 1982.[4] The story underlying these statistics is the remarkable success of worldwide campaigns to treat acute diarrhea with oral rehydration therapy (ORT), as well as improved hygiene, medical care including immunizations, and access to drinking water. The development of ORT is among the most successful collaborations ever between basic and applied biomedical research and one of the most significant applications of nutritional therapy to the management of acute disease. Interestingly, it is also a case of reverse technology transfer,[5] in which work originally carried out for benefit of those in countries with less developed economies has changed the standard of practice, even in industrialized settings.

In addition to these successes worldwide, there has also been significant progress made in the United States. With the introduction of the rotavirus vaccine in 2006, the number of U.S. hospitalizations for acute

diarrhea has decreased dramatically. In 2006, before administration of the vaccine was widespread, an estimated 74 per 10,000 children under age 5 years were hospitalized for gastroenteritis or rotavirus, a figure that decreased to 50 hospitalizations per 10,000 children in 2009.[6]

However, challenges remain in the treatment of diarrhea both in the United States and abroad. It is likely that the full benefits of the fluid, electrolyte, and nutritional therapy of acute diarrhea have not been realized. Shortcomings in treatment are significant, as they result in added morbidity and costs in both developing and industrialized societies. It may be argued that the full implementation of ORT for treatment of acute diarrhea in countries with developed economies has lagged behind its use in less developed countries, perhaps because of the ingrained use of intravenous therapy or the reduced appeal of a technologically simple solution.[7] In addition, it has been difficult to establish continued feeding during diarrheal episodes as normative therapy, despite a wealth of in vitro and in vivo data supporting the role of continued nutrition in improving measurements of gastrointestinal function, as well as anthropometric, biochemical, and clinical outcomes.[8,9]

In addition, while the mortality attributed to diarrheal illness is decreasing worldwide, significant morbidity continues. Diarrheal morbidity includes dehydration, micronutrient deficiencies, altered growth, and cognitive deficits. Episodes of diarrhea predispose to stunting, with an estimated 5% increased risk of stunting for each diarrheal episode before age 2 years.[10] Diarrhea and enteric infections early in life predispose to deficits in cognitive development and result in significant burden of disability-adjusted life years, including decreased work productivity and IQ.[11] Therefore, there continues to be a great need to identify gaps in current knowledge and promote implementation research with the ultimate goal of decreasing both the morbidity and mortality of diarrheal illnesses.

This chapter reviews the historical background and scientific basis of ORT and provides a framework for assessing and treating the dehydrated patient or the patient at risk for dehydration. The discussion focuses on common clinical scenarios and traditional practices, especially with regard to feeding. The limitations of ORT are discussed, as well as areas of ongoing research.

HISTORICAL BACKGROUND

Early attempts at treating dehydration resulting from diarrhea were first described in medical literature in the 1830s, during epidemics of *Vibrio*

cholerae infections.[12] It was not until 100 years later that the use of intravenous fluids became widespread. Accurate chemical analysis of diarrheal stools eventually permitted the formulation of physiologically appropriate replacement solutions, leading to the successful treatment of cholera with intravenous fluids by the 1940s.[13] Further research into intestinal electrolyte transport led to the development of oral solutions for rehydration.[14,15] In 1971, these were put to test in the field with the large-scale treatment of refugees from the Bangladesh war of independence.[16] The remarkable success of oral solutions hastened the development of the World Health Organization (WHO) guidelines for ORT and production of standard packets of oral rehydration salts. It may be argued that the initial application of ORT to cholera epidemics in the developing world hindered the acceptance of this therapy's relevance in the United States and Europe. With time, however, ORT has become accepted as the standard of care for clinically efficacious and cost–effective management of acute diarrhea,[17,18] although it remains underused in settings in which parenteral rehydration has been the established practice. One national survey of physicians in emergency care facilities in the United States showed that many would treat mild dehydration with intravenous therapy, and half would always or almost always use intravenous therapy for a moderately (5–10%) dehydrated child under age 2.[19]

PHYSIOLOGIC BASIS FOR THE USE OF ORAL REHYDRATION SOLUTIONS

Fluid must be absorbed and reabsorbed for survival. Under the best of circumstances, the adult intestinal epithelium must handle 6,500 mL fluid per day, the combination of oral intake and salivary, gastric, pancreatic, biliary, and upper intestinal secretions. This is typically reduced to 1,500 mL by the distal ileum and is further reduced in the colon to stool output less than 250 mL/day.[20] During disease, the volume of intestinal fluid output is remarkably increased, which overwhelms reabsorptive capacity and leads to diarrhea.

Applied clinical research, first carried out among cholera patients, showed that diarrhea in cholera is not the result of a failure of intestinal reabsorption but rather a state of extreme output in which reabsorptive mechanisms remain intact.[21] Not only *V. cholerae* but many strains of *Escherichia coli, Shigella, Salmonella,* and other pathogenic bacteria have been shown to produce toxins that bind to enterocyte receptors. This

results in chloride-mediated secretion stimulated by second messengers such as cyclic adenosine monophosphate, cyclic guanosine monophosphate, and calcium.[22] Even those infectious agents typically classified as causing osmotic diarrhea may, in some cases, also increase enterocyte secretion. *Rotavirus,* for instance, damages the villous brush border, causing osmotic diarrhea, but also produces an enterotoxin that causes a Ca^{++}-mediated secretory diarrhea.[23]

Basic scientific studies of intestinal solute transport mechanisms were also crucial in outlining the processes by which solute absorption is maintained. Water passively follows the osmotic gradient generated by the transcellular transport of electrolytes and nutrients. Although three principal mechanisms of sodium absorption have been described,[20] that essential to the efficacy of oral rehydration solutions (ORSs) was shown to result from the stoichiometric co-transport of sodium and glucose molecules at the intestinal brush border.[24] Figure 60-1 provides a schematic of the cotransport process. Cotransport across the luminal membrane is facilitated by the protein SGLT1.[25] Once in the enterocyte, the transport of glucose into the blood is facilitated by GLUT 2 in the basolateral membrane. The Na^+-K^+ adenosine triphosphatase provides the gradient that drives the process. Clinical studies have demonstrated that this mechanism remains intact even in patients with severe diarrhea.[21]

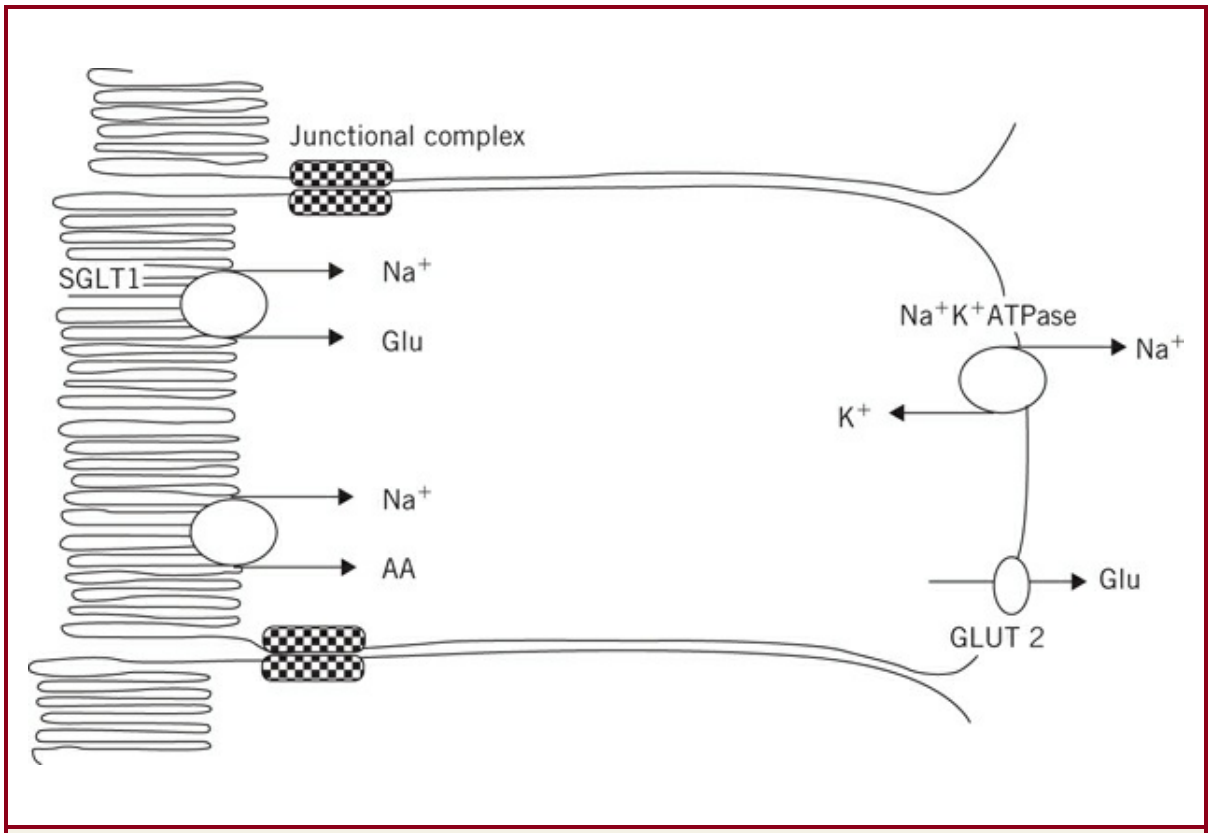


Figure 60-1 Solute-coupled sodium absorption. Adapted from King et al.[18] AA, amino acid; ATPase, adenosine triphosphatase; SGLT1, sodium glucose transporter-1; Glu, glucose.

In 1975, the WHO and the United Nations Children's Fund (UNICEF) agreed to promote a single solution (WHO-ORS) containing (in millimol per liter) sodium 90, potassium 20, chloride 80, base 30, and glucose 111 (2%). This composition was selected to allow a single solution to be used among diverse populations in different countries. Although this solution performed well over 25 years, subsequent clinical research, documented in numerous controlled trials and summarized in a meta-analysis,[26] has favored the adoption of a lower osmolarity standard rehydration solution. Based on these findings, UNICEF and WHO now recommend an ORS formulation with sodium concentration 75 mEq/L, glucose 75 mmol/L, and total osmolarity 245 mmol/L.[27] See Table 60-1 for a comparison of common oral solutions.

TABLE 60-1 Composition of Commercial Oral Rehydration Solutions As Well As Commonly Consumed Beverages

Solution	CHO (g/L)	Glucose (mmol/L	Na (mmol/L)	Na:Glu	K (mmol/L)	Cl (mmol/L)	Ba
WHO-ORS (2002)	13.5	75	75	1:1	20	65	10
ESPGHAN ORS	16	89	60	0.7:1	20	60	10
ReSoMal	22.5	125	45	0.4:1	40	70	21
Enfalyte**	30	167	50	0.3:1	25	45	34
Pedialyte†	25	139	45	0.3:1	20	35	30
Apple juice‡	120	—	0.4	—	44	45	—
Coca Cola§	112	—	1.6	—	—	—	13
Gatorade‖	58	—	23	—	3	17	3
Chicken broth‡	8	—	260	—	0.5	260	—
Brewed Tea‡	4	—	—	—	6	—	—

CHO, carbohydrate; Glu, glucose; ESPGHAN, European Society of Paediatric Gastroenterology, Hepat rehydration solution; WHO, World Health Organization.

[*]Actual or potential bicarbonate (e.g., lactate, citrate, or acetate)

[**]Mead-Johnson Laboratories, Princeton, NJ.

[†]Ross Laboratories, Columbus, OH (data for Flavored and Freezer Pop Pedialyte are identical).

[‡]U.S Department of Agriculture.

[§]Coca-Cola Corporation, Atlanta, GA (figures do not include electrolytes, which may be present in local phosphate).

[‖]The Gatorade Company, Chicago, IL.

Source: Adapted from King et al.[18]

HOME MANAGEMENT OF ACUTE DIARRHEA

The simplicity of treatment with ORS makes the management of uncomplicated cases of diarrhea at home an important component of optimal care. As long as caretakers are properly instructed regarding the need for clinical assessment when children appear significantly ill or appear to be failing treatment, then it is appropriate to initiate therapy at home. Early intervention may reduce complications such as dehydration and poor nutrition. In both developed and developing settings, early administration of ORS may lead to fewer office or emergency room visits,[28] hospitalizations, or deaths.

In fact, it has frequently been the case that mothers and other caretakers begin treatment of diarrhea at home.[29] The advent of ORT allows a therapy that is more effective and less harmful than many traditional home therapies. Families should be encouraged to have a supply of ORS in the home at all times, much in the same way that acetaminophen and adhesive bandages are viewed as staples of the medicine chest. As soon as diarrhea begins, one of the commercially available products can be started at home. Although it is possible to produce a homemade solution with appropriate concentrations of glucose and sodium, possibilities for serious error in preparation exist. Therefore, standard commercial oral rehydration preparations should be recommended where they are readily available and not prohibitively expensive. Regardless of the fluid used, an age-appropriate, unrestricted diet should be encouraged.[9] The most important principles underlying home management of diarrhea is the need to give increased volumes of fluid as well as to maintain adequate energy intake. Infants should be more frequently breast-fed or bottle-fed, and children should be offered fluids frequently.

Caretakers should be educated to recognize signs of illness or

treatment failure that necessitate medical intervention. Infants with acute diarrhea are more prone to dehydration than are older children because they have a higher body surface-to-volume ratio, a higher metabolic rate, and relatively smaller fluid reserves. In addition, they depend on their caretakers for fluid provision. For this reason, parents should seek medical care promptly for infants with diarrhea, even before the first signs of dehydration are evident. Indications for prompt medical evaluation are summarized in Table 60-2. No guidelines have established a specific age under which evaluation is mandated, but, in general, infants should be evaluated by a health professional when there is risk of dehydration; in general, the smaller the child, the lower the threshold for hands-on evaluation. When fever is present, infants and children should be evaluated according to current guidelines. Naturally, underlying conditions, including history of prematurity, metabolic disorders, immune compromise, and recent recovery from surgical interventions, may prompt early evaluation, as may concurrent illnesses such as a respiratory infection. Children with dysentery (visible blood or mucus in stool) should also be evaluated by medical staff promptly.

TABLE 60-2 Indications for Medical Evaluation

Young age, with low threshold for evaluating all children under 10 kg and lowest threshold for those under 6 months
History of prematurity, underlying chronic medical conditions, or concurrent illness
Fever ≥ 38°C for infants under 3 months, ≥ 39°C for infants aged 3–36 months
Visible blood in the stools
High output, including large-volume diarrhea and persistent vomiting
Caretaker's report of signs consistent with dehydration, including sunken eyes, decreased tears, dry mucous membranes
Poor apparent response to ORT or inability to administer ORT

ORT, oral rehydration therapy.

Source: Adapted from King et al.[18]

Naturally, parents or other caretakers cannot be assumed to have skills of assessment comparable to a clinician, but their report of any of the easily recognized signs of dehydration may indicate the need for immediate evaluation. Reports of changing mental status are particularly concerning. When the child's condition is in doubt, there should be a low threshold for recommending evaluation. The hands-on visit provides an opportunity for physical assessment including vital signs, more detailed history-gathering, and family education.

CLINICAL ASSESSMENT

Diarrhea is generally defined as the passage of three or more loose or watery stools per day. The volume of fluid lost through the stools can vary from 5 mL/kg/day (near normal) to 200 mL/kg or more.[30] Dehydration and electrolyte losses follow untreated diarrhea and cause the primary morbidity of acute gastroenteritis. In addition, loose stools may be among the presenting signs of non-gastrointestinal illnesses, including meningitis, malaria, bacterial sepsis, pneumonia, otitis media, and urinary tract infection. Vomiting alone can be the first symptom of metabolic disorders, congestive heart failure, toxic ingestions, or trauma. To rule out other serious illnesses, a detailed history and physical examination should be performed in the setting of acute gastroenteritis.

History taking should include questions about the onset, frequency, quantity, and character (including the presence of bile, blood, mucous) of vomiting and diarrhea. Recent oral intake including food, fluids, and breast-feeding, as well information about urine output should be carefully noted. A recent weight measurement, if available, should be compared to the current weight to estimate total body fluid deficit. Associated symptoms, including fever or changes in mental status, should be recorded. Past medical history should include questions regarding underlying significant medical problems, history of other recent infections, and human immunodeficiency virus (HIV) status, if known. Relevant social history may include the number and nature of caretakers and their ability to return for re-evaluation, which may affect instructions regarding follow-up care.

As part of the complete physical examination, an accurate body weight must be obtained, along with temperature, heart rate, respiratory rate, and blood pressure. When premorbid weight is not known but a previous growth curve is available, an estimate of fluid loss may be obtained by subtracting current weight from expected weight based on previous weight-for-age percentile.[31] The quality of this estimate will vary, depending on the number and variability of prior data points. The general condition of the patient should be assessed, with special concern given to infants and children who appear listless, apathetic, or minimally interactive. The appearance of the eyes should be noted, including the degree to which they are sunken and the presence or absence of tears. The condition of the lips, mouth, and tongue may yield important clues as to the degree of dehydration, even if the patient has recently taken fluid. Deep respirations may be suggestive of metabolic acidosis. Faint or absent

bowel sounds may suggest ileus. Extremities should be assessed for signs of general perfusion, capillary refill, and skin turgor. Visual examination of the stool can confirm abnormal consistency and determine the presence of blood or mucus.

The clinical signs and symptoms of dehydration are outlined in Table 60-3. A comprehensive literature review found that the three most useful individual signs for predicting 5% dehydration in pediatric patients are abnormal capillary refill time, abnormal skin turgor, and an abnormal respiratory pattern.[32] The assessment of the anterior fontanel, though helpful in select instances, can be unreliable or misleading.[33] In infants and children, a fall in blood pressure is a late sign that signifies the development of uncompensated shock and may correspond to fluid deficits greater than 10%. Increases in heart rate and slowed peripheral perfusion may be more sensitive indicators of moderate dehydration, although both may be difficult to interpret as they can vary with fever. Decreased urine output is a sensitive but nonspecific sign,[31] since urine output may be difficult to assess in infants with diarrhea.

TABLE 60-3 Signs of Dehydration

	Minimal or No Dehydration	Mild to Moderate Dehydration	Severe Dehydration
Mental status	Well, alert	Normal, fatigued or restless, irritable	Apathetic, lethargic, unconscious, floppy
Thirst	Drinks normally, may refuse	Thirsty, eager to drink	Drinks poorly, unable to drink
Heart rate	Normal	Normal to increased	Tachycardia, with bradycardia in most severe cases
Quality of pulses	Normal	Normal to decreased	Weak, thready, or impalpable
Breathing	Normal	Normal, fast	Deep
Eyes	Normal	Slightly sunken	Deeply sunken
Tears	Present	Decreased	Absent
Mouth and tongue	Moist	Dry	Parched
Skin fold	Instant recoil	<two seconds	>two seconds
Capillary refill	Normal	Prolonged	Prolonged, minimal
Extremities	Warm	Cool	Cold, mottled, cyanotic

Urine output	Normal to decreased	Decreased	Minimal
Loss of body weight	<3%	3–9%	>9%

Source: Adapted from King et al.[18]

Previous guidelines, including the CDC's 2003 recommendations[18] divide patients into subgroups for mild (3–5% fluid deficit), moderate (6–9% fluid deficit), or severe dehydration (>10% shock or near shock). Other classification schemes, including 2005 WHO,[34] divide patients into those showing no signs of dehydration (<5%), some signs of dehydration (5–10%), and severe dehydration (>10%). Studies that have carefully evaluated the correlation of clinical signs of dehydration with posttreatment weight gain suggest that the first signs of dehydration are evident at 3–4% dehydration, with more numerous clinical signs evident at 5% dehydration and those signs indicating severe dehydration not evident until fluid loss of 9–10%.[31] Because of this threshold effect, it may be difficult to distinguish between mild and moderate dehydration based on clinical signs alone. For this reason, Table 60-3 groups together patients with mild to moderate dehydration and notes that these signs may be seen over a relatively wide range of fluid loss, from 3 to 9%. The magnitude of ongoing losses and the patient's oral intake will determine the success of ORT as much as current status. The goal of assessment is not to solely determine the patient's hydration status but rather to provide a starting point for treatment. In this way, the goal is to determine which patients may be safely sent home for therapy, which should remain for observation during therapy, and which should receive more immediate intensive therapy.

Supplementary laboratory studies in the assessment of the patient with acute diarrhea are usually unnecessary, including serum electrolytes.[34] Stool cultures are indicated in the case of dysentery but are generally not indicated in acute, watery diarrhea in the immunocompetent patient. However, laboratory studies should be performed where they may give important clues as to underlying or alternative causes of illness. For instance, urine cultures should be obtained when there is reason to be concerned for urinary tract infection, and blood cultures and white blood cell count with differential are indicated where the clinical presentation suggests possible sepsis. Children with severe dehydration may require serum electrolyte evaluation and monitoring.

THERAPY OF ACUTE GASTROENTERITIS BASED ON DEGREE OF DEHYDRATION

Table 60-4 provides a summary of specific treatment recommendations synthesized from CDC, WHO, and ESPGHAN guidelines.[18,34,35] Treatment of dehydration should include two phases: rehydration and maintenance. In the rehydration phase, the fluid deficit is replaced quickly, over 3–4 hours, and clinical hydration is attained. In the maintenance phase, maintenance calories and fluids are given. Rapid realimentation should follow rapid rehydration, with a goal of rapid return to an age-appropriate unrestricted diet, including solids. Gut rest is not indicated. If anything, caloric provision should be increased as soon as tolerated to make up for lost caloric intake during the acute illness. In general, lactose restriction and formula changes are not necessary. Full-strength formula is generally tolerated and allows more rapid return to full caloric intake. Breast-feeding should be continued throughout each treatment phase, including rehydration. In both phases, fluid losses from vomiting and diarrhea are replaced in an ongoing manner. No unnecessary medications should be used, and laboratory studies should be limited to those necessary to guide management.

Minimal Dehydration

For those patients with minimal or no dehydration, treatment is aimed at replacing ongoing losses. Children with diarrhea require increased fluid intake to make up for losses and to provide for maintenance needs. In principle, 1 mL of fluid should be given for each gram output. In hospital settings, soiled diapers can be weighed (without urine) and the estimated dry weight of the diaper can be subtracted. When losses are not easily measured, a reasonable rule of thumb is to provide 10 mL/kg additional fluid for each watery stool or 2 mL/kg for each episode of emesis. Nutrition should not be restricted. Instructions regarding dietary therapy are summarized in Table 60-4.

Mild to Moderate Dehydration

Children who are found on assessment to have mild to moderate dehydration should receive rapid replacement of their estimated fluid deficit, with additional fluid given for ongoing losses. Current guidelines recommend giving 50–100 mL/kg of ORS over 2–4 hours to replace estimated fluid deficit, with additional ORS given to replace ongoing losses. WHO guidelines recommend a maximum rate of 20 mL/kg/hour,[34]

but the rate may be individualized. Using a teaspoon, syringe, or medicine dropper, small volumes of fluid (e.g., one teaspoon) should be offered at first, with the amount gradually increased as tolerated. If a child appears to want more than the estimated amount of ORS, more can be offered. Although it is safe to give ORS very rapidly, vomiting may be increased with larger boluses. Frequent, small bolus feedings will generally be tolerated. Nasogastric (NG) feeding allows continuous administration of ORS at a slow, steady rate. Clinical experience supports the use of NG feedings even in vomiting patients. Rehydration via NG tube may be particularly useful in an emergency department setting where rapid correction of hydration may prevent unnecessary hospitalization. Although rapid intravenous hydration may also prevent unnecessary hospital admissions, one study showed that rapid NG rehydration was well tolerated, more cost–effective, and associated with fewer complications.[36] In addition, emergency department caregivers reported being more familiar with intravenous hydration as compared to NG rehydration when oral rehydration failed.[37]

TABLE 60-4 Summary of Treatment Plans

Degree of Dehydration	Rehydration Therapy	Replacement of Losses	Nutrition
Minimal or no dehydration	N/A	1 mL ORS for each gram lost to vomiting or diarrhea or 10 mL/kg body weight for each diarrheal stool, 2 mL/kg for each episode emesis	Continue breast-feeding; resume age-appropriate normal diet after initial hydration, including adequate caloric intake for maintenance*
Mild to moderate dehydration	ORS 50–100 mL/kg body weight over 3–4 h	Same as above	Same as above
Severe dehydration	LR or NS in 20 mL/kg IV boluses until perfusion and mental status improve. Then give 100 mL/kg ORS over 4 h or 100 mL/kg D5 ½ NS IV over 8 h	Same as above; if unable to drink, give via NG tube or give IV D5 ¼ NS with 20 mmol/L KCl/L	Begin oral or enteral feedings after initial hydration, as soon as acidosis corrected

IV, intravenous; LR, lactated Ringer's; N/A, not applicable; NS, normal saline; ORS, oral rehydration solution.

* Overly restricted diets should be avoided during acute diarrhea. Breast-fed infants should continue to nurse ad libitum even during acute rehydration. Infants too weak to eat may be given breast milk or

formula via NG tube. Full-lactose formulas are generally well tolerated. If lactose malabsorption appears clinically significant, lactose-free formulas may be used. Complex carbohydrates, fresh fruits, lean meats, yogurt, and vegetables are all recommended.

Source: Adapted from King et al.[18]

OUTPATIENT OBSERVATION

Because a certain percentage of children with mild to moderate dehydration will fail to improve with ORT, it is prudent to observe dehydrated children until signs of dehydration resolve. Similarly, children who do not show clinical signs of dehydration but who demonstrate unusually high output may be held for observation. Hydration status should be reassessed on a regular basis, with more frequent monitoring given to those patients whose status is more tenuous. This may be carried out in an emergency room, office, or other outpatient setting. Once dehydration is corrected, further management may be carried out at home if the following conditions are met: current ORT is well tolerated and the child's caretakers demonstrate comprehension of home rehydration techniques and understand indications for returning for further evaluation, and have adequate means of transportation to return to care if needed. Even among those whose illness appears uncomplicated on initial assessment, a small percentage may fail ORT and, therefore, a plan for possible reassessment must be in place. Caretakers should be encouraged to return to medical attention should they have any concerns, if they are not sure that rehydration is proceeding well, or for the development of any new or worsening symptoms.

MANAGEMENT IN THE HOSPITAL

Indications for hospital management of a dehydrated child include (1) inability of caretakers to manage ORT at home; (2) significant difficulties administering ORT, including vomiting, ORS refusal, or inadequate intake; (3) concern for possible comorbidity; (4) failure of treatment, including worsening diarrhea or dehydration despite ORT; (5) severe (>9%) dehydration; (6) social or logistical issues that may prevent return evaluation if necessary; or (7) any other factors that may indicate close observation, such as very young age of the patient, unusual irritability or drowsiness, progressive course of symptoms, or uncertainty of diagnosis.

SEVERE DEHYDRATION

Severe dehydration constitutes a medical emergency. Intravenous rehydration should begin immediately. Twenty milliliters per kilogram boluses of lactated Ringer's (LR) solution, normal saline, or a similar isotonic solution should be given until pulse, perfusion, and mental status return to normal. This may require two intravenous lines or even alternative access sites (e.g., intraosseous infusion, venous cutdown). The patient should be closely observed during this period and vital signs monitored on a regular basis. Where available, serum electrolytes, bicarbonate, blood urea nitrogen, creatinine, and serum glucose should be obtained, although it is safe to commence rehydration therapy without these results, as normal saline or LR infusion is the appropriate first step in either hyponatremic or hypernatremic dehydration. Hypotonic solutions should not be used for acute parenteral rehydration.[38]

The severely dehydrated patient may require several boluses, which may be carried out in short succession. Practically speaking, overly rapid rehydration is unlikely to occur as long as weight-based boluses are given with close observation. Errors occur most commonly in settings in which adult dosing is given to infants. For example, if a practitioner were to order "500 cc of normal saline intravenous bolus × ii," this would provide 200 mL/kg for the average 2- to 3-month-old, which would be incorrect. Edema of the eyelids and extremities may indicate overhydration. Diuretics should not be given. Once the edema has subsided, the patient may be reassessed for continued therapy. With very frail or malnourished infants, it may be prudent to proceed with 10 mL/kg boluses because of their poorer ability to increase cardiac output and because it may be especially difficult to distinguish dehydration from sepsis in these patients. The smaller boluses will also facilitate closer evaluation. Please see Chapter 14: "Protein-Energy Malnutrition: Pathophysiology, Clinical Consequences and Treatment" for details. In general, frequent reassessment of hydration status should be performed to follow the adequacy of replacement therapy. Failure to respond to fluid bolus should raise the suspicion of alternative or concurrent diagnoses, including, septic shock, as well as metabolic, cardiac, or neurologic disorders.

As soon as the patient's level of consciousness returns to normal, therapy can often be changed to the oral route, with the patient taking orally the remaining estimated deficit. An NG tube may be helpful for patients too weak to drink adequately but with normal mental status. Although no studies have specifically documented increased aspiration risk with NG tube use in obtunded patients, intravenous therapy is ordinarily favored in these patients. Although it is reasonable to leave

intravenous access in place in these patients should it be needed, early reintroduction of ORS is safer. The use of intravenous catheters is associated with frequent minor complications, including extravasation of intravenous fluid, and rare significant complications, including the inadvertent administration of inappropriate fluid such as those containing excessive potassium. In addition, early ORS will likely encourage earlier resumption of feeding, and data suggest that resolution of acidosis may be more rapid with ORS compared with intravenous fluid.[36]

DIETARY THERAPY

Recommendations for maintenance dietary therapy depend on the age and diet history of the patient. Breast-fed infants should continue nursing on demand. Those receiving formula should continue their usual formula immediately on rehydration in amounts sufficient to satisfy energy and nutrient requirements. Lactose-free or lactose-reduced formulas are usually not necessary. A meta-analysis of clinical trials shows no advantage of lactose-free formulas over lactose-containing formulas for most infants, although some infants with malnutrition or severe dehydration recover more quickly when given lactose-free formula.[39] Patients with true lactose intolerance may have exacerbation of diarrhea when a lactose-containing formula is introduced. The presence of a stool pH below 6.0 or the presence of reducing substances in the stool (>0.5%) in the absence of clinical symptoms is not diagnostic of lactose intolerance. Although medical practice has often favored beginning feeds with diluted (e.g., half or quarter strength) formula, there is insufficient evidence to justify this practice; full caloric intake should be restored as soon as possible.

Soy formulas containing soy fiber, specifically Isomil® DF, have been widely marketed to physicians and consumers in the United States. Brown et al. showed that added soy fiber reduced the duration of liquid stools without changing overall stool output.[40] This “cosmetic” effect might have some benefits with regard to diminishing diaper rash and encouraging early resumption of normal diet but is probably not sufficient to merit its use as standard of care.

Children receiving semisolid or solid foods should continue to receive their usual diet during diarrhea. Foods high in simple sugars should be avoided, as the osmotic load may worsen diarrhea. For this reason, large amounts of juice, gelatin desserts, and other highly sugared liquids should be avoided. Some guidelines have recommended avoiding fatty foods, but it is difficult to maintain adequate calories without some fat. In addition,

fat may reduce intestinal motility. The practice of withholding food for 24 hours is inappropriate. Early feeding may decrease changes in intestinal permeability brought about by infection,[41] reduce illness duration, and improve nutritional outcomes.[8,9] Highly specific diets, such as the BRAT (bananas, rice, applesauce, and toast) diet, are commonly recommended. Although one study has shown some benefit from green bananas and pectin in persistent diarrhea (PD),[42] the BRAT diet is unnecessarily restrictive and, like juice-centered diets, provides suboptimal nutrition for the patient's nourishment and recovering gut.

Many children in developing countries have multiple episodes of diarrhea in a single year. In this way, losses from diarrhea contribute to poor nutrition, which may, in turn, worsen the severity of subsequent episodes of diarrhea.[43] For this reason, WHO guidelines recommend caloric supplements following episodes of diarrhea, such as added cereal in milk, oil in cereal, and foods with high nutrient density.[34] In general, current guidelines call for age-appropriate unrestricted diets, including complex carbohydrates, meats, yogurt, fruits, and vegetables.

LIMITATIONS OF ORT

Although ORT is recommended for all age groups and for diarrhea of any etiology, some limitations exist. In children presenting in shock, the administration of oral solutions is contraindicated, as airway protective reflexes may be impaired. Likewise, patients with abdominal ileus should not be given oral fluids until bowel sounds are audible. It should be remembered that intestinal intussusception may present with diarrhea, including bloody diarrhea. Radiographic and surgical evaluations are warranted when the diagnosis is in question.

Stool output in excess of 10 mL/kg/hour has been associated with a lower rate of success of oral rehydration.[44] No patient, however, should be denied ORT simply because of a high purging rate because most patients will respond well if given adequate replacement fluid.

A small proportion of infants with acute diarrhea experience carbohydrate malabsorption, with a dramatic increase in stool output following the administration of ORS. The presence of stool-reducing substances alone is not sufficient to make the diagnosis because this is a common finding in patients with diarrhea and does not in itself predict failure of oral therapy. Patients with true glucose malabsorption will also show an immediate reduction in stool output when ORS is replaced with

intravenous therapy. The incidence of clinically significant glucose malabsorption during acute diarrhea is probably less than 1%, although rates as high as 8% have been reported in selected populations.[45]

Many patients with acute diarrhea have concomitant vomiting. Most, however, can be successfully rehydrated with oral fluids if small volumes of ORS (5 mL) are given every five minutes, with a gradual increase in the amount consumed. Administration via a spoon or syringe with close supervision helps guarantee a gradual progression in the amount administered. Often, correction of acidosis and dehydration lessens the frequency of vomiting. Continuous slow NG infusion of ORS via a feeding tube may be helpful. Even if some emesis occurs after NG administration of fluid, treatment may not be adversely affected.[36]

HYPERNATREMIC DEHYDRATION

Patients with hypernatremic dehydration respond well to ORT. Those with severe dehydration should first receive intravenous hydration, as outlined above. Subsequent hydration may be achieved with ORS.[46] As with normonatremic dehydration, ORS should be given both to replace the calculated deficit and for ongoing losses. ORS is safer than intravenous therapy because it is less likely to lead to a precipitous decline in serum sodium, which may increase intracranial pressure.

PHARMACOLOGIC THERAPY

Antibiotics

Because viral agents are the predominant cause of acute diarrhea (e.g., *Rotavirus*, astrovirus, enteric adenovirus, *Norovirus*, and *Sapovirus*), the routine use of antimicrobial agents for the treatment of diarrhea misuses resources and may lead to increased microbial resistance. Especially in the hospital setting, bacterial causes of diarrhea are unusual. A survey of stool cultures submitted over several years to the microbiology laboratory at Boston Children's Hospital revealed such infrequent occurrence of bacterial infections that the practice of sending stool cultures of patients who develop diarrhea while hospitalized has been abandoned.[47] Hospitalized patients and patients with a history of recent antibiotic use are, however, at risk for infections with *Clostridium difficile*. Infections with *C. difficile* documented by toxin assay or PCR should be treated with metronidazole 30 mg/kg divided three times daily given orally. Oral

vancomycin is also effective but should be reserved as a second-line agent, given its expense and concern over the possible emergence of vancomycin-resistant *Enterococcus.*

Even when a bacterial cause is suspected, antibiotic therapy is generally not indicated because the majority of cases of acute diarrhea are self-limited and not shortened by antibiotics. In addition, antibiotics may cause harm. There is evidence that antibiotic use may increase the risk of hemolytic uremic syndrome from *E. coli* infections.[48] There remains some concern that treatment of *Salmonella enteritis* may increase the risk of the carrier state, although patients below 3 months of age or with bacteremia should be treated.[49] Patients with underlying disorders, including immunodeficiency, provide exceptions to these rules. Detailed information regarding the appropriate use of antimicrobial therapy for infectious causes of acute diarrhea is beyond the scope of this chapter and is available in a variety of other resources. [35,50]

Non-Antibiotic Drug Therapies

The use of nonspecific antidiarrheal agents such as adsorbents (e.g., kaolin-pectin), antimotility agents (e.g., loperamide), antisecretory drugs, or toxin binders (e.g., cholestyramine) is a common practice. Few data are available to support their efficacy. The side effects of these drugs are well known, in particular among the antimotility agents, including opiate-induced ileus, drowsiness, and nausea owing to atropine effects and binding of nutrients and other drugs. One report from Pakistan detailed 18 cases of severe abdominal distention in association with use of loperamide, including at least 6 deaths.[51] Bismuth subsalicylate has shown some efficacy in traveler's diarrhea and other causes of acute gastroenteritis in children,[52] and although the side effects are less than antimotility agents, some theoretic concerns over the potential toxicity of salicylate remain. In any event, reliance on antidiarrheal agents shifts the therapeutic focus away from appropriate fluid, electrolyte, and nutritional therapy and adds unnecessarily to the economic cost of the illness.

It should be noted that none of the drugs mentioned above specifically address the underlying causes of diarrhea, specifically increased secretion by intestinal crypt cells. Racecadotril, an enkephalinase inhibitor, preserves the antisecretory activity of enkephalins, was first discovered in 1975. It does not slow intestinal transit or promote bacterial overgrowth. Its use has shown promise in two controlled clinical trials in children, significantly reducing acute watery diarrhea compared to placebo.[53] As

long as its cost is reasonable and its use does not distract from ORT and education regarding home management of diarrhea, racecadotril may prove to be a useful adjunct to the treatment of watery diarrhea.

Similarly, the use of antiemetics may be warranted in some cases. Phenothiazines can interfere with oral rehydration by causing sleepiness. Oral ondansetron, a 5HT3 receptor antagonist, has been shown to be effective in decreasing the need for intravenous hydration and hospitalization in children with actue gastroenteritis.[54] A single oral dose of ondansetron, given to pediatric emergency department patients with gastroenteritis and dehydration, significantly decreased the likelihood and frequency of vomiting. Although in this trial there was no significant reduction in the rate of hospitalization, the ondansetron group was significantly less likely to require intravenous hydration, had increased oral intake, and had a shorter mean length of stay in the emergency department.[55]

FUNCTIONAL FOODS

Functional foods may be defined as those that have an effect on physiologic processes separate from their established nutritional function.[56] Probiotics, live microorganisms found in fermented foods and intended to promote intestinal health, are the classic functional food. The mechanism of action of probiotics may include competition with pathogenic bacteria for receptor sites or intraluminal nutrients, production of antibiotic substances, and enhancement of host immune defenses.[57,58]. Chapter 12 provides a detailed discussion of this topic; the following discussion focused on the effects of functional foods solely on diarrhea.

Probiotics seem to be particularly effective in instances of diarrhea due to antibiotic use and rotavirus. In 2011, a systematic review examined 16 studies using various species of probiotics, including *Lactobacillus, Lactococcus, Saccharomyces, Bacillus, Bifidobacterium, and Leuconostoc cremoris*, for the prevention of antibiotic-associated diarrhea in children. Results indicated that probiotics prevented antibiotic-associated diarrhea in children, with higher doses of probiotics (>five billion colony forming units daily) showing a greater protective effect. There were no serious adverse events reported.[59] A second comprehensive review concluded that there is evidence for the efficacy of a wide variety of probiotics in acute gastroenteritis, especially rotavirus.[60] Probiotics may be effective in terms of prevention of diarrhea as well. A meta-analysis of studies

predominately executed in developed countries concluded that probiotics reduced the risk of acute diarrhea among children by 57% and that of antibiotic-associated diarrhea by 52%.[61] In this analysis, the protective effect did not vary significantly among the variety of probiotic strains examined.

Despite these proven benefits, there are several limitations of probiotics. First, because these products are generally not regulated, there is potential for great variability of species concentration and other aspects among different products. In addition, concerns about safety exist, such as the potential for bacterial translocation and subsequent bacteremia or endocarditis.[62] With few exceptions, however, including administration to immunocompromised patients or those with indwelling central venous catheters, the safety of probiotics for the general population has been demonstrated and may confer the benefits discussed above.

Prebiotics differ from probiotics in that they are nutrients, rather than organisms, used to preferentially stimulate the growth of health-promoting intestinal flora.[63] Oligosaccharides found in breast milk have been called the prototypic prebiotic in that they foster the growth of lactobacilli and bifidobacteria in the colon of breast-fed neonates. The prebiotic effects of inulin and fructose oligosaccharides have been studied primarily in vitro and in animal model studies.[64] Two randomized, blinded trials on prebiotic-supplemented infant cereal in an urban, economically depressed area did not demonstrate a reduced incidence of diarrheal disease.[65] Specific recommendations regarding the use of prebiotics should await further well-controlled human trials. In the meantime, it should be noted that dietary fiber may serve as both a prebiotic and a modifying agent of intestinal motility. Current guidelines recommend fiber-containing fruits and vegetables as part of the diet of children recovering from acute gastroenteritis.

SUPPLEMENTAL ZINC ADMINISTRATION

Many reports have linked diarrhea and abnormal zinc status, including increased stool zinc loss,[66] and negative zinc balance.[67] It has long been observed that diarrhea may occur with severe zinc deficiency, as in acrodermatitis enteropathica, but the broader clinical question is whether more incremental deficiencies of zinc may play a role in childhood diarrhea and whether supplementation may be of benefit for either improved outcomes in acute or chronic diarrhea or as prophylaxis against

diarrheal disease. In this way, zinc therapy can be considered both as an adjunct treatment during acute episodes of diarrhea as well as a preventive therapy for subsequent diarrheal episodes.

When given during an acute episode of diarrhea, zinc decreases the severity and duration of diarrhea.[68] In children older than 6 months of age, oral zinc supplementation has been shown to decrease the duration of acute diarrhea by 10.4 hours and reduces the probability of developing diarrhea for more than 7 days. Moreover, this effect was more pronounced in children with moderate malnutrition, whose duration of diarrhea was decreased by 27 hours.[69] There were no serious adverse effects reported, although some children experienced vomiting with oral zinc supplementation.

WHO and UNICEF now recommend that all children with acute diarrhea receive zinc supplementation for a total of 10–14 days during and after diarrhea (10 mg per day for infants under 6 months of age and 20 mg per day for children greater than or equal to 6 months of age).[34,70] Zinc supplementation in more developed settings may not be as efficacious, as has been demonstrated in trials of children (one among children with diarrhea and one among healthy children) from the United States and Poland.[71,72] Further research is needed to identify the mechanism of action and to determine optimal schemes for delivery of zinc to those populations who will gain the most benefit.

Oral zinc supplementation has also been studied as a prophylactic measure for preventing episodes of acute diarrhea. The Zinc Investigators Collaborative Group reviewed evidence that zinc supplementation is beneficial as a prophylactic supplement for decreasing the incidence of diarrheal disease and pneumonia. Among infants and young children who received supplemental zinc for 5 or 7 days per week for 12 to 54 weeks, the pooled odds ratio for diarrhea incidence was 0.82 (95% confidence interval [CI] 0.72–0.93) and that for pneumonia incidence was 0.59 (95% CI 0.41–0.83).[73]

OTHER CLINICAL SCENARIOS

Acute Bloody Diarrhea (Dysentery)

Dysentery is defined as acute bloody diarrhea caused by invasive microbial infection. This does not include occult blood (detected by guaiac card only), streaks of blood on the surface of formed stool, or melena. The reader is referred to WHO guidelines, which discuss the evaluation and

management of dysentery at length.[30,34]

The treatment of dehydration in dysentery follows the same principles as the treatment of acute watery diarrhea. The child with bloody diarrhea is at higher risk for complications, including sepsis and other systemic disease, so that the threshold for admission to the hospital for close observation is lower. Stool cultures are indicated in the setting of acute bloody diarrhea and are helpful for guiding therapy. Food should not be withheld in dysentery any more than in other cases of diarrhea. Because patients with dysentery may have significant anorexia, they should be coaxed to eat.[30] More frequent, smaller meals may be better tolerated. Children convalescing from dysentery should be given extra food to help them regain nutrition lost during the acute illness.

Cholera

Cholera differs from other causes of acute diarrhea in that (1) it may occur in large epidemics involving adults and children (as seen in the recent epidemic in Haiti); (2) it produces voluminous diarrhea, which may lead to shock and death by dehydration in a very short period; and (3) certain antibiotics may shorten the course.[34] Management of cholera may be carried out using the methods for assessment of dehydration and treatment plans outlined above and summarized in Tables 60-4 and 60-5. The health worker must be prepared to deliver prodigious quantities of fluid, either orally or intravenously, to overcome ongoing losses. These losses should be quantified so that treatment may be updated and modified under continuous close observation. Unlike diarrhea from other causes, where cereal-based ORSs are equivalent to standard ORSs, there may be some advantages in using rice-based ORS in treating cholera.[74] Traditional wisdom held that sodium concentrations of ORS used in treating cholera should be higher than those for other causes of diarrhea (based on measured stool sodium losses), but a CHOICE study group trial showed no clinical difference between those treated with the lower osmolarity solution compared with a standard solution, apart from some increased incidence of asymptomatic hyponatremia.[75] In Bangladesh, a study of over 50,000 monitored patients with uncomplicated watery diarrhea found no evidence for an increase in symptomatic hyponatremia in those treated with reduced osmolarity ORS. In fact, the incidence rate of developing symptomatic hyponatremia was significantly lower in those treated the newer, reduced osmolarity formulation.[76]

Persistent Diarrhea

Persistent diarrhea (PD) may be defined as diarrhea of acute onset that lasts more than 14 days. PD is also frequently associated with poor weight gain or weight loss.[77] Globally, PD accounts for a substantial proportion of diarrhea-related morbidity and between 36% and 54% of all diarrhea-related deaths.[78] A community-based study suggested that PD is experienced by roughly one in five young children per year in the United States, though with far less morbidity and mortality.[79]

In developing countries, the most important epidemiological risk factor for acquiring PD is malnutrition, further emphasizing the importance of aggressive nutritional therapy in the treatment of acute diarrhea. Other risk factors for PD include immune status (including HIV infection), zinc deficiency, lack of breast-feeding, male sex, and infection with enteropathogenic or enteroaggregative *Escherichia coli* or *Cryptosporidium*.[80] Malnutrition can also significantly increase mortality from PD, with a study of diarrheal deaths in Bangladesh demonstrating a 17-fold higher relative risk of death from PD in malnourished children compared to those lesser degrees of malnutrition.[81] Another study in Kenya showed that children who are hospitalized for severe acute malnutrition who have diarrhea are at an increased risk of dying compared to those hospitalized children without diarrhea.[82]

TABLE 60-5 ORS for the Severely Malnourished Child

Simple ORS for the Severely Malnourished Child	More Effective ORS for the Severely Malnourished Child
Dilute 1 L WHO-ORS to make 2 L	Dilute 1 L WHO-ORS to make 2 L
Add 45 mL KCl solution from stock solution containing 100 g KCl/L	Add the following salts:
Add and dissolve 50 g sucrose	3.6 g KCl
	1.3 g K citrate
Children given this solution should also receive:	1.2 g MgCl
2 mL 50% MgSO4 solution (4 mEq Mg/mL) IM once	130 mg Zn acetate
Zinc chloride solution (10 g/L) 1 mL/kg/d until diarrhea stops	22 mg CuSO4
	0.44 mg NaSeO4
	0.20 mg KI

	Add and dissolve 50 g sucrose

IM, intramuscularly; ORS, oral rehydration solution.

Source: Adapted from World Health Organization.[34]

The approach to the patient with PD should include prompt assessment and treatment of the diarrhea. Worldwide, the most important predisposing factor for PD is an acute diarrheal episode triggered by an enteric infection.[80] Particularly in the developing world, bacterial and parasitic infections are common and warrant aggressive diagnosis and therapy. In areas where HIV infection is endemic, chronic gastrointestinal infections with organisms such as *Cryptosporidium parvum* are even more likely.

When infection has been carefully excluded, other specific etiologies must be considered. These include food allergy and dietary protein intolerances, celiac disease, disaccharide (i.e., lactose) intolerance, cystic fibrosis and other causes of pancreatic insufficiency, and inflammatory bowel disease. For further details on the diagnosis and nutritional management of these disorders, please refer to Chapters 52 (Inflammatory Bowel Disease), 53 (Celiac Disease), 54 (Food Allergies), and 55 (Exocrine Pancreatic Disease).

In the developed world, chronic nonspecific diarrhea of childhood (or "toddler's diarrhea") is a common, nonpathologic phenomenon seen in children between the ages of 6 and 36 months. It is characterized by the passage of two or more loose, voluminous stools per day for over four weeks, in the absence of abdominal pain or growth inhibition. Diets either low in fat (which modulates intestinal motility), low in fiber (which may promote balanced intestinal flora) and/or high in high-osmolarity carbohydrate drinks, particularly sorbitol-containing fruit juice, may all contribute to PD in the developed world. In such cases, dietary modification should be attempted before undertaking an extensive diagnostic evaluation.

Attention to nutrition is critical in the treatment of PD, with restoration of a nutritious diet, including adequate calories, being a key element of therapy. As with acute diarrhea, recommended foods include age-appropriate unrestricted diets, including complex carbohydrates, meats, yogurt, fruits, and vegetables.[18] As previously discussed, zinc supplementation has been shown to be effective in reducing the duration and severity of PD and should be continued for prophylaxis against additional episodes after appropriate therapy is complete.[73] In cases of infectious etiologies, education regarding sanitation and hygiene may reduce rates of re-infection.

For details of PD in the setting of protein energy malnutrition, please refer to Chapter 14.

DIARRHEA WITH SEVERE MALNUTRITION

Assessment of diarrhea and dehydration in the malnourished child is difficult because many of the signs outlined in Table 60-3 may be unreliable. Skin turgor may appear poor owing to the absence of subcutaneous fat. Eyes may be sunken from loss of periorbital fat. Irritability or apathy from malnutrition may complicate the assessment of mental status. When possible, malnourished children with diarrhea should be referred to a hospital. A severely malnourished child with signs of dehydration but without a history of increased stool output should be treated for septic shock.[34] Because of the increased risk of bacteremia in severely malnourished children, and, in fact, in a broad range of children with diarrhea in a less developed setting,[83] the use of empiric antibiotics is not unreasonable where blood cultures cannot be obtained, although the literature regarding choice of antibiotics, doses, and duration is sparse.

Because intravenous therapy may cause overhydration and heart failure in the severely malnourished child, except for treatment of shock, slow oral rehydration is the treatment of choice. An NG tube may be used for children who drink poorly. Rehydration should begin with 10 mL/kg over 2 hours. This rate may be adjusted based on the child's thirst or ongoing stool losses. Increasing edema may be evidence of overhydration.

Full-strength WHO-ORS contains excessive sodium and inadequate potassium for the severely malnourished child. WHO guidelines recommend modified solutions such as ReSoMal, as outlined in Tables 60-1 and 60-5. Feeding should begin as soon as possible once hydration has been achieved and should continue every two to three hours day and night. Malnourished children often exhibit anorexia and require coaxing to eat. For more details, the reader is referred to Chapter 14.

CHOICE OF ORS

Table 60-1 outlines the composition of several commonly available ORSs, as well as other beverages frequently but inappropriately used for rehydration. The vast differences between the composition of the WHO ORS and beverages inappropriately recommended for rehydration should be noted. In July 2002, WHO and UNICEF recommended a significant change in ORS composition, including a reduction in sodium and glucose

to maintain a 1:1 molar ratio but to decrease total osmolarity to 245 mOsm/L. The change was prompted by clinical trials showing that this solution resulted in less use of supplemental intravenous fluid therapy as well as lower stool output and less vomiting compared with the previous WHO standard ORS.[26,84] Although the differences in measured stool output resulting from different ORS formulations may be modest, reduction in intravenous fluid use is a substantial economic and health benefit of the new solution. The composition of the new solution is also more in line with solutions more commonly used in industrialized countries.

NEW SOLUTIONS

Numerous improved ORSs have been attempted. These have generally included additional substrates for sodium cotransport (such as the amino acids glycine, alanine, and glutamine)[85,86] or, as reviewed by Fontaine and colleagues,[74] substituting complex carbohydrates for the glucose (rice and other cereal-based ORSs) to reduce osmolarity while preserving glucose-sodium cotransport. Given trials to date, the amino acid preparations do not appear to be more effective than traditional ORSs and are more costly. Rice-based ORS may be recommended where training is adequate and home preparation is preferable and appears to be particularly effective in treating diarrhea from cholera.[74,87] Nevertheless, given the simplicity and safety of ORS packets in developing countries and commercially available ORSs in developed countries, these remain the first choice for most clinicians.

Other potential additives to ORS include substances capable of liberating short-chain fatty acids. These include amylase-resistant starch derived from corn[88] and partially hydrolyzed guar gum.[89] The presumed mechanism of action is the enlistment of increased colonic sodium uptake coupled to short-chain fatty acid transport. A controlled trial of children with acute diarrhea demonstrated significantly decreased diarrhea duration in those given amylase-resistant starch-fortified ORS compared to those receiving the standard WHO ORS formulation.[90] In addition, data from adults with acute cholera-like diarrhea in India showed that duration of acute diarrhea was reduced by 55% (19 hours versus 42 hours, $P < .001$) in the cohort given ORS with amylase-resistant starch versus standard ORS.[91] Currently, guidelines do not support the routine incorporation of any potential ORS additives, including guar gum, nondigestible

carbohydrates, probiotics, zinc, glutamine, citing insufficient evidence.[35]

BARRIERS TO ORT

Barriers to the use of ORS and continued nutrition during diarrheal disease include, among patients, cultural practices[29] and lack of information[92] and, among physicians, preference for intravenous hydration, even where evidence suggests improved results from oral rehydration.[93] At the other extreme from the development of the ultimate ORS, it remains distressingly common for patients, even at times under physician supervision, to attempt rehydration with solutions bearing no resemblance to physiologically based ORS. Table 60-1 includes a number of fluids commonly used in treating diarrhea, which do not contain physiologically sound concentrations of carbohydrates and electrolytes. An informal survey of numerous hospital websites reveals outdated recommendations for the treatment of diarrhea that include nonstandard fluids. A case report of one child whose care was compromised by following advice obtained from a prominent hospital's Internet site highlights the continued gap between knowledge and practice and the ongoing need to disseminate accurate information regarding oral rehydration.[94]

CONCLUSION

The treatment of acute diarrhea has for many years been shown to rely on the simple but overwhelmingly effective therapy of oral rehydration. The important co-principle of early refeeding of children immediately on rehydration has also gained wider acceptance. The combination of oral rehydration and early nutritional support is a safe and effective treatment for the vast majority of children with diarrhea. In addition, the advances made in industrialized countries toward reducing hospitalizations for *Rotavirus*-associated gastroenteritis (due to the successful use of the rotavirus vaccine) are promising. If the principles of therapy outlined above are accepted by all levels of the medical and public health community, and if education of parents includes beginning ORT at home, then numerous unnecessary hospitalizations and deaths could be avoided. In addition, if ORT, zinc, and vaccines were combined with several other important and well-known public health interventions (exclusive breast-feeding, hand washing, improved waste disposal, and access to clean drinking water), diarrheal deaths could be drastically reduced.[95] In an age

where governments and nongovernmental organizations are dedicated to improving health outcomes in children with AIDS, tuberculosis and malaria, the world awaits strong action against a more common but often overlooked cause of child death: diarrheal diseases.

REFERENCES

1. Liu L, Johnson HL, Cousens S, et al. Global, regional, and national causes of child mortality: An updated systematic analysis for 2010 with time trends since 2000. *Lancet*. 2012;379(9832):2151–2161.
2. Kosek M, Bern C, Guerrant RL. The global burden of diarrhoeal disease, as estimated from studies published between 1992 and 2000. *Bull World Health Organ*. 2003;81(3):197–204.
3. Bern C, Martines J, de Zoysa I, Glass R. The magnitude of the global problem of diarrhoeal disease: A ten year update. *Bull World Health Organ*. 1992;70(6):705–714.
4. Snyder JD, Merson MH. The magnitude of the global problem of acute diarrhoeal disease: A review of active surveillance data. *Bull World Health Organ*. 1982;60(4):605–613.
5. Santosham M, Keenan EM, Tulloch J, Broun D, Glass R. Oral rehydration therapy for diarrhea: An example of reverse transfer of technology. *Pediatrics*. 1997;100(5):E10.
6. Desai R, Curns AT, Steiner CA, Tate JE, Patel MM, Parashar UD. All-cause gastroenteritis and rotavirus-coded hospitalizations among US children, 2000-2009. *Clin Infect Dis*. 2012;55(4):e28–34.
7. Hoekstra JH. Acute gastroenteritis in industrialized countries: Compliance with guidelines for treatment. *J Pediatr Gastroenterol Nutr*. 2001;33 Suppl 2:S31–35.
8. Duggan C, Nurko S. "Feeding the gut": The scientific basis for continued enteral nutrition during acute diarrhea. *J Pediatr*. 1997;131(6):801–808.
9. Sandhu BK. Rationale for early feeding in childhood gastroenteritis. *J Pediatr Gastroenterol Nutr*. 2001;33 Suppl 2:S13–16.
10. Black RE, Allen LH, Bhutta ZA, et al. Maternal and child undernutrition: Global and regional exposures and health consequences. *Lancet*. 2008;371(9608):243–260.
11. Guerrant RL, Oria RB, Moore SR, Oria MO, Lima AA. Malnutrition as an enteric infectious disease with long-term effects on child development. *Nutr Rev*. 2008;66(9):487–505.
12. Latta T. Malignant cholera: Documents communicated by the Central Board of Health London, relative to the treatment of cholera by copious injection of aqueous and saline fluids into the veins. *Lancet*. 1832;2:274.
13. Phillips RA. Twenty years of cholera research. *JAMA*. 1967;202(7):610–614.
14. Nalin D, Cash R, Islam R, et al. Oral maintenance therapy for cholera in adults. *Lancet*. 1968;2(7564):370–372.
15. Hirschhorn N, Kinzie J, Sachar D, et al. Decrease in net stool output in cholera during intestinal perfusion with glucose-containing solutions. *N Engl J Med*. 1968;279:176–181.
16. Hirschhorn N, Greenough WB, 3rd. Progress in oral rehydration therapy. *Sci Am*. 1991;264(5):50–56.
17. Centers for Disease Control and Prevention. Managing acute gastroenteritis

among children: Oral rehydration, maintenance, and nutritional therapy. *Pediatrics*. 2004;114(2):507.
18. King CK, Glass R, Bresee JS, Duggan C, Centers for Disease Control and Prevention. Managing acute gastroenteritis among children: Oral rehydration, maintenance, and nutritional therapy. *MMWR Recomm Rep*. 2003;52(RR-16):1–16.
19. Ozuah PO, Avner JR, Stein RE. Oral rehydration, emergency physicians, and practice parameters: A national survey. *Pediatrics*. 2002;109(2):259–261.
20. Acra SA, Ghishan GK. Electrolyte fluxes in the gut and oral rehydration solutions. *Pediatr Clin North Am*. 1996;43(2):433–449.
21. Pierce N, Banwell J, Mitra R, et al. Effect of intragastric glucose- electrolyte infusion upon water and electrolyte balance in Asiatic cholera. *Gastroenterology*. 1968;55:333–342.
22. Field M, Rao MC, Chang EB. Intestinal electrolyte transport and diarrheal disease (1). *N Engl J Med*. 1989;321(12):800–806.
23. Huang H, Schroeder F, Zeng C, Estes MK, Schoer JK, Ball JM. Membrane interactions of a novel viral enterotoxin: Rotavirus nonstructural glycoprotein NSP4. *Biochemistry*. 2001;40(13):4169–4180.
24. Curran P. NaCl and water transport by rat ileum in vitro. *J Gen Physiol*. 1960;43:1137–1148.
25. King JC. The risk of maternal nutritional depletion and poor outcomes increases in early or closely spaced pregnancies. *J Nutr*. 2003;133(5 Suppl 2):1732S–1736S.
26. Hahn S, Kim Y, Garner P. Reduced osmolarity oral rehydration solution for treating dehydration due to diarrhoea in children: Systematic review. *BMJ*. 2001;323(7304):81–85.
27. World Health Organization. *Oral rehydration salts (ORS): A new reduced osmolarity formulation*. Geneva: WHO; 2002.
28. Duggan C, Lasche J, McCarty M, et al. Oral rehydration solution for acute diarrhea prevents subsequent unscheduled follow-up visits. *Pediatrics*. 1999;104:e29.
29. Mull JD, Mull DS. Mothers' concepts of childhood diarrhea in rural Pakistan: What ORT program planners should know. *Soc Sci Med*. 1988;27(1):53–67.
30. World Health Organization. *The treatment of diarrhoea: A manual for physicians and other senior health workers*. Geneva: WHO; 1995. Report No.: WHO/CDR/95.3.
31. Gorelick M, Shaw K, Murphy K. Validity and reliability of clinical signs in the diagnosis of dehydration in children. *Pediatrics*. 1997;99(5):E6. http://www.pediatrics.org/cgi/content/full/99/5/e6.
32. Steiner MJ, DeWalt DA, Byerley JS. Is this child dehydrated? *JAMA*. 2004;291(22):2746–2754.
33. MacKenzie A, Barnes G, Shann F. Clinical signs of dehydration in children. *Lancet*. 1989;2:605–607.
34. World Health Organization. Department of Child and Adolescent Health and Development. *The Treatment of Diarrhoea: A Manual for Physicians and Other Senior Health Workers*. 4th rev. ed. Geneva: World Health Organization; 2005.
35. Guarino A, Albano F, Ashkenazi S, et al. European Society for Paediatric Gastroenterology, Hepatology, and Nutrition/European Society for Paediatric Infectious Diseases evidence-based guidelines for the management of acute gastroenteritis in children in Europe. *J Pediatr Gastroenterol Nutr*. 2008;46 Suppl 2:S81–122.
36. Nager AL, Wang VJ. Comparison of nasogastric and intravenous methods of

rehydration in pediatric patients with acute dehydration. *Pediatrics*. 2002;109(4):566–572.
37. Freedman SB, Keating LE, Rumatir M, Schuh S. Health care provider and caregiver preferences regarding nasogastric and intravenous rehydration. *Pediatrics*. 2012;130(6):e1504–e1511.
38. Jackson J, Bolte RG. Risks of intravenous administration of hypotonic fluids for pediatric patients in ED and prehospital settings: Let's remove the handle from the pump. *Am J Emerg Med*. 2000;18(3):269–270.
39. Brown K, Peerson J, Fontaine O. Use of nonhuman milks in the dietary management of young children with acute diarrhea: A meta-analysis of clinical trials. *Pediatrics*. 1994;93:17–27.
40. Brown K, Perez F, Peerson J, et al. Effect of dietary fiber (soy polysaccharide) on the severity, duration and nutritional outcome of acute, watery diarrhea in children. *Pediatrics*. 1993;92:241–247.
41. Isolauri E, Juntunen M, Wiren S, Vuorinen P, Koivula T. Intestinal permeability changes in acute gastroenteritis: Effects of clinical factors and nutritional management. *J Pediatr Gastroenterol Nutr*. 1989;8:466–473.
42. Rabbani GH, Teka T, Zaman B, Majid N, Khatun M, Fuchs GJ. Clinical studies in persistent diarrhea: Dietary management with green banana or pectin in Bangladeshi children. *Gastroenterology*. 2001;121(3):554–560.
43. Checkley W, Gilman R, Black R, et al. Effects of nutritional status on diarrhea in Peruvian children. *J Pediatr*. 2002;140(2):210–218.
44. Sack DA, Islam S, Brown KH, et al. Oral therapy in children with cholera: A comparison of sucrose and glucose electrolyte solutions. *J Pediatr*. 1980;96(1):20–25.
45. Salazar-Lindo E, Sack RB, Chea-Woo E, et al. Bicarbonate versus citrate in oral rehydration therapy in infants with watery diarrhea: A controlled clinical trial. *J Pediatr*. 1986;108(1):55–60.
46. Santosham M, Daum RS, Dillman L, et al. Oral rehydration therapy of infantile diarrhea: A controlled study of well-nourished children hospitalized in the United States and Panama. *N Engl J Med*. 1982;306(18):1070–1076.
47. Zaidi AK, Macone A, Goldmann AD. Impact of simple screening criteria on utilization of low-yield bacterial stool cultures in a Children's Hospital. *Pediatrics*. 1999;103(6 Pt 1):1189–1192.
48. Wong CS, Jelacic S, Habeeb RL, Watkins SL, Tarr PI. The risk of the hemolytic-uremic syndrome after antibiotic treatment of *Escherichia coli* O157:H7 infections. *N Engl J Med*. 2000;342(26):1930–1936.
49. American Academy of Pediatrics. Salmonella. In: Pickering L, editor. *2000 Red Book: Report of the Committee on Infectious Diseases*. 25th ed. Elk Grove Village, IL: American Academy of Pediatrics; 2000. pp. 501–506.
50. Guerrant RL, Van Gilder T, Steiner TS, et al. Practice guidelines for the management of infectious diarrhea. *Clin Infect Dis*. 2001;32(3):331–351.
51. Bhutta TI, Tahir KI. Loperamide poisoning in children. *Lancet*. 1990;335(8685):363.
52. Gorbach SL. Bismuth therapy in gastrointestinal diseases. *Gastroenterology*. 1990;99(3):863–875.
53. Salazar-Lindo E, Santisteban-Ponce J, Chea-Woo E, Gutierrez M. Racecadotril in the treatment of acute watery diarrhea in children. *N Engl J Med*. 2000;343(7):463–467.
54. Fedorowicz Z, Jagannath VA, Carter B. Antiemetics for reducing vomiting related to acute gastroenteritis in children and adolescents. *Cochrane Database Syst Rev*.

2011;(9):CD005506.
55. Freedman SB, Adler M, Seshadri R, Powell EC. Oral ondansetron for gastroenteritis in a pediatric emergency department. *N Engl J Med*. 2006;354(16):1698–1705.
56. Duggan C, Gannon J, Walker WA. Protective nutrients and functional foods for the gastrointestinal tract. *Am J Clin Nutr*. 2002;75(5):789–808.
57. Lu L, Walker WA. Pathologic and physiologic interactions of bacteria with the gastrointestinal epithelium. *Am J Clin Nutr*. 2001;73(6):1124S–1130S.
58. Walker WA. Role of nutrients and bacterial colonization in the development of intestinal host defense. *J Pediatr Gastroenterol Nutr*. 2000;30(Suppl 2):S2–7.
59. Johnston BC, Goldenberg JZ, Vandvik PO, Sun X, Guyatt GH. Probiotics for the prevention of pediatric antibiotic-associated diarrhea. *Cochrane Database Syst Rev*. 2011;(11):CD004827.
60. Szajewska H, Mrukowicz JZ. Probiotics in the treatment and prevention of acute infectious diarrhea in infants and children: A systematic review of published randomized, double-blind, placebo-controlled trials. *J Pediatr Gastroenterol Nutr*. 2001;33 Suppl 2: S17–25.
61. Sazawal S, Hiremath G, Dhingra U, Malik P, Deb S, Black RE. Efficacy of probiotics in prevention of acute diarrhoea: A meta-analysis of masked, randomised, placebo-controlled trials. *Lancet Infect Dis*. 2006;6(6):374–382.
62. Snydman DR. The safety of probiotics. *Clin Infect Dis*. 2008;46 Suppl 2:S104–111; discussion S44–51.
63. Roberfroid MB. Prebiotics: Preferential substrates for specific germs? *Am J Clin Nutr*. 2001;73(2 Suppl):406S–409S.
64. Roberfroid MB, Delzenne NM. Dietary fructans. *Annu Rev Nutr*. 1998;18:117–143.
65. Duggan C, Penny ME, Hibberd P, et al. Oligofructose-supplemented infant cereal: 2 randomized, blinded, community-based trials in Peruvian infants. *Am J Clin Nutr*. 2003;77(4):937–942.
66. Castillo-Duran C, Vial P, Uauy R. Trace mineral balance during acute diarrhea in infants. *J Pediatr*. 1988;113:452–457.
67. Castillo-Duran C, Vial P, Uauy R. Oral copper supplementation: Effect on copper and zinc balance during acute gastroenteritis in infants. *Am J Clin Nutr*. 1990;51(6):1088–1092.
68. Santosham M, Chandran A, Fitzwater S, Fischer-Walker C, Baqui AH, Black R. Progress and barriers for the control of diarrhoeal disease. *Lancet*. 2010;376(9734):63–67.
69. Lazzerini M, Ronfani L. Oral zinc for treating diarrhoea in children. *Cochrane Database Syst Rev*. 2012;6:CD005436.
70. WHO/UNICEF. *WHO/UNICEF Joint Statement: Clinical Management of Acute Diarrhoea*. New York, NY, and Geneva, Switzerland: World Health Organization/The United Nations Childrens Fund; 2005.
71. Heinig MJ, Brown KH, Lonnerdal B, Dewey KG. Zinc supplementation does not affect growth, morbidity, or motor development of US term breastfed infants at 4-10 mo of age. *Am J Clin Nutr*. 2006;84(3):594–601.
72. Patro B, Szymanski H, Szajewska H. Oral zinc for the treatment of acute gastroenteritis in Polish children: A randomized, double-blind, placebo-controlled trial. *J Pediatr*. 2010;157(6):984–988 e1.
73. Zinc Investigators' Collaborative Group, Bhutta ZA, Black RE, et al. Prevention of diarrhea and pneumonia by zinc supplementation in children in developing countries: Pooled analysis of randomized controlled trials. *J Pediatr*.

1999;135(6):689–697.
74. Fontaine O, Gore SM, Pierce NF. Rice-based oral rehydration solution for treating diarrhoea. *Cochrane Database Syst Rev*. 2000;(2):CD001264.
75. Alam NH, Majumder RN, Fuchs GJ, CHOICE Study group. Efficacy and safety of oral rehydration solution with reduced osmolarity in adults with cholera: A randomised double-blind clinical trial. *Lancet*. 1999;354(9175):296–269.
76. Alam NH, Yunus M, Faruque ASG, et al. Symptomatic hyponatremia during treatment of dehydrating diarrheal disease with reduced osmolarity oral rehydration solution. *JAMA*. 2006;296(5):567–573.
77. Bhutta ZA, Nelson EA, Lee WS, et al. Recent advances and evidence gaps in persistent diarrhea. *J Pediatr Gastroenterol Nutr*. 2008;47(2):260–265.
78. Anonymous. Evaluation of an algorithm for the treatment of persistent diarrhoea: A multicentre study. International Working Group on Persistent Diarrhoea. *Bull World Health Organ*. 1996;74(5):479–489.
79. Vernacchio L, Vezina RM, Mitchell AA, Lesko SM, Plaut AG, Acheson DW. Characteristics of persistent diarrhea in a community-based cohort of young US children. *J Pediatr Gastroenterol Nutr*. 2006;43(1):52–58.
80. Bhutta ZA, Ghishan F, Lindley K, Memon IA, Mittal S, Rhoads JM. Persistent and chronic diarrhea and malabsorption: Working Group report of the second World Congress of Pediatric Gastroenterology, Hepatology, and Nutrition. *J Pediatr Gastroenterol Nutr*. 2004;39 Suppl 2:S711–716.
81. Fauveau V, Henry FJ, Briend A, Yunus M, Chakraborty J. Persistent diarrhea as a cause of childhood mortality in rural Bangladesh. *Acta Paediatr Suppl*. 1992;381:12–14.
82. Talbert A, Thuo N, Karisa J, et al. Diarrhoea complicating severe acute malnutrition in Kenyan children: A prospective descriptive study of risk factors and outcome. *PLoS One*. 2012;7(6):e38321.
83. Mitra AK, Albert MJ, Alam AN. Bacteraemia and meningitis among hospital patients with diarrhoea. *Trans R Soc Trop Med Hyg*. 1993;87(5):560–563.
84. CHOICE Study Group. Multicenter, randomized, double-blind clinical trial to evaluate the efficacy and safety of a reduced osmolarity oral rehydration salts solution in children with acute watery diarrhea. *Pediatrics*. 2001;107(4):613–618.
85. Silva AC, Santos-Neto MS, Soares AM, Fonteles MC, Guerrant RL, Lima AA. Efficacy of a glutamine-based oral rehydration solution on the electrolyte and water absorption in a rabbit model of secretory diarrhea induced by cholera toxin. *J Pediatr Gastroenterol Nutr*. 1998;26(5):513–519.
86. Duggan C. Glutamine-based oral rehydration solutions: The magic bullet revisited? *J Pediatr Gastroenterol Nutr*. 1998;26(5):533–535.
87. Bhan MK, Mahalanabis D, Fontaine O, Pierce NF. Clinical trials of improved oral rehydration salt formulations: A review. *Bull World Health Organ*. 1994;72(6):945–955.
88. Ramakrishna BS, Venkataraman S, Srinivasan P, Dash P, Young GP, Binder HJ. Amylase-resistant starch plus oral rehydration solution for cholera. *N Engl J Med*. 2000;342(5):308–313.
89. Alam NH, Meier R, Schneider H, et al. Partially hydrolyzed guar gum-supplemented oral rehydration solution in the treatment of acute diarrhea in children. *J Pediatr Gastroenterol Nutr*. 2000;31(5):503–507.
90. Raghupathy P, Ramakrishna BS, Oommen SP, et al. Amylase-resistant starch as adjunct to oral rehydration therapy in children with diarrhea. *J Pediatr Gastroenterol Nutr*. 2006;42(4):362–368.
91. Ramakrishna BS, Subramanian V, Mohan V, et al. A randomized controlled trial

of glucose versus amylase resistant starch hypo-osmolar oral rehydration solution for adult acute dehydrating diarrhea. *PLoS One*. 2008;3(2):e1587.
92. Anidi I, Bazargan M, James FW. Knowledge and management of diarrhea among underserved minority parents/caregivers. *Ambul Pediatr*. 2002;2(3):201–206.
93. Szajewska H, Hoekstra JH, Sandhu B. Management of acute gastroenteritis in Europe and the impact of the new recommendations: A multicenter study. The Working Group on acute Diarrhoea of the European Society for Paediatric Gastroenterology, Hepatology, and Nutrition. *J Pediatr Gastroenterol Nutr*. 2000;30(5):522–527.
94. Crocco AG, Villasis-Keever M, Jadad AR. Two wrongs don't make a right: Harm aggravated by inaccurate information on the Internet. *Pediatrics*. 2002;109(3):522–523.
95. Fischer Walker CL, Friberg IK, Binkin N, et al. Scaling up diarrhea prevention and treatment interventions: A Lives Saved Tool analysis. *PLoS Med*. 2011;8(3):e1000428.

61 Intestinal Failure, Short Bowel Syndrome, and Intestinal Transplantation

Daniel Kamin, MD

Care for infants and children with intestinal failure (IF) challenges nutritional medicine fundamentally: it attempts to provide nutrition that supports normal growth and development, even while the very organ fulfilling this role has failed. Before the advent of intravenous nutrition, patients with IF often succumbed to malnutrition and dehydration. Patients are now able to grow and develop normally on long-term parenteral nutrition (PN), and methods exist to transition patients from parenteral to enteral nutrition. For those suffering severe complications, novel medical and surgical therapies increase hope for successful transition to enteral feeding. Outcomes after intestinal transplantation have substantially improved since its inception so that motivation for choosing this therapy is moving beyond a “last ditch” effort to rescue very ill children. With these points in mind, we review the etiology, complications, and medical management of pediatric IF including intestinal transplantation; elaborate on the nutritional physiologic principles motivating this management; and highlight nutritional outcomes of various management strategies.

DEFINITIONS AND DISEASES

Definitions

The term “intestinal failure” is now often used to describe a state wherein gastrointestinal function is insufficient to support normal growth in children and physiologic balance in both adults and children. Gastrointestinal function refers to the ability to absorb enteral nutrients and fluid. Although this definition is broadest, its consistent application relies on uniformity in defining normal growth or physiologic balance. Some have suggested that IF should be defined by the extent of energy loss in stool rather than the inability to provide enteral energy. Another practical approach is to identify IF when there is ongoing need (some have argued >90 days) for PN or intravenous fluids to maintain normal growth in children and physiologic balance in children and adults. For patients with short bowel syndrome (SBS), length or percentage of small bowel remaining might also figure into a definition.

Laboratory assessment has promise for helping to define IF. For instance, serum citrulline level has been proposed as a marker for functional small intestinal mass. It is a nonstructural amino acid net produced only in presumably functionally capable enterocytes.[1] Clinical studies in enteropathy and SBS support its use as a valid marker of functional enterocyte mass and absorptive function. Retrospective studies have suggested that serum levels[2,3] of 15 μmol/L or 19 μmol/L have reasonably good discriminating value for differentiating between SBS patients who will or will not become PN independent. Prospective and serial evaluation of citrulline in neonates[4] found that citrulline level distinguished necrotizing enterocolitis (NEC) (<17 μmol/L) from age-matched healthy infants and that over time rising citrulline concentration correlated with increasing amounts of enteral nutrition. Citrulline has not enjoyed uniform success[5] as a proxy for intestinal adaptation or function.

A consensus report settled on the following: “IF results from obstruction, dysmotility, surgical resection, congenital defect, or disease-associated loss of absorption and is characterized by the inability to maintain protein-energy, fluid, electrolyte, and micronutrient balance [without intravenous support].” For children, an additional criterion would be the inability to preserve normal growth.[6]

Underlying Diseases

Most cases of IF (Table 61-1) are caused by surgical resection of the small intestine. The most common conditions are necrotizing enterocolitis,

gastroschisis, and congenital intestinal atresias. Resections may be classified according to the amount of residual small intestine, as measured distal to the ligament of Treitz along the antimesenteric border. It may be appropriate to consider the remnant small bowel as a percentage of total normal small intestinal length.[7]

Congenital diarrheal diseases lead to IF because of the severe relentless electrolyte and nutrient losses, which prevent successful enteral nutrition. These diseases result from defects in enterocyte structure and/or function, and often there is villus atrophy. Microvillus inclusion disease (MVID) is an autosomal recessive condition characterized by microvillus "inclusions" in enterocytes and colonocytes when viewed with an electron microscope. Severe watery diarrhea usually begins in the newborn period, and IV support may be required from the outset. The disease is clearly inherited, as multiple siblings may be affected and ethnic clustering exists among individuals of Navajo decent. A disease-causing gene has been found (*MYO5B*), which encodes a cytoplasmic protein critical for the apical localization of membrane components.[8] The prognosis is generally unfavorable without transplantation, but case reports have described relative stability by halting progression of liver diseases and careful attention to hydration.

TABLE 61-1 Causes of Intestinal Failure in Children

Category	Disorder	
SBS	Infant	Older Child
	Gastroschisis Necrotizing enterocolitis Small intestinal atresia Malrotation with volvulus	Crohn's disease Mesenteric infarction Radiation enteritis Tumors Trauma
Motility	Hirschsprung's disease Pseudo-obstruction Neuronal intestinal dysplasia	
Mucosa	Primary Epithelial	Immune Mediated
	Microvillus inclusion disease Tufting enteropathy	Autoimmune enteropathy Immunodeficiency syndromes
Miscellaneous	Enteroendocrine cell hypoplasia	

Source: Reproduced with permission from Kamin D, Duggan C. Enteral support for children with intestinal failure. In: Langnas A, Goulet O, Quigley E, Tappenden K, editors. *Intestinal Failure: Diagnosis, Management and Transplantation*. Malden, MA: Blackwell Publishing; 2007.

Tufting enteropathy (or intestinal epithelial dysplasia) is also an

autosomal recessive condition that presents in infancy with watery diarrhea and impaired growth. Focal epithelial “tufts” of closely packed enterocytes with rounded apical plasma membranes constitute the distinguishing features of this condition. Ethnic clustering is noted. In 2008, a gene defect was identified in epithelial adhesion molecule (EpCAM).[9] Prognosis is felt to be less dire than MVID[10]; indeed, reports are increasing of patients being weaned off PN,[11] including an adult affected woman who gave birth to a healthy infant.[12]

Severe motility disorders include long-segment Hirschprung’s disease (LSHD) and chronic idiopathic intestinal pseudo-obstruction (CIPO). LSHD refers to the characteristic aganglionosis usually isolated to the colon extending for various lengths into the small bowel. This condition causes functional SBS because the involved “paralyzed” intestine will not support aboral flow of intestinal contents. Additionally, the colon will always be affected and cannot be used for electrolyte and water recovery. Significant difficulty in weaning from PN may be expected with residual functional small bowel lengths less than 50 cm.[13]

CIPO is a heterogeneous mix of conditions sharing a common clinical presentation of apparent intestinal obstruction without surgical findings.[14] CIPO is often further classified according to histologic and manometric criteria. Myopathic CIPO refers to the primary defect localized in the smooth muscle; often, these patients have dysfunction of other smooth muscle, most commonly in the genitourinary tract (e.g., megacystis-microcolon-intestinal hypoperistalsis syndrome). This form of CIPO is generally thought to be severer, resulting in more debilitating symptoms because dysmotility is more extensive or more complete. Neuropathic CIPO indicates that the enteric nervous system is disordered, resulting in motor responses that poorly propel intestinal contents forward. In both conditions, presentation is usually in the first five years of life; patients often undergo multiple laparotomies before diagnosis; and an attempt to establish intestinal function by avoiding diseased intestine usually results in gastrostomies, jejunostomies, and diverting ileostomies. Nevertheless, the most severely affected individuals will still not be able to tolerate sufficient enteral nutrition to support growth, and long-term PN is required.

Epidemiology

IF is a relatively rare condition. Unlike rare cancers, bona fide prospective registries do not yet exist in North America to track patient numbers and

outcomes. A standard proxy is the use of home PN. Using Medicare and Oley Foundation information, Howard et al. estimated that 40,000 people were using home PN in 1992.[15] Extrapolation based on the 1994 Oley Foundation report demonstrates that roughly 40% of patients used home PN because of a primary gastrointestinal cause of IF. Children 14 years and younger accounted for approximately 25% of this group. Based on the aforementioned 40,000 estimate, approximately 4,000 US children aged 14 years and younger were on home PN because of primary gastrointestinal IF in 1992.[15] A more recent multicenter European study estimated the prevalence of pediatric home PN use at 2–6.8 children per million population.[16] Finally, a 2005 Italian survey that recorded all patients receiving artificial home nutrition reported 1.2 children/million inhabitants as using home PN.[17]

It is difficult to estimate the prevalence of individual causes of IF. Very limited published data exist for pediatrics. Certainly, SBS constitutes the majority diagnosis, and all are relatively rare. Table 61-2 displays the causes and proportions of pediatric IF from a recent multi-institutional report.[18]

TABLE 61-2 Distribution of Causes of Pediatric Intestinal Failure

Total (%)	**272**
Necrotizing enterocolitis	71 (26)
Intestinal atresia	27 (10)
Gastroschisis	44 (16)
Malrotation/volvulus	24 (9)
LSHD	11 (4)
Congenital enteropathy	3 (1)
Other single diagnoses	14 (5)
Unknown	1
Multiple single diagnoses[a]	77 (28)

[a]Includes combinations such as gastroschisis and atresia, or necrotizing enterocolitis and atresia.

Source: Adapted with permission from Squires RH, Duggan C, Teitelbaum DH, et al. Natural history of pediatric intestinal failure: initial report from the Pediatric Intestinal Failure Consortium. *J Pediatr*. 2012;161:723-8e2.[18]

PHYSIOLOGY AND PATHOPHYSIOLOGY

Normal Intestinal Growth

In utero intestinal growth is most marked during the last trimester, nearly doubling over this period of time.[19] This has obvious implications for prognosis, as acquisition of IF in early infancy or after premature birth (e.g., necrotizing enterocolitis) may lead to improved outcomes because of the growth potential of residual small intestine.

Historically, average small intestinal length (from the ligament of Treitz to the ileocecal valve) for full-term infants was estimated to be 250−300 cm,[19,20] with maximal increase in length occurring in the first year of life.[20] Normal adult small intestinal lengths ranged from[21] Generous spans are the results of variable contexts (e.g. radiologic, intra-operative, post-mortem) and/or methods used to determine intestinal length.

Recently, prospective evaluation of bowel length during laparotomy in infants and young children without gastrointestinal surgical disease identified height/length as the best predictor of extant bowel length (R^2 = 0.852; Figure 61-1), and even proposed a formula to estimate bowel length in children: ln(SBL) = 6.741 – 80.409/height.[22]

Adaptation

Intestinal adaptation refers to the gross anatomic, histologic, and functional changes that occur in the intestine that maximize digestive and absorptive capacity in the context of loss of intestinal function from either resection or disease. The phenomenon has been recognized for over a century.[23] Experimental work in dogs and clinical experience led Flint[24] to conclude in 1912 that as much as 50% of the small intestine might be removed without inexorable weight loss and death and that neither colon nor stomach could compensate for the loss. Today, while this estimate can be pushed further, the principles and observations derived from experiments conducted years ago are still apt.[21]

Adaptive changes in humans may begin 12–24 hours after massive intestinal resection and will continue for a year or longer after resection. It had once been thought that maximal adaptation occurs after 1–3 years in children and 1–4 years in adults. However, medical, surgical and nutritional innovations have extended these ranges.[18]

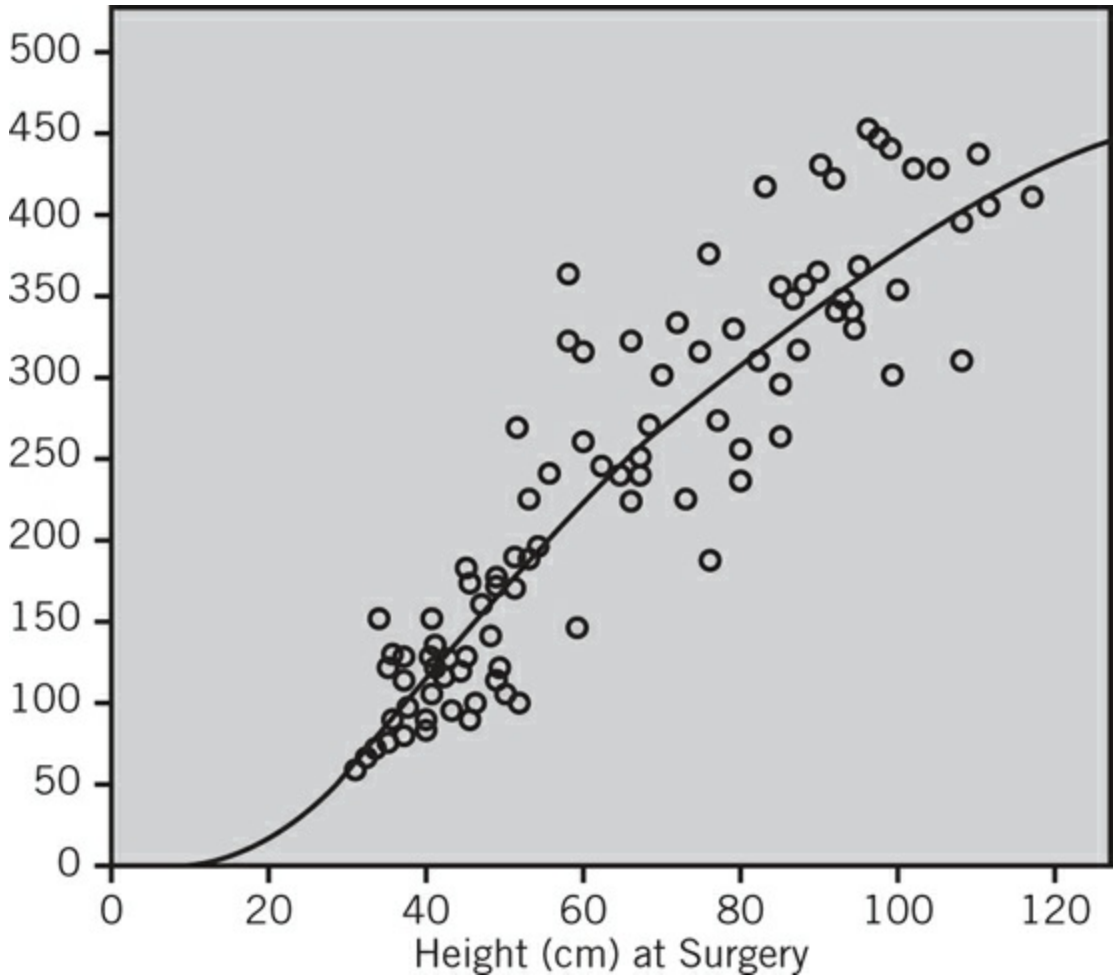


Figure 61-1 Small-intestine length versus height in 108 children without intestinal disease evaluated at laparotomy, $R^2 = 0.852$. Reproduced with permission from Struijs et al.[22]

Structural Adaptation

Laboratory data. Rodent data indicate that structural adaptation can be marked. Gross morphologic changes occur after extensive small intestinal resections (>70%) such as thickening, increase in length, and dilation.[30] Adult female Sprague-Dawley rats were examined one week after 80% small bowel resections. The wet weight of the small intestine increased fivefold, up to 70% of the preresection weight. However, there were no further increases in intestinal mass when assessed 5 and 10 weeks post resection, and there was no recovery of the preresection intestinal length (Figure 61-2). In the juvenile domestic pig, 16 weeks after surgery bowel diameter and length increased proportionally more in those that received

75% mid-small intestinal resections compared to animals who received transections.[26] Discrepancies between studies showing recovery of intestinal length may depend on species and timing. It is not clear whether intestinal length can be recovered only within species-specific "window periods."

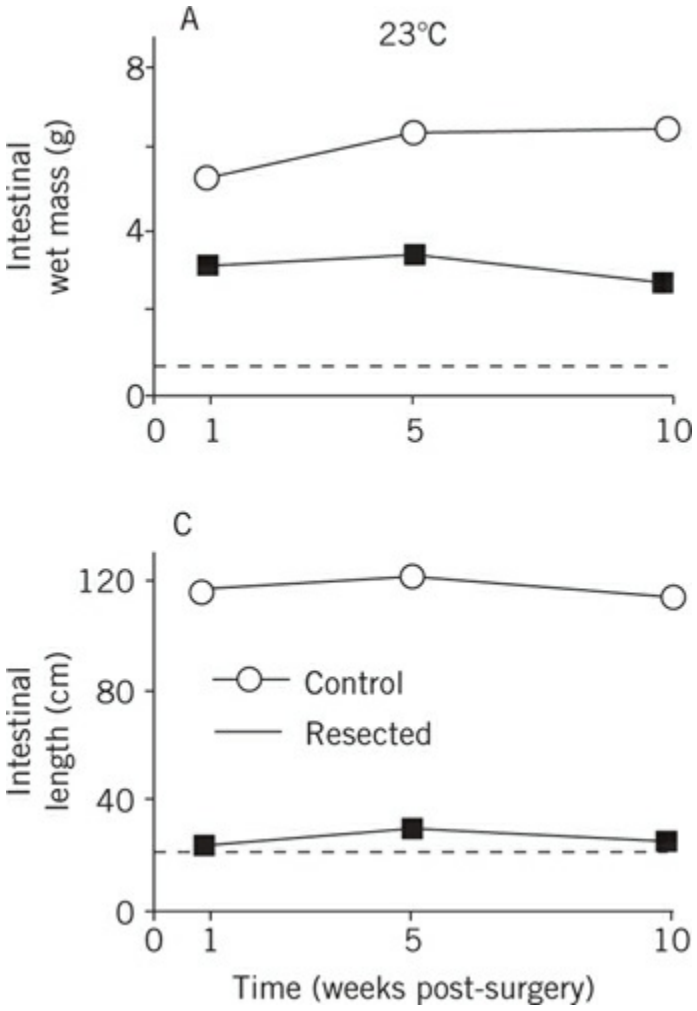


Figure 61-2 Effect of resection on wet mass (upper graph) and intestinal length (lower graph) in female Sprague-Dawley rats. Dotted line represents estimated wet mass or length immediately after resection. In this figure, 80% small intestinal resections were performed (dark squares), whereas control animals (open circles) had sham operations. (Reproduced with permission from O'Connor.[41])

Adapted rodent intestinal mucosa is characterized by elongated crypts and wider, taller villi.[27] There is increased cellularity, which is manifest by

increased DNA and protein content in adapted intestine.[28] These processes are more marked when resection is more extensive,[29] and when the residual intestine is ileum.[30,31]

Mucosal responses to adaptation likely differ between species. For instance, resection often causes crypt-villus elongation in rodents, while similar structural changes have not been routinely noted in humans.[32] The neonatal piglet model is a reasonable alternative, and has been used to show efficacy of surgical and medical therapies for intestinal failure.[33]

Human data. There is adequate documentation that human intestine can adapt to improve nutrient absorption following injury or resection.[24,34] The macroscopic changes of intestinal hypertrophy and lengthening have been observed in many case series,[32] but the crypt-villus architectural modifications observed in rodents have not been recapitulated.[34,35] For example, duodenal mucosa from an adult patient with significant SBS demonstrated hyperplasia of crypts and villi compared to specimens from normal adult humans, but cell proliferation, crypt depth, and villus height were equivalent.[36] However, a small case series of patients who received segmental small intestinal transplants found that villus length did increase by 50% over baseline at six months, suggesting that intestinal adaptation in the human may include morphologic changes in the villus that maximize surface area.[37]

Functional Adaptation

Animal data: Functional adaptation is characterized by an increase in the absorption of carbohydrate, protein, water, or electrolytes per unit length of intestine. Within six hours of intestinal resection, upregulation of the sodium-glucose cotransporter occurs in enterocytes, leading to enhanced electrolyte and water absorption.[38] Physiologic mechanisms underlying these changes may be upregulation of total available transporters, a larger number of functional enterocytes, or upregulation of efficiency of individual nutrient transporters.[39] Work using a rabbit model of SBS that examined nutrient transport across jejunal sections isolated from animals with and without significant distal small bowel resections indicates that changes in the lipid content of the brush border membrane may account for enhanced passive absorption of free fatty acids in small intestine.[40] Dowling and Booth[30] in their classic paper on rat SBS found that per unit length of remaining intestine glucose absorption was increased two weeks later and that this effect was more pronounced in ileum than jejunum.

It is worth noting that structural changes may not strictly correlate with

functional adaptations. For instance, total mucosal glucose absorption reached 33% of the pre-resection value after 80% small bowel resection in the Sprague-Dawley rat, while mucosal weight increased to an average two-thirds that of the pre-resection weight.[41]

Delayed transit through the GI tract, affording luminal nutrients more contact time with the intestinal absorptive surface, may be considered an adaptive mechanism after small intestinal resection. Few studies have documented this well,[42] while others have found transit time normal or shortened.[43] Moreover, the lack of correlation between small intestinal transit time and residual bowel length complicates generation of simple theories.[43] Transit time may even be delayed because of bowel dilation and stasis, both concerns that could contribute to maladaptive physiology.

Human data: Segmental intestinal transplant from living human donors provides some insight on the adaptive process. When 200 cm of ileum was transplanted into a human leukocyte antigen-matched sibling who suffered traumatic loss of his entire jejunum, ileum, and right colon, *d*-xylose and fat balance studies were markedly abnormal one month after transplant but were considered normal at six months.[44] Ziegler et al.[45] compared small intestinal biopsies from 13 adult SBS patients on long-term PN to those obtained from 33 adult control subjects undergoing endoscopy without histories of intestinal resection, inflammation, or infection. All 13 SBS patients were also consuming oral nutrition. Small intestinal villus height, width, and crypt depth were no different between the groups, suggesting that intestinal adaptation in humans does not depend on sheer size of the crypt-villus structure. Interestingly, Ziegler did identify upregulated expression of the peptide transporter PepT1 in colonic mucosa of SBS patients, a finding that colonic salvage not just of amino acids as well as carbohydrates (see later) in patients with small intestinal malabsorptionmay be an important component of adaptation.

Mineral absorption may adapt over an extended period of time. Gouttebel et al. investigated calcium absorption in 30 adults with SBS, comparing those whose resections were less than two years ago to those greater than two years. While *d*-xylose absorption was equivalent, calcium absorption continued to increase in the latter group, suggesting that adaptation may be a prolonged, slowly evolving process.[46]

Little information exists that characterizes intestinal adaptation with respect to motility in humans. Moreover, much of the phenotype of intestinal adaptation in humans has not been fully described (e.g., bowel flora and immunologic function). Of note, a recent paper[47] identified a

particular D/L stool lactate ratio in patients with SBS, suggesting that patients may harbor a particular microbiota that could be adaptive or maladaptive. In summary, in humans structural adaptations are less obvious than functional; adaptation may take months and possibly years; and the colon can be adapted to absorb not just fluid and electrolytes but also calories, possibly even beyond short-chain fatty acids (SCFAs).

Molecular Adaptation

Adaptation can be characterized on the molecular level.[48–50] For instance, cDNA isolated from residual ileum 48 hours after 70% resection in rats suggested significant upregulation (compared to sham-operated rats) of genes encoding proteins responsible for nutrient trafficking, cell cycle control, and protein processing.[50] Further, Stern et al.[49] used exogenous epidermal growth factor (EGF) in a similar experiment and detected upregulation of a gene product (proline-rich protein 2) not previously thought to play a role in intestinal adaptation. Current and future studies using microarray analysis and other sophisticated molecular techniques will soon offer a much more complete picture of adaptation, and possibly lead to more rational identification of therapies to promote adaptation.

Determinants of Adaptation

By far, the most powerful and consistent determinant of adaptation is provision of enteral nutrition. Directly and/or indirectly, nutrient flow through the intestinal tract promotes structural and functional integrity of the intestine; the amount of enteral nutrition that is tolerated is a robust predictor of successful adaptation and thus freedom from parenteral therapies.

Earlier Observations

There has been great interest in isolating the components linking provision of enteral nutrition with enhanced intestinal function. Observations included the effects of direct nutrient contact with the mucosa, pancreatic and hepatobiliary secretions, and circulating hormones. Ileum that is transposed into a proximal position in jejunum will develop larger villi and crypts,[51] suggesting that direct mucosal contact with richer chyme is important for maintaining mucosal structure. When the ampulla of Vater is surgically transposed to distal small intestine, mucosal hyperplasia will occur distally, highlighting the possible importance of hepatobiliary and pancreatic secretions in adaptation.[52] Finally, animal studies support the

notion that humoral factors are released secondary to enteral feeding that maintain structure and function of intestinal mucosa. Feeding an animal with a Thiry-Vella fistula, in which a portion of small intestine is removed completely from the luminal circuit while maintaining physiologic blood supply, will produce mucosal hyperplasia in the fistula when nutrients are administered enterally but not parenterally.[53] Parabiotic animal studies are also revealing. Here, two animals have blood supplies that are surgically cross-circulated, while only one animal has an intestinal resection. Both animals will then develop mucosal hyperplasia of small intestine when the resected animal is fed enterally.[54]

Nutritive Factors

Lipids: The amount, nature, and route of administration of lipids may influence the adaptive process. Studies in rats imply that both saturated and polyunsaturated fatty acids (PUFAs) have trophic effects on small intestine. Maxton[55] gave female Wistar rats four weeks of Vivonex or Vivonex with 50% of calories replaced with either saturated or essential fatty acid-substituted triglycerides and found that compared to Vivonex small bowel mucosal weight, protein, and DNA content were all higher in the supplemented groups. Alternatively, using a rat model Vanderhoof et al.[56] found that menhaden oil (a marine source of ω-3 PUFAs), compared to safflower oil, stimulated greater small bowel mucosal regeneration during recovery from mucositis induced with subcutaneous methotrexate.

Admittedly, nonresection models may have limited applicability for understanding adaptation after intestinal resection. Manipulation of dietary fat has been explored in rodents with experimental SBS. Kollman[57] investigated the effect of various combinations of docosahexaenoic acid (DHA) and arachidonic acid (AA) (major constituents of marine oil) in the diets of small bowel-resected Sprague-Dawley rats. Rats received 80% small bowel resections, leaving distal ileum and ICV intact. Rats were fed varying amounts of DHA and AA (from 5% AA and 3.3% DHA to 45% AA and 30% DHA) while energy from fat was kept constant at 30%. At 14 days, the ileal mucosa removed from rats receiving the highest amount of DHA and AA was significantly heavier (87.38 vs. 71.55 mg/cm) than the mucosa removed from animals who received normal diet (10% soy oil and 90% hydrogenated coconut oil). However, sucrase activity did not differ between the groups nor did mucosal characteristics from the duodenum.

Changes in nutrient transport have also been explored. A rat model of SBS suggests that a high-fat diet, compared to isocaloric regular chow, enhances fatty acid transport in the remaining small intestine. Three days

after 75% small intestinal resection, male Sprague-Dawley rats given a diet with saturated fat-enhanced chow had enhanced fat clearance (based on fat balance studies) versus rats that had received usual chow.[58] Earlier, Keelan et al.[59] resected the distal half of the small intestine in adult female Sprague-Dawley rats, leaving the ICV intact. Animals were fed regular chow for three weeks and then two additional weeks with isocaloric saturated fatty acid (SFA)- or PUFA-enriched chow. Glucose transport was twice as high across mucosa isolated from animals fed the SFA diet compared to that measured in tissue derived from animals fed the PUFA-enriched diet. Heterogeneity of positive findings, differing models (e.g., resection vs. chemical injury), and various protocols (e.g., age, sex, amount, and type of resection) limit the ability to draw firm conclusions. It is clear that the amount and type of enteral fat (most convincingly saturated long-chain triglycerides [LCTs] or fatty acids) can modify the nature and extent of adaptation.

Carbohydrates: Dietary carbohydrate absorption may be enhanced by altering the amount of administered carbohydrate (the functional "workload" hypothesis). This has been demonstrated carefully in nonresection rat models. Diamond et al.[60] isolated intestinal sleeves from mice and demonstrated in vitro that dietary carbohydrate significantly increased the V_{max} across the mucosa, suggesting carbohydrate induction of monosaccharide carriers in the intestine. Later, Shu et al.[61] found that increasing dietary fructose or sucrose induced a parallel higher expression of GLUT5 mRNA, sucrase activity, and brush border fructose transport. Moreover, enterocytes may acquire transport characteristics only while they are still in the crypt. Using a time-sequenced freeze fraction and transporter staining technique, Ferraris et al.[62] found that glucose-dependent regulation of Na^+/D-glucose cotransport (assessed by functional staining technique, which quantifies glucose transporter density) was possible only when cells were still in the crypts. Disaccharides may be more trophic on intestinal mucosa than equivalent amounts of monosaccharides, an effect abolished when disaccharide hydrolysis was inhibited.[63]

Other dietary constituents may influence the transport of carbohydrates. Using a rat model of ethanol mucosal injury, Thomson et al.[64] found that SFA-enriched diet protected the in vitro jejunal and ileal transport of glucose, whereas mucosa derived from rats fed PUFA-enriched diet demonstrated decrements in glucose transport when exposed to ethanol. Alternatively, fish oil seemed to have an opposite influence on

Na^+-dependent glucose transport in proximal small intestine membrane vesicles isolated from rats maintained for four weeks on menhaden oil-enriched (unsaturated fat) or butter fat (SFA)-enriched diets.[65] Compared to the SFA diet, Vmax for glucose was significantly higher in vesicles derived from rats fed the fish oil–enriched diet.

Taken together, these data do not clearly support a specific nutrient formulation to enhance carbohydrate absorption; enteral administration of nutrients in general, including a disaccharide or polysaccharide carbohydrate source, positively influences intestinal absorption of carbohydrates.

Protein: Similar to "reflexive" influence of macronutrients on their own absorption, dietary protein influences intestinal amino acid transport. Isocalorically fed mice on high-nitrogen diets will increase uptake by 77%–81% for nonessential amino acids, while only increasing uptake by 32%–61% for more toxic but essential amino acids.[66] Conversely, nitrogen-deficient diets lead to decreased uptake of nonessential amino acids while uptake for essential amino acids was maintained, a finding that suggests independent regulation of individual or groups of amino acids.[67]

The amino acid glutamine has received significant attention as a nutrient that enhances intestinal adaptation. Ziegler et al.[68] gave rats receiving 80% intestinal resections either low or glutamine-supplemented diets. Using intestinal DNA content as proxy for intestinal adaptation, Ziegler suggested that feeds supplemented with glutamine for seven days enhanced intestinal adaptation compared to rats receiving deficient diets. This is not a uniform finding, however,[69] which is corroborated by the mixed results seen in clinical trials of glutamine supplementation.[70,71] Some amino acids may in fact have detrimental effects on the adaptive response. Parenteral arginine administration led to lower proliferative and higher apoptotic indexes in 75% resected rats 15 days following resection.[72]

Miscellaneous: Pectin and other soluble fibers may facilitate intestinal adaptation. Animal studies document improved caloric and fluid absorption when diets are supplemented with soluble fiber.[73] A rodent SBS study found that the weight, DNA, and protein content of ileum and colonic sections were significantly higher in rats fed diets supplemented with 2% pectin compared to animals with similar small bowel resections consuming an unsupplemented, isocaloric diet.[74] Proposed mechanisms of action include increased lumen/nutrient contact time, "carbohydrate salvage" via colonic assimilation of SCFAs derived by fermentation of the

soluble fiber, and direct effects of SCFAs on tissue trophism and nutrient absorption.

Salvage occurs when anaerobic bacteria in the colon ferment unabsorbed carbohydrate, producing SCFAs. Studies of human subjects with SBS have shown that carbohydrate fermentation does occur.[75] SCFAs are used by the colon as a fuel source,[76] thus increasing the amount of calories assimilated from enteral nutrition.

SCFAs may also directly enhance intestinal tissue growth and function. SCFAs given intravenously prevented total parenteral nutrition (TPN)-induced small intestinal mucosal atrophy in rats.[77] Rodent work also suggests that SCFA supplementation augments intestinal assimilation of nutrients and electrolytes. Tappenden et al.[78] gave male Sprague-Dawley rats who underwent an 80% jejunoileal resection either isocaloric SCFA-supplemented or unsupplemented PN. Three or seven days later, residual small intestinal mucosal/submucosal weight and DNA content were significantly greater in the supplemented group compared to unsupplemented rats. Later, using a similar model this group found that ileal uptake of D-glucose was substantially higher in the SCFA-supplemented animals, whereas expressions of membrane glucose (GLUT2) and ion transporter (sodium-dependent glucose transporter 1) mRNA were also higher in ileal tissue from rats receiving SCFA-supplemented TPN.[79]

Intestinal trophic factor secretion could be augmented by SCFA supplementation. Tappenden et al.[78] found that ileal proglucagon mRNA was higher among SBS rats who received SCFA-supplemented versus unsupplemented TPN. More recently, a randomized SBS rodent trial found that compared to rodents fed fiber-free elemental diets rats given the same diet supplemented with 2.5% pectin had significantly increased lower intestinal weight and length (ileum, cecum, and colon), and serum samples from supplemented rats had significantly increased glucagon-like peptide 2 (GLP-2) concentrations.[80] Taken together, these data suggest that SCFAs enhance bowel adaptation via multiple mechanisms, and further work may help direct how these molecules can be harnessed most usefully in the clinical setting.

Polyamines are dietary constituents that play an important role in cellular growth and differentiation.[81] Polyamines are polycationic compounds present in all prokaryotic and eukaryotic cells, and they must be present in sufficient quantities to support cell division and differentiation. Rodent intestinal perfusion studies suggest that polyamines

enhance glucose absorption via heightened expression of *SGLT1*.[82] Moreover, experiments suggest that polyamines exert trophic effect on intestinal mucosa and muscle. Enteral diets supplemented with the polyamine precursor ornithine α-ketoglutarate lead to mucosal hyperplasia and heightened muscle glutamine repletion in rodents with 80% small intestinal resections.[83] Ornithine decarboxylase, an enzyme responsible for enterocyte polyamine synthesis, is upregulated in hyperplastic ileal mucosa isolated from 80% intestinal resected rodents given SCFA-supplemented TPN.[78] Trophic effects are seen even in nonresected animals fed polyamine-supplemented diets.[84]

These data indicate that polyamines may mediate some important elements of intestinal adaptation. Well-designed prospective clinical trials can determine the nature of their role in human adaptation, with particular attention to the possibility that polyamines may be "conditionally essential" and thus more or less helpful depending on age and underlying disease.

Clinical studies: Clinical data suggest that enteral nutrition maintains and/or promotes intestinal function. In 1975, Greene et al.[85] gave 16 infants with protracted diarrhea either PN for two to three weeks followed by semi-elemental oral formula (n = 10) or dilute PN and continuous enteral feeding with elemental formula (n = 6). After two to three weeks of treatment, sucrase and maltase activity in jejunal biopsies improved in both groups, but they improved to the normal range only in the group that received early enteral nutrition. Additionally, the enteral feeding group stayed in hospital for fewer days (mean 34 vs. 46 days; $P < .05$), suggesting that enteral nutrition brings about recovery more effectively and more quickly than regimens that rely largely on PN. Orenstein[86] later extended these findings by studying 13 infants with intractable diarrhea, malnutrition, and malabsorption. The severe group (defined as D-xylose < 10 mg/dL, n = 8) was randomized to receive continuous enteral nutrition using semi-elemental formula or PN for three to four weeks followed by enteral nutrition. Diarrhea resolved more quickly for those receiving enteral nutrition versus PN (2–4 weeks vs. 7–12 weeks; P = .02), and malabsorption, assessed by achieving D-xylose greater than 15 mg/dL, also resolved more quickly for those receiving enteral nutrition (2–4 weeks vs. 7–12 weeks; P = .02).

Early introduction of enteral nutrition influences adaptation. Sondheimer et al.[87] identified 44 neonates retrospectively with SBS on PN for at least three months. Independent predictors of PN duration included

percentage of enteral nutrition at 12 weeks after surgery, suggesting that enteral nutrition may influence outcomes. These findings were corroborated by Andorsky et al.,[88] who reported on 30 neonates who had received at least three months of PN because of SBS. A higher percentage of enteral calories at six weeks after surgery (breast milk or elemental enteral feedings) was associated with a shorter duration of PN. Although multivariate analysis found that only residual small intestinal length was an independent predictor of PN duration, these data suggest that attention to early introduction of enteral nutrition may positively influence adaptation.

Nonnutritive Factors

Patient- and organ-related factors: The age of the patient, amount of residual intestine, type of residual intestine, presence of a proximal ostomy, lack of colon in continuity, absence of ICV, and underlying disease in residual intestine have all been noted as organ- and patient-specific characteristics that can influence the success of intestinal adaptation. Many of the relationships were identified or suspected first by Wilmore in 1972.[89]

Most studies demonstrate that residual intestinal length robustly predicts the duration of PN, a proxy for the tenor of intestinal adaptation. Based on retrospective studies[7,87,90,91] and one prospective study,[92] residual small intestinal length less than 15–40 cm in an infant, especially with lack of colon in continuity, predicts inability to wean from PN. With patients living longer with ultra-short-segment SBS, exceptions to this rule are accumulating.

Ileum appears to adapt more readily than jejunum. In 1967, Dowling and Booth[30] performed 70% distal or proximal small intestinal resection in rats and then measured efficiency of glucose absorption over time as well as mucosal morphological characteristics in the residual small intestine. Ileal villus height increased on average by 53% in rats with residual ileum, whereas villi from residual jejunum increased only by 14%. At the same time, glucose absorption from residual ileum increased by 96% compared to baseline, but there was only an 18% increase in sugar absorption from residual jejunum. Human data support the notion that jejunum has at best modest adaptive capacity. Patients with jejunocolic anastomosis after extensive resections (>75% small bowel resection) did demonstrate mucosal hyperplasia in residual jejunum,[34,35] whereas others have reported no hyperplasia or in fact hypoplasia following jejunocolic anastomosis[93] or

end jejunostomy.[94]

Debate continues with respect to the importance of the ICV in continuity. Whether ICV promotes intestinal adaptation or prevents complications that impede adaptation is inescapably confounded by the usual absence of some or all colon when the ICV is removed. Clinical experience and some observational data[95] support the notion that colon in continuity decreases time to PN independence. In any case, most studies report[7,90,91] or suggest[92] that presence of ICV improves the likelihood of achieving PN independence. Some of these studies also suggest that colonic resections[90] or lack of continuity[92] predict prolonged PN courses. Corroboration comes from adult series,[96] which clearly indicate that patients lacking colon in continuity or ileum/ICV are at higher risk for requiring long-term PN.

Growth hormone (GH) and related factors: Intuition likely resulted in early consideration of GH as an adaptation-enhancing hormone. Strong animal data support its role in either promoting mucosal growth/function or preventing mucosal atrophy.[97] Intestinal mucosa atrophies when mice are hypophysectomized, and supplemental GH restores mucosal adaptation in these mice that have had intestinal resections.[98] Insulin-like growth factor-1 (IGF-1) signaling likely accounts for GH effect in small intestine. While not all studies show that GH administration causes mucosal growth,[99] the impact may be more on nutrient or ion transport.[100]

Mouse small intestinal resection models have found that GH and glutamine-supplemented diet together synergistically increase plasma IGF-1 levels, mucosal DNA content, and villus growth in the residual small intestine.[68,101] Thus, many clinical trials evaluating GH in SBS have included glutamine supplementation. The physiologic basis for this interaction may be provision of a substrate for ornithine carboxylase, an enzyme responsible for nucleotide production and polyamine synthesis, both important for tissues with rapidly diving cells.[102]

Clinical evaluation of GH has yielded variable results. Adult studies using moderate- to high-dose GH, glutamine supplementation (enterally or parenterally), and regulated diets have not been promising.[103] For example, Scolapio et al.[104] conducted a randomized, blinded, and placebo-controlled crossover trial testing the adaptive impact of GH (0.15 mg/kg/d), oral glutamine (0.63 g/kg/d), and high-carbohydrate diet on eight adult SBS patients (average small bowel length, 71 cm; mean duration of TPN, 12.9 years). Treatment with GH transiently increased total body water and electrolyte absorption but made no difference with

respect to multiple measures of nutrient transport or villus morphology. Hyperphagia and normal body composition could be important mediators of optimal adaptation; negative studies may be explained by a lack of consideration of these, and other, factors.[105]

Positive results with GH have been reported. In a randomized double-blind trial, 12 adult SBS patients (duration of PN, 7 ± 1 years; small bowel remnant length, 48 ± 11 cm) on TPN and unrestricted hyperphagic diets were given three weeks of low-dose (0.05 mg/kg/d) GH or placebo, followed by a crossover period of three additional weeks.[106] Compared to the control group, those receiving GH had improved lean body mass and significantly increased nutrient absorption. Absorption of protein increased by 15% ± 5% ($P < .002$), whereas enteral caloric absorption improved to 37% ± 16% of total parenteral energy delivery. Later, Byrne et al.[107] conducted a randomized placebo-controlled inpatient trial in 41 adult PN-dependent subjects with SBS (average duration of PN, 4.6 years; average length of small bowel, 73 cm) comparing glutamine-supplemented diets with and without GH. Compared to those receiving glutamine only, subjects who received GH plus glutamine-supplemented diet were able to wean from PN more substantially during the four-week treatment trial (mean reduction in PN volume, 7.7 L/wk ± 3.2 vs. 3.8 L/wk ± 2.4; $P < .001$); these effects were durable three months later but only for the group that received both GH and glutamine diet. It does not appear that any patient was able to completely wean off PN.

There is very limited published experience regarding the GH axis in pediatric IF. PN-dependent children with neonatal SBS received rhGH 0.14 mg/kg/d and glutamine for three months.[108] Results suggested a beneficial effect of rhGH by decreasing the need for PN but with mild effects on body composition and gut mucosa. In addition, there was no apparent benefit from glutamine treatment. More recently, an open-label trial involved eight PN-dependent children with neonatal SBS receiving more than 50% of their protein-energy requirements from PN.[109] They received 0.13 mg/kg/d of rhGH for 3 months and were followed for an additional 12 months. During treatment, plasma citrulline significantly increased (baseline 16.5 ± 14.8 μmol/L vs. on treatment 25.2 ± 18.3 μmol/L; $P < .05$), median enteral energy intake increased by 54% ($P < .001$), and net energy balance significantly increased. However, after 12 months of follow-up only two patients remained independent of PN. Guo et al.[110] recently reported on their retrospective experience in administering rhGH 0.05 mg/kg/d for three weeks; enteral glutamine for

two years; and a low-fat, high-carbohydrate diet in seven children with SBS and PN dependence. After a median follow-up of 15 months since treatment with rhGH, all but one patient were weaned from PN.

These data show that late exogenous treatment with GH and glutamine supplementation may promote intestinal adaptation. Safety concerns notwithstanding, any future prospective investigation will be more meaningful when differences in anatomy, age, dietary compliance, nutritional status, and mucosal disease processes are minimized or accounted for a priori.

IGF-1 physiology may have an important role in intestinal adaptation following resection. This has been established in animal studies. Small and large intestinal resections induce IGF-1 mRNA expression in residual colon and ileum. Compared to resection alone, exogenous IGF-1 produced heightened IGF-1 mRNA expression in residual ileum[68] and colon[111]; larger increases in small intestinal weight, DNA, and protein content[68]; and enhanced crypt cellular proliferation.[112] Administration of IGF-1 with supplemental glutamine likely has synergistic influences on these processes.[68] Interestingly, IGF-1 signaling from within the smooth muscle layers of the small intestine may exert some control over intestinal adaptation.[113]

Differential physiologic responses have been identified with respect to IGF-1 and intestinal resection. In rat models of SBS, residual colon[114] and ileum[68] underwent adaptive changes in response to intestinal resection, whereas jejunum had no response to resection alone; changes in colon and ileum correlated with tissue protein expression of IGF-1-binding protein 3. In contrast, exogenous IGF-1 produced adaptive changes in jejunum and ileum but not colon.[115] This has not been a consistent finding.[111] This complex physiology with respect to IGF-1 provides additional insight into the aforementioned heterogeneous GH findings.

Exogenous administration of IGF-1 may facilitate transition to enteral nutrition in animal models of SBS. Gillingham et al.[116] gave rats who received 60% jejunoileal resection plus cecectomy either IGF-1 or vehicle for 7 days while receiving PN and then gave 100% enteral nutrition for an additional 10 days. Rats who received IGF-1 had significantly higher amounts of lean body mass and intestinal mucosal hyperplasia compared to animals receiving vehicle.

Barksdale et al.[117] investigated the relationship between growth failure (defined as weight and length for age < 5%) and serum hormone profiles in 40 patients (29 boys; mean age, 5.3 years; range, 0.5–18.6 years) with

SBS (73% receiving PN). In the cohort, 38% (11 patients) with height measurements (n = 29) met criteria for growth failure, and significantly more children with growth failure had low IGF-1 compared to those without growth failure (64% vs. 17%; $P < .05$). GH levels were generally normal in all patients. Although patient numbers were small, Barksdale et al. speculated that these data serve as indirect evidence that in a select group of children with poorly adapted intestine inadequate IGF-1 may have caused growth failure even in the context of adequate enteral nutrition and/or PN. Larger, prospective studies accounting for confounders such as comorbid illness will be required to validate this potentially important claim.

EGFs: EGF is well recognized for its intestinal trophic properties.[118] Characterized molecular events that occur with EGF stimulation include redistribution of *SGLT1* from microsomal pools to the brush border membrane[119] and intestinal resection-induced movement of EGF receptors from basolateral to the brush border membrane.[120]

In vivo support for EGF's trophic properties mainly comes from animal models of SBS. Mice that receive sialoadenectomy (salivary gland is the major source for EGF in mice) and proximal small bowel resection, compared to mice with small bowel resection with sham sialoadenectomy, have significantly attenuated ileal villus height and tissue weight gain, differences abolished by exogenous administration of enteral or parenteral EGF.[121] Ileal brush border surface area and glucose transport were enhanced when EGF was administered to rabbits with proximal surgical SBS compared to those that received saline.[122] Conversely, compared to mice receiving saline mice receiving 50% small bowel resections who also received orogastric EGF receptor inhibitor demonstrated poor adaptation (reduced intestinal wet weight, crypt depth, and villus height) in residual small intestine.[123] Barrier function and carbohydrate absorption were enhanced in juvenile rats with surgical SBS administered recombinant human EGF compared to rats with similar operations administered saline.[124] Lastly, one human study[125] is consistent with the notion that EGF plays a role in human intestinal mucosal integrity. EGF levels in multiple body fluids from 25 infants with NEC were significantly lower than levels found in 19 gestational-age- and weight-matched neonates without NEC.

A pilot study has been conducted in five children with severe SBS (defined as <25% small bowel length predicted for age) using exogenous, enterally administered EGF. Compared to baseline, after six weeks of

therapy carbohydrate absorption was significantly higher, as was tolerance of enteral feedings. Although the trial was successful, soon after it ended three patients developed sepsis.[126] Other human observational studies suggest that EGF is important for the maintenance of adequate barrier[125] and reparative function. It is important to emphasize the role of oral feeding and subsequent salivary gland EGF release in promoting intestinal adaptation. Gastric tube feeding by shunting the oral step reduces the contribution of EGF to intestinal adaptation.

Glucagon-like peptide 2 (GLP-s): GLP-2 is a pro-glucagon-derived peptide produced in distal ileum or proximal colonic L-cells in response to luminal nutrient exposure, providing feedback information to the upper intestine so that fluid and nutrient absorption may be optimal.[127] Animal models of SBS strongly suggest that GLP-2 plays an important role in intestinal adaptation following resection or injury. Rats subjected to 70% jejunal-ileal resections followed by oral feeding sustained persistent elevation in plasma GLP-2 levels compared to those receiving transection alone, and measures of adaptive growth in the resected animals positively correlated with levels of circulating GLP-2 and proglucagon mRNA in residual ileum and colon.[128] Martin et al.[129] extended these findings in rats by demonstrating that the magnitude of GLP-2 elevation directly correlated with the extent of intestinal resection, mucosal enterocyte proliferation, and protein and fat malabsorption. This suggested that heightened GLP-2 release may induce intestinal trophism in response to nutrient exposure in distal small and large intestines. Mechanisms for enhanced tissue tropism include increasing local blood flow[130] and inducing local expression of growth factors and growth factor receptors.[131]

Heightened adaptation occurs when GLP-2 or analog is administered to animals with induced SBS[132,133] and served to motivate a number of excellent studies in humans. Jeppesen published the first pilot study in 2001,[134] showing that GLP-2 administered to patients with SBS could increase intestinal absorption and promote accretion of lean body mass. The commercial development of peptidase-resistant GLP-2 analog teduglutide motivated further clinical study, culminating in high-quality clinical trials[135,136] that have confirmed its effectiveness in promoting intestinal adaptation.

Jeppesen et al. performed a multicenter, prospective, and double-blind 24-week trial comparing daily injected teduglutide (0.05 mg/kg/d; $N = 43$) to placebo ($N = 43$) in adults with SBS and PN dependence. At the end of the study period (Figure 61-3), significantly ($P = .002$) more subjects

given teduglutide (63%) met the primary end point (>20% reduction in PN volume) than did subjects in the control group (30%). Further, the proportion of subjects who were able to wean one or more days of parenteral support was significantly greater in the treatment than control group (54% vs. 23%; *P* = .005). Adverse events requiring study termination were no more common in the treatment group (two patients in the treatment group and three in the control group). Prolonged therapy (to 52 weeks) has been described.[135]

The Food and Drug Administration (FDA) approved teduglutide in 2012 for use in adults with SBS and PN dependence. It has not yet been evaluated in children. Concern does remain regarding cost, loss of effect once the drug is stopped, and long-term safety if the drug is continued. Adverse effects of the drug have included the development of intestinal polyps (screening colonoscopy is recommended before the drug is started), and positive fluid balance. Best practice approaches have already been described guidelines are available.[137]

Insulin has been shown to influence intestinal structure and absorptive function. The effects of parenteral insulin (INS) on structural intestinal adaptation, cell proliferation, and apoptosis were assessed in rats divided into three experimental groups: sham operated rats underwent bowel transection and reanastomosis, SBS rats underwent a 75% small bowel resection, and SBS-INS rats underwent a 75% small bowel resection and were treated with insulin (1 U/kg, twice daily subcutaneously) from day 3 through day 14.[138] SBS-INS animals demonstrated higher jejunal and ileal bowel and mucosal weights, jejunal and ileal mucosal DNA and protein, and jejunal and ileal crypt depth compared with SBS animals. SBS-INS rats also had a greater cell proliferation index in both jejunum and ileum and a trend toward a decrease in enterocyte apoptotic index in jejunum and ileum, compared with the SBS untreated group. Oral insulin may also promote adaptive changes in rats with SBS.[139]

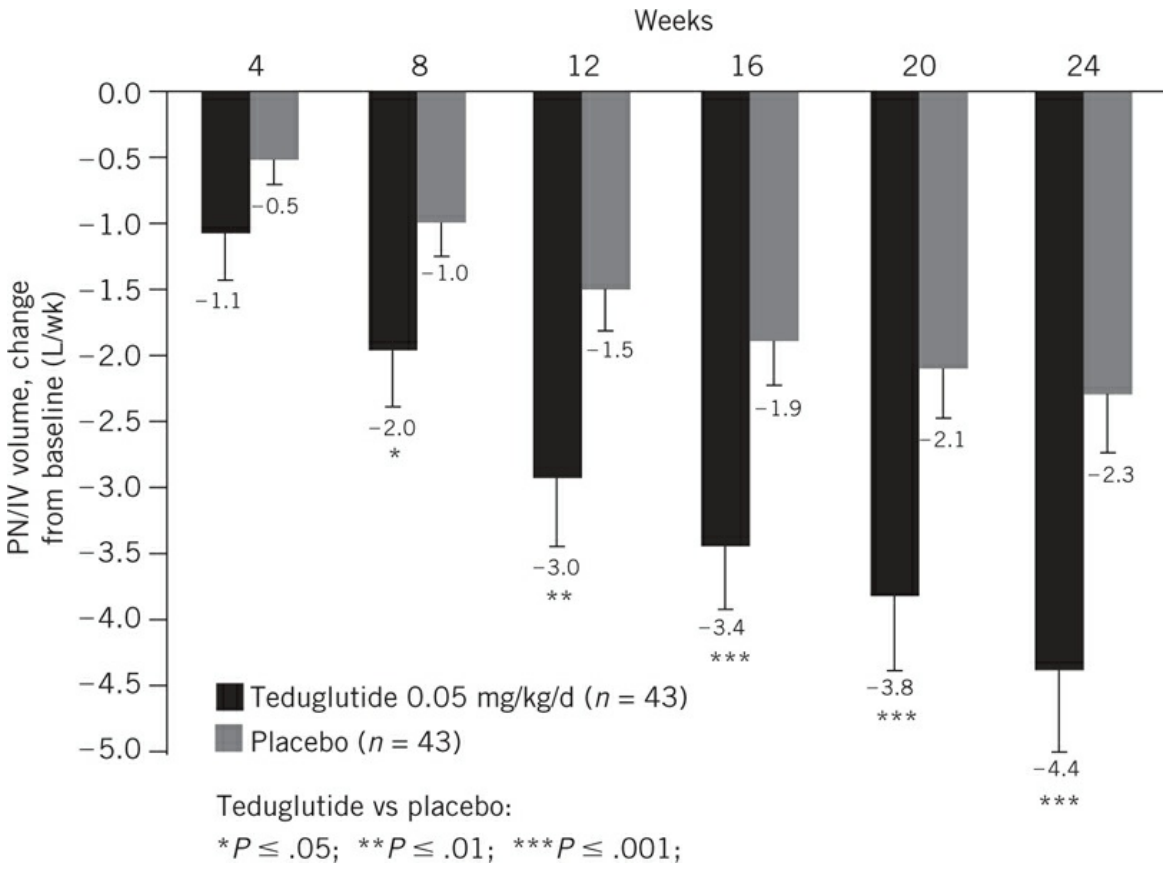


Figure 61-3 Parenteral nutrition volume reductions among subjects receiving teduglutide versus placebo over 24 weeks. (Reproduced with permission from Jeppesen PB et al.[136])

Other hormones. A variety of other circulating factors have been considered for trophic effect in intestine. See Table 61-3 for a comprehensive list.

Intracellular observations. Differential gene expression certainly underlies the structural and functional changes characterizing intestinal adaptation. Investigations using cDNA microarray technology have recently begun to elucidate these pathways. Erwin et al.[60] resected 50% of proximal intestine in male mice and extracted RNA from residual ileum three days later. Compared to ileum from mice receiving sham operations, resected ileum had upregulation of genes that regulate apoptosis, cell cycle control, stress responses, ion flux, and transcription. The same group extended these findings by giving EGF, with demonstration that a novel gene product, small proline-rich protein 2, was significantly induced above baseline, with further enhancement after EGF administration.[49] Other

molecules have been considered, such as peroxisome proliferation-activated receptor α (PPAR α).[105] Molecular techniques will be instrumental in understanding the molecular correlates of adaptation, adaptation failure, and identifying logical pathways amenable to manipulation.

Summary

Animal and clinical data indicate that enteral nutrition, directly or indirectly, enhances intestinal adaptation. Complex macronutrients may enhance adaptation to a larger extent than single amino acid or monosaccharide-containing nutrition. Specific fatty acids or triglycerides may offer advantages over others, but discrepant findings disallow formulation of an ideal fat composition. Nonnutrient factors such as peptide hormones may exert important paracrine or endocrine affects; indeed, both GH and a GLP-2 analog are FDA approved for the promotion of intestinal adaptation in adults with SBS and PN dependence.

Pathophysiology

Parenteral Nutrition Cholestasis

Intestinal failure associated liver disease or (IFALD) is a leading cause of mortality in both adults and children with chronic IF and has been a primary indication for liver-intestinal transplantation in pediatrics. The prevalence, natural history, and liver histology differ between adults and children. The etiology is still unclear and likely multifactorial (see Table 61-4).

Cholestasis has generally been a problem for pediatric patients with IF. Most series have reported that 40%–60% of infants on chronic PN develop cholestasis, whereas 40%–55% of adults develop steatosis or steatohepatitis.[140] Yet, Cavicchi et al.[141] conducted a prospective investigation on PN cholestasis in their adult population of 90 home PN patients receiving PN between 1985 and 1996. The group reported that fully 65% of home PN patients developed chronic cholestasis after an average of 6 months on PN (range 3–132 months), with complications associated with portal hypertension developing in 41% of patients. A much lower burden of liver disease is suggested in more recent adult reports.[96]

TABLE 61-3 Peptides and Hormones with Demonstrated Effects (Animal or Human Studies) on Intestinal Adaptation

Following Resection			
Hormone	**Source**	**Receptor/Location**	**Action/Effects**
Gastrin	G cells (predominantly antrum and stomach)	CCK-B receptor on ECL cells	Increased gastric acid secretion gastric hyperplasia
Leptin	Fat cells, gastric mucosa	Surface receptors, many kids/concentrated in hypothalamus but also diffuse distribution	Enhances carbohydrate absorption in small intestine
GH	Anterior pituitary	GHR expressed in stomach, small intestine, colon	Increased cellular proliferation, increased absorption of water, sodium, and amino acids
IGF-1	Liver	IGF-IR (ubiquitous)	Increased cellular proliferation, increased weight
Peptide YY	L cell in ileum, colon	Y receptor family/diffusely distributed in intestine and CNS	Decreased gastric emptying, prolonged intestinal transit
GLP-2	L cells in ileum and colon	GLP-2 receptor	Increased cellular proliferation, increased weight
Hepatocyte growth factor	Intestinal fibroblasts	c-Met receptors/intestinal epithelial cells	Increased cellular proliferation, differentiation
Epidermal growth factor	Brunner's glands of duodenum, salivary glands, pancreatic secretions	TGF-α/EGFR receptor found throughout the intestinal epithelium	Increased cellular proliferation
Transforming growth factor-α	Epithelial cell throughout the GI tract	TGF-α/EGFR receptor found throughout the intestinal epithelium	Increased cellular proliferation
Keratinocyte growth factor	Stromal cells	KGFR/intestinal epithelial cells	Increased cellular proliferation
Interleukin-11	Stromal cells	IL-11R/intestinal epithelial cells	Increased intestinal mass, enhanced absorption
Bombesin	Brain and intestinal tissue	Several cell surface receptors/diffuse in GI tract including enteric nerves	Increased enterocyte proliferation, mucosal weight

Retinoic acid	Diet, stored in liver	Nuclear receptors	Stimulate crypt cell proliferation and enterocyte migration
Insulin	Released from enteral feeding		greater cell proliferation index decrease in enterocyte apoptotic

Abbreviations: CCK-B, cholecystokinin B; ECL, enterochromaffin-like cell; GHR, growth hormone receptor; GI, gastrointestinal; KGFR, keratinocyte growth factor receptor.

Source: Adapted with permission from Cisler JJ, Buchman AL. Intestinal adaptation in short bowel syndrome. *J Investig Med*. 2005;53:402–413.[94]

The burden of IFALD among children with IF is likely changing. Even a decade ago, approximately 60%–70% of patients with IF acquired in infancy had IFALD.[18,92] while up to one-third of infant cohorts went on to develop life-threatening liver disease.[92,142] Patients referred to transplant centers continue to have a high burden of IFALD.[143] We expect that modern cohorts will demonstrate lower burdens of IFALD; indirect evidence comes from the Intestinal Transplant Registry, which notes a recent drop in liver-inclusive grafts in children.[144]

IFALD has not been clearly defined. Clinical studies have typically used a direct bilirubin of greater than 2 mg/dL for more than two months in the context of PN.[18] A consensus conference in 2007[145] recommended that IFALD should be categorized according to severity, early, established, and late (Table 61-5).

IFALD is a serious complication, leading to increased mortality. Nasr et al.[146] reviewed all infants in their SBS registry (n = 70) born between 1997 and 2003 and found that infants (n = 23) whose direct bilirubin rose above 100 μmol/L (5.8 mg/dL) had a 70% chance of developing end-stage liver disease, on average 60 days (range 10–365 days) subsequent to reaching this threshold. With the rising tide of multidisciplinary centers of excellence and the use of more appropriate PN constituents, it is likely that IFALD and, more importantly, late IFALD will become much less frequent.

TABLE 61-4 Intestinal Failure-Associated Liver Disease: Etiology and Management

Predisposing Factor	Management Response
Prematurity	
Duration of PN	Cycling PN, vigilance in transition to drip enteral nutrition and

	oral nutrition
Length of bowel remnant	
Reduced bile flow	Ursodeoxycholic acid 20–30 mg/kg/d
Lack of enteral feeding	Early introduction of drip feeds
Recurrent sepsis	Daily line care, rigorous education, and reinforcement
Deficiencies	
Essential fatty acid	Balanced fatty acid supply (ω-6/ω-3)
Choline, taurine, cysteine	Pediatric amino acid formulations
Antioxidants	Supplementing α-tocopherol (vitamin E) in lipid emulsions
Toxicity	
Dextrose	Limit excessive administration; cycling
Phytosterols	Substitute other lipid sources for pure soy bean; if using pure soy oil-based lipid emulsion, administer 1 g/kg/d or less in face of worsening cholestasis and thrombocytopenia
Manganese	Monitor levels
Bacterial overgrowth	Targeted antibiotic therapy, surgical tapering, probiotics

Source: Adapted with permission from Goulet O.[310]

TABLE 61-5 Intestinal Failure-Associated Liver Disease Severity

Severity Grade	Clinical	Laboratory	Imaging	Histology
Early	NN	Alkaline phosphatase and/or GGT >1.5 × ULN, total bilirubin abnormal and <3mg/dl for >6 weeks in children	Ultrasound shows an echogenic appearance	Up to 25% steatosis and up to 50% of portal tracts show fibrosis
Established	NN	Total bilirubin 3-6mg/dl, Alkaline phosphatase, GGT and/or transaminases > 1.5x ULN	Ultrasound shows highly echogenic liver with sludge, +/- stones, splenomegaly	Significant steatosis (>25) and fibrosis affecting >50% of portal tracks
Late	Splenomegaly,	Total Bilirubin	NN	Considerable

	spider angiomata, varices, ascites	>6mg/dl, transaminases and alkaline phosphtase >3x ULN, platelets <100, INR >1.5	fatty change, areas of intense fibrosis with architectural distortion

Abbrevations: NN none noted; GGT gamma glutamyl transferase

Source: Adapted with permission from Beath et al.[145]

Cholestasis and/or portal hypertension may hamper intestinal adaptation. Weber and Keller[147] stratified a group of 42 infants with SBS/IF by the severity of liver disease, assessed both clinically and with biopsy or measurement of portal venous pressure. Compared to infants with minimal clinical or objective evidence of liver disease, time on PN was markedly longer in those with marked clinical liver disease (80 vs. 100 days) or abnormal portal pressures (72 vs. 184 days). These data formed the basis for the recommendation that select patients with SBS and late IFALD should be considered for liver transplantation.[148]

Risk factors: Experimental studies suggest that lack of enteral feeding may indirectly lead to liver toxicity. Intestinal stasis and biliary stasis develop during fasting, likely influenced by a gastrointestinal hormonal milieu dramatically different from the fed state. Intestinal stasis may lead to overgrowth, translocation, and sepsis, whereas biliary stasis may cause accumulation of hepatotoxic bile acids. Biliary sludge may form in the context of biliary stasis from the lack of cholecystokinin (CCK) stimulation. However, CCK administration to pediatric patients with IF did not influence frequency or severity of cholestasis.[149] Retrospective analysis of clinical data suggests that early introduction of enteral feeding may be protective in this context.[88]

Early and repeated blood stream infections likely influence the severity or progression of IFALD. Hermans et al.[150] reviewed their experience with IFALD in children on prolonged PN (n = 30; mean PN duration, 65 months) who had received liver biopsies. Those children with severe fibrosis were more likely to have had more frequent (average 3.2 vs. 1.5 events/y) and earlier (average 1 vs. 4 months) sepsis events compared to those with mild to moderate fibrosis. Earlier retrospective reviews of infant IF have demonstrated strong relationships between cholestasis and early and/or frequency of sepsis events.[151,152]

Gestational age or diagnosis may influence risk for IFALD. Christensen et al.[153] reviewed a regional database and extracted data on all neonates receiving PN for more than 14 days (n = 1366). Groups at highest

risk for developing a persistently elevated direct bilirubin greater than 2 mg/dL (operational definition of IFALD) were those with birth weight less than 750 g and diagnoses of jejunal atresia or gastroschisis.

Clinical evidence suggests that hypertriglyceridemia (>150 mg/dL) may cause or be caused by hepatic dysfunction in neonates receiving more than two weeks of PN.[154] Colomb et al.[155] found that for 10 infants on chronic PN, more than half of 23 episodes of cholestasis were preceded by increases in IV lipid delivery. Improvements were noted with temporary discontinuation or decreasing lipid administration. Among adults, Cavicchi et al.[141] found that lipid administration in excess of 1 g/kg/d was an independent predictor of chronic cholestasis in 90 adults with IF followed between 1985 and 1996.

The nature of this lipid-related pathophysiology has been explored in vitro and in animals. Mechanisms include lack of enteral processing, direct hepatocyte toxicity from lipid peroxidation, and cytokine-induced injury. Compared to mice fed both PN solution and Intralipid® orally, mice given PN solution orally and Intralipid IV developed significantly greater fatty liver and hepatocellular damage after 19 days.[156] In vitro investigations found that Intralipid alters cholesterol metabolism when added directly to hepatocyte cell cultures.[157] Rabbits with PN-induced liver injury will show histologic and ultrastructural improvements when supplemented with IV glutathione, an intracellular antioxidant.[158] Vitamin E, particularly α-tocopherol, may also play a role in preventing hepatocyte lipid peroxidation.[159] Although the α-tocopherol content of different lipid emulsions is widely variable,[160] the implications of laboratory and clinical data are somewhat difficult to interpret.[161]

Phytosterols, which are natural components of the soy oil used to produce intravenous lipid emulsions, may alter cell membrane properties and decrease bile flow in neonatal pigs.[162] A mechanism was later suggested by Carter et al.,[163] who demonstrated that phytosterols are potent antagonists for the farnesoid X receptor, a critical regular of bile acid synthesis. Serum phytosterols have been noted to be significantly higher in neonates with IFALD compared to neonates with IF but without IFALD, and values were correlated with duration of PN, markers of liver injury, and histological damage.[164,165]

Lastly, rat models of inflammatory injury in the liver[166] show that endotoxemia via the portal vein may upregulate liver macrophages, which elaborate injurious cytokines. These findings were extended by El Kasmi et al.,[167] who noted that endotoxemia and intravenous lipid were both

necessary to activate liver macrophages and induce liver injury via a toll-like receptor 4-mediated pathway.

Management considerations: Knowledge of these risk factors and laboratory investigations give rise to the principles of IFALD management (Table 61-4). A critical strategy is the prevention of sepsis. Daily line care, intensive education and reinforcement, and multidisciplinary teams with dedicated staff to manage patients on home PN form the ideal system to prevent infection, monitor nutritional variables carefully, and address potential problems quickly. Introduction of small amounts of oral or enteral nutrition may help promote normal bile flow, although data suggest that CCK secretion may not be adequate in SBS/IF in response to enteral stimulus.[168]

Significantly decreasing the amount of soy-based lipid may be successful. Cober et al.[169] prospectively instituted a very low intravenous lipid (1gram/kg/day 2× per week) regimen in 31 infants with IF and IFALD (direct bilirubin >2.5mg/dl on >2 weeks of PN typically with soy-based lipid 3grams/kg/day). After 60 days, direct bilirubin had fallen significantly, compared to an age, diagnosis, and birth-weight matched historical control group. Mild essential fatty acid deficiency developed in 8 children, which corrected with increased intravenous lipid. Growth was similar between the treatment and control groups, and no adverse events were recorded.

Lower dose soy-based lipid has been studied prospectively.[170] Nineteen infants with intestinal failure were randomized in double blind fashion to initiate PN with either Intralipid 1gram/kg/day or Omegaven 1gram/kg/day. The trial was stopped early after 2 years because of very low incidence of cholestasis in either group. However, retrospective evaluation by these investigators found the same burden of cholestasis in patients who were receiving Intralipid 1gram/k/day as compared 2-3grams/kg/day.[171]

Increased attention to the kind of infused intravenous lipid has coincided with a dramatic change in the severity and burden of IFALD. Gura et al.[172] reported on two infants with severe IFALD whose biochemical cholestasis resolved after the substitution of an ω-3-rich, fish oil-derived IV lipid emulsion (Omegaven®) for soy-based lipid emulsion (Intralipid). Subsequently, this group[173] (see Figure 61-4) and other groups[174] have observed marked improvement in cholestasis and indicators of impending decompensated liver disease using this or related strategies. Recently, Omegaven was reported to improve cholestasis in an

adult.[175]

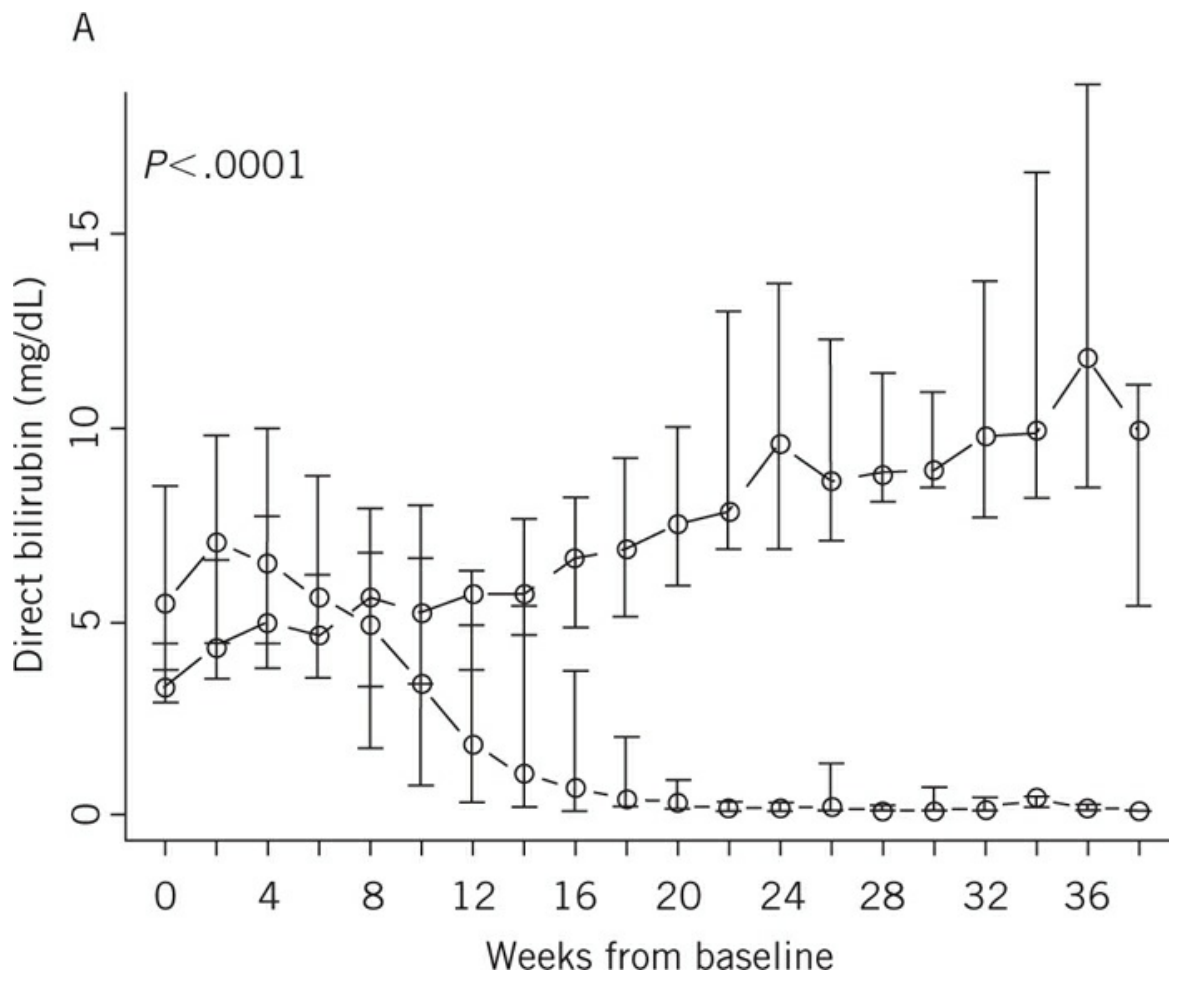


Figure 61-4 Trend in direct bilirubin for children given ω-3 fatty acid-based lipid mixture (Omegaven; *n* = 42; lower line) compared to a historical cohort of patients receiving soy oil-based lipid emulsion (Intralipid; *n* = 49; upper line). Points represent medians and vertical bars represent interquartile range. (Reproduced from Puder et al.[173])

Other lipid strategies may also successfully prevent or improve IFALD. Goulet et al.[176] prospectively demonstrated the efficacy and lack of hepatic toxicity of a lipid product containing a mixture containing soy, olive, medium-chain triglyceride (MCT), and fish-derived lipid (SMOFlipid®) compared to standard soy oil-derived lipid emulsion (Intralipid) among a cohort of children (*n* = 26) with IF over the course of one month. Finally, a retrospective evaluation of a single center's use of SMOFlipid (*n* = 8), compared to an historical cohort of children on

Intralipid (n = 9), suggested improvement in cholestasis after switching from Intralipid.[177] There has yet to be a prospective trial comparing the safety and efficacy of SMOFlipid and Omegaven. For this and other reasons, consensus guidelines[178] have not recommended a particular lipid emulsion strategy.

Biochemical and even clinical improvement does not equal resolution of disease. For instance, serial liver biopsies in six children with IFLAD on ω-3-predominant lipid mixtures demonstrated persistence of fibrosis even with resolution of biochemical liver abnormalities.[179] Fitzgibbons et al.[180] retrospectively evaluated liver biopsies in 66 patients with exposure to PN for greater than 30 days and found that liver fibrosis and cirrhosis did not correlate with biochemical markers of liver disease. Ongoing prospective evaluation of patients with a history of IFALD will be critical in determining the true natural history of this condition, setting the stage for recommendations regarding optimal strategies to prevent complications of chronic liver injury. Prospective randomized trials are needed.

Line Infections and Thrombosis

Any child with a central venous catheter (CVC) is at risk for developing catheter-related bacteremia or fungemia. Patients with IF/SBS may be at particularly high risk[181] because of the abnormal intestinal milieu. Intestinal stasis, bacterial overgrowth, and intestinal inflammation may increase the risk for bacterial translocation from the intestine to blood stream. The organisms can “seed” the venous catheter and not only lead to bacteremia or sepsis but also the need for removal of the line.

Although this scenario has been demonstrated in animal models,[182] definitive evidence in humans does not exist.[183] Suggestive evidence includes isolation of enteric organisms at a much higher frequency from the blood of patients with IF than from patients with CVCs but without IF.[184] Significant concordance has also been found between species isolated from stool or ostomy output and blood culture isolates.[185] Lastly, Weber[186] found that there was a statistically significant increase in enteric organism bacteremia during feeding advancement in 21 infants with IF/SBS compared to baseline as well as a reference population of infants with CVCs but without IF.

Thrombosis occurs when intravascular clotting occurs about the catheter. Scant literature indicates that IF/SBS is a hypercoagulatory state.[187] Whether IF patients compared to other patients with long-term central access have increased incidence of thrombosis has not directly been

studied. Moukarzel et al.[188] found that for long-term PN-dependent pediatric patients the mean life span for a CVC was 22.4 ± 14.7 months (range, 1.5–178 months); 25% of CVC removals were caused by catheter occlusion, which may or may not be due to thrombosis alone. Recurrent vascular occlusion/thrombosis limiting central venous access is particularly worrisome for patients who require central venous nutrition for survival, and this is one of the indications for intestinal transplantation. Nevertheless, true loss of all catheter insertion sites is rare.[189]

Bone Disease

Osteopenia and osteomalacia frequently develop in patients with IF. Ubesie et al.[190] retrospectively reviewed a cohort of 123 children with IF (defined as the need for PN for >30 days) and found that vitamin D deficiency and low bone mineral density occurred in 40% and 12% percent of subjects, respectively. Causative factors include inadequate vitamin D, inability to provide enough calcium and phosphorus parenterally without causing precipitation, magnesium malabsorption, aluminum toxicity, recurrent acidosis, and physical inactivity.[191] In the past, aluminum accumulation may have occurred because of the relatively large amounts found in the parenteral protein source.[192]

Management considerations include monitoring of serum alkaline phosphatase and circulating vitamin D, early consideration of supplemental enteral calcium while monitoring urine for hypercalciuria (see later), and reasonable weight-bearing physical activity. For more extreme cases, pamidronate might be considered.[193] Bone mineral ought to be monitored even after PN has been discontinued, as deficits have been noted in children with a history of PN use.[194] Long-term outcomes have not yet been well characterized.

Bacterial Overgrowth

Small intestinal bacterial overgrowth (SIBO) occurs frequently in patients with IF. The prevalence of SIBO in SBS/IF is widely variable and primarily depends on the methods chosen to make the diagnosis.[182]

Risk factors for SIBO include abnormal intestinal anatomy, decreased secretion of acid in the stomach and enzymes from the pancreas, and possibly an underlying immune defect. Motility in SBS is abnormal[195]; lack of effective migrating motor complexes during fasting because of dilated segments of intestine may promote retention of bacteria in the small intestine. The distal ileum, and not the ICV per se, may be

responsible for the propulsive function in the distal small intestine [196]; as such, resection of the ICV may be more a marker for SIBO risk than its cause.

SIBO may cause disease as a result of malabsorption and compromised intestinal epithelial barrier. Bacteria in the upper small intestine may deconjugate bile salts, leading to impaired micelle formation and increased risk for fat malabsorption. Vitamin B12 deficiency is thought to occur in SIBO because of sequestration by anaerobic bacteria.

The sequence of SIBO, mucosal inflammation, compromised intestinal epithelial barrier, bacterial translocation, and bacteremia/sepsis has been carefully studied in animals.[182,197] Human experiments have not found clear relationships. A small study of 10 infants with SBS noted a relationship between SIBO, markers of mucosal inflammation, and risk of bacteremia.[198] More recently, Gutierrez[199] reviewed 57 patients with IF who had duodenal cultures obtained to evaluate for bacterial overgrowth. Of them, 40 patients (70%) had bacterial overgrowth, and no association was found with bacteremia. In adults, O'Boyle[200] failed to demonstrate any relationship between humoral measures of permeability, mucosal inflammation or atrophy, and bacterial counts in mesenteric lymph nodes.

Nevertheless, SIBO may influence other organs or organ systems. IFALD[201] and sepsis[202] may be influenced not by bacterial translocation directly but instead via cell and humoral immunologic phenomena originating in the intestinal mucosa affected by SIBO.

The diagnosis of SIBO can be challenging as both invasive and noninvasive determinations have important limitations. Traditionally, SIBO has been defined as greater than 100,000 CFU/mL of fluid isolated from the proximal small intestine.[182] Bacteria cultured usually include flora typical of the oropharynx or colon, such as streptococci, members of *Enterobacteriaceae*, and staphylococci. Endoscopic jejunal cultures, although being the gold standard, are both difficult to obtain and poorly reproducible. Hydrogen breath testing has become the noninvasive test of choice for diagnosing SIBO. Hydrogen is a by-product of carbohydrate fermentation by intestinal bacteria, which can be quantified in the breath. Elevated background levels or early peaks after administration of a simple carbohydrate such as lactulose can be considered consistent with SIBO. Multiple reasons for falsely positive and negative tests exist. Moreover, testing will oftentimes be impractical or cumbersome in pediatrics. Thus, empiric antibiotic trials are the norm in the clinical setting.[182]

Management includes selective and periodic use of enteral antibiotics,

prokinetic agents, and surgical therapy to relieve partial obstructions or dilated loops of bowel (see later). Antibiotic therapy remains primarily empiric. Single 7- to 10-day courses may be prescribed, but more typical for patients with SBS/IF would be repeated regimens (e.g., first week of every month) or a continuous, rotating antibiotic regimen. Pro- and prebiotics likely have a role, but safety and efficacy have not been demonstrated clearly enough in clinical studies to recommend routine use. Further discussion and information regarding antibiotic and probiotic use in SIBO can be found elsewhere in the literature.[182]

Renal Calculi

Calcium oxalate nephrolithiasis may occur in patients with SBS because of the hyperabsorption of oxalate.[191] Oxalate is usually complexed with residual calcium, forming nonabsorbable salts. However, in the setting of fatty acid malabsorption with colon in continuity calcium is instead complexed with fatty acids, leaving the oxalate free to diffuse through the colonic mucosa. Nonabsorbed bile salts may also increase colonic permeability to oxalate.[203] Obligate renal excretion will lead to hyperoxaluria and increased risk for stone formation. Management may include low-oxalate diet, calcium supplementation, adequate hydration, resins such as cholestyramine, and medications to alkalinize the urine.

D-Lactic Acidosis

D-Lactic acidosis was first described in 1983,[204] and it is an uncommon but distinctive complication of SBS. The clinical presentation is highly variable but usually features altered mental status and anion gap metabolic acidosis. The mental status changes may be subtle, such as apathy or irritability, or dramatic, such as lethargy or even coma. Suspicion for this diagnosis should be raised in any patient with SBS and altered mental status, especially in a patient with known bowel dilation. Confirmation of the diagnosis requires the ability to measure *d*-lactate level in blood or urine. The pathophysiology is presumed to entail the overgrowth of *d*-lactate-producing bacterial organisms in the small bowel. In the setting of carbohydrate-rich feeding, large amounts of *d*-lactate are generated and promptly absorbed. D-Lactate is poorly metabolized in humans, such that toxic amounts may accumulate easily.[205]

Treatment primarily involves discontinuing enteral feeding, selective bowel decontamination, and sometimes bicarbonate infusions. Prevention may include surgical tapering of dilated bowel loops aimed at reducing

stasis and overgrowth.[206] Medical maneuvers using a mixture of antibiotics and long-term probiotics may also be successful.[207] However, antibiotics may occasionally promote the growth of *d*-lactic acid–producing organisms.[208]

MANAGEMENT AND OUTCOMES

Understanding the driving forces behind intestinal adaptation motivates a principled approach to what is primarily nutritional therapy for children needing intestinal rehabilitation. To be sure, the outcomes are slow to evolve. The results, however, are nothing short of momentous. Success demands patience, persistence, and attention to detail. This can best be achieved through a multidisciplinary approach, providing broad-based medical, nutritional, and surgical support for the patient and longitudinal social support for patients and caregivers.[142] More in-depth and comprehensive review of SBS management is provided in recently published texts.[209]

Medical Support

What to Feed

Although the general role of enteral nutrition in intestinal function and structure is well supported, the specific composition, especially in IF syndromes, has been less well delineated.

Breast milk: Breast milk's special immunologic and anti- infective properties ought to be especially advantageous to infants with IF. Breast milk contains growth factors, nucleotides, long-chain fatty acids, glutamine, and other amino acids that may promote intestinal adaptation. In addition, breast-feeding has been associated with improved immune function, as well as the genesis of a fecal microbiota rich in lactobacilli and bifidobacteria,[210] both of which might improve IF prognosis. On the other hand, breast milk contains lactose, which is sometimes not well tolerated in patients with reduced intestinal surface area. In a population of 30 infants with SBS (defined as >90 days of dependency on PN), the percentage of days that infants received breast milk was strongly correlated with fewer days of PN use ($r = -0.821$, $P < .03$).[88] Breast milk may be associated with the prevention of IFALD.[211] As a result of these theoretical and practical data, best practice may be to use breast milk, if available, for most infants with IF receiving enteral support.

Enteral formulas: For infants with SBS for whom breast milk is not

available, the choice of enteral formulas is controversial. On the one hand, the functional workload presented by complex nutrients may promote mucosal cell proliferation[212] via direct contact with disaccharides[63] or indirectly, whereby distal small intestinal or colonic exposure to luminal nutrients promotes release of trophic factors that stimulate small bowel adaptation.[54,78] On the other hand, the presence of a limited mucosal surface area can lead to lactose, protein, and long-chain fatty acid malabsorption. If this malabsorption is severe, fluid, electrolyte, and metabolic balance can be difficult to achieve.

Carbohydrates may be poorly tolerated by patients with IF, as they are broken down by gastrointestinal bacteria into small, osmotically active organic acids that can present a major osmotic load to the distal small intestine and colon.[213] For this reason, carbohydrate content should generally not exceed 40% of calories, and formulas given to IF patients are usually lactose free. While lactase activity is significantly lower in the jejunum compared with sucrase-isomaltase and maltase-glucoamylase activity,[214] unless there is an extensive jejunal resection most infants and children with SBS will tolerate small to moderate amounts of lactose.[189]

Fiber supplementation may be helpful in the older child with SBS with intact colon. Colonic bacterial fermentation of the unabsorbed fiber produces SCFAs, such as butyrate, which appear to induce trophic changes in animal small intestinal mucosa[77,215] and serve as a calorie source for colonocytes.[216] Moreover, pectin, a fermentable but nondigestable oligosaccharide, increased nitrogen absorption and decreased transit time in a child with SBS,[217] whereas supplementation improved electrolyte absorption and mucosal permeability for infants with protracted diarrheal illnesses.[218]

Although fat tends to be poorly absorbed in SBS, it is a dense energy source relative to carbohydrate or protein. Considering the relatively greater adverse effect of carbohydrate on osmotic diarrhea, it is usually advantageous for patients with SBS to include at least moderate amounts of fat in their diets.

MCTs may be more reliably absorbed than LCTs. MCTs are efficiently hydrolyzed by gastric and pancreatic lipases, giving rise to relatively water-soluble component fatty acids and monoglycerides, which readily diffuse into enterocytes and enter the portal blood directly.[219] This is relevant because infants with dysmotility or SBS often have dilated, poorly motile segments of intestine, conditions that encourage SIBO.[182] Bacteria metabolize and inactivate bile acids, preventing the solubilization

necessary for LCT digestion and absorption. On the other hand, excessive provision of MCT oil can cause diarrhea and ketosis, while MCTs do not provide essential fatty acids. Thus, MCT content should not be the sole source of dietary lipid.

Moreover, as previously noted, free fatty acids derived from hydrolyzed LCTs may have strong trophic effects on small intestinal mucosa. Specific fatty acids may influence intestinal adaptation, as animal data suggest that ω-3-, ω-6-,[56,57] and PUFA-supplemented[220] diets promote mucosal growth and/or function.

An ideal protein source for infants with IF has not yet been determined. Protein itself may have trophic properties. Colostrum protein concentrate improved weight gain and morphometric measures of intestinal adaptation in a piglet model of SBS using polymeric diet plus colostrum versus polymeric diet alone.[221]

Nevertheless, it is not yet clear if the type of protein in infant formulas influences outcomes. Bines et al.[222] reported a series of four SBS patients with persistent feeding intolerance. After receiving an elemental amino acid–based formula, all four patients discontinued PN within 15 months. In addition, histologic and physiologic measures of small bowel function improved concurrently. One retrospective review also found better outcomes (shorter duration of PN dependency) with the use of an amino acid–based formula.[88]

Hydrolyzed protein formulas have also been evaluated clinically. Intact protein infant formula was compared to hydrolyzed protein formula in a crossover study of 60 days' duration in 10 infants with SBS.[223] No effect of formula type was observed on growth, nitrogen absorption, or mucosal permeability. An earlier study in adult jejunostomy patients produced similar findings.[224]

Concern for the development of IgE-mediated[225] and non-IgE-mediated sensitization and allergic enteritis has favored the use of protein hydrolysate or amino acid–based formulas for children with IF. Allergic enteritis may occur in children with IF because of sensitization associated with abnormal mucosal permeability.[222,226] Retrospective small series have found eosinophilic gastrointestinal disease in patients with SBS who were not on hypoallergenic diets,[227] while small retrospective reports note[222,228] improved bowel function in patients who switched to amino acid–based formulas.

In summary, animal and some human data support an important role of enteral nutrition in children with SBS/IF. Modest data support the use of

protein hydrolysate or amino acid–based formulas. Most formulas are lactose free and many include a portion of their fat source as MCTs. Infants with diarrhea and protein-losing enteropathies may require formulas with enhanced protein content. Appendix 3 provides important details on enteral products used in infants and children with IF.

Supplementary fluids, electrolytes, and micronutrients: Excess fluid and electrolyte losses may complicate the management of IF patients, particularly in patients with high-output jejunostomies, in pseudo-obstruction patients who incur large gastric/duodenal fluid losses, or in those with persistent secretory diarrhea. Glucose-containing electrolyte solutions (also called oral rehydration solutions) with sodium concentrations of 75–90 mEq/L are typically used to replace jejunostomy and diarrheal losses. Replacement fluids, be they enteral or intravenous, may be adjusted based on electrolyte concentration of the lost fluids. The intravenous route should be used when there is still net loss of fluid following enteral replacement. Monitoring urine sodium concentration for excessive retention (<10 mEq/L) may indicate the need to provide more sodium-containing fluids, even if serum values are near normal. Excessive magnesium losses can occur with large ostomy volumes, and appropriate magnesium replacement may also improve calcium nutriture.[229]

Zinc[230] and selenium[231] losses increase with watery diarrhea and high ostomy output. Zinc supplements are often used empirically, given that serum values do not reliably reflect body stores. Fat-soluble vitamin supplements should be considered, as fat malabsorption is common when absorptive surface area or micellar function is limited. Serum vitamin B12 methylmalonic acid monitoring and/or parenteral supplementation are necessary for infants with ileal resection or diversion. Serum methylmalonic acid is considered a more sensitive and specific marker for B12 nutriture than serum B12 levels, owing to variable B12 assay performance.[232] Persistent methylmalonic acid elevation despite appropriate supplementation may indicate bacterial production of the metabolite.[233]

How to Feed

Continuous enteral feeding via a nasogastric or gastrostomy tube is advantageous in the patient with SBS or diarrhea,[86,234] as this permits constant saturation of carrier transport proteins, thus taking full advantage of the available absorptive surface area. A controlled study of intragastric feeds in infants with chronic diarrhea and/or SBS found that continuous feeding was associated with improved nutrient balance and weight gain

compared to intermittent feeding.[235]

Devices: Most centers recommend percutaneous enteral access for children who will require enteral nutrition for greater than 3 months. Nevertheless, most infants start with nasogastric or nasoenteric (transpyloric) feedings. Table 61-6 displays the methods, contraindications, and important features of enteral feeding strategies.

TABLE 61-6 Devices Used for Enteral Feeding

Devices	Indications	Contraindications	Advantages	Disadva
Gastric Feeding				
Nasogastric	Nutritional support <3 months	Severe GE reflux, aspiration, slow gastric emptying	Easy to place at home	Frequent dislodgem nasal sym
PEG	Nutritional support >3 months	History of abdominal surgery, abnormal abdominal anatomy, portal hypertension with dilated abdominal vessels	Fewer occlusions with larger bore, one-step low-profile devices available	
Surgical gastrostomy	Nutritional support >3 months and higher risk for injury to abdominal organs	Coagulopathy, poor candidate for surgery (cardiac or respiratory insufficiency)	Immediate placement of low-profile device, direct visualization of stomach	Open surg
Transpyloric Feeding				
GJ	Longer-term enteral nutrition for patients with severe GERD, gastric dysmotility, or need for gastric decompression	Recent proximal surgical anastomoses	Radiologic placement, transpyloric tube can be passed through existing gastrostomy tube	Requires h of gastrost tract prior replaceme GJ, intussusce around tub frequent occlusions jejunal por
Nasojejunal	Short-term nutritional support for patients with severe GERD, gastric	Recent proximal surgical anastomoses	Radiologic or bedside placement techniques, noninvasive	Frequent dislodgem intussusce nasal sym

	dysmotility, pancreatitis			
Jejunal	Long-term enteral nutrition for patients with severe GERD, upper intestinal dysmotility	Generalized dysmotility, poor wound healing	Direct access to small intestine	Open surg procedure mechanica problems

Abbreviations: GE, gastroesophageal; GERD, gastroesophageal reflux disease; GJ, gastrojejunal; PEG percutaneous endoscopic gastrostomy.

Source: Reproduced with permission from Kamin D, Duggan C. Enteral support for children with intestir In: Langnas A, Goulet O, Quigley E, Tappenden K, editors. *Intestinal Failure: Diagnosis, Management a Transplantation*. Malden, MA: Blackwell Publishing; 2007.

Methods for advancement: A protocol for enteral advancement in SBS, or in IF in general, is outlined in Table 61-7. Strict adherence to an ostomy/stool output maximum is unwise, and enteral advancement can occur so long as fluid and electrolyte balance is maintained. Small quantities of oral feedings should be introduced in infants two or three times a day to stimulate sucking and swallowing and to minimize the chances of feeding aversion in the future. Frequent setbacks are not unusual. Enteral feedings may eventually be transitioned to oral/bolus feedings, or oral/bolus and nocturnal feedings, to allow more freedom from the feeding pump.

TABLE 61-7 Enteral Advancement Methodology

A. Feeding Initiation

Audible bowel signs?

Functioning ostomy (if applicable)?

No contraindications?*

No, then continue IVF/PN/NPO [intravenous fluid/parenteral nutrition/nil per os]

Yes, then start feeds (NG/NJ/G-tube/J-tube) [nasogastric/nasojejunal]

Day 1: Breast milk (full strength) or formula (20 cal/oz.) @ 10–20 mL/kg/d continuously** × 24 h

*Contraindications to enteral feeding:

1) Paralytic or drug-induced ileus;
2) Grossly bloody stools or ostomy output and/or radiological changes of intestinal ischemia;
3) Shock/poor perfusion due to cardiac or respiratory insufficiency;
4) Bilious and/or persistent vomiting (defined as more than 3 episodes of emesis in 12 h);

5) Clinical suspicion of obstruction or ileus (severe abdominal distension, decreased ostomy or stool output, and/or radiologic changes of obstruction or ileus).
6) Electrolyte instability
7) Ostomy or stool volumes > 3 mL/kg/h

**Oral feeds may only be offered as follows:

1) Infant is developmentally able to feed by mouth (po).
2) One hour's worth of continuous feeds may be offered PO QD-TID after 5 days of continuous feeds. During this time, tube feeds should be held.
3) More than one hour's worth of continuous feeds may be offered PO once the infant has reached full volume of feeds by continuous route OR at least seven days have passed on the feeding advancement protocol.

B. Feeding Advancement

Principle 1: Quantify feeding intolerance primarily by stool or ostomy output and secondarily by reducing substances. Reducing substances should be measured twice daily. Persistent vomiting may also be indication of intolerance.
Principle 2: Tolerance assessed no more than twice per 24 h. No more than one advance per 24 h period.
Principle 3: Ultimate goals: 150–200 mL/kg/d, 100–140 kcal/kg/d
Principle 4: If ostomy/stool output precludes advancement at 20 cal/oz. for 7 days, then increasing caloric density of the formula can be performed.
Principle 5: Isocaloric reductions in PN support should be undertaken simultaneous with feeding advancement.

1. Stool output

If <10 g/kg/d or <10 stools/d----------> advance rate by 10–20 mL/kg/d
If 10–20 g/kg/d or 10–12 stools/d-----> no change
If >20 g/kg/d or >12 stools/d--------> reduce rate or hold feeds*

2. Ileostomy output

If <2 g/kg/h ----------> advance rate by 10-20 mL/kg/d
If 2–3 g/kg/h ----------> no change
If >3 g/kg/h ----------> reduce rate or hold feeds*

3. Stool-reducing substances

If <1% ----------> advance feeds per stool or ostomy output
If =1% ----------> no change
If >1% ----------> reduce rate or hold feeds*

*Feeds should generally be held for 8 h, then restarted at 3/4 the previous rate.

Source: Reproduced with permission from Kamin D, Duggan C. Enteral support for children with intestinal failure. In: Langnas A, Goulet O, Quigley E, Tappenden K, editors. *Intestinal Failure:*

Surgical Support

Surgery plays a crucial role in the lives of children with IF/SBS.[18] The underlying gastrointestinal disease may demand urgent or emergent operative repair. Subsequent surgical management of IF/SBS alters the bowel physically to effect improvement in bowel function or to address complications. Reestablishment of intestinal continuity, relief of strictures (stricturoplasty), resections employing serosal patches, and bowel tapering for reestablishment of normal intestinal caliber (tapering enteroplasty) are essential surgical tools designed to repair common intestinal complications while preserving intestinal length.

Surgery may also assuage intestinal pathophysiology by altering normal intestine. Patients with SBS may have malabsorption of nutrients and electrolytes because of insufficient contact time with intestinal mucosa. Intestinal valves using sutures or synthetic material, denervation of intestinal segments, and intussusception of intestinal loops have been attempted in animals, but there are minimal published clinical data to support their use in increasing transit time in humans.[13] Segments of intestine may be reversed, which create antiperistaltic "valves" that slow transit enough to improve absorption. Colon segments have been transposed into small intestine for similar reasons, in either iso- or antiperistaltic fashion. A recently published retrospective series of 38 adult patients with SBS and prolonged IF describes a postsurgical PN weaning rate of 45%, using reversed small bowel segments 10–12 cm in length.[236] Nevertheless, this and other techniques have not gained wide acceptance because short and dilated intestine would not be well suited for this approach, reversal segment length choice is unclear, and disrupting normal bowel continuity is counterintuitive.

Longitudinal intestinal lengthening and tailoring (LILT) (Figure 61-5) and serial transverse enteroplasty (STEP) (Figure 61-6) represent advanced techniques that at once taper dilated segments of bowel and increase intestinal length. Digestive function may improve not only because aboral flow is facilitated by the normal caliber lumen but also because subsequent adaptation may lead to increased intestinal surface area.

LILT was first described in 1980 by Bianchi[237] and has since been used at many centers.[13] The technique involves separating the leaflets of mesentery, preserving the segmental blood supply to each side of the intestine. The intestine is then split longitudinally, maintaining the blood

supply to each half. Two lumens are formed by closing the halves along the longitudinal axis, finally forming an end-to-end anastomosis in the isoperistaltic fashion. The resultant bowel is half in diameter and up to twice as long.

Data regarding efficacy come from retrospective and prospective observational case series. A review of 19 pediatric IF patients undergoing LILT between 1984 and 2004 found that 79% were living, 60% were sent for intestinal transplantation, and 44% were able to completely wean from TPN after LILT. Nearly half of patients completely weaned from TPN require supplemental intravenous fluids, and bowel dilation in the LILT segment was common.[238] Satisfactory[239,240] and modest[241,242] success rates with LILT have been the norm. Standardized indications, contraindications, and surgical guidelines have not been developed, which may explain the inconsistent results reported in the literature.[243] Complications such as anastomotic leaking, obstruction, and dilatation are not rare and lead to significant ongoing morbidity or mortality.[244] Criteria for LILT vary but generally require greater than 20 cm of symmetrically dilated intestine in the context of residual intestine greater than 40 cm.[13]

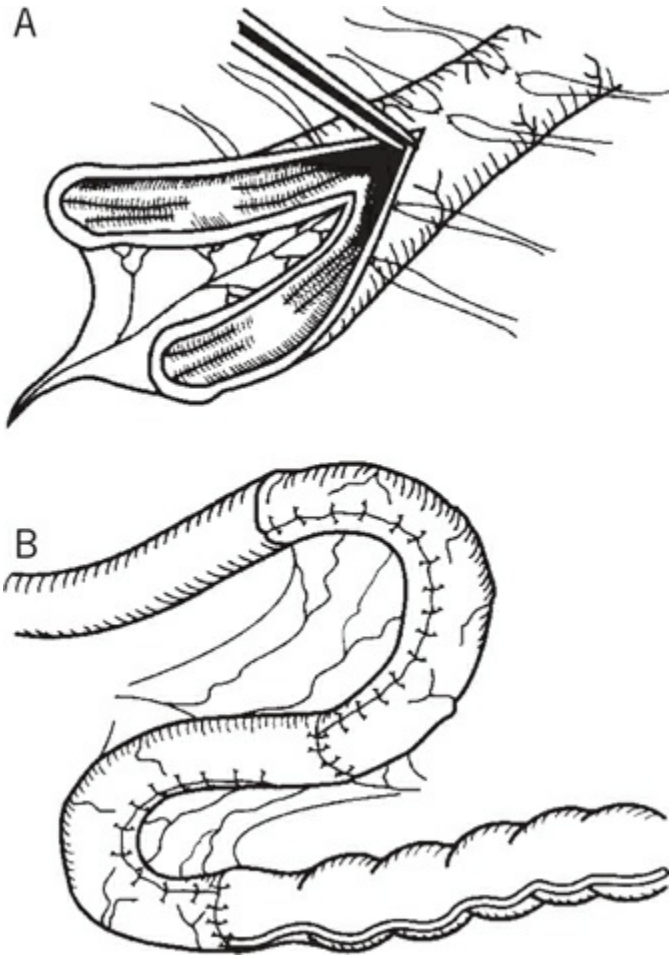


Figure 61-5 Longitudinal intestinal lengthening and tailoring (LILT). (A) Depiction of the separation of the two leaflets of mesentery each attached to longitudinally split piece of small intestine. (B) Final results after each piece of intestine has been sewed together longitudinally, and then end-to-end anastomosis made in series. (Reproduced with permission from Bianchi A.[243])

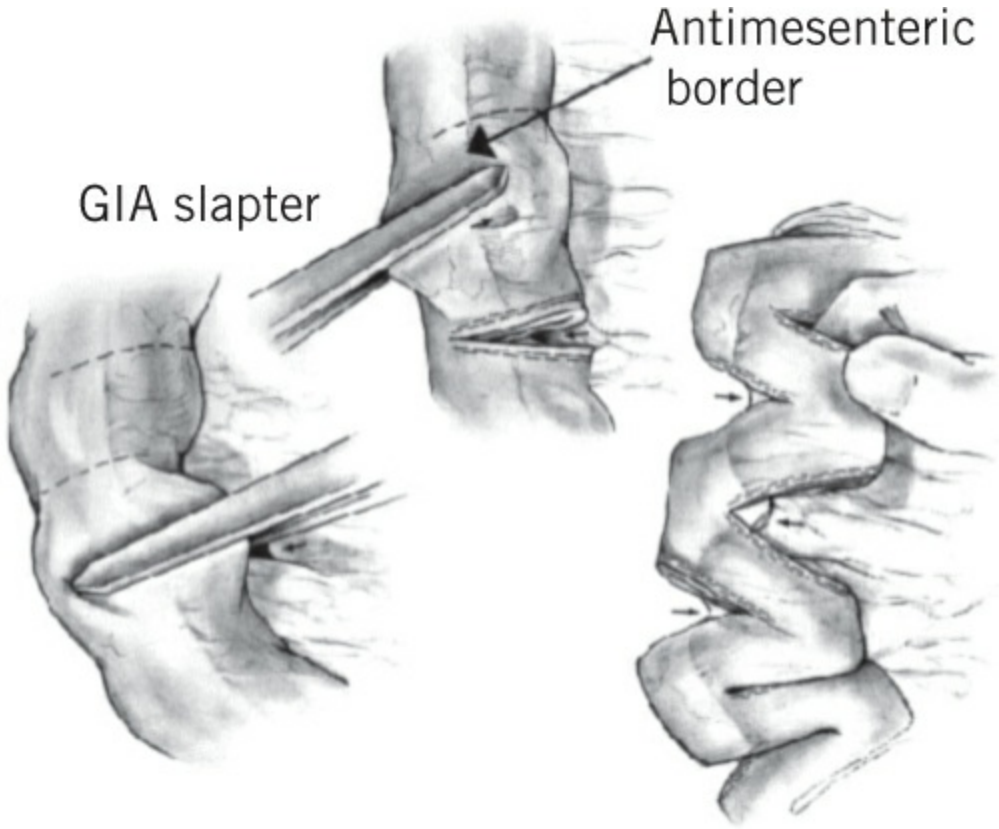


Figure 61-6 Depiction of surgical technique used to perform serial tranverse enteroplasty (STEP). (Reproduced with permission from Javid PJ et al.[309])

STEP involves the application of a staple gun incompletely across the intestine, on opposite sides in serial fashion, creating a zigzag pattern that results in a lumen both narrower and longer. Since its description by Kim et al.[245] in 2003, it has become an accepted technique at many IF centers. Potential advantages over LILT include no enterotomies, the ability to modulate lumen diameter based on the density of staple firings, and the feasibility even if intestinal dilation is in unusual locations or dilation is not of uniform caliber.[33]

A STEP registry has prospectively characterized the pooled experience with the procedure from 50 centers in 13 countries between September 2004 and January 2010.[246,247] Ninety-seven patients (mean age 6.6 years) underwent STEP and had adequate demographic and follow-up data for analysis. Survival was 89%. Of the 78 patients who underwent STEP for PN dependence, 47% achieved enteral autonomy after a median of 21

months. Longer preoperative bowel length predicted achievement of enteral autonomy, whereas higher preoperative serum bilirubin and shorter pre-STEP bowel length predicted death or transplant after first STEP. Fourteen patients in the registry had a "re-STEP" or a third STEP, with enteral autonomy achieved in six of these patients. Longer-term follow-up is emerging and generally encouraging[248]; vigilance is warranted with respect to acute anastomotic leaking and chronic anastomotic bleeding and bowel redilation.[249,250]

The nature of and mechanisms for specific physiologic adaptations have not been well characterized in SBS. Intestine subjected to lengthening procedures has only rarely been studied in this manner.[251] Chang et al. investigated mechanisms in young pigs (n = 10) who received 90% intestinal resections, half of whom had STEP to remaining bowel.[33] After six weeks on oral diet, STEP animals showed improved D-xylose absorption, higher serum citrulline levels (a biomarker for intestinal mass), and no gram-negative bacterial overgrowth. In pigs with surgically created SBS, Modi et al.[252] found that phase III contractions in the small bowel were increased after STEP, providing an additional mechanism for improved bowel function using this surgical technique. The impact of nontransplant surgery on mucosal immune/barrier function and gastroendocrine function is not known.

Long-Term Outcomes

Quality of life (QOL), mortality, and morbidity have been characterized in patients with IF; but, generally speaking, the retrospective nature of ascertainment and single-center experiences have limited the ability to apply findings confidently to individual patients.

QOL has been assessed mostly in adults. Not surprisingly, scoring on QOL tests is usually lowest during the first year on chronic PN, especially if before acquisition of IF the patient had been healthy, for example, mesenteric infarction. QOL slowly improves over the next year, leveling off at 60–70 (100 is "best quality of life") after five years of PN.[253] Factors associated with more favorable QOL include strong self-esteem in the patient and good family support.[254] Moreover, participation in strong peer support networks is associated with reduced incidence of sepsis and depression.[255] Personal stories demonstrating the possibility for meaningful life on chronic PN[256] imbue these statistics with meaning.

Pironi et al.[257] compared health-related QOL (using the SF-36 questionnaire) between adult patients on stable chronic PN (n = 18) and

those with intestinal transplants (n = 12). The results were nearly equivalent, with the exception of subjective physical health, which was significantly higher in the transplant group because of less body pain.

Gottrand et al.[258] conducted a cross-sectional, national multicenter QOL survey of children and adolescents on home PN because of digestive disease. Of the 104 eligible patients, 72 (70%) agreed to participate. QOL was high at all ages, and comparable to scores from a healthy pediatric reference population. QOL was significantly impaired in parents, especially mothers, and in adolescents with ileostomies. This study suggests that children with IF on home PN cope reasonably well, while parents perceive their own lives as significantly limited because of their children's illnesses. A more recent qualitative study of seven school agers and adolescents with lifelong IF found that affected children were resilient, family function was high, and the burdens of chronic illness were manageable.[259]

The growth pattern in children on chronic PN for gastrointestinal disorders can be favorable but may be difficult to consistently achieve. Colomb et al.[260] examined school-age girls and boys (n = 16) on home PN for an average of 10 years. Only four subjects had consistently normal weight and height velocities, whereas the others had fluctuating periods of regular (velocity > 50%) and slow (velocity < 25%) growth, not necessarily associated with illness. Analysis did suggest that inadequate nitrogen provision was closely associated with periods of slower growth. Consistent with earlier reports,[261] normal growth can be achieved on PN, but careful and repeated evaluation and biochemical monitoring are essential.

It is critical to confirm that normal growth can be maintained once PN and/or artificial nutrition are gradually discontinued. Goulet et al.[91] reviewed all cases (n = 87; average follow up time 15 years) of neonatal SBS at their institution in infants born between 1975 and 1991. The subgroup that had weaned from artificial nutrition (parenteral and enteral) grew up with normal puberty and achievement of expected height based on the parents. Gonzalez et al.[262] evaluated 10 children in cross-sectional fashion with history of SBS (mean age, 7.4 years; range, 3–12 years) who had been off PN for at least 2 years. Eight children had normal weight for height, although height for age was low in five children who had SBS in early life. The values were not adjusted for parental height. Body fat mass calculated from anthropometric measures was low in five subjects. These data are motivation enough for all patients with a history of IF to be

followed prospectively; publications based on prospectively obtained data will be instrumental for assessing nutritional outcomes in this population.

Cognitive development and outcomes have been sporadically studied in infants and children on long-term PN. It is difficult to separate the effect of the artificial nutrition from that of the underlying illness. Ralston et al.[263] examined a group of nine infants who were receiving home PN through to age 3 years. Although somatic growth was normal by 2 years of age, neurocognitive development in two patients worsened inexplicably to "mildly retarded" at 3 years of age, whereas for the others development appeared normal. More recently, Beers et al.[264] performed detailed neuropsychiatric testing in eight school-age patients with SBS, comparing results to eight patients with cystic fibrosis. Specific deficits were noted in visual-spatial performance, including left- but not right-sided upper extremity deficits in dexterity and psychomotor speed. The authors suggest that right-hemispheric central nervous system impairments may be present in patients with SBS. Earlier work also suggested the same pattern of deficits.[265] Studies confirming and/or extending these findings have not been published.

In the recent past, outcomes for children with long-standing IF were somewhat guarded, especially those with established or late IFALD. In the last five years, with the decreasing development of IFALD and improvement in intravenous catheter safety, this outlook has changed substantially.

Mortality for patients with IF on home PN is of particular interest, because decisions with respect to referring for intestinal transplant depend heavily on comparative mortality. Studies have typically been retrospective, spanning experiences over many years.[92,266–268]

Recently, Pironi[269] prospectively collected information on 854 patients (children n = 166 and adults n = 688) with IF from European centers between 2004 and 2008. Overall pediatric survival was approximately 90%. The only predictors of death on PN were presence of IFALD or intra-abdominal desmoid tumor, whereas those with ultra-short-segment SBS, intractable diarrhea, or less than 2 years of age statistically faired no differently from the overall group. This survey included only 22 patients who received intestinal transplants, with posttransplant survival notably poor at approximately 65% after up to five years of follow-up. Larger series from the United States report higher survival rates (see more later).

This compares reasonably well to the largest published experience in children. Colomb et al.[268] reviewed their experience in Paris with 302

children on home PN between 1980 and 1999. Survival rates were 97%, 89%, 81%, and 72% after 2, 5, 10, and 15 years of home PN, respectively. For children on home PN because of gastrointestinal disorders, half of deaths were attributable to PN complications such as liver disease or line sepsis, although only 9% (21 patients) of children with gastrointestinal diagnoses died during the study period.

Patients with IF continue to adapt over time. Amiot et al. noted in their cohort of 268 adults that some individuals achieved enteral autonomy up to 5 years after establishing full intestinal continuity.[96] In the latest retrospective review of young children with IF (*n* = 272 infants),[18] a few children achieved enteral autonomy up to 5 years after developing IF. As established IFALD and late IFALD become less common, the window of opportunity for developing full intestinal adaptation has widened.

In summary, long-term survival and QOL on PN are excellent. Care obtained in centers of excellence with multidisciplinary teams can make a marked difference in ensuring excellent and improving outcomes. Life-threatening complications related to the provision of PN have become much less frequent, while the hope of achieving enteral autonomy may be possible even years after developing IF.

TRANSPLANTATION

Intestinal transplantation was introduced into clinical practice in 1989 with the advent of modern immunosuppressive agents. The purpose was to save the lives of children and adults with IF dying from complications related to PN. Experience, surgical technique, immunosuppression, and medical/nutritional management have evolved significantly since then, as have organ and patient survival. Between 1985 and May 2011, the international Intestinal Transplant Registry has recorded approximately 2500 intestinal transplants, spanning 79 worldwide transplant centers, with approximately 200 operations performed per year. About half of organ recipients have been children, although in recent years the proportion of child recipients has fallen to about 30%.[144] Medicare began covering intestinal transplantation in 2001.

Indications

Because intestinal transplantation still has formidable immediate and long-term risks, strict criteria have been proposed[189,270] or are even required by insurance companies.[271] Current consensus regards intestinal transplant as

appropriate therapy for permanent IF when one or more of the following complications related to PN have occurred: (1) serious liver disease, with evidence of portal hypertension and/or coagulopathy; (2) thrombosis of major venous access points (>2 thromboses in jugular, subclavian, or femoral veins), (3) recurrent catheter-related sepsis events; (4) frequent hospitalizations caused by severe water and electrolyte abnormalities or surgical emergencies, (5) high risk of death attributable to underlying disease; and (6) IF with high morbidity or low acceptance of PN.[189,272] Common underlying conditions for patients receiving transplants are displayed in Table 61-8.

The definition of "permanent" is sometimes obvious. For instance, conditions such as severe congenital diarrhea from MVID, SBS with duodenocolic anastomosis, and extensive Hirschsprung's disease with minimal functional small intestine have been unequivocally associated with permanent IF. On the other hand, CIPOS and SBS that is less extensive may have rehabilitation potential that is more or less obvious depending on local experience and over time, as surgical and supports improve.

It would be best to evolve toward an objective score that uses clinical and biological data to predict survival on chronic PN.[273,274] Akin to model for end-stage liver disease, patients could be referred early before irreversible liver disease ensues and/or illness ensues that jeopardizes survival post transplantation. Alternatively, a valid scoring system would prevent premature or inappropriate referrals. Achieving this degree of sophistication is problematic, because intestinal transplantation is rare and the patient population is so heterogeneous.

The current criteria for intestinal transplantation have been questioned. Pironi et al.[272] prospectively followed approximately 300 adults and 90 children with IF on PN for five years and compared survival among transplant "candidates" and "noncandidates" (candidacy defined as meeting criteria for intestinal transplantation according to consensus guidelines[270]). For patients remaining on PN, five-year survival rates for candidates because of PN failure (impending or over liver failure, CVC-related thrombosis in two or more central veins, frequent or severe CVC-related sepsis, or frequent episodes of severe dehydration) and noncandidates were 73% (confidence interval, 62%–84%) and 87% (confidence interval, 83%–91%; difference, $P < .003$), respectively. However, as already noted, in comparison to noncandidates, the only patient-related factors (including indication for transplant, age, and diagnosis) among candidates that predicted death were liver failure and

intra-abdominal desmoid tumor.

TABLE 61-8 Indications for Intestinal Transplantation in Children and Adults, 1985–2011

Worldwide Cases (*N*)	2384 Patients	
Underlying Diagnosis	**Children, % of Worldwide Cases**	**Adults, % of Worldwide Cases**
SBS	63	65
Gastroschisis	25	7
Volvulus	13	11
Necrotizing enterocolitis	15	2
Intestinal atresias	8	2
Crohn's disease	0	13
Ischemia	0	24
Trauma	1	6
Motility	19	15
Mucosal defects	10	2
Other causes	0	0
Retransplant	8	10
Tumor	0	8

Source: Adapted from Intestinal Transplant Registry.[144]

These data motivated the conclusion that intestinal transplantation ought to be offered expeditiously as lifesaving treatment for patients in IFALD-related liver failure or with desmoid tumors, whereas preemptive or rehabilitative intestinal transplantation might be considered for patients who meet other traditional criteria, with careful attention to the comparative costs of continued PN versus transplantation.[272] With improvement in the ability to prevent development of late IFALD, it is instructive to note that throughout Europe between 2004 and 2008 the study recorded 22 transplant procedures, only 6 of which included liver. Similar data do not exist in North America, but it is likely that in Europe the threshold for referral for transplant has been higher.[275]

Graft Considerations

The intestine may be transplanted alone or in combination with other abdominal organs. Isolated intestinal transplantation (ITx) is performed when significant liver disease is not present. These grafts (usually entire jejunum and ileum) may include ICV and some amount of colon, but the majority of transplants include only the small intestine. Living donor ITx can be performed,[276] in which a segment (approximately 200 cm) would be used.

The proportion of liver-containing grafts has fallen in the past five years.[144] Modern techniques have evolved so that en bloc methods are used to implant the graft without having to do complex biliary and vascular anastomoses. Thus, when there is serious liver disease but normal function of stomach and duodenum, an en bloc organ unit composed of graft duodenum, pancreas, small intestine, and liver can be implanted after removal of native small intestine and liver (LITx). The graft duodenum exists in addition to the native duodenum but serves only as a conduit for biliary secretions.

Multivisceral transplantation (MVTx) consists of removal and replacement of the entire foregut and midgut. The graft consists of stomach, duodenum, pancreas, jejunoileum, sometimes spleen, and most often the liver. This graft type would be reserved for patients with abdominal catastrophes so extensive that other graft types would be impossible or too difficult to implant. However, this graft type has been used in younger patients who might only have needed LITx.[277] Advantages include more available space for placement of organs, and improved hemostasis. Coagulopathy and portal hypertension can cause major hemorrhage during native organ explantation. Bleeding can be effectively controlled by ligating the celiac and superior mesenteric arteries during surgery when hemorrhage is severe.[278] This necessitates full foregut removal as the blood supply has been severed.

Children with SBS and significant liver disease have received isolated liver transplants.[279] As mentioned earlier, significant liver disease may hamper intestinal adaptation so that isolated liver transplant may allow more complete adaptation and avoid the need for intestinal transplantation. However, with modern methods to treat and prevent IFALD this approach may become obsolete.

Posttransplantation Management and Care

Care for pediatric intestinal transplant recipients comprises

immunosuppression and allograft rejection monitoring, prevention or early detection and treatment of immune and nonimmune complications related to immunosuppression, infection prevention, prophylaxsis and treatment, and nutrition support. Aside from nutrition support, the other aspects of care are in principle identical to care for children with other solid organ transplants, and they have been reviewed elsewhere.[280] There are no studies demonstrating superiority of any method of nutritional management post intestinal transplantation. What follows is an amalgam of personal experience and expert opinion deduced from informal communication at conferences and review of consensus statements.

PN is resumed usually in the first one to two days post transplantation. Most transplant programs will introduce enteral fluids within seven days of transplant and usually sooner if recovery from surgery is demonstrable with good ileostomy output and bowel sounds.[280] Many centers will secure access into the upper graft with a gastrojejunal tube, such that the tip of the feeding catheter lies well beyond the anastomosis and sits in the graft jejunum. Gastroparesis is expected after intestinal or multivisceral transplantation, and recovery may take weeks. The jejunal access enables increasing enteral nutrition in this context, so that full volume and calorie provision may be achieved enterally while the stomach and upper native intestine recover function. If gastric secretions are significantly bile tinged and pH is 5 or greater, some or all of the lost fluids can be “re-fed” into the jejunal port along with the enteral nutrition. Without surgical or medical complications, PN can typically be discontinued between three and four weeks after transplantation.

Hypoallergenic, MCT oil-supplemented formulas are considered standard for intestinal transplant recipients. These recommendations flow from suppositions that anticipated abnormal barrier function[281] and lack of physiologic lymphatic drainage may increase the chance of allergic sensitization and fat malabsorption, respectively. Maternal breast milk is rarely available by the time young children receive transplants, but administration of banked breast milk has not been well documented. Full-strength feeds are usually commenced after one to two days of clear fluid administration. Advancement is homologous to standards set forth for children with SBS.

Transition to gastric or oral feeds is based on individual patient tolerance and clinical context. The nature and timing of the recommended foods vary considerably between transplant centers. For infants or young children, it may be prudent to avoid highly allergenic foods,[282] as symptoms of gastrointestinal allergy may be confused with allograft

rejection. Nevertheless, food avoidance may have no influence on whether patients ultimately develop allergy or not. Food allergies have developed after intestinal transplantation.[283,284] However, in addition to risk from abnormal mucosal barrier function, immunosuppression may also increase the potential for allergic responses to environmental stimuli.[285]

Some centers prefer that patients consume low-fat diets, as fat absorption is expected to be abnormal with disruption of lymphatic drainage and loss of neural input to exocrine pancreas. Mean coefficient of fat absorption was 86% ± 12% among 22 pediatric intestinal transplant recipients receiving full enteral nutrition between 43 and 1000 days post transplantation.[286] Adequate vitamin E absorption could not be demonstrated until four months post transplantation. Individual case reports indicate that fat excretion normalizes by six months.[44,287] Finally, pancreatic insufficiency has not commonly developed in patients after MVTx.[288] Fat absorption may equally be a function of frequency and severity of infectious and immune complications. Thus, motivation for strict adherence to low-fat diet is likely less important than careful liberalization and monitoring for individual intolerance.

Similar to patients with SBS, sugar malabsorption can lead to osmotic diarrhea; most centers recommend that patients avoid excessive intake of fruit juices for instance. Sugar absorption has indirectly been evaluated by Kaufman et al.[286] Disaccharidase activities were all within normal ranges from small intestinal mucosal biopsies obtained 15-412 days post transplantation in 22 transplant recipients. Breath testing as a means of obtaining a functional index of carbohydrate tolerance will usually not be helpful, because of lack of colon in continuity and bacterial overgrowth.

As with all immunocompromised patients, good food safety practices should be reviewed carefully and periodically with parents and patients to reduce the risk of food-borne illnesses. Excellent resources are available[289] for seeking expert advice on this important subject.

Most intestinal transplant patients receive micronutrient supplements. Patients should receive supplemental fat-soluble vitamins, with attention also to zinc, calcium, magnesium, and iron status. Venick et al.[290] found in their cohort of 19 pediatric intestinal transplant recipients who survived greater than six months post transplantation that 4 had laboratory evidence of essential fatty acid deficiency and 17 had zinc deficiency. Iron deficiency was also common. Laboratory monitoring for fat-soluble vitamin, zinc, and iron deficiency should occur two times per year unless clinical context warrants more frequent testing.

Bone density will likely be abnormal in intestinal transplant patients for multiple reasons. One published case report[291] noted low bone density in the femur and femur neck of a pediatric patient who received a living donor graft from an identical twin. Annual bone density scans with appropriate bone-age, size, and sex standards may be prudent, although vigilance with respect to adequate calcium and vitamin D nutriture is appropriate in all cases.

"Functional foods," or nutrients that are beneficial to a bodily structure or function beyond mere caloric value, have not been evaluated clinically. Rodent work suggests that glutamine supplementation enterally[292,293] or parenterally[294] enhances restoration of normal mucosal architecture following intestinal transplantation, which may decrease bacterial translocation as determined by bacterial counts in mesenteric lymph node samples. Preservation injury may also be prevented by administering glutamine to the donor prior to removal of the organ.[295] ω-3 Fatty acids may exert an immunomodulatory effect at the mucosa[296,297] and small vessel[298] level to promote allograft function and prevent allograft vasculopathy. Concern exists with respect to giving probiotics to transplant patients, because immunocompromised patients or premature neonates make up the majority of cases of bacteremia with probiotics in the literature. A mouse study looking at acute graft versus host disease of the bowel found that administration of *Lactobacillus rhamnosus* GG was associated with reduced severity and frequency of graft versus host disease, as well as reduced bacterial translocation assessed in mesenteric lymph nodes.[299]

Because intestinal rejection causes significant organ dysfunction and increases the likelihood for bacterial translocation and sepsis, resumption of PN is not uncommon especially during moderate or severe episodes of rejection. Severe acute cellular rejection may result in loss of the intestinal graft, which would necessitate resuming lifelong PN unless another transplant is attempted. Chronic intestinal rejection results in the slow but progressive and irreversible loss of graft function and ultimately leads to requirement for long-term PN as well. Venous access may be challenging,[300] and invasive measures may be required to maintain access because of stenosis and collateralization of the usual large venous structures.

Outcomes

In the 1990s, graft and patient survival after intestinal transplantation were

poor, with patient survival rates less than 50% and 40% at one and three years post transplantation, respectively.[301] However, beginning in 2002–2003, overlapping with the usage of lymphocyte-depleting agents for induction, survival rates improved markedly. Experienced centers have reported one- and five-year patient survival rates of 92% and 70%, respectively.[302] The worldwide experience since the year 2000 is not as favorable, with 1-, 5-, and 10-year posttransplant patient and graft survival rates of 76%/71%, 58%/50%, and 44%/40%, respectively. Multiple regression analysis identified home location at time of organ offer, first transplant, and liver-inclusive graft as positive predictors of graft survival, whereas nonpredictive factors included age, diagnosis, and center volume. Graft function is also a concern, with approximately 25% of recipients needing intravenous fluids or PN six months and greater after transplantation.[144]

QOL after intestinal transplantation in children appears favorable. Sudan et al.[303] administered the validated Child Health Questionnaire to 29 pediatric intestinal transplant recipients (at least one year post transplantation) and their parents. Comparison was made to published norms for healthy children. Intestinal recipients (average, 5 years post transplantation; mean, 11 years of age) rated QOL the same as healthy children in all domains. More recently, the UCLA group[304] reported their experience with 24 recipients (median time since transplant, 2.8 years; median age, 6 years) assessing QOL using two validated instruments of determining pediatric health-related QOL. Particular deficits were identified in the self-reported school functioning and psychosocial health domains, while parents reported broad deficits in QOL. These results indicated that self-reported QOL was generally good, while particular domains and parental perception are important components that clinical teams can proactively address.

Nutritional outcomes following intestinal transplantation and predictors thereof have not yet been well characterized. Until the past decade, survival limited interest and ability to identify relationships and trends. Five centers have published growth outcomes after intestinal transplantation, as displayed in Table 61-9. It is expected that at transplant many patients would be stunted and underweight. Post transplantation, improvements in z scores can be appreciated, but it is not yet clear whether and how much length/height catch-up growth can be achieved. Rigorous prospective assessment controlling for bone age and parental height will be essential. In the UCLA cohort, Venick[305] identified low steroid dosage,

infrequent hospitalizations, and peptide-based formulas as positive predictors of linear growth.

TABLE 61-9 Growth Parameters in Children After Intestinal Transplantation

	Nucci et al.[284]	Venick et al.[305]	Rivas et al.[306]	Iyer et al.[307]	Lacaille et al. 2008[308]
N	24	33	23	46	31
Median age, years (range)	3.2 (0.7–17.4)	2.2	1.1	3.7 (0.4–16.6)	5.0 (0.4–13 years)
Mean height z score					
Transplant	−1.9	−3.0	−3.8	−1.9	−1.2
1 Year post transplant	−2.4[a]	−2.2	NA	−1.9	−1.3
2 Years post transplantation	−1.8	−2.0	−2.2	−2.0	−1.2
Mean weight z score					
Transplant	−0.7	−1.0	−-2.3	−0.8	−0.2
1 Year post transplant	−1.0	−0.2	NA	−0.8	+0.1
2 Years post transplantation	−1.0	−0.3	−1.0	−0.8	+0.3

Abbreviation: NA, not available.

[a]Significantly different from transplant, $P \leq .04$.

CONCLUDING REMARKS

IF is a debilitating, life-threatening condition that predisposes children to significant medical, nutritional, and social disorders. Repair and rehabilitation require intimate knowledge of the process of intestinal adaptation, the nutritional principles guiding management of intravenous support for children of all ages (Chapter 70), and vigilance for medical and nutritional complications associated with IF. Medical and surgical innovations have changed the therapeutic landscape for children with IF. Attention to the composition of parenteral lipid, among other factors, has dramatically decreased the number of children who develop life-threatening IFALD, while early and persistent enteral nutrition remains a cornerstone of intestinal rehabilitation and IFALD prevention. Intestinal

transplantation has become a viable lifesaving treatment option when PN fails, and in select circumstances preemptive or rehabilitative transplantation is appropriate for patients with permanent IF and morbid diagnoses. Patients with intestinal transplants have different but nevertheless significant nutritional needs post transplantation, demanding ongoing monitoring and repair of abnormal growth and micronutrient deficiencies. In the future, "immunonutrition" will combine the goals of graft tolerance and nutrition support, with the prospect of withdrawal of traditional medications that negatively impact growth, glucose homeostasis, and cardiovascular health.

REFERENCES

1. Wu G. Synthesis of citrulline and arginine from proline in enterocytes of postnatal pigs. *Am J Physiol*. 1997;272:G1382–G1390.
2. Fitzgibbons S, Ching YA, Valim C, et al. Relationship between serum citrulline levels and progression to parenteral nutrition independence in children with short bowel syndrome. *J Pediatr Surg*. 2009;44:928–932.
3. Rhoads JM, Plunkett E, Galanko J, et al. Serum citrulline levels correlate with enteral tolerance and bowel length in infants with short bowel syndrome. *J Pediatr*. 2005;146:542–547.
4. Ioannou HP, Diamanti E, Piretzi K, et al. Plasma citrulline levels in preterm neonates with necrotizing enterocolitis. *Early Hum Dev*. 2012;88:563–566.
5. Luo M, Fernandez-Estivariz C, Manatunga AK, et al. Are plasma citrulline and glutamine biomarkers of intestinal absorptive function in patients with short bowel syndrome? *J Parenter Enteral Nutr*. 2007;31:1–7.
6. O'Keefe SJ, Buchman AL, Fishbein TM, et al. Short bowel syndrome and intestinal failure: consensus definitions and overview. *Clin Gastroenterol Hepatol*. 2006;4:6–10.
7. Spencer AU, Neaga A, West B, et al. Pediatric short bowel syndrome: redefining predictors of success. *Ann Surg*. 2005;242:403–409; discussion 9–12.
8. Muller T, Hess MW, Schiefermeier N, et al. MYO5B mutations cause microvillus inclusion disease and disrupt epithelial cell polarity. *Nat Genet*. 2008;40:1163–1165.
9. Sivagnanam M, Mueller JL, Lee H, et al. Identification of *EpCAM* as the gene for congenital tufting enteropathy. *Gastroenterology*. 2008;135:429–437.
10. Goulet O, Salomon J, Ruemmele F, et al. Intestinal epithelial dysplasia (tufting enteropathy). *Orphanet J Rare Dis*. 2007;2:20.
11. Lemale J, Coulomb A, Dubern B, et al. Intractable diarrhea with tufting enteropathy: a favorable outcome is possible. *J Pediatr Gastroenterol Nutr*. 2011;52:734–739.
12. Cameron DJ, Barnes GL. Successful pregnancy outcome in tufting enteropathy. *J Pediatr Gastroenterol Nutr*. 2003;36:158.
13. Goulet O, Ruemmele F, Lacaille F, Colomb V. Irreversible intestinal failure. *J Pediatr Gastroenterol Nutr*. 2004;38:250–269.
14. Stanghellini V, Cogliandro RF, de Giorgio R, et al. Chronic intestinal pseudo-obstruction: manifestations, natural history and management. *Neurogastroenterol Motil*. 2007;19:440–452.

15. Howard L, Ament M, Fleming CR, et al. Current use and clinical outcome of home parenteral and enteral nutrition therapies in the United States. *Gastroenterology*. 1995;109:355–365.
16. Van Gossum A, Vahedi K, Abdel M, et al. Clinical, social and rehabilitation status of long-term home parenteral nutrition patients: results of a European multicentre survey. *Clin Nutr*. 2001;20:205–210.
17. Pironi L, Hebuterne X, Van Gossum A, et al. Candidates for intestinal transplantation: a multicenter survey in Europe. *Am J Gastroenterol*. 2006;101:1633–1643; quiz 79.
18. Squires RH, Duggan C, Teitelbaum DH, et al. Natural history of pediatric intestinal failure: initial report from the Pediatric Intestinal Failure Consortium. *J Pediatr*. 2012;161:723–8e2.
19. Touloukian RJ, Smith GJ. Normal intestinal length in preterm infants. *J Pediatr Surg*. 1983;18:720–723.
20. Siebert JR. Small-intestine length in infants and children. *Am J Dis Child*. 1980;134:593–595.
21. Nightingale J, Woodward JM. Guidelines for management of patients with a short bowel. *Gut*. 2006;55(Suppl 4):iv1–12.
22. Struijs MC, Diamond IR, de Silva N, Wales PW. Establishing norms for intestinal length in children. *J Pediatr Surg*. 2009;44:933–938.
23. Senn N. An experimental contribution to intestinal surgery with special reference to the treatment of intestinal obstruction. *Ann Surg*. 1888;7:99–115.
24. Flint J. The effect of extensive resections of the small intestine. *B Johns Hopkins Hosp*. 1912;23:127–144.
25. Nygaard K. Resection of the small intestine in rats. 3. Morphological changes in the intestinal tract. *Acta Chir Scand*. 1967;133:233–248.
26. Sigalet DL, Lees GM, Aherne F, et al. The physiology of adaptation to small bowel resection in the pig: an integrated study of morphological and functional changes. *J Pediatr Surg*. 1990;25:650–657.
27. Haxhija EQ, Yang H, Spencer AU, et al. Influence of the site of small bowel resection on intestinal epithelial cell apoptosis. *Pediatr Surg Int*. 2006;22:37–42.
28. Weale AR, Edwards AG, Bailey M, Lear PA. Intestinal adaptation after massive intestinal resection. *Postgrad Med J*. 2005;81:178–184.
29. Hanson WR, Osborne JW, Sharp JG. Compensation by the residual intestine after intestinal resection in the rat. I. Influence of amount of tissue removed. *Gastroenterology*. 1977;72:692–700.
30. Dowling RH, Booth CC. Structural and functional changes following small intestinal resection in the rat. *Clin Sci*. 1967;32:139–149.
31. Booth CC, Evans KT, Menzies T, Street DF. Intestinal hypertrophy following partial resection of the small bowel in the rat. *Br J Surg*. 1959;46:403–410.
32. Tavakkolizadeh A, Whang EE. Understanding and augmenting human intestinal adaptation: a call for more clinical research. *J Parenter Enteral Nutr*. 2002;26:251–255.
33. Chang RW, Javid PJ, Oh JT, et al. Serial transverse enteroplasty enhances intestinal function in a model of short bowel syndrome. *Ann Surg*. 2006;243:223–228.
34. Weinstein LD, Shoemaker CP, Hersh T, Wright HK. Enhanced intestinal absorption after small bowel resection in man. *Arch Surg*. 1969;99:560–562.
35. Porus RL. Epithelial hyperplasia following massive small bowel resection in man. *Gastroenterology*. 1965;48:753–757.
36. Pironi L, Paganelli GM, Miglioli M, et al. Morphologic and cytoproliferative

patterns of duodenal mucosa in two patients after long-term total parenteral nutrition: changes with oral refeeding and relation to intestinal resection. *J Parenter Enteral Nutr*. 1994;18:351–354.
37. Jao W, Sileri P, Holaysan J, et al. Morphologic adaptation following segmental living related intestinal transplantation. *Transplant Proc*. 2002;34:924.
38. Hines OJ, Bilchik AJ, Zinner MJ, et al. Adaptation of the Na+/glucose cotransporter following intestinal resection. *J Surg Res*. 1994;57:22–27.
39. Helliwell PA, Richardson M, Affleck J, Kellett GL. Regulation of GLUT5, GLUT2 and intestinal brush-border fructose absorption by the extracellular signal-regulated kinase, p38 mitogen-activated kinase and phosphatidylinositol 3-kinase intracellular signalling pathways: implications for adaptation to diabetes. *Biochem J*. 2000;350 (Pt 1):163–169.
40. Thomson AB. Resection of rabbit ileum: effect on jejunal structure and carrier-mediated and passive uptake. *Q J Exp Physiol*. 1986;71:29–46.
41. O'Connor TP, Lam MM, Diamond J. Magnitude of functional adaptation after intestinal resection. *Am J Physiol*. 1999;276:R1265–R1275.
42. Althausen TL, Doig RK, Uyeyama K, Weiden S. Digestion and absorption after massive resection of the small intestine. II. Recovery of the absorptive function as shown by intestinal absorption tests in two patients and a consideration of compensatory mechanisms. *Gastroenterology*. 1950;16:126–139.
43. Fallingborg J, Pedersen P, Jacobsen BA. Small intestinal transit time and intraluminal pH in ileocecal resected patients with Crohn's disease. *Dig Dis Sci*. 1998;43:702–705.
44. Benedetti E, Baum C, Cicalese L, et al. Progressive functional adaptation of segmental bowel graft from living related donor. *Transplantation*. 2001;71:569–571.
45. Ziegler TR, Fernandez-Estivariz C, Gu LH, et al. Distribution of the H+/peptide transporter PepT1 in human intestine: up-regulated expression in the colonic mucosa of patients with short-bowel syndrome. *Am J Clin Nutr*. 2002;75:922–930.
46. Gouttebel MC, Saint Aubert B, Colette C, et al. Intestinal adaptation in patients with short bowel syndrome. Measurement by calcium absorption. *Dig Dis Sci*. 1989;34:709–715.
47. Mayeur C, Gratadoux JJ, Bridonneau C, et al. Faecal D/L lactate ratio is a metabolic signature of microbiota imbalance in patients with short bowel syndrome. *PloS One*. 2013;8:e54335.
48. Erwin CR, Falcone RA, Jr., Stern LE, et al. Analysis of intestinal adaptation gene expression by cDNA expression arrays. *J Parenter Enteral Nutr*. 2000;24:311–316.
49. Stern LE, Erwin CR, Falcone RA, et al. cDNA microarray analysis of adapting bowel after intestinal resection. *J Pediatr Surg*. 2001;36:190–195.
50. Dodson BD, Wang JL, Swietlicki EA, et al. Analysis of cloned cDNAs differentially expressed in adapting remnant small intestine after partial resection. *Am J Physiol*. 1996;271:G347–G356.
51. Menge H, Robinson JW. Functional and structural characteristics of the rat intestinal mucosa following ileo-jejunal transposition. *Acta Hepatogastroenterol (Stuttg)*. 1978;25:150–154.
52. Altmann GG. Influence of bile and pancreatic secretions on the size of the intestinal villi in the rat. *Am J Anat*. 1971;132:167–177.
53. Dworkin LD, Levine GM, Farber NJ, Spector MH. Small intestinal mass of the rat is partially determined by indirect effects of intraluminal nutrition. *Gastroenterology*. 1976;71:626–630.

54. Williamson RC, Buchholtz TW, Malt RA. Humoral stimulation of cell proliferation in small bowel after transection and resection in rats. *Gastroenterology*. 1978;75:249–254.
55. Maxton DG, Cynk EU, Jenkins AP, Thompson RP. Effect of dietary fat on the small intestinal mucosa. *Gut*. 1989;30:1252–1255.
56. Vanderhoof JA, Blackwood DJ, Mohammadpour H, Park JH. Effect of dietary menhaden oil on normal growth and development and on ameliorating mucosal injury in rats. *Am J Clin Nutr*. 1991;54:346–350.
57. Kollman KA, Lien EL, Vanderhoof JA. Dietary lipids influence intestinal adaptation after massive bowel resection. *J Pediatr Gastroenterol Nutr*. 1999;28:41–45.
58. Sukhotnik I, Gork AS, Chen M, et al. Effect of a high fat diet on lipid absorption and fatty acid transport in a rat model of short bowel syndrome. *Pediatr Surg Int*. 2003;19:385–390.
59. Keelan M, Walker K, Thomson AB. Resection of rabbit ileum: effect on brush border membrane enzyme markers and lipids. *Can J Physiol Pharmacol*. 1985;63:1528–1532.
60. Diamond JM, Karasov WH, Cary C, et al. Effect of dietary carbohydrate on monosaccharide uptake by mouse small intestine in vitro. *J Physiol*. 1984;349:419–440.
61. Shu R, David ES, Ferraris RP. Dietary fructose enhances intestinal fructose transport and GLUT5 expression in weaning rats. *Am J Physiol*. 1997;272:G446–G453.
62. Ferraris RP, Diamond JM. Crypt/villus site of substrate–dependent regulation of mouse intestinal glucose transporters. *Proc Natl Acad Sci USA*. 1993;90:5868–5872.
63. Weser E, Babbitt J, Hoban M, Vandeventer A. Intestinal adaptation. Different growth responses to disaccharides compared with monosaccharides in rat small bowel. *Gastroenterology*. 1986;91:1521–1527.
64. Thomson AB, Keelan M, Clandinin MT. Feeding rats a diet enriched with saturated fatty acids prevents the inhibitory effects of acute and chronic ethanol exposure on the in vitro uptake of hexoses and lipids. *Biochim Biophys Acta*. 1991;1084:122–128.
65. Brasitus TA, Dudeja PK, Bolt MJ, et al. Dietary triacylglycerol modulates sodium-dependent D-glucose transport, fluidity and fatty acid composition of rat small intestinal brush-border membrane. *Biochim Biophys Acta*. 1989;979:177–186.
66. Karasov WH, Solberg DH, Diamond JM. Dependence of intestinal amino acid uptake on dietary protein or amino acid levels. *Am J Physiol*. 1987;252:G614–G625.
67. Vanderhoof JA, Grandjean CJ, Burkley KT, Antonson DL. Effect of casein versus casein hydrolysate on mucosal adaptation following massive bowel resection in infant rats. *J Pediatr Gastroenterol Nutr*. 1984;3:262–267.
68. Ziegler TR, Mantell MP, Chow JC, et al. Gut adaptation and the insulin-like growth factor system: regulation by glutamine and IGF-I administration. *Am J Physiol*. 1996;271:G866–G875.
69. Wiren ME, Permert J, Skullman SP, et al. No differences in mucosal adaptive growth one week after intestinal resection in rats given enteral glutamine supplementation or deprived of glutamine. *Eur J Surg*. 1996;162:489–498.
70. Duggan C, Stark AR, Auestad N, et al. Glutamine supplementation in infants with gastrointestinal disease: a randomized, placebo-controlled pilot trial. *Nutrition*.

2004;20:752–756.
71. Albers MJ, Steyerberg EW, Hazebroek FW, et al. Glutamine supplementation of parenteral nutrition does not improve intestinal permeability, nitrogen balance, or outcome in newborns and infants undergoing digestive-tract surgery: results from a double-blind, randomized, controlled trial. *Ann Surg*. 2005;241:599–606.
72. Sukhotnik I, Mogilner JG, Lerner A, et al. Parenteral arginine impairs intestinal adaptation following massive small bowel resection in a rat model. *Pediatr Surg Int*. 2005;21:460–465.
73. Roth JA, Frankel WL, Zhang W, et al. Pectin improves colonic function in rat short bowel syndrome. *J Surg Res*. 1995;58:240–246.
74. Koruda MJ, Rolandelli RH, Settle RG, et al. Harry M. Vars award. The effect of a pectin-supplemented elemental diet on intestinal adaptation to massive small bowel resection. *J Parenter Enteral Nutr*. 1986;10:343–350.
75. Royall D, Wolever TM, Jeejeebhoy KN. Evidence for colonic conservation of malabsorbed carbohydrate in short bowel syndrome. *Am J Gastroenterol*. 1992;87:751–756.
76. Marsman KE, McBurney MI. Dietary fiber increases oxidative metabolism in colonocytes but not in distal small intestinal enterocytes isolated from rats. *J Nutr*. 1995;125:273–282.
77. Koruda MJ, Rolandelli RH, Bliss DZ, et al. Parenteral nutrition supplemented with short-chain fatty acids: effect on the small-bowel mucosa in normal rats. *Am J Clin Nutr*. 1990;51:685–689.
78. Tappenden KA, Thomson AB, Wild GE, McBurney MI. Short-chain fatty acids increase proglucagon and ornithine decarboxylase messenger RNAs after intestinal resection in rats. *J Parenter Enteral Nutr*. 1996;20:357–362.
79. Tappenden KA, Thomson AB, Wild GE, McBurney MI. Short-chain fatty acid-supplemented total parenteral nutrition enhances functional adaptation to intestinal resection in rats. *Gastroenterology*. 1997;112:792–802.
80. Fukunaga T, Sasaki M, Araki Y, et al. Effects of the soluble fibre pectin on intestinal cell proliferation, fecal short chain fatty acid production and microbial population. *Digestion*. 2003;67:42–49.
81. Tabor CW, Tabor H. 1,4-Diaminobutane (putrescine), spermidine, and spermine. *Annu Rev Biochem*. 1976;45:285–306.
82. Uda K, Tsujikawa T, Ihara T, et al. Luminal polyamines upregulate transmural glucose transport in the rat small intestine. *J Gastroenterol*. 2002;37:434–441.
83. Dumas F, De Bandt JP, Colomb V, et al. Enteral ornithine alpha- ketoglutarate enhances intestinal adaptation to massive resection in rats. *Metabolism*. 1998;47:1366–1371.
84. Raul F, Gosse F, Galluser M, et al. Functional and metabolic changes in intestinal mucosa of rats after enteral administration of ornithine alpha-ketoglutarate salt. *J Parenter Enteral Nutr*. 1995;19:145–150.
85. Greene HL, McCabe DR, Merenstein GB. Protracted diarrhea and malnutrition in infancy: changes in intestinal morphology and disaccharidase activities during treatment with total intravenous nutrition or oral elemental diets. *J Pediatr*. 1975;87:695–704.
86. Orenstein SR. Enteral versus parenteral therapy for intractable diarrhea of infancy: a prospective, randomized trial. *J Pediatr*. 1986;109:277–286.
87. Sondheimer JM, Cadnapaphornchai M, Sontag M, Zerbe GO. Predicting the duration of dependence on parenteral nutrition after neonatal intestinal resection. *J Pediatr*. 1998;132:80–84.
88. Andorsky DJ, Lund DP, Lillehei CW, et al. Nutritional and other postoperative

management of neonates with short bowel syndrome correlates with clinical outcomes. *J Pediatr*. 2001;139:27–33.
89. Wilmore DW. Factors correlating with a successful outcome following extensive intestinal resection in newborn infants. *J Pediatr*. 1972;80:88–95.
90. Quiros-Tejeira RE, Ament ME, Reyen L, et al. Long-term parenteral nutritional support and intestinal adaptation in children with short bowel syndrome: a 25-year experience. *J Pediatr*. 2004;145:157–163.
91. Goulet O, Baglin-Gobet S, Talbotec C, et al. Outcome and long-term growth after extensive small bowel resection in the neonatal period: a survey of 87 children. *Eur J Pediatr Surg*. 2005;15:95–101.
92. Wales PW, de Silva N, Kim JH, et al. Neonatal short bowel syndrome: a cohort study. *J Pediatr Surg*. 2005;40:755–762.
93. De Francesco A, Malfi G, Delsedime L, et al. Histological findings regarding jejunal mucosa in short bowel syndrome. *Transplant Proc*. 1994;26:1455–1456.
94. Cisler JJ, Buchman AL. Intestinal adaptation in short bowel syndrome. *J Investig Med*. 2005;53:402–413.
95. Nightingale JM, Lennard-Jones JE, Gertner DJ, et al. Colonic preservation reduces need for parenteral therapy, increases incidence of renal stones, but does not change high prevalence of gall stones in patients with a short bowel. *Gut*. 1992;33:1493–1497.
96. Amiot A, Messing B, Corcos O, et al. Determinants of home parenteral nutrition dependence and survival of 268 patients with non-malignant short bowel syndrome. *Clin Nutr*. 2012.
97. Lund PK. Molecular basis of intestinal adaptation: the role of the insulin-like growth factor system. *Ann N Y Acad Sci*. 1998;859:18–36.
98. Taylor B, Murphy GM, Dowling RH. Pituitary hormones and the small bowel: effect of hypophysectomy on intestinal adaptation to small bowel resection in the rat. *Eur J Clin Invest*. 1979;9:115–127.
99. Ljungmann K, Grofte T, Kissmeyer-Nielsen P, et al. GH decreases hepatic amino acid degradation after small bowel resection in rats without enhancing bowel adaptation. *Am J Physiol Gastrointest Liver Physiol*. 2000;279:G700–G706.
100. Berni Canani R, Cirillo P, Mallardo G, et al. Growth hormone regulates intestinal ion transport through a modulation of the constitutive nitric oxide synthase-nitric oxide-cAMP pathway. *World J Gastroenterol*. 2006;12:4710–4715.
101. Gu Y, Wu ZH, Xie JX, et al. Effects of growth hormone (rhGH) and glutamine supplemented parenteral nutrition on intestinal adaptation in short bowel rats. *Clin Nutr*. 2001;20:159–166.
102. Dowling RH. Cellular and molecular basis of intestinal and pancreatic adaptation. *Scand J Gastroenterol Suppl*. 1992;193:64–67.
103. Jeppesen PB, Szkudlarek J, Hoy CE, Mortensen PB. Effect of high-dose growth hormone and glutamine on body composition, urine creatinine excretion, fatty acid absorption, and essential fatty acids status in short bowel patients: a randomized, double-blind, crossover, placebo-controlled study. *Scand J Gastroenterol*. 2001;36:48–54.
104. Scolapio JS, Camilleri M, Fleming CR, et al. Effect of growth hormone, glutamine, and diet on adaptation in short-bowel syndrome: a randomized, controlled study. *Gastroenterology*. 1997;113:1074–1081.
105. Drozdowski L, Thomson AB. Intestinal mucosal adaptation. *World J Gastroenterol*. 2006;12:4614–4627.
106. Seguy D, Vahedi K, Kapel N, et al. Low-dose growth hormone in adult home parenteral nutrition-dependent short bowel syndrome patients: a positive study.

Gastroenterology. 2003;124:293–302.
107. Byrne TA, Wilmore DW, Iyer K, et al. Growth hormone, glutamine, and an optimal diet reduces parenteral nutrition in patients with short bowel syndrome: a prospective, randomized, placebo-controlled, double-blind clinical trial. *Ann Surg*. 2005;242:655–661.
108. Lifschitz C, Duggan C, Langston C, et al. Growth hormone therapy in children with short bowel syndrome: a randomized, placebo controlled trial. Digestive Diseases Week, 2003. *Gastroenterology*. 2003;124:A–64.
109. Goulet O, Dabbas-Tyan M, Talbotec C, et al. Effect of recombinant human growth hormone on intestinal absorption and body composition in children with short bowel syndrome. *J Parenter Enteral Nutr*. 2010;34:513–520.
110. Guo M, Li Y, Li J. Role of growth hormone, glutamine and enteral nutrition in pediatric short bowel syndrome: a pilot follow-up study. *Eur J Pediatr Surg* [et al] = Zeitschrift fur Kinderchirurgie. 2012;22:121–126.
111. Mantell MP, Ziegler TR, Adamson WT, et al. Resection-induced colonic adaptation is augmented by IGF-I and associated with upregulation of colonic IGF-I mRNA. *Am J Physiol*. 1995;269:G974–G980.
112. Dahly EM, Guo Z, Ney DM. IGF-I augments resection-induced mucosal hyperplasia by altering enterocyte kinetics. *Am J Physiol Regul Integr Comp Physiol*. 2003;285:R800–R808.
113. Knott AW, Juno RJ, Jarboe MD, et al. Smooth muscle overexpression of IGF-I induces a novel adaptive response to small bowel resection. *Am J Physiol Gastrointest Liver Physiol*. 2004;287:G562–G570.
114. Gillingham MB, Kritsch KR, Murali SG, et al. Resection upregulates the IGF-I system of parenterally fed rats with jejunocolic anastomosis. *Am J Physiol Gastrointest Liver Physiol*. 2001;281:G1158–G1168.
115. Gillingham MB, Dahly EM, Carey HV, et al. Differential jejunal and colonic adaptation due to resection and IGF-I in parenterally fed rats. *Am J Physiol Gastrointest Liver Physiol*. 2000;278:G700–G709.
116. Gillingham MB, Dahly EM, Murali SG, Ney DM. IGF-I treatment facilitates transition from parenteral to enteral nutrition in rats with short bowel syndrome. *Am J Physiol Regul Integr Comp Physiol*. 2003;284:R363–R371.
117. Barksdale EM, Jr., Koehler AN, Yaworski JA, et al. Insulinlike growth factor 1 and insulinlike growth factor 3: indices of intestinal failure in children. *J Pediatr Surg*. 1999;34:655–661; discussion 61–62.
118. Yang H, Teitelbaum DH. Novel agents in the treatment of intestinal failure: humoral factors. *Gastroenterology*. 2006;130:S117–S121.
119. Chung BM, Wallace LE, Winkfein RK, et al. The effect of massive small bowel resection and oral epidermal growth factor therapy on SGLT-1 distribution in rabbit distal remnant. *Pediatr Res*. 2004;55:19–26.
120. Avissar NE, Wang HT, Miller JH, et al. Epidermal growth factor receptor is increased in rabbit intestinal brush border membrane after small bowel resection. *Dig Dis Sci*. 2000;45:1145–1152.
121. Helmrath MA, Shin CE, Erwin CR, Warner BW. The EGF\EGF-receptor axis modulates enterocyte apoptosis during intestinal adaptation. *J Surg Res*. 1998;77:17–22.
122. Hardin JA, Chung B, O'Loughlin E V, Gall DG. The effect of epidermal growth factor on brush border surface area and function in the distal remnant following resection in the rabbit. *Gut*. 1999;44:26–32.
123. O'Brien DP, Nelson LA, Williams JL, et al. Selective inhibition of the epidermal growth factor receptor impairs intestinal adaptation after small bowel resection. *J*

Surg Res. 2002;105:25–30.
124. Sham J, Martin G, Meddings JB, Sigalet DL. Epidermal growth factor improves nutritional outcome in a rat model of short bowel syndrome. *J Pediatr Surg*. 2002;37:765–769.
125. Shin CE, Falcone RA, Jr., Stuart L, et al. Diminished epidermal growth factor levels in infants with necrotizing enterocolitis. *J Pediatr Surg*. 2000;35:173–176; discussion 7.
126. Sigalet DL, Martin GR, Butzner JD, et al. A pilot study of the use of epidermal growth factor in pediatric short bowel syndrome. *J Pediatr Surg*. 2005;40:763–768.
127. Wallis K, Walters JR, Forbes A. Review article: glucagon-like peptide 2—current applications and future directions. *Aliment Pharmacol Ther*. 2007;25:365–372.
128. Dahly EM, Gillingham MB, Guo Z, et al. Role of luminal nutrients and endogenous GLP-2 in intestinal adaptation to mid-small bowel resection. *Am J Physiol Gastrointest Liver Physiol*. 2003;284:G670–G682.
129. Martin GR, Wallace LE, Hartmann B, et al. Nutrient-stimulated GLP-2 release and crypt cell proliferation in experimental short bowel syndrome. *Am J Physiol Gastrointest Liver Physiol*. 2005;288:G431–G438.
130. Hoyerup P, Hellstrom PM, Schmidt PT, et al. Glucagon-like peptide-2 stimulates mucosal microcirculation measured by laser Doppler flowmetry in end-jejunostomy short bowel syndrome patients. *Regul Pept*. 2012.
131. Brinkman AS, Murali SG, Hitt S, et al. Enteral nutrients potentiate glucagon-like peptide-2 action and reduce dependence on parenteral nutrition in a rat model of human intestinal failure. *Am J Physiol Gastrointest Liver Physiol*. 2012;303:G610–G622.
132. Scott RB, Kirk D, MacNaughton WK, Meddings JB. GLP-2 augments the adaptive response to massive intestinal resection in rat. *Am J Physiol*. 1998;275:G911–G921.
133. Martin GR, Wallace LE, Sigalet DL. Glucagon-like peptide-2 induces intestinal adaptation in parenterally fed rats with short bowel syndrome. *Am J Physiol Gastrointest Liver Physiol*. 2004;286:G964–G972.
134. Jeppesen PB, Hartmann B, Thulesen J, et al. Glucagon-like peptide 2 improves nutrient absorption and nutritional status in short-bowel patients with no colon. *Gastroenterology*. 2001;120:806–815.
135. O'Keefe SJ, Jeppesen PB, Gilroy R, et al. Safety and efficacy of teduglutide after 52 weeks of treatment in patients with short-bowel syndrome intestinal failure. *Clin Gastroenter Hepatol*. 2013.
136. Jeppesen PB, Pertkiewicz M, Messing B, et al. Teduglutide reduces need for parenteral support among patients with short bowel syndrome with intestinal failure. *Gastroenterology*. 2012;143:1473–81e3.
137. Seidner DL, Schwartz LK, Winkler MF, et al. Increased intestinal absorption in the era of teduglutide and its impact on management strategies in patients with short bowel syndrome-associated intestinal failure. *J Parenter Enteral Nutr*. 2013;37:201–211.
138. Sukhotnik I, Mogilner J, Shamir R, et al. Effect of subcutaneous insulin on intestinal adaptation in a rat model of short bowel syndrome. *Pediatr Surg Int*. 2005;21:132–137.
139. Ben Lulu S, Coran AG, Shehadeh N, et al. Oral insulin stimulates intestinal epithelial cell turnover following massive small bowel resection in a rat and a cell culture model. *Pediatr Surg Int*. 2012;28:179–187.
140. Kelly DA. Intestinal failure-associated liver disease: what do we know today?

Gastroenterology. 2006;130:S70–S77.
141. Cavicchi M, Beau P, Crenn P, et al. Prevalence of liver disease and contributing factors in patients receiving home parenteral nutrition for permanent intestinal failure. *Ann Intern Med*. 2000;132:525–532.
142. Modi BP, Langer M, Ching YA, et al. Improved survival in a multidisciplinary short bowel syndrome program. *J Pediatr Surg*. 2008;43:20–24.
143. Cowles RA, Ventura KA, Martinez M, et al. Reversal of intestinal failure-associated liver disease in infants and children on parenteral nutrition: experience with 93 patients at a referral center for intestinal rehabilitation. *J Pediatr Surg*. 2010;45:84–87; discussion 7–8.
144. Grant D. Intestinal Transplant Registry report: global availability of intestine transplantation, observed trends in clinical practice and survival. In: XIIIth Symposium of the Intestinal Transplant Association. Washington DC; 2011.
145. Beath S, Pironi L, Gabe S, et al. Collaborative strategies to reduce mortality and morbidity in patients with chronic intestinal failure including those who are referred for small bowel transplantation. *Transplantation*. 2008;85:1378–1384.
146. Nasr A, Avitzur Y, Ng VL, et al. The use of conjugated hyperbilirubinemia greater than 100 micromol/L as an indicator of irreversible liver disease in infants with short bowel syndrome. *J Pediatr Surg*. 2007;42:359–362.
147. Weber TR, Keller MS. Adverse effects of liver dysfunction and portal hypertension on intestinal adaptation in short bowel syndrome in children. *Am J Surg*. 2002;184:582–586; discussion 6.
148. Horslen SP, Sudan DL, Iyer KR, et al. Isolated liver transplantation in infants with end-stage liver disease associated with short bowel syndrome. *Ann Surg*. 2002;235:435–439.
149. Teitelbaum DH, Tracy TF, Jr., Aouthmany MM, et al. Use of cholecystokinin-octapeptide for the prevention of parenteral nutrition-associated cholestasis. *Pediatrics*. 2005;115:1332–1340.
150. Hermans D, Talbotec C, Lacaille F, et al. Early central catheter infections may contribute to hepatic fibrosis in children receiving long-term parenteral nutrition. *J Pediatr Gastroenterol Nutr*. 2007;44:459–463.
151. Sondheimer JM, Asturias E, Cadnapaphornchai M. Infection and cholestasis in neonates with intestinal resection and long-term parenteral nutrition. *J Pediatr Gastroenterol Nutr*. 1998;27:131–137.
152. Beath SV, Davies P, Papadopoulou A, et al. Parenteral nutrition-related cholestasis in postsurgical neonates: multivariate analysis of risk factors. *J Pediatr Surg*. 1996;31:604–646.
153. Christensen RD, Henry E, Wiedmeier SE, et al. Identifying patients, on the first day of life, at high-risk of developing parenteral nutrition-associated liver disease. *J Perinatol*. 2007.
154. Toce SS, Keenan WJ. Lipid intolerance in newborns is associated with hepatic dysfunction but not infection. *Arch Pediatr Adolesc Med*. 1995;149:1249–1253.
155. Colomb V, Jobert-Giraud A, Lacaille F, et al. Role of lipid emulsions in cholestasis associated with long-term parenteral nutrition in children. *J Parenter Enteral Nutr*. 2000;24:345–350.
156. Javid PJ, Greene AK, Garza J, et al. The route of lipid administration affects parenteral nutrition-induced hepatic steatosis in a mouse model. *J Pediatr Surg*. 2005;40:1446–1453.
157. Whitfield PD, Clayton PT, Muller DP. Effect of intravenous lipid emulsions on hepatic cholesterol metabolism. *J Pediatr Gastroenterol Nutr*. 2000;30:538–546.
158. Hong L, Wu J, Cai W. Glutathione decreased parenteral nutrition- induced

hepatocyte injury in infant rabbits. *J Parenter Enteral Nutr*. 2007;31:199–204.
159. Goulet O, Joly F, Corriol O, Colomb-Jung V. Some new insights in intestinal failure-associated liver disease. *Curr Opin Organ Transplant*. 2009;14:256–261.
160. Wanten G, Beunk J, Naber A, Swinkels D. Tocopherol isoforms in parenteral lipid emulsions and neutrophil activation. *Clin Nutr*. 2002;21:417–422.
161. Mercer DF. Fish oil emulsions in the management of intestinal failure- associated liver disease. *J Clin Gastroenterol*. 2012;46:823–827.
162. Clayton PT, Whitfield P, Iyer K. The role of phytosterols in the pathogenesis of liver complications of pediatric parenteral nutrition. *Nutrition*. 1998;14:158–164.
163. Carter BA, Taylor OA, Prendergast DR, et al. Stigmasterol, a soy lipid- derived phytosterol, is an antagonist of the bile acid nuclear receptor FXR. *Pediatr Res*. 2007;62:301–306.
164. Kurvinen A, Nissinen MJ, Gylling H, et al. Effects of long-term parenteral nutrition on serum lipids, plant sterols, cholesterol metabolism, and liver histology in pediatric intestinal failure. *J Pediatr Gastroenterol Nutr*. 2011;53:440–446.
165. Kurvinen A, Nissinen MJ, Andersson S, et al. Parenteral plant sterols and intestinal failure-associated liver disease in neonates. *J Pediatr Gastroenterol Nutr*. 2012;54:803–811.
166. Doi F, Goya T, Torisu M. Potential role of hepatic macrophages in neutrophil-mediated liver injury in rats with sepsis. *Hepatology*. 1993;17:1086–1094.
167. El Kasmi KC, Anderson AL, Devereaux MW, et al. Toll-like receptor 4-dependent Kupffer cell activation and liver injury in a novel mouse model of parenteral nutrition and intestinal injury. *Hepatology*. 2012;55:1518–28.
168. Ling PR, Sheikh M, Boyce P, et al. Cholecystokinin (CCK) secretion in patients with severe short bowel syndrome (SSBS). *Dig Dis Sci*. 2001;46:859–864.
169. Cober MP, Killu G, Brattain A, Welch KB, Kunisaki SM, Teitelbaum DH. Intravenous fat emulsions reduction for patients with parenteral nutrition-associated liver disease. *Journal of Pediatrics*. 2012;160:421–7.
170. Nehra D, Fallon EM, Potemkin AK, et al. A comparison of 2 intravenous lipid emulsions: interim analysis of a randomized controlled trial. *J Parenter Enteral Nutr*. 2014;38(6):693–701.
171. Nehra D, Fallon EM, Carlson SJ, et al. Provision of a soy-based intravenous lipid emulsion at 1 g/kg/d does not prevent cholestasis in neonates. *J Parenter Enteral Nutr*. 2013;37:498–505.
172. Gura KM, Duggan CP, Collier SB, et al. Reversal of parenteral nutrition-associated liver disease in two infants with short bowel syndrome using parenteral fish oil: implications for future management. *Pediatrics*. 2006;118:e197–e201.
173. Puder M, Valim C, Meisel JA, et al. Parenteral fish oil improves outcomes in patients with parenteral nutrition-associated liver injury. *Ann Surg*. 2009;250:395–402.
174. Diamond IR, Sterescu A, Pencharz PB, et al. Changing the paradigm: omegaven for the treatment of liver failure in pediatric short bowel syndrome. *J Pediatr Gastroenterol Nutr*. 2009;48:209–215.
175. Burns DL, Gill BM. Reversal of parenteral nutrition-associated liver disease with a fish oil-based lipid emulsion (Omegaven) in an adult dependent on home parenteral nutrition. *J Parenter Enteral Nutr*. 2012.
176. Goulet O, Antebi H, Wolf C, et al. A new intravenous fat emulsion containing soybean oil, medium-chain triglycerides, olive oil, and fish oil: a single-center, double-blind randomized study on efficacy and safety in pediatric patients receiving home parenteral nutrition. *J Parenter Enteral Nutr*. 2010;34:485–495.
177. Muhammed R, Bremner R, Protheroe S, et al. Resolution of parenteral nutrition-

associated jaundice on changing from a soybean oil emulsion to a complex mixed-lipid emulsion. *J Pediatr Gastroenterol Nutr*. 2012;54:797–802.
178. Vanek VW, Seidner DL, Allen P, et al. A.S.P.E.N. position paper: clinical role for alternative intravenous fat emulsions. *Nutr Clin Pract*. 2012;27:150–192.
179. Mercer DF, Hobson BD, Fischer RT, et al. Hepatic fibrosis persists and progresses despite biochemical improvement in children treated with intravenous fish oil emulsion. *J Pediatr Gastroenterol Nutr*. 2013;56:364–369.
180. Fitzgibbons SC, Jones BA, Hull MA, et al. Relationship between biopsy-proven parenteralnutrition-associated liver fibrosis and biochemical cholestasis in children with short bowel syndrome. *J Pediatr Surg*. 2010;45:95–99; discussion 9.
181. Diamanti A, Basso MS, Castro M, et al. Prevalence of life-threatening complications in pediatric patients affected by intestinal failure. *Transplant Proc*. 2007;39:1632–1633.
182. Quigley EM, Quera R. Small intestinal bacterial overgrowth: roles of antibiotics, prebiotics, and probiotics. *Gastroenterology*. 2006;130:S78–S90.
183. Lichtman SN. Translocation of bacteria from gut lumen to mesenteric lymph nodes—and beyond? *J Pediatr Gastroenterol Nutr*. 1991;13:433–434.
184. Piedra PA, Dryja DM, LaScolea LJ, Jr. Incidence of catheter-associated gram-negative bacteremia in children with short bowel syndrome. *J Clin Microbiol*. 1989;27:1317–1319.
185. Kurkchubasche AG, Smith SD, Rowe MI. Catheter sepsis in short-bowel syndrome. *Arch Surg*. 1992;127:21–24; discussion 4–5.
186. Weber TR. Enteral feeding increases sepsis in infants with short bowel syndrome. *J Pediatr Surg*. 1995;30:1086–1088; discussion 8–9.
187. Inaki I, Paloma P, Iske G, Hill S. Long-term parenteral nutrition outcome and incidence of pulmonary emboli. In: IXth International Small Bowel Transplantation Symposium. Brussels, Belgium; 2005.
188. Moukarzel AA, Haddad I, Ament ME, et al. 230 Patient years of experience with home long-term parenteral nutrition in childhood: natural history and life of central venous catheters. *J Pediatr Surg*. 1994;29:1323–1327.
189. American Gastroenterological Association medical position statement. Short bowel syndrome and intestinal transplantation. *Gastroenterology*. 2003;124:1105–1110.
190. Ubesie AC, Heubi JE, Kocoshis SA, et al. Vitamin D deficiency and low bone mineral density in pediatric and young adult intestinal failure. *J Pediatr Gastroenterol Nutr*. 2013.
191. Nightingale JM. Hepatobiliary, renal and bone complications of intestinal failure. *Best Pract Res Clin Gastroenterol*. 2003;17:907–929.
192. Vargas JH, Klein GL, Ament ME, et al. Metabolic bone disease of total parenteral nutrition: course after changing from casein to amino acids in parenteral solutions with reduced aluminum content. *Am J Clin Nutr*. 1988;48:1070–1078.
193. Pastore S, Londero M, Barbieri F, et al. Treatment with pamidronate for osteoporosis complicating long-term intestinal failure. *J Pediatr Gastroenterol Nutr*. 2012;55:615–618.
194. Dellert SF, Farrell MK, Specker BL, Heubi JE. Bone mineral content in children with short bowel syndrome after discontinuation of parental nutrition. *J Pediatr*. 1998;132:516–519.
195. Quigley EM, Thompson JS. The motor response to intestinal resection: motor activity in the canine small intestine following distal resection. *Gastroenterology*. 1993;105:791–798.
196. Quigley EM, Phillips SF, Dent J. Distinctive patterns of interdigestive motility at

the canine ileocolonic junction. *Gastroenterology*. 1984;87:836–844.
197. Zhang SC, Wang W, Ren WY, et al. Effect of cisapride on intestinal bacterial and endotoxin translocation in cirrhosis. *World J Gastroenterol*. 2003;9:534–538.
198. Cole CR, Frem JC, Schmotzer B, et al. The rate of bloodstream infection is high in infants with short bowel syndrome: relationship with small bowel bacterial overgrowth, enteral feeding, and inflammatory and immune responses. *J Pediatr*. 2010;156:941–947, 7e1.
199. Gutierrez IM, Kang KH, Calvert CE, et al. Risk factors for small bowel bacterial overgrowth and diagnostic yield of duodenal aspirates in children with intestinal failure: a retrospective review. *J Pediatr Surg*. 2012;47:1150–1154.
200. O'Boyle CJ, MacFie J, Dave K, et al. Alterations in intestinal barrier function do not predispose to translocation of enteric bacteria in gastroenterologic patients. *Nutrition*. 1998;14:358–362.
201. Solga SF, Diehl AM. Non-alcoholic fatty liver disease: lumen-liver interactions and possible role for probiotics. *J Hepatol*. 2003;38:681–687.
202. Alverdy J, Holbrook C, Rocha F, et al. Gut-derived sepsis occurs when the right pathogen with the right virulence genes meets the right host: evidence for in vivo virulence expression in *Pseudomonas aeruginosa*. *Ann Surg*. 2000;232:480–489.
203. Kathpalia SC, Favus MJ, Coe FL. Evidence for size and charge permselectivity of rat ascending colon. Effects of ricinoleate and bile salts on oxalic acid and neutral sugar transport. *J Clin Invest*. 1984;74:805–811.
204. Perlmutter DH, Boyle JT, Campos JM, et al. D-Lactic acidosis in children: an unusual metabolic complication of small bowel resection. *J Pediatr*. 1983;102:234–238.
205. Halperin ML, Kamel KS. D-lactic acidosis: turning sugar into acids in the gastrointestinal tract. *Kidney Int*. 1996;49:1–8.
206. Modi BP, Langer M, Duggan C, et al. Serial transverse enteroplasty for management of refractory D-lactic acidosis in short-bowel syndrome. *J Pediatr Gastroenterol Nutr*. 2006;43:395–397.
207. Uchida H, Yamamoto H, Kisaki Y, et al. D-lactic acidosis in short-bowel syndrome managed with antibiotics and probiotics. *J Pediatr Surg*. 2004;39:634–636.
208. Coronado BE, Opal SM, Yoburn DC. Antibiotic-induced D-lactic acidosis. *Ann Intern Med*. 1995;122:839–842.
209. Duggan C, Jaksic T, Gura K. *Clinical Mangement of Intestinal Failure*. Boca Raton, FL: CRC Press; 2012.
210. Garrido D, Barile D, Mills DA. A molecular basis for bifidobacterial enrichment in the infant gastrointestinal tract. *Adv Nutr*. 2012;3:415S–421S.
211. Kulkarni S, Mercado V, Rios M, et al. Breast milk is better than formula milk in preventing parenteral nutrition associated liver disease [PNALD] in infants on prolonged parenteral nutrition. *J Pediatr Gastroenterol Nutr*. 2013.
212. Bines JE, Taylor RG, Justice F, et al. Influence of diet complexity on intestinal adaptation following massive small bowel resection in a preclinical model. *J Gastroenterol Hepatol*. 2002;17:1170–1179.
213. Jirapinyo P, Young C, Srimaruta N, et al. High-fat semielemental diet in the treatment of protracted diarrhea of infancy. *Pediatrics*. 1990;86:902–908.
214. Gray G, Santiago N. Disaccharide absorption in normal and diseased human intestine. *Gastroenterology*. 1966;51:489.
215. Pirman T, Ribeyre MC, Mosoni L, et al. Dietary pectin stimulates protein metabolism in the digestive tract. *Nutrition*. 2007;23:69–75.
216. Roediger WE, Millard S. Colonocyte metabolism. *Gut*. 1996;38:792–793.

217. Finkel Y, Brown G, Smith HL, et al. The effects of a pectin-supplemented elemental diet in a boy with short gut syndrome. *Acta Paediatr Scand*. 1990;79:983–986.
218. Rabbani GH, Teka T, Saha SK, et al. Green banana and pectin improve small intestinal permeability and reduce fluid loss in Bangladeshi children with persistent diarrhea. *Dig Dis Sci*. 2004;49:475–484.
219. Jeppesen PB, Mortensen PB. Colonic digestion and absorption of energy from carbohydrates and medium-chain fat in small bowel failure. *J Parenter Enteral Nutr*. 1999;23:S101–S105.
220. Sagher FA, Dodge JA, Moore R, et al. Modulation of fluid absorption and the secretory response of rat jejunum to cholera toxin by dietary fat. *Gut*. 1990;31:1256–1261.
221. Nagy ES, Paris MC, Taylor RG, et al. Colostrum protein concentrate enhances intestinal adaptation after massive small bowel resection in juvenile pigs. *J Pediatr Gastroenterol Nutr*. 2004;39:487–492
222. Bines J, Francis D, Hill D. Reducing parenteral requirement in children with short bowel syndrome: impact of an amino acid-based complete infant formula. *J Pediatr Gastroenterol Nutr*. 1998;26:123–128.
223. Ksiazyk J, Piena M, Kierkus J, Lyszkowska M. Hydrolyzed versus nonhydrolyzed protein diet in short bowel syndrome in children. *J Pediatr Gastroenterol Nutr*. 2002;35:615–618.
224. McIntyre PB, Fitchew M, Lennard-Jones JE. Patients with a high jejunostomy do not need a special diet. *Gastroenterology*. 1986;91:25–33.
225. Mazon A, Solera E, Alentado N, et al. Frequent IgE sensitization to latex, cow's milk, and egg in children with short bowel syndrome. *Pediatr Allergy Immunol*. 2007.
226. Vanderhoof JA, Young RJ. Hydrolyzed versus nonhydrolyzed protein diet in short bowel syndrome in children. *J Pediatr Gastroenterol Nutr*. 2004;38:107.
227. Ching YA, Modi BP, Jaksic T, Duggan C. High diagnostic yield of gastrointestinal endoscopy in children with intestinal failure. *J Pediatr Surg*. 2008;43:906–910.
228. De Greef E, Mahler T, Janssen A, et al. The influence of neocate in paediatric short bowel syndrome on PN weaning. *J Nutr Metab*. 2010;2010:1–6.
229. Buchman AL. Etiology and initial management of short bowel syndrome. *Gastroenterology*. 2006;130:S5–S15.
230. Hambidge KM. Zinc and diarrhea. *Acta Paediatr Suppl*. 1992;381:82–86.
231. Rannem T, Hylander E, Ladefoged K, et al. The metabolism of [75Se]selenite in patients with short bowel syndrome. *J Parenter Enteral Nutr*. 1996;20:412–416.
232. Stabler SP. Vitamin B12 deficiency. *N Engl J Med*. 2013;368:2041–2042.
233. Sentongo TA, Azzam R, Charrow J. Vitamin B12 status, methylmalonic acidemia, and bacterial overgrowth in short bowel syndrome. *J Pediatr Gastroenterol Nutr*. 2009;48:495–497.
234. Weizman Z, Schmueli A, Deckelbaum RJ. Continuous nasogastric drip elemental feeding. Alternative for prolonged parenteral nutrition in severe prolonged diarrhea. *Am J Dis Child*. 1983;137:253–255.
235. Parker P, Stroop S, Greene H. A controlled comparison of continuous versus intermittent feeding in the treatment of infants with intestinal disease. *J Pediatr*. 1981;99:360–364.
236. Beyer-Berjot L, Joly F, Maggiori L, et al. Segmental reversal of the small bowel can end permanent parenteral nutrition dependency: an experience of 38 adults with short bowel syndrome. *Ann Surg*. 2012;256:739–744; discussion 44-45.

237. Bianchi A. Intestinal loop lengthening—a technique for increasing small intestinal length. *J Pediatr Surg*. 1980;15:145–151.
238. Walker SR, Nucci A, Yaworski JA, Barksdale EM, Jr. The Bianchi procedure: a 20-year single institution experience. *J Pediatr Surg*. 2006;41:113–119; discussion 9.
239. Sudan D, DiBaise J, Torres C, et al. A multidisciplinary approach to the treatment of intestinal failure. *J Gastrointest Surg*. 2005;9:165–176; discussion 76–77.
240. Reinshagen K, Kabs C, Wirth H, et al. Long-term outcome in patients with short bowel syndrome after longitudinal intestinal lengthening and tailoring. *J Pediatr Gastroenterol Nutr*. 2008;47:573–578.
241. Hosie S, Loff S, Wirth H, et al. Experience of 49 longitudinal intestinal lengthening procedures for short bowel syndrome. *Eur J Pediatr Surg* [et al] = Zeitschrift fur Kinderchirurgie. 2006;16:171–175.
242. Bianchi A. Longitudinal intestinal lengthening and tailoring: results in 20 children. *J R Soc Med*. 1997;90:429–432.
243. Bianchi A. From the cradle to enteral autonomy: the role of autologous gastrointestinal reconstruction. *Gastroenterology*. 2006;130:S138–S146.
244. Waag KL, Hosie S, Wessel L. What do children look like after longitudinal intestinal lengthening. *Eur J Pediatr Surg* [et al] = Zeitschrift fur Kinderchirurgie. 1999;9:260–262.
245. Kim HB, Fauza D, Garza J, et al. Serial transverse enteroplasty (STEP): a novel bowel lengthening procedure. *J Pediatr Surg*. 2003;38:425–429.
246. Modi BP, Javid PJ, Jaksic T, et al. First report of the international serial transverse enteroplasty data registry: indications, efficacy, and complications. *J Am Coll Surg*. 2007;204:365–371.
247. Jones BA, Hull MA, Potanos KM, et al. Report of 111 consecutive patients enrolled in the International Serial Transverse Enteroplasty (STEP) Data Registry: a retrospective observational study. *J Am Coll Surg*. 2013;216:438–446.
248. Oliveira C, de Silva N, Wales PW. Five-year outcomes after serial transverse enteroplasty in children with short bowel syndrome. *J Pediatr Surg*. 2012;47:931–937.
249. Miyasaka EA, Brown PI, Teitelbaum DH. Redilation of bowel after intestinal lengthening procedures—an indicator for poor outcome. *J Pediatr Surg*. 2011;46:145–149.
250. Fusaro F, Hermans D, Wanty C, et al. Post-serial transverse enteroplasty bowel redilatation treated by longitudinal intestinal lengthening and tailoring procedure. *J Pediatr Surg*. 2012;47:e19–e22.
251. Weber TR. Isoperistaltic bowel lengthening for short bowel syndrome in children. *Am J Surg*. 1999;178:600–604.
252. Modi BP, Ching YA, Langer M, et al. Preservation of intestinal motility after the serial transverse enteroplasty procedure in a large animal model of short bowel syndrome. *J Pediatr Surg*. 2009;44:229–235; discussion 35.
253. Howard L. Home parenteral nutrition: survival, cost, and quality of life. *Gastroenterology*. 2006;130:S52–S59.
254. Smith CE. Quality of life in long-term total parenteral nutrition patients and their family caregivers. *J Parenter Enteral Nutr*. 1993;17:501–506.
255. Smith CE, Curtas S, Werkowitch M, et al. Home parenteral nutrition: does affiliation with a national support and educational organization improve patient outcomes? *J Parenter Enteral Nutr*. 2002;26:159–163.
256. Fairman J, Compher C, Morris J, Mullen JL. Living long with short bowel syndrome: a historical case of twenty-nine years of living with home parenteral

nutrition. *J Parenter Enteral Nutr*. 2007;31:127–134.
257. Pironi L, Paganelli F, Lauro A, et al. Quality of life on home parenteral nutrition or after intestinal transplantation. *Transplant Proc*. 2006;38:1673–1675.
258. Gottrand F, Staszewski P, Colomb V, et al. Satisfaction in different life domains in children receiving home parenteral nutrition and their families. *J Pediatr*. 2005;146:793–797.
259. Emedo MJ, Godfrey EI, Hill SM. A qualitative study of the quality of life of children receiving intravenous nutrition at home. *J Pediatr Gastroenterol Nutr*. 2010;50:431–440.
260. Colomb V, Dabbas M, Goulet O, et al. Prepubertal growth in children with long-term parenteral nutrition. *Horm Res*. 2002;58(Suppl 1):2–6.
261. Amarnath RP, Fleming CR, Perrault J. Home parenteral nutrition in chronic intestinal diseases: its effect on growth and development. *J Pediatr Gastroenterol Nutr*. 1987;6:89–95.
262. Gonzalez HF, Perez NB, Malpeli A, et al. Nutrition and immunological status in long-term follow up of children with short bowel syndrome. *J Parenter Enteral Nutr*. 2005;29:186–191.
263. Ralston CW, O'Connor MJ, Ament M, et al. Somatic growth and developmental functioning in children receiving prolonged home total parenteral nutrition. *J Pediatr*. 1984;105:842–846.
264. Beers SR, Yaworski JA, Stilley C,. Cognitive deficits in school-age children with severe short bowel syndrome. *J Pediatr Surg*. 2000;35:860–865.
265. O'Connor MJ, Ralston CW, Ament ME. Intellectual and perceptual-motor performance of children receiving prolonged home total parenteral nutrition. *Pediatrics*. 1988;81:231–236.
266. Messing B, Crenn P, Beau P, et al. Long-term survival and parenteral nutrition dependence in adult patients with the short bowel syndrome. *Gastroenterology*. 1999;117:1043–1050.
267. Lloyd DA, Vega R, Bassett P, et al. Survival and dependence on home parenteral nutrition: experience over a 25-year period in a UK referral centre. *Aliment Pharmacol Ther*. 2006;24:1231–1240.
268. Colomb V, Dabbas-Tyan M, Taupin P, et al. Long-term outcome of children receiving home parenteral nutrition: a 20-year single-center experience in 302 patients. *J Pediatr Gastroenterol Nutr*. 2007;44:347–353.
269. Pironi L, Goulet O, Buchman A, et al. Outcome on home parenteral nutrition for benign intestinal failure: a review of the literature and benchmarking with the European prospective survey of ESPEN. *Clin Nutr*. 2012;31:831–845.
270. Kaufman SS, Atkinson JB, Bianchi A, et al. Indications for pediatric intestinal transplantation: a position paper of the American Society of Transplantation. *Pediatr Transplant*. 2001;5:80–87.
271. Cigna Healthcare Coverage Position-Intestinal and Multivisceral Transplant. 2007. http://www.cigna.com/customer_care/healthcare_professional/index.html. Accessed July 5, 2007.
272. Pironi L, Joly F, Forbes A, et al. Long-term follow-up of patients on home parenteral nutrition in Europe: implications for intestinal transplantation. *Gut*. 2011;60:17–25.
273. Fishbein TM, Matsumoto CS. Intestinal replacement therapy: timing and indications for referral of patients to an intestinal rehabilitation and transplant program. *Gastroenterology*. 2006;130:S147–S151.
274. O'Keefe SJ. Candidacy for intestinal transplantation. *Am J Gastroenterol*. 2006;101:1644–6.

275. D'Antiga L, Goulet O. Intestinal failure in children: the European view. *J Pediatr Gastroenterol Nutr*. 2013;56:118–126.
276. Benedetti E, Holterman M, Asolati M, et al. Living-related segmental bowel transplantation: from experimental to standardized procedure. *Ann Surg*. 2006;244:694–699.
277. Kato T, Tzakis AG, Selvaggi G, et al. Intestinal and multivisceral transplantation in children. *Ann Surg*. 2006;243:756–764; discussion 64–66.
278. Ruiz P, Kato T, Tzakis A. Current status of transplantation of the small intestine. *Transplantation*. 2007;83:1–6.
279. Botha JF, Grant WJ, Torres C, et al. Isolated liver transplantation in infants with end-stage liver disease due to short bowel syndrome. *Liver Transpl*. 2006;12:1062–1066.
280. Horslen SP. Optimal management of the post-intestinal transplant patient. *Gastroenterology*. 2006;130:S163–S169.
281. Gabe SM, Bjarnason I, Tolou-Ghamari Z, et al. The effect of tacrolimus (FK506) on intestinal barrier function and cellular energy production in humans. *Gastroenterology*. 1998;115:67–74.
282. Nucci AM, Barksdale EM, Jr., Yaworski JA, et al. Enteral formula use in children after small bowel transplant. *Nutr Clin Pract*. 2002;17:113–117.
283. Chehade M, Nowak-Wegrzyn A, Kaufman SS, et al. De novo food allergy after intestinal transplantation: a report of three cases. *J Pediatr Gastroenterol Nutr*. 2004;38:545–547.
284. Nucci AM, Barksdale EM, Jr., Beserock N, et al. Long-term nutritional outcome after pediatric intestinal transplantation. *J Pediatr Surg*. 2002;37:460–463.
285. Nowak-Wegrzyn AH, Sicherer SH, Conover-Walker MK, Wood RA. Food allergy after pediatric organ transplantation with tacrolimus immunosuppression. *J Allergy Clin Immunol*. 2001;108:146–147.
286. Kaufman SS, Lyden ER, Brown CR, et al. Disaccharidase activities and fat assimilation in pediatric patients after intestinal transplantation. *Transplantation*. 2000;69:362–365.
287. Berney T, Genton L, Buhler LH, et al. Five-year follow-up after pediatric living related small bowel transplantation between two monozygotic twins. *Transplant Proc*. 2004;36:316–318.
288. Loinaz C, Mittal N, Kato T, et al. Multivisceral transplantation for pediatric intestinal pseudo-obstruction: single center's experience of 16 cases. *Transplant Proc*. 2004;36:312–313.
289. Diagnosis and management of foodborne Illnesses—A Primer for Physicians and Other Health Care Professionals. In: Agriculture USDO, ed.; 2004. pp. 1–33.
290. Venick RS, Farmer DG, Saikali D, et al. Nutritional outcomes following pediatric intestinal transplantation. *Transplant Proc*. 2006;38:1718–1719.
291. Genton L, Raguso CA, Berney T, et al. Four year nutritional follow up after living related small bowel transplantation between monozygotic twins. *Gut*. 2003;52:659–662.
292. Yagi M, Sakamoto K, Hasebe K, et al. Effect of a glutamine-enriched diet on small bowel allograft during immunosuppressive therapy. *Nutrition*. 1997;13:778–782.
293. Zhang W, Frankel WL, Bain A, et al. Glutamine reduces bacterial translocation after small bowel transplantation in cyclosporine-treated rats. *J Surg Res*. 1995;58:159–164.
294. Zhang W, Frankel WL, Singh A, et al. Improvement of structure and function in orthotopic small bowel transplantation in the rat by glutamine. *Transplantation*.

1993;56:512–517.
295. Lew JI, Zhang W, Koide S, et al. Glutamine improves cold-preserved small bowel graft structure and function following ischemia and reperfusion. *Transplant Proc*. 1996;28:2605–2606.
296. Wu XT, Li JS, Zhao XF, et al. Effects of n-3 fatty acid, fructose-1,6- diphosphate and glutamine on mucosal cell proliferation and apoptosis of small bowel graft after transplantation in rats. *World J Gastroenterol*. 2003;9:1323–1326.
297. Ogita K, Suita S, Taguchi T, et al. Effects of omega-3 fatty acids in rat allogenic small intestinal transplantation. *Pediatr Surg Int*. 2003;19:157–161.
298. Ma H, Wang J, Wang J, et al. Fish oil ameliorates the allograft arteriosclerosis of intestine on rats. *Pediatr Transplant*. 2007;11:173–179.
299. Gerbitz A, Schultz M, Wilke A, et al. Probiotic effects on experimental graft-versus-host disease: let them eat yogurt. *Blood*. 2004;103:4365–4367.
300. Rodrigues AF, van Mourik ID, Sharif K, et al. Management of end-stage central venous access in children referred for possible small bowel transplantation. *J Pediatr Gastroenterol Nutr*. 2006;42:427–433.
301. Grant D, Abu-Elmagd K, Reyes J, et al. 2003 Report of the intestine transplant registry: A new era has dawned. *Ann Surg*. 2005;241:607–613.
302. Abu-Elmagd KM, Costa G, Bond GJ, et al. Five hundred intestinal and multivisceral transplantations at a single center: major advances with new challenges. *Ann Surg*. 2009.
303. Sudan D, Horslen S, Botha J, et al. Quality of life after pediatric intestinal transplantation: the perception of pediatric recipients and their parents. *Am J Transplant*. 2004;4:407–413.
304. Ngo KD, Farmer DG, McDiarmid SV, et al. Pediatric health-related quality of life after intestinal transplantation. *Pediatr Transplant*. 2011;15:849–854.
305. Venick RS, Wozniak LJ, Colangelo J, et al. Long-term nutrition and predictors of growth and weight gain following pediatric intestinal transplantation. *Transplantation*. 2011;92:1058–1062.
306. Rivas K, Kato T, Ueno T, et al. Growth after intestinal transplantation in children. In: IXth International Small Bowel Transplantation Symposium. Brussels; 2005.
307. Iyer K, Horslen S, Iverson A, et al. Nutritional outcome and growth of children after intestinal transplantation. *J Pediatr Surg*. 2002;37:464–6.
308. Lacaille F, Vass N, Sauvat F, et al. Long-term outcome, growth and digestive function in children 2 to 18 years after intestinal transplantation. *Gut* 2008:In press.
309. Javid PJ, Kim HB, Duggan CP, Jaksic T. Serial transverse enteroplasty is associated with successful short-term outcomes in infants with short bowel syndrome. *J Pediatr Surg*. 2005;40:1019–23; discussion 23–4.
310. Goulet O, Ruemmele F. Causes and management of intestinal failure in children. *Gastroenterology*. 2006;130:S16–28.

The Critically Ill Child

62

Eric A. Sparks, MD, Jeremy G. Fisher, MD, Faraz A. Khan, MD, Nilesh M. Mehta, MD, DCH, MRCPCH, and Tom Jaksic, MD, PhD

INTRODUCTION

Malnutrition among critically ill infants and children has remained prevalent with estimates ranging from 19% to 70%.[1–3,5] Mortality and outcomes including length of stay are influenced by nutritional status.[3,4] Unfortunately, the metabolic needs of critically ill children are frequently not met. Accurate individual assessment of the requirement of protein, energy, vitamin, and micronutrient and selection of the appropriate route of nutrient administration are important goal of pediatric critical care.

Sir David Cuthbertson first described the fundamental aspects of the metabolic response to injury in the adult.[6] Following insult, an acute catabolic phase precedes the metabolic process of recovery and repair. This metabolic response to trauma can be conceptually divided into two phases. The initial *ebb phase* is characterized by decreased enzymatic activity and oxygen consumption, low cardiac output with core temperature that may be subnormal. This is followed by the hypermetabolic *flow phase* characterized by increased cardiac output, oxygen consumption, and glucose production. During this phase, protein and lipid mobilization is associated with increased urinary nitrogen excretion and weight loss.[7]

The catabolic response is mediated by a surge in cytokines and the characteristic endocrine response associated with trauma or surgery. This results in the availability of substrates essential for wound healing and inflammatory response. Research regarding the pediatric stress response

has shown that although neonates and children demonstrate similar qualitative metabolic sequelae of illness as adults, significant quantitative differences exist.[8] An understanding of the metabolic events that accompany critical illness and surgery of the child is the first step in implementing adequate nutritional support. The goal of nutritional supportive therapy in this setting is to augment the short-term benefits of the pediatric stress response while minimizing the long-term harmful consequences. Ultimately, an individualized determination of nutrient requirements must be made to provide appropriate amounts of both macro- and micronutrients.

The metabolic response to critical illness differs from the response during prolonged nutrient deprivation or starvation. In starvation, ketone bodies are available as an energy source that spares protein catabolism. However, during illness, high insulin levels inhibit ketone formation. Furthermore, the exogenous administration of glucose does not quell protein catabolism in critical illness as it does during starvation.[9]

METABOLIC RESPONSE TO CRITICAL ILLNESS: AN OVERVIEW OF THE HYPERMETABOLIC OR STRESS RESPONSE

Children, similar to adults, rely on the breakdown and transfer of protein, carbohydrates, and lipid to meet the catabolic demands of the metabolic stress response. Figure 62-1 illustrates the basic pathways involved in the pediatric metabolic response. In general, this metabolic stress response is characterized by an increase in net muscle protein degradation that results in enhanced movement of free amino acid products through the circulation. These amino acids serve as the building blocks for rapid synthesis of proteins that act as the building blocks for tissue repair and the inflammatory response. Remaining amino acids not used in this way are channeled to the liver, where their carbon skeletons are used in gluconeogenesis. In injury, glucose is a necessary substrate for the brain, adrenal, and erythrocytes. Although the catabolism of muscle protein is an effective short-term adaptation for the child, it is of limited duration and potentially damaging owing to relative reduction in lean body mass.

Carbohydrate and lipid turnover is also increased several fold during the pediatric metabolic response. Although these metabolic alterations would be expected to increase overall energy requirements, evidence suggests that any increase is variable if present at all.[10–12] Overall, the

energy needs of the critically ill child are governed by the severity and persistence of the underlying illness. Accurate assessment of energy requirement in individual patients will allow optimal caloric supplementation and avoid the deleterious effects of both under- and overfeeding.

Finally, a unique hormonal and cytokine profile is found in the child with critical illness. These patients demonstrate an elevation in serum levels of insulin, catabolic hormones (glucagons, cortisol, catecholamines), and cytokines that are known to interact with the inflammatory process.[14,15] Current research in this field is focused on ways to manipulate these hormonal and cytokine alterations to minimize the deleterious consequences induced by the metabolic stress response.

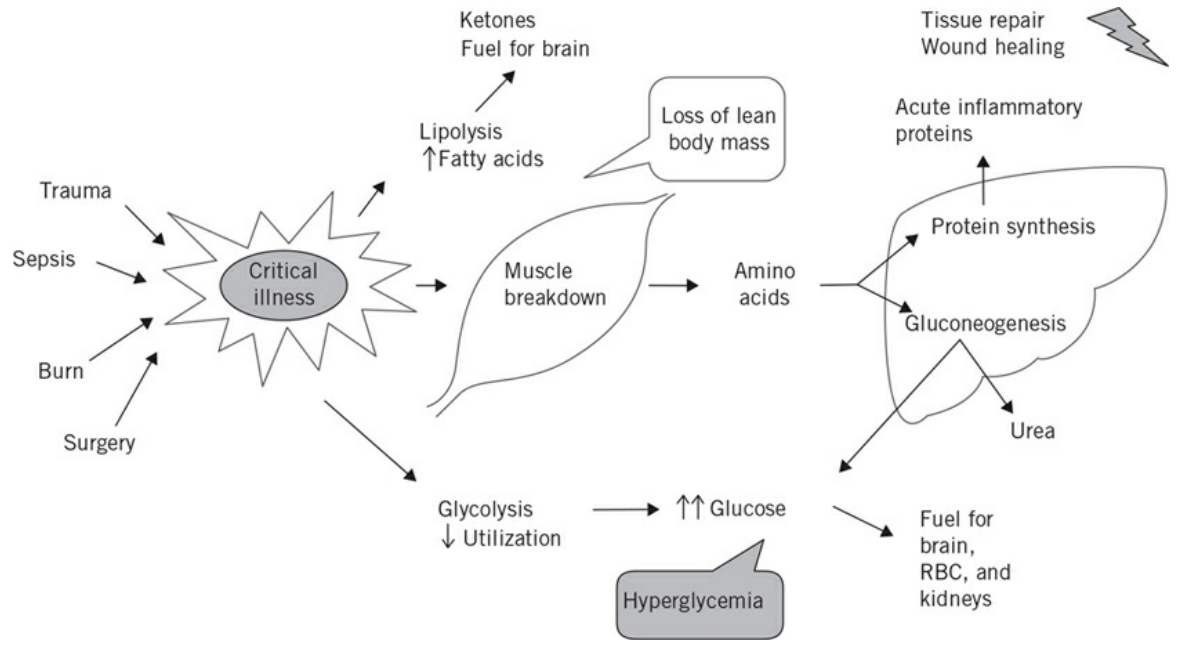


Figure 62-1 Basic pathways of the metabolic stress response.

MECHANISMS OF THE METABOLIC RESPONSE

Many of the deleterious consequences associated with the metabolic stress response can potentially be circumvented. Two integrated and complex systems are involved: neuroendocrine pathways and inflammatory cytokines. The neuroendocrine response to critical illness is relatively well defined, and its features are common to both children and adults. Beginning soon after the initial insult, a unique hormonal profile takes

hold. Catecholamines are released from the adrenal medulla on stimulation by the sympathetic nervous system. The secretion of adrenocorticotropic hormone mediated by the anterior pituitary results in an increase in serum cortisol and aldosterone levels. There are also increased concentrations of growth hormone and antidiuretic hormone, which are secreted by the anterior and posterior pituitary glands, respectively. In addition, glucagon levels rise with the surge in catecholamines. After a brief, initial decline, a rapid increase in serum insulin concentration is found.[16–18]

Overall, the hormonal changes in neonates and infants have been shown to be of greater magnitude but shorter duration than those found in older children and adults. The alterations in intermediary metabolism inherent in the metabolic response can be attributed, at least in part, to hormonal changes. The mobilization of amino acids from muscle, the increased rate of gluconeogenesis in the liver, and the increased lipolysis from adipocyte stores can be explained by the elevations in glucagon, cortisol, and the catecholamines. The increase in net protein degradation, however, is not consistent with the persistent elevation of insulin, a known anabolic hormone. It has been postulated that injury-induced protein degradation is the result of a postreceptor defect in insulin action.[19] This hypothesis is further supported by the finding that burn patients do not demonstrate the same degree of inhibition of protein breakdown compared with normal controls when subjected to hyperinsulinemic euglycemic conditions.[20]

The presence of inflammatory cytokines can be found in the circulation within hours of elective surgery or trauma. These peptides are secreted by leukocytes in a variety of settings and help to mediate the inflammatory response. In general, the prototypical cytokine response in neonates does not markedly differ from that of older children or adults. Interleukin (IL)-6 has been found in increased concentrations in the serum of children within hours after surgery, and this increase persists up to 24–48 hours postoperatively.[21] IL-6 levels have also been shown to be significantly elevated in critically ill children requiring extracorporeal membrane oxygenation (ECMO).[22] IL-6 levels have been directly correlated with increased protein turnover and catabolism as well as a higher incidence of postoperative complications and mortality.[18,19] Tumor necrosis factor (TNF) has also been shown to increase in neonatal critical illness such as necrotizing enterocolitis and sepsis. It is hypothesized that TNF is involved in the release of prostaglandins, thereby mediating the hypotension associated with the septic state. In addition, the release of IL-

1, IL-2, IL-8 interferon-γ, and other growth factors is known to augment the immunologic and hormonal response to injury.

PROTEIN METABOLISM

Amino acids are the key components of growth and tissue repair. The vast majority (98%) is found in existing proteins, with the remainder residing in the free amino acid pool. Proteins themselves are not static as they are continually degraded and synthesized in the process of protein turnover. The reuse of amino acids released by protein breakdown is extensive; protein turnover contributes more than two times the amino acids derived from protein intake. As a reference, healthy newborns have a protein turnover of approximately 6.7 g/kg/d, and adults have a protein turnover of approximately 3.5 g/kg/d.[23] An advantage of high protein turnover is that a continuous flow of amino acids is available for synthesis of new proteins. In this way, maximal physiologic adaptability is present in times of injury or illness.

In the metabolically stressed patient, such as adults with severe burn injury or children with significant cardiorespiratory failure requiring ECMO, protein turnover is double that of normal subjects.[24,25] Specifically, this process involves a redistribution of amino acids from skeletal muscle to the liver, wound, and other tissues taking part in the inflammatory response.[26] The mediators of the inflammatory response—enzymes, serum proteins, and glucose (via gluconeogenesis)—are thus synthesized. Evidence for this process can be seen in the serum by the marked increase in hepatically derived acute-phase reactants (C-reactive protein, fibrinogen, α1-antitrypsin, α-1-acid glycoprotein) and the reciprocal reduction in transport proteins such as albumin and retinol-binding protein (RBP).

Importantly, although children with critical illness have increases in both whole-body protein degradation and whole-body protein synthesis, it is the former that predominates during the stress response. Thus, these patients manifest net negative protein and nitrogen balance, skeletal muscle wasting, weight loss, and immune dysfunction.

Apart from the need for acute-phase protein production in the inflammatory response, another driving force for increased protein catabolism is the obligate production of glucose from amino acids through gluconeogenesis. This process is essential as glucose is the preferred substrate for the brain, erythrocyte, and adrenal medulla and is a versatile energy source for tissues involved in the inflammatory response. In this

process, free alanine and glutamine are produced by muscle protein degradation.[27] Glutamine is first converted to alanine in the intestinal mucosa and then transported through the portal vein to the liver, to be used in glucose production.[28] Alanine is deaminated to pyruvate and contributes a three-carbon unit toward gluconeogenesis, in the glucose-alanine cycle.[27]

Although in fasting subjects, alanine and glutamine account for over 60% of total amino acids released from muscle, this ratio is further accentuated in critical illness. From neonates to adults, increased gluconeogenesis is known to occur during illness and injury. On a per kilogram basis, this process is particularly prominent in smaller neonates, presumably because of an elevated brain-to-body weight ratio and hence an increased requirement for glucose as an energy source.[29]

Because hepatic gluconeogenesis incorporates deamination of these amino acids to cleave the carbon skeleton, the ammonia moiety by-product is detoxified through the urea cycle. Urea is a soluble metabolite that can be safely excreted in the urine. In addition, glycolysis is often activated for energy production given the anaerobic environment of injured tissue. Although this process is exceedingly inefficient compared to the aerobic oxidation of glucose, its end product, lactate, can eventually be reduced to pyruvate and shunted into gluconeogenesis via the Cori cycle.

Although the catabolism of muscle protein to generate glucose and inflammatory response proteins is an excellent short-term adaptation, it is ultimately limited because of the reduced protein stores available in children and neonates. Interestingly, unlike during starvation, the provision of dietary glucose alone is ineffective in reducing the endogenous glucose production via gluconeogenesis in the metabolically stressed state.[30] Therefore, without elimination of the inciting stress for catabolism (i.e., the critical illness or injury) the progressive breakdown of muscle leads to untoward consequences. Loss of diaphragmatic and intercostal muscle mass results in respiratory compromise, and the subsequent loss of cardiac muscle predisposes to fatal arrhythmia. Fortunately, the provision of amino acid nutritional supplementation has been shown to improve overall protein balance in the premature neonate.[31,32] The mechanism for this change appears to be an increase in protein synthesis, whereas the rate of protein degradation remains relatively constant.[33]

As expected, the amount of protein required to optimally enhance protein accretion is higher in critically ill than in healthy children. Infants demonstrate 25% higher protein degradation after surgery, a 100% increase in urinary nitrogen excretion with bacterial sepsis and a 100%

increase in net protein breakdown while on ECMO.[24,33] Children under treatment for cancer have also been shown to have increased net protein breakdown.[34] The provision of dietary protein sufficient to optimize protein synthesis, facilitate wound healing and the inflammatory response, and preserve skeletal muscle protein mass is the single most important nutritional intervention in critically ill children. The quantities of protein recommended for critically ill neonates and children are outlined in Table 62-1.[24] Certain severely stressed states, such as significant burn injury or patients requiring ECMO, may require additional protein supplementation to meet metabolic demands. Hence, growth rates and other indices for protein accretion (i.e., prealbumin levels) must be monitored closely in chronically ill patients. Excessive protein administration should be avoided as toxicity has been documented, particularly in children with marginal renal and hepatic function. Studies using high protein allotments of 4–6 g/kg/d have been associated with adverse effects such as azotemia, metabolic acidosis, and neurodevelopmental abnormalities.[35,36] Further research is needed to evaluate the safety of very high protein regimens in infants.

Although the precise amino acid composition to best increase whole-body protein balance has yet to be fully determined, stable isotope techniques have been described to study this issue.[37–39] Neonates, in particular, have immature biosynthetic pathways that may alter their ability to synthesize specific amino acids. For example, histidine is a conditionally essential amino acid until 6 months of age, and data suggest that biosynthetic pathways for cysteine, taurine, and proline may be of limited capacity in the premature neonate.[37–41] These immature amino acid pathways may have clinical relevance. Given that cysteine is essential for the production of the antioxidant glutathione in very low birth weight neonates, glutathione concentrations in erythrocytes are only 30%–40% of that in normal infants, suggesting that they may be more susceptible to oxidant stress.[41] In addition, proline demand has been shown to increase significantly in patients with severe burns.[25] Other studies of critically ill adults have linked citrulline deficiency to outcomes of morbidity and mortality. These authors propose correction of acquired citrulline and/or arginine deficiency as potential therapeutic aims. However, further research is needed to delineate these biochemical pathways as well as the safety and efficacy of replacement.[42–44]

TABLE 62-1 Estimated Protein Requirements for

Injured Children of Various Age Groups	
Age (yr)	**Estimated Protein Requirements (g/kg/d)**
0–2	2.0–3.0
2–13	1.5–2.0
13–18	1.5

In summary, during critical illness and recovery from trauma or surgery, there is increased protein catabolism. The short-term adaptive benefit of this response may, in time, be outweighed by the loss of protein in critical organs and the consequent morbidity associated with exhaustion of limited protein reserves. This sustained protein breakdown cannot be ameliorated by optimal caloric provision alone (as in starvation), but protein balance may be aided by optimal protein provision.

ENERGY METABOLISM (ASSESSMENT OF ENERGY EXPENDITURE)

During critical illness, increases in the metabolic turnover of protein, carbohydrate, and lipid result in an increased basal energy requirement for the pediatric patient. This question is important as both insufficient and excessive caloric provision has significant consequences for the critically ill child. Inadequate nonprotein caloric provision can accentuate negative nitrogen balance.[45] Conversely, the dietary provision of excess calories from glucose results in an increased production of carbon dioxide (CO_2), with no change in loss of lean body mass.[45,46] Recent data in neonates on ECMO therapy have also shown a possible paradoxical increase in net protein degradation from excess carbohydrate intake.[47] Administration of high glucose loads in critical illness may exacerbate hyperglycemia, increase carbon dioxide generation, and promote hyperlipidemia.

On the other hand, hypocaloric diet appears to have demonstrable benefits in critically ill obese adults. Obesity may be a risk factor for higher mortality in critically ill children.[48] In critically ill obese adults, permissive underfeeding consisting of a high-protein enteric diet providing 60%–70% of daily energy requirements is recommended. This strategy has been shown to aid weight loss while maintaining nitrogen balance, increasing insulin sensitivity, and reducing comorbidities.[49,50] Some critically ill obese children may benefit from a hypocaloric diet, although there is no data supporting this practice.

A careful appraisal of energy requirements in the critically ill pediatric patient is mandatory. The components of total energy expenditure (TEE) for a sedentary individual are (1) resting energy expenditure (REE)—70%, (2) diet-induced thermogenesis (DIT)—10%, and (3) energy expended during physical activity—20%. The sum of these components determines the energy requirement for an individual. Previous recommendations for energy requirements were based on estimates of basal metabolic rate (BMR) or REE derived either by indirect calorimetry or by standard equations.[51] REE can be measured using direct or indirect methods. The direct calorimetric method measures the heat released by a subject at rest and is based on the principle that all energy is eventually converted to heat. In practice, the patient is placed in a thermally isolated chamber, and the heat dissipated is measured for a given period.[52] This method is the true gold standard for measured energy expenditure. Although accurate, direct calorimetry is not practical for hospitalized critically ill patients. On the other hand, REE stimates using standardized World Health Organization (WHO) predictive equations are unreliable, particularly in the underweight subjects.[53–55] Newer equations have attempted to improve the prediction of REE in children by accounting for weight-based groups or including pubertal staging, with variable success.

The challenges of accurate REE estimation are further compounded in critically ill children. Their energy requirements show individual variation and are dependent on severity of injury. For instance, an infant with respiratory distress on pressure support is likely to have a high energy requirement due to increased work of breathing. The same patient when placed on mechanical ventilatory support, with muscle relaxants, is unlikely to have as high an energy requirement. Infants with congenital diaphragmatic hernia on ECMO support have been shown to have energy expenditures approximately 90 kcal/kg/d. Following extubation, the same patients may have energy requirements as high as 140 kcal/kg/d. Although stress factors ranging from 1.0 to 2.7 have been suggested to correct for these variations,[56,57] calculated standardized energy expenditure equations have not been satisfactorily validated in critically ill children.[57–60]

Indirect methods quantify energy production based on O_2 consumption and CO_2 production by the whole body during a specific time interval. Specifically, *indirect calorimetry*, measures VO_2 (the volume of oxygen consumed) and VCO_2 (the volume of CO_2 produced) and uses a correlation factor based on urinary nitrogen excretion to calculate the overall rate of energy production.[61] Historically, indirect calorimetry has

been utilized for energy expenditure estimation. This method employs a *metabolic cart*, a leak-free system with a computer-controlled gas exchange measurement device as well as 24-hour urine collection. Indirect calorimetry provides a measurement of the overall respiratory quotient (RQ), defined as the ratio of CO_2 produced to O_2 consumed, for a given patient. However, this method is not accurate in the setting of air leaks around the endotracheal tube, in some ventilatory circuits, in the setting of a chest tube for pneumothorax, or in subjects on ECMO support. High inspired oxygen fraction (FiO_2 0.6) will also adversely affect indirect calorimetry.

Another method of determining energy expenditure is based on the use of doubly labeled water (DLW), which has been used in humans since the early 1980s.[62] In this technique, orally administered stable isotopes of water ($2H_2O$ and H_2 18O) mix with the body water and the 18O is lost from the body as both water and CO2, whereas the 2H is lost from the body as water. The difference in the rates of loss of the isotopes 18O and 2H from the body reflects the rate of CO_2 production, which can be used to calculate the TEE. This method is noninvasive and isotope dilution is measured over two half-lives of the isotope, giving an average estimate of TEE over a period of few days. Isotope studies using DLW have since been validated and have been followed with intense and skeptical scrutiny.[63–65] However, the DLW method has its limitations and is not widely used.[63] The critically ill child with active capillary leak, decreased urine output, fluid overload, and diuretic use would pose major challenges to the assumptions made in the use of this technique. Moreover, this technique is not widely available, and due to the technical challenges, it is not often used outside dedicated laboratories.

Despite the limitations of available methods, experimental studies have tried to address the issue of estimating energy expenditure in the heterogeneous pediatric intensive care unit (PICU) population. Using ^{13}C-labeled bicarbonate and DLW ($2H_2$ 18O), studies in pediatric surgical patients have successfully measured TEE.[24,65] These techniques have been shown to correlate well with indirect calorimetry and have been used to measure energy expenditure of critically ill neonates in a variety of settings.[66] A common theme in these studies is the observation that the severity and duration of the critical illness govern the energy needs of the ill child. In general, any increase in energy expenditure during illness or after surgery is variable, and recent studies suggest that the increase is far

less than originally hypothesized.

In children with severe burns, the initial REE during the flow phase of injury is increased by 50% but then returns to normal during convalescence.[67] In neonates with bronchopulmonary dysplasia, in which the illness increases the patient's work of breathing, a 25% elevation in energy requirement is evident.[68] Newborns undergoing major surgery have only a transient 20% increase in energy expenditure that returns to baseline values within 12 hours postoperatively provided that no major complications develop.[69,70] Stable, extubated neonates, five days following surgery, have been shown to have REEs comparable to normal infants.[71] Effective anesthetic and analgesic management may play a significant role in muting the stress response of the neonate. Studies have demonstrated no discernible increase in REE in neonates undergoing patent ductus arteriosus ligation who receive intraoperative fentanyl anesthesia and postoperative intravenous analgesic regimens.[70] Finally, by using stable isotopic methods, it has been shown that the mean energy expenditures of critically ill neonates on ECMO are nearly identical to age- and diet-matched nonstressed controls.[22]

In critically ill mechanically ventilated children, use of sedation and muscle paralysis decreases the component of energy requirement related to physical activity,[72] and caloric needs in the critically ill child may be lower than previously considered. Thus, energy requirement estimates in some select groups of patients remain variable and possibly overestimated, leading to risk of overfeeding. Many studies reporting the energetic needs of critically ill children have been conducted on heterogeneous populations under various conditions. For practical purposes, the recommended dietary caloric intake for healthy children represents a reasonable starting point for the upper limit of caloric allotment in the critically ill child.[73]

Since predictive equations, used with stress factors to account for the degrees of illness, have proven inaccurate in determining individual energy expenditures, the preferred method to measure energy expenditure remains indirect calorimetry, when available.[54] This is particularly germane in children with identifiable risk factors for malnutrition. However, due to practical limitations of indirect calorimetry, current recommendation focus on using indirect calorimetry for children with identifiable risk factors for metabolic derangement.[74,75] Once protein needs have been met, safe caloric provisions using carbohydrate and lipid energy sources have similar beneficial effects on net protein synthesis and overall protein balance in critically ill patients. The rational partitioning of these

substrates requires knowledge of carbohydrate and lipid handling in critical illness.

CARBOHYDRATE METABOLISM

Glucose availability is crucial during critical illness. Glycogen stores are limited and quickly depleted resulting in the need for gluconeogenesis. In injured and septic adults, a threefold increase in glucose turnover and oxidation has been demonstrated as well as an elevation in gluconeogenesis.[30,76]

As mentioned earlier, a significant feature of the metabolic stress response is that the provision of dietary glucose does not halt gluconeogenesis. Consequently, the catabolism of muscle protein to produce glucose continues unabated.[30] However, it is clear that a combination of dietary glucose and amino acids effectively improves net protein balance in critical illness. This occurs primarily through the augmentation of protein synthesis and has little, if any, effect on protein breakdown.[33,77]

In early nutritional support regimens for critically ill patients, high glucose allotments were used in an effort to quell gluconeogenesis. The excess glucose was converted to fat, resulting in the net generation of CO_2. The synthesis of fat from glucose has an RQ of approximately 8.7. Clinically, this high RQ is not attained, as glucose is never purely used for fatty acid synthesis. Nonetheless, the provision of excess glucose results in an elevated RQ and thus increases the ventilatory burden on the child. The mean RQ in postsurgical neonates fed a high-glucose diet is approximately 1.0, whereas comparable neonates fed with reduced quantities of glucose in combination with a standard concentration of lipid have an RQ of 0.83.[78] Adult patients fed excess parenteral nutrition calories have a 30% increase in O_2 consumption, a 57% increase in CO_2 production, and a 71% elevation in minute ventilation.[46]

Thus, avoidance of excess calories from glucose provisions and the use of a mixed-fuel system employing both carbohydrates and lipid are theoretically and practically useful in critically ill children, many of whom already have respiratory challenges from the nature of their illness. This approach also helps to alleviate difficulties with hyperglycemia in the relatively insulin-resistant stressed child.

HYPERGLYCEMIA IN CRITICAL ILLNESS: ROLE OF TIGHT GLYCEMIC CONTROL

Hyperglycemia is prevalent in critically ill children including those in the posttraumatic and postsurgical state.[6,79–81] Multiple studies have demonstrated an association of the severity and duration of this hyperglycemia with morbidity and mortality.[79,82,83] Stess hyperglycemia develops in critical illness as a result of increased gluconeogenesis, glycogenolysis, and insulin resistance with subsequent impairment of cellular glucose uptake. These shifts are produced as a result of known changes in circulating counter-regulatory hormones and proinflammatory cytokines. The net increase in serum glucose concentration is an adaptive response to provide fuel to organs and injured tissues during illness. Upregulation of glucose-independent GLUC transport proteins in peripheral tissues leads to cellular glucose excess, which does not occur during normal homeostasis. This, in turn, leads to increased cellular glucose metabolism; increased production of reactive oxygen species; and results in increased apoptosis, systemic inflammation, and phagocyte dysfunction.[84]

Multiple investigators have attempted to curtail the dangerous effects of stress hyperglycemia using strategies of tight glycemic control (TGC). Some prospective studies of critically ill adults have demonstrated reductions in mortality using TGC. However, these studies have also reported increases in hypoglycemic episodes when lower blood glucose levels are targeted.[85,86] Two published prospective studies have evaluated the use of TGC in the pediatric critically ill population. A Belgian study randomized 700 infants and children to either a TGC strategy or a conventional glucose control. They found reductions in hyperglycemia, inflammatory response, secondary infections, PICU length of stay, and mortality in the TGC cohort, but at the cost of a significant increase in hypoglycemia.[87] Another trial from Boston evaluated 980 children after cardiac surgery randomized to standard care or TGC with the aid of continuous glucose monitoring. This study demonstrated a reduction of hyperglycemia and a significant but relatively low risk of hypoglycemia in the TGC group. However, the primary outcome variable of infection was unaffected and mortality, length of stay, and incidence of organ failure were not reduced.[88]

Thus, there appears to be a narrow therapeutic window of glycemic control in which negative sequelae of stress hyperglycemia may be avoided without causing complications of hypoglycemia. Although it is

prudent to avoid prolonged hyperglycemia in the PICU patient, in the presence of conflicting data, aggressive glycemic control cannot be recommended as yet.

LIPID METABOLISM

Analogous to protein and carbohydrate metabolism, lipid turnover is generally accelerated by critical illness, surgery, and trauma.[77] Although lipid use is compromised in the initial, brief ebb phase following acute trauma or sepsis, overall lipid turnover increases dramatically during the predominant flow phase. Adult patients demonstrate two- to fourfold increases in lipid turnover compared with healthy controls, and this increase is proportional to the degree of illness.[89] This process involves the recycling of free fatty acids and glycerol into and from triglycerides. In critically ill children, this increase in lipid turnover has been shown by several studies. Stressed infants and children demonstrate a reduction in RQ, an increased rate of fat oxidation, and a decline in plasma triglyceride levels.[60,90]

This evidence suggests that fats are the prime source of energy in metabolically stressed children. Glycerol, released along with free fatty acids from the breakdown of triglycerides, may be converted to pyruvate and thereby shunted into the gluconeogenesis pathway. Again similar to protein catabolism, the provision of dietary glucose does not decrease glycerol clearance or overall lipid turnover.

Because of the increased demand for lipid use in critical illness coupled with the limited lipid stores in the pediatric patient, critically ill children are susceptible to the evolution of biochemically detected essential fatty acid deficiency if administered a fat-free diet.[91,92] In infants, linoleic and linolenic acid are considered essential, whereas arachidonic acid and docosahexaenoic acid are thought to be conditionally essential. If the body lacks dietary linoleic acid, the formation of arachidonic acid (a tetraene) by desaturation and chain elongation cannot take place. The same pathway then converts available oleic acid to 5,8,11-eicosatrienoic acid (a triene), resulting in an elevated triene to tetraene ratio, which is empirically characteristic of essential fatty acid deficiency.[93] Clinically, this syndrome presents as dermatitis, alopecia, thrombocytopenia, and increased susceptibility to bacterial infection. To avoid essential fatty acid deficiency in critically ill or injured infants, the allotment of linoleic and linolenic acid is recommended at concentrations of 4.5% and 0.5% of total

calories, respectively.[94,95] The provision of commercially available lipid solutions to parenterally fed critically ill children reduces the risk of essential fatty acid deficiency, results in improved protein use, and does not significantly increase CO_2 production or metabolic rate.[96]

There are disadvantages, however, with lipid administration, including hypertriglyceridemia, increased rates of infection, and decreased alveolar oxygen diffusion capacity.[97–99] Most centers start lipid supplementation in ill children at 1.0 g/kg/d and advance over a period of days to 2–3 g/kg/d, with monitoring of triglyceride levels and screening for essential fatty acid deficiency where appropriate. Lipid administration is generally restricted to a maximum of 30%–40% of total calories, although this practice has not been validated by clinical trials.

ELECTROLYTE METABOLISM

Requirements for the basic electrolytes—sodium, potassium, chloride, bicarbonate, and calcium—must be evaluated frequently in the critically ill patient. In addition to routine electrolyte monitoring, careful attention to phosphate and magnesium levels is recommended. Hypophosphatemia may lead to hemolytic anemia and respiratory muscle dysfunction and may also be seen with refeeding syndrome in the ill child. Renal failure can result in the retention of phosphate, and nutritional allotments must be reduced accordingly. Deficiency of magnesium can cause fatal cardiac arrhythmia. Abnormalities of acid–base physiology in the critically ill child can also influence the nutritional regimen. For example, head-injured patients may develop an iatrogenically induced respiratory alkalosis. If a metabolic alkalosis secondary to active diuresis or gastric suction is also present, chloride administration should be used to correct the alkalosis. Untreated alkalemia tends to inhibit the respiratory drive, shift potassium intracellularly, and decrease ionized calcium concentrations by increasing the affinity of albumin for calcium. Metabolic acidosis can also be seen in critically ill children, often from associated hypotension or ischemic etiologies. In this case, the provision of acetate instead of Cl in the PN regimen should be used so as not to worsen the existing acidemia. The provision of excess acetate at 1 mEq/kg/d is usually a safe adjunct to limit metabolic acidosis.

VITAMIN AND TRACE MINERAL METABOLISM

Vitamin and trace mineral metabolism in critically ill and postoperative patients has not been extensively studied. For the neonate and the child, required vitamins include fat-soluble vitamins (A, D, E, and K) and water-soluble vitamins (ascorbic acid, thiamin, riboflavin, pyridoxine, niacin, pantothenate, biotin, folate, and vitamin B_{12}), and these are routinely administered. Because vitamins are not stoichiometrically consumed in biochemical reactions but rather act as catalysts, the administration of large supplements of vitamins in stressed states is not logical from a nutritional standpoint. The trace minerals required for normal development are zinc, iron, copper, selenium, manganese, iodide, molybdenum, and chromium. Trace minerals are used in the synthesis of active sites of a ubiquitous and physiologically essential class of enzymes termed metalloenzymes. As with vitamins, the role of metalloenzymes is to act as catalysts. Hence, unless there are excessive losses, such as increased zinc loss with severe diarrhea, large nutritional requirements in critical illness should not be anticipated. The Dietary Reference Intake of vitamin and trace mineral needs of healthy children and neonates are reviewed periodically and are well defined in the literature.[100] These recommended levels have been used in critically ill patients, and little evidence exists that they are nutritionally inadequate. In children with severe hepatic failure, copper and manganese accumulation can occur; thus, parenteral trace mineral supplementation is often reduced.

BODY COMPOSITION AND METABOLIC RESERVES

Table 62-2 outlines the body composition of the neonate, child, and adult patient.[101–103] The most striking difference in body composition between the adult and the child is the quantity of protein in reserve and available at times of illness. Table 62-3 outlines the relative protein stores in humans at various stages of development.[104] As a percentage of body weight, the protein stores of adults are nearly twice those of the neonate. In contrast, carbohydrate stores remain constant and do not afford significant reserve at any age. Lipid concentrations are low at birth and increase gradually during development; premature infants have even lower proportions of stored lipid at birth. In addition to the reduced macronutrient reserves in the child, both neonates and children have much higher baseline requirements. The REE for neonates is up to three times that for adults when standardized for weight.[105,106]

TABLE 62-2 Body Composition of Neonates, Children, and Non-obese Adults as a Percentage of Total Body Weight

Age	Protein (%)	Fat (%)	Carbohydrate (%)
Neonates	11	14	0.4
Children (Age 10 yr)	15	17	0.4
Adults	18	19	0.4

TABLE 62-3 Proportion of Skeletal Muscle in the Body of Humans at Various Stages of Development

Stage	Proportion of Skeletal Muscle (%)
Birth	21
Weaning	18
Adolescent	36
Adult	45
Elderly	27

This inverse relationship between energy expenditure and body weight is constant throughout mammalian species and is partially related to body surface area. The recommended intakes of macronutrients and water for healthy children and adults can be found in Table 62-4. The protein requirements of the full-term neonate (per kg body weight) are nearly three times that of the adult. Moreover, premature neonates require a minimum of 2.9–3.3 g/kg/d of protein to maintain growth rates similar to *those* in utero, and this is 3.5 times the requirement for protein balance in the adult.[107,108] Thus, the critically ill child has the potential to be much more susceptible to the harmful effects of protracted catabolic stress, and the prompt implementation of adequate nutritional support must be a priority in the patient's care.

Accurate assessment of body composition is necessary for planning ongoing nutritional intake, monitoring dynamic changes in the body compartments (such as ongoing loss of lean body mass), and assessing the adequacy of nutritional supportive regimens during critical illness. Ongoing loss of lean body mass is an indicator of inadequate dietary supplementation and may have clinical implications in the critically ill child.

However, standard measurements such as weight and anthropometrics

are frequently invalid surrogates for lean body mass in the critical care setting. This is largely due to the presence of capillary leak, manifesting as edema and large fluid shifts. Other techniques for assessing body composition (such as dual-energy X-ray absorptiometry (DEXA), biochemical parameters, ultrasound assessment, stable isotope studies, and air displacement plethysmography) are either impractical for clinical use, inaccurate in the setting of critical care, or have not yet been validated in the setting of critical care.[109] Ongoing studies attempt to identify the most appropriate body composition measures in the ICU.

TABLE 62-4 Dietary Reference Intakes (DRIs); Recommended Intakes of Macronutrients and Total Water

Age	Total Water (L/day)	Carbohydrate (g/day)	Fat (g/day)	Linoleic Acid (g/day)	α-Linoleic Acid (g/day)	Protein (g/day)
0–6 mo	0.7	60	3	4.4	0.5	9.1
7–12 mo	0.8	95	30	4.6	0.5	11
1–3 yr	1.3	130	30–40	7	0.7	13
4–8 yr	1.7	130	25–35	10	0.9	19
Males						
9–13 yr	2.4	130	25–35	12	1.2	34
14–18 yr	3.3	130	25–35	16	1.6	52
19–30 yr	3.7	130	25–35	17	1.6	56
31–50 yr	3.7	130	25–35	17	1.6	56
Females						
9–13 yr	2.1	130	25–35	10	1	34
14–18 yr	2.3	130	25–35	11	1.1	46
19–30 yr	2.7	130	25–35	12	1.1	46

31–50 yr	2.7	130	25–35	12	1.1	46

Dietary reference intakes (DRIs) are given and may be used as goals for individual intake.

Source: Adapted from Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein, and Amino Acids (2002/2005). Dietary Reference Intakes for Water, Potassium, Sodium, Chloride, and Sulfate (2005). These reports may be accessed via http://www.nap.edu.

LABORATORY TESTS TO ASSESS NUTRITIONAL STATUS

Serum albumin is frequently used as a tool for nutritional assessment in the ICU. Levels 2.2 g/dL reflect malnutrition. However, the long half-life of albumin (14–20 days) makes it less responsive to acute changes in nutritional status. Serum albumin concentrations may be affected by albumin infusion, dehydration, sepsis, trauma, and liver disease, independent of nutritional status. Thus, its reliability as a marker of visceral protein status is questionable.

Prealbumin (also known as transthyretin or thyroxine- binding prealbumin) is a stable circulating glycoprotein synthesized in the liver. It binds with RBP and is involved in the transport of thyroxine as well as retinol. Prealbumin, so named by its proximity to albumin on an electrophoretic strip, has a half-life of 24–48 hours and reflects more acute nutritional changes. Prealbumin concentration is diminished in renal and liver disease and may be an unreliable marker for nutritional status in these settings. Chemical profile should be monitored on admission and repeated periodically.

Other laboratory tests that help in overall nutritional assessment include serum electrolytes, blood urea nitrogen, glucose, coagulation profile, iron, magnesium, calcium, and phosphate. The adequacy of cellular immunity can be estimated through the measurement of total lymphocyte count (TLC) and by delayed-type hypersensitivity testing with a series of common antigens (e.g., *Candida, Trichophyton*, and tuberculin). However, critically ill children in the postsurgical state may be anergic for many days, regardless of nutritional status.

ROUTES OF NUTRITIONAL PROVISION

Enteral Nutrition

Following the estimation of energy expenditure and requirement in the critically ill child, the next challenge is to facilitate the provision of this

nutritional support. In the critically ill child with a functioning gastrointestinal tract, the enteral route of nutrient administration is preferable to PN. Enteral nutrition is physiologic and has been shown to be more cost–effective without the added risk of nosocomial infection inherent in PN.[110,111] Gastric and postpyloric enteral feeding routes are used. No large studies have demonstrated a benefit of postpyloric feeding tubes over nasogastric tubes. Continuous feedings using standard formulas can adequately nourish the majority of injured patients. Diarrhea can often be avoided by carefully controlling the infusion rate until tolerance is established.

The enteral route has been successfully used for nutritional support of the critically ill child.[112–114] Enterally administered feeds meet nutritional requirements in critically ill children with functional gastrointestinal system and have the advantages of cost, manageability, safety, and preservation of gastrointestinal function. Early introduction of enteral feeds in critically ill patients has been shown to decrease infectious episodes and decrease length of hospital stay in critically ill patients.[115,116] This practice also helps to achieve positive protein and energy balance and restores nitrogen balance during the acute state of illness. It maintains gut integrity and elicits release of growth factors and hormones that maintain gut integrity and function.[117]

Despite its perceived benefits, current studies in ICUs show that a significant proportion of eligible patients do not receive enteral feeds.[118–120] Prolonged fluid resuscitation, interruption of feeds for procedures, and enteral feed intolerance are major factors hindering the achievement of estimated energy requirements despite maximizing the energy content of feeds. Consistently underachieved enteral nutrition goals are thought to be perhaps responsible for the absence of beneficial effect in multiple studies and meta-analysis of the efficacy of immunonutrition in preventing infection.[121]

Enteral feedings are indicated early on the course of critical illness if peristalsis has been established. Feeds can be administered into the stomach or jejunum with the aid of feeding tubes inserted nasally or orally. Intragastric or intrajejunal tip placement is often confirmed by radiograph. Postpyloric feeding is increasingly adopted to feed children with reflux or delayed gastric emptying who are at risk of aspiration.

Surgical placement of gastrostomy or jejunostomy tube allows long-term enteral feeding and administration of drugs in selected patients during intensive care and after discharge from the ICU. Stoma site infection,

obstruction, and tube dislodgement are common complications and must be identified and managed early. Tube tip malposition is frequently encountered with any of these devices either at placement or during the course of its use. Bedside screening methods for correct tip position range from auscultation during air insufflation to ultrasound-guided tip localization. However, feeds should be held when malposition of tip is suspected and when in doubt, radiographic confirmation of correct tip position should be obtained before recommencing feeds.

At the time of extubation, tube feeds are held for six hours to lower the corresponding risk of aspiration. It is also recommended not to use enteral feeds with patients who are hypotensive or who have evidence of bowel ischemia so as to limit the risk of small-bowel necrosis associated with rapid enteral feeding.[122] Intolerance to enteral feeds may be a limiting factor and supplementation with PN in this group of patients allows optimal nutritional intake. In a subgroup of critically ill patients, PN may be required for a period before initiation of enteral feeds. However, many studies have shown inability to achieve the set nutritional goal.

Parenteral Nutrition

PN allows intravenous administration of macro- and micronutrients to meet the nutritional requirements of the body. PN is indicated in children who are unable to tolerate enteral feeds for a prolonged period. Intravenous feeding may supplement enteral feeding or replace it entirely as in PN. Although widespread in its application, PN is associated with mechanical, infectious, and metabolic complications and hence should only be used in carefully selected patients. In the setting of an intact intestinal function, PN is not indicated if enteral feeds alone can maintain nutrition.

Fluid and electrolyte status will guide the initial PN prescription. The patient's hydration, size, age, and underlying disease will dictate the amount of the fluid to be administered. PN should not be used for replacing ongoing losses. PN should be prescribed daily and after reviewing electrolytes and blood sugar to allow adjustments in the macro- and micronutrient composition.

Careful biochemical monitoring is mandatory to prevent acute and long-term complications from PN therapy. A PN profile is recommended at initiation of therapy and then weekly. It includes serum levels of sodium, potassium, chloride, glucose, carbon dioxide, blood urea nitrogen, creatinine, albumin, magnesium, phosphate, total and direct bilirubin, and transaminases. For children requiring PN longer than 30 days, selenium,

iron, zinc, copper, and carnitine levels may be checked. It is essential to monitor daily vital statistics and routine anthropometry to ensure adequate growth and development. Critical care units benefit from the expertise of a dedicated nutritionist, who should be consulted on a regular basis to guide optimal nutritional intake of patients.

Central venous access is required to facilitate the delivery of hyperosmolar PN solutions into a large-bore vein with high- volume blood flow, to prevent thrombosis and phlebitis. The incidences of infective and life-threatening complications related to indwelling central lines have necessitated extreme caution when using central PN.[123–125] Central line tips are located in the superior vena cava, right atrium, or inferior vena cava. Central line tips must be placed by experienced operators and line tip confirmed on radiographs before using them for PN delivery. At our institution, only central lines are recommended for the delivery of infusates with osmolarity higher than 900 mosm/L (10% dextrose, 2% amino acids with standard additives). (Please see Chapters 67, "Enteral Nutrition," and 68, "Parenteral Nutrition," for further reading on enteral and parenteral nutrition.)

IMMUNITY-ENHANCING DIETS FOR THE CRITICALLY ILL CHILD

Immune-enhancing diets (IEDs) have been available for many years and their role in the care of critically ill patients remains controversial. Studies examining their role in critically ill patients suffer from heterogeneous clinical populations, methodological flaws, and the heterogeneous nature of the IED formulations used. Thus meta-analyses of these studies have resulted in conflicting conclusions that do not allow for dispelling current doubts regarding the safety and efficacy of these diets. The commercially available diets contain varied compounds in different doses and the role of individual compounds is impossible to interpret. Furthermore, there is little research evaluating the role of immunonutrition in critically ill children.

A recent meta-analysis of randomized clinical trials examining the efficacy of enteral immunonutrients (fish oil, glutamine, and arginine) in adult ICU, trauma, and burn patients included 3013 subjects from 24 studies.[126] The authors concluded that IEDs have no benefit in the critically ill adult ICU, burn, or trauma patients. In this analysis, only the subgroup of medical ICU patients receiving formulas high in fish oil saw a reduction in mortality, infections, and length of stay.

Arginine is a conditionally essential amino acid. It becomes essential in the setting of sepsis. Its effects, safety, and efficacy in the setting of critical care are ill-defined in the literature, with significant controversy. Some authors have suggested, with experimental support, that arginine supplementation results in nitric oxide–mediated protection against microvascular injury, which is typical of sepsis.[126] Some studies, however, have shown an increase in mortality as a result of arginine supplementation.[127–129] Thus, further research is needed before a definitive recommendation for or against arginine supplementation in critical illness can be made.

Recent studies have also investigated the role of glutamine supplementation in PN. Glutamine, cysteine, and glycine are precursors for glutathione, a major antioxidant. Glutamine is also a precursor for nucleotide synthesis and although a nonessential amino acid, it can become conditionally essential in catabolic states such as sepsis and trauma. Its applicability may be widespread and it has been introduced in PN solutions for its presumed benefits, such as restoration of protein and nitrogen balance, attenuation of gastrointestinal mucosal atrophy, reduced bacterial translocation, and reduction in bacteremia after chemotherapy. Pediatric burn patients have been shown to have deficient peripheral glutamine production. Glutamine supplementation was associated with reduced gram-negative bacteremia in severely burned patients in a double-blinded randomized control trial.[130] Glutamine has also been supplemented in enteral formulations, although only 50% is absorbed into splanchnic circulation when given enterally. The National Institute of Child Health and Development (NICHD) neonatal research network did not find any significant difference in outcomes in a multicenter randomized study of 1430 extremely low birth weight neonates who were randomized to PN containing 20% glutamine or isonitrogenous control.[131] Another large randomized trial of glutamine and antioxidant supplementation in 1223 critically ill adults did not demonstrate any improvement in mortality or secondary outcomes.[132]

OMEGA-3 FATTY ACIDS

Substitution of the intralipid component of PN (rich in proinflammatory omega-6 fatty acids) with fish oil has been demonstrated to alleviate some of the toxic effects of long-term PN on the liver. The beneficial effects of omega-3 fatty acids have been shown in animal and human models and this is an area of great interest. Omega-3 fatty acids have an anti-

inflammatory effect, with decreased cytokine production shown in some models. Several studies have demonstrated that initiation of parenteral fish oil in PN-dependent children with intestinal failure-associated cholestatic liver damage is associated with resolution of hyperbilirubinemia while avoiding essential fatty acid deficiency.[133–136] Other research has shown similar reversal of intestinal failure-associated cholestasis by employing a vegetable-based lipid emulsion at a restricted dose (≤1g/kg/d).[137,138] Prospective randomized clinical trials regarding these lipid management strategies are needed.

CONCLUSIONS

The pediatric metabolic response to critical illness is associated with a significant increase in the whole-body turnover of protein, carbohydrate, and lipid substrates. This process allows for the production of proteins necessary for the inflammatory response and wound healing. Protein loss is commensurate with the severity of illness and injury and is the hallmark of this metabolic process. Interestingly, energy expenditure is not markedly increased during this metabolic response.

Implementing an appropriate nutritional regimen to minimize the protein loss while facilitating recovery is of paramount importance in the long-term management of children suffering from critical illness or major injury. Maintaining positive protein balance in the face of protein loss requires substantially increased protein allotments, especially in neonates. Excessive carbohydrate provisions do not blunt the metabolic response and, indeed, have been shown to have deleterious consequences. In addition, because the energy expenditure of these patients does not appear to increase significantly, the recommended caloric intake for healthy children in a similar age group represents a reasonable starting point for the upper limit of caloric allotment in critically ill children. Nutritional therapy in the critically ill child should be provided via the enteral route where possible. PN may be employed to supplement or replace enteral nutrition in cases where the GI tract is unavailable. Careful monitoring, judicious application, and prompt recourse to enteral feeds, as feasible, will allow safe use of PN in the critically ill child. An interdisciplinary team approach is needed for the ideal nutritional management of the critically ill child.

REFERENCES

1. Pollack MM, Wiley JS, Kanter R, Holbrook PR, et al. Malnutrition, in critically ill infants and children. *JPEN J Parenter Enteral Nutr*. 1982;6(1):20–24.
2. Hulst J, Joosten K, Zimmermann L, et al. Malnutrition in critically ill children: From admission to 6 months after discharge. *Clin Nutr*. 2004;23(2):223–232.
3. de Souza Menezes F, Leite HP, Koch Nogueira PC. Malnutrition as an independent predictor of clinical outcome in critically ill children. *Nutrition*. 2012;28(3):267–270.
4. Pollack MM, Ruttimann UE, Wiley JS. Nutritional depletions in critically ill children: Associations with physiologic instability and increased quantity of care. *JPEN J Parenter Enteral Nutr*. 1985;9(3):309–313.
5. López-Herce J, Prieto MB. Malnutrition in the critically ill child: The importance of enteral nutrition. *Int J Environ Res Public Health*. 2011;8(11):4353–4366.
6. Cuthbertson D. Intensive-care-metabolic response to injury. *Br J Surg*. 1970;57(10):718–721.
7. Cuthbertson DP. Post-shock metabolic response. *The Lancet*. 1942;239(6189):433–437.
8. Agus MSD, Jaksic T. Nutritional support of the critically ill child. *Curr Opin Pediatr*. 2002;14(4):470–481.
9. O'Connell RC, Morgan AP, Aoki TT, et al. Nitrogen conservation in starvation: graded responses to intravenous glucose. *J Clin Endocrinol Metab*. 1974;39(3):555–563.
10. Framson CMH, LeLeiko NS, Dallal GE, et al. Energy expenditure in critically ill children. *Pediatr Crit Care Med*. 2007;8(3):264–267.
11. Vazquez Martinez JL, Martinez-Romillo PD, Diez Sebastian J, Ruza Tarrio F. Predicted versus measured energy expenditure by continuous, online indirect calorimetry in ventilated, critically ill children during the early postinjury period. *Pediatr Crit Care Med*. 2004;5(1):19–27.
12. Hardy CM, Dwyer J, Snelling LK, et al. Pitfalls in predicting resting energy requirements in critically ill children: A comparison of predictive methods to indirect calorimetry. *Nutr Clin Pract*. 2002;17(3):182–189.
13. Klein GW, Hojsak JM, Schmeidler J, Rapaport R. Hyperglycemia and outcome in the pediatric intensive care unit. *J Pediatr*. 2008;153(3):379–384.
14. Old LJ. Tumor necrosis factor (TNF). *Science*. 1985;230(4726):630–632.
15. Baracos V, Rodemann HP, Dinarello CA, Goldberg AL. Stimulation of muscle protein degradation and prostaglandin E2 release by leukocytic pyrogen (interleukin-1). A mechanism for the increased degradation of muscle proteins during fever. *N Engl J Med*. 1983;308(10):553–558.
16. Anand KJ, Hickey PR. Pain and its effects in the human neonate and fetus. *N Engl J Med*. 1987;317(21):1321–1329.
17. Anand KJ, Brown MJ, Bloom SR, Aynsley-Green A. Studies on the hormonal regulation of fuel metabolism in the human newborn infant undergoing anaesthesia and surgery. *Horm Res* 1985;22(1–2):115–128.
18. Srinivasan G, Jain R, Pildes RS, Kannan CR. Glucose homeostasis during anesthesia and surgery in infants. *J Pediatr Surg*. 1986;21(8):718–721.
19. Black PR, Brooks DC, Bessey PQ, et al. Mechanisms of insulin resistance following injury. *Ann Surg*. 1982;196(4):420–435.
20. Jahoor F, Shangraw RE, Miyoshi H, et al. Role of insulin and glucose oxidation in mediating the protein catabolism of burns and sepsis. *Am J Physiol*. 1989;257(3 Pt 1):E323–331.
21. Sweed Y, Puri P, Reen DJ. Early induction of IL-6 in infants undergoing major abdominal surgery. *J Pediatr Surg*. 1992;27(8):1033–1036.

22. Jaksic T, Shew SB, Keshen TH, et al. Do critically ill surgical neonates have increased energy expenditure? J *Pediatr Surg*. 2001; 36(1):63–67.
23. Denne SC, Kalhan SC. Leucine metabolism in human newborns. *Am J Physiol*. 1987;253(6 Pt 1):E608–615.
24. Keshen TH, Miller RG, Jahoor F, Jaksic T. Stable isotopic quantitation of protein metabolism and energy expenditure in neonates on- and post-extracorporeal life support. *J Pediatr Surg*. 1997;32(7):958–962.
25. Jaksic T, Wagner DA, Burke JF, Young VR. Proline metabolism in adult male burned patients and healthy control subjects. *Am J Clin Nutr*. 1991;54(2):408–413.
26. Williamson DH, Farrell R, Kerr A, Smith R. Muscle-protein catabolism after injury in man, as measured by urinary excretion of 3-methylhistidine. *Clin Sci Mol Med*. 1977;52(5):527–533.
27. Felig P. The glucose-alanine cycle. *Metabolism*. 1973;22(2):179–207.
28. Windmueller HG, Spaeth AE. Uptake and metabolism of plasma glutamine by the small intestine. *J Biol Chem*. 1974;249(16):5070–5079.
29. Keshen T, Miller R, Jahoor F, et al. Glucose production and gluconeogenesis are negatively related to body weight in mechanically ventilated, very low birth weight neonates. *Pediatr Res*. 1997;41(1):132–138.
30. Long CL, Kinney JM, Geiger JW. Nonsuppressability of gluconeogenesis by glucose in septic patients. *Metabolism*. 1976;25(2):193–201.
31. van Lingen RA, van Goudoever JB, Luijendijk IH, et al. Effects of early amino acid administration during total parenteral nutrition on protein metabolism in preterm infants. *Clin Sci* (Lond). 1992;82(2):199–203.
32. Rivera A, Bell EF, Bier DM. Effect of intravenous amino acids on protein metabolism of preterm infants during the first three days of life. *Pediatr Res*. 1993;33(2):106–111.
33. Duffy B, Pencharz P. The effects of surgery on the nitrogen metabolism of parenterally fed human neonates. *Pediatr Res*. 1986;20(1):32–35.
34. Daley SE, Pearson AD, Craft AW, et al. Whole body protein metabolism in children with cancer. *Arch Dis Child*. 1996;75(4):273–281.
35. Premji S, Fenton T, Sauve R. Does amount of protein in formula matter for low-birthweight infants? A Cochrane systematic review. *JPEN J Parenter Enteral Nutr*. 2006;30(6):507–514.
36. Premji SS, Fenton TR, Sauve RS. Higher versus lower protein intake in formula-fed low birth weight infants. *Cochrane Database Syst Rev*. 2006(1):CD003959.
37. Reeds PJ, Berthold HK, Boza JJ, et al. Integration of amino acid and carbon intermediary metabolism: Studies with uniformly labeled tracers and mass isotopomer analysis. *Eur J Pediatr*. 1997;156 Suppl 1:50–58.
38. Miller RG, Jahoor F, Jaksic T. Decreased cysteine and proline synthesis in parenterally fed, premature infants. *J Pediatr Surg*. 1995;30(7):953–957; discussion 957–958.
39. Miller RG, Keshen TH, Jahoor F, et al. Compartmentation of endogenously synthesized amino acids in neonates. *J Surg Res*. 1996;63(1):199–203.
40. Zlotkin SH, Anderson GH. The development of cystathionase activity during the first year of life. *Pediatr Res*. 1982;16(1):65–68.
41. Shew SB, Keshen TH, Jahoor F, Jaksic T. Assessment of cysteine synthesis in very low-birth weight neonates using a [^{13}C6]glucose tracer. *J Pediatr Surg*. 2005;40(1):52–56.
42. Piton G, Manzon C, Monnet E, et al. Plasma citrulline kinetics and prognostic value in critically ill patients. *Intensive Care Med*. 2010; 36(4):702–706.
43. Ware LB, Magarik JA, Wickersham N, et al. Low plasma citrulline levels are

associated with acute respiratory distress syndrome in patients with severe sepsis. *Crit Care*. 2013;17(1:R10).
44. Cynober L. Citrulline: Just a biomarker or a conditionally essential amino acid and a pharmaconutrient in critically ill patients? *Crit Care*. 2013;17(2):122.
45. Calloway DH, Spector H. Nitrogen balance as related to caloric *Am J Clin Nutr*. 1954;2(6):405–412.
46. Askanazi J, Rosenbaum SH, Hyman AI, et al. Respiratory changes induced by the large glucose loads of total parenteral nutrition. *JAMA*. 1980; 243(14):1444–1447.
47. Shew SB, Keshen TH, Jahoor F, Jaksic T. The determinants of protein catabolism in neonates on extracorporeal membrane oxygenation. *J Pediatr Surg*. 1999;34(7):1086–1090.
48. Choban PS, Dickerson RN. Morbid obesity and nutrition support: is bigger different? *Nutr Clin Pract*. 2005;20(4):480–487.
49. McClave SA, Martindale RG, Vanek VW, et al. Guidelines for the provision and assessment of nutrition support therapy in the adult critically ill patient: Society of Critical Care Medicine (SCCM) and American Society for Parenteral and Enteral Nutrition (A.S.P.E.N.). *JPEN J Parenter Enteral Nutr*. 2009;33(3):277–316.
50. Bechard LJ, Rothpletz-Puglia P, Touger-Decker R, et al. Influence of obesity on clinical outcomes in hospitalized children: A systematic review. *JAMA Pediatr*. 2013;167(5): 476–482.
51. Schofield WN. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr*. 1985;39 Suppl 1:5–41.
52. Seale JL, Rumpler WV. Comparison of energy expenditure measurements by diet records, energy intake balance, doubly labeled water and room calorimetry. *Eur J Clin Nutr*. 1997;51(12):856–863.
53. Daly JM, Heymsfield SB, Head CA, et al. Human energy requirements: Overestimation by widely used prediction equation. *Am J Clin Nutr*. 1985;42(6):1170–1174.
54. Hunter DC, Jaksic T, Lewis D, et al. Resting energy expenditure in the critically ill: Estimations versus measurement. *Br J Surg*. 1988; 75(9):875–878.
55. Muller MJ, Bosy-Westphal A, Klaus S, et al. World Health Organization equations have shortcomings for predicting resting energy expenditure in persons from a modern, affluent population: generation of a new reference standard from a retrospective analysis of a German database of resting energy expenditure. *Am J Clin Nutr*. 2004;80(5):1379–1390.
56. Elwyn DH, Kinney JM, Askanazi J. Energy expenditure in surgical patients. *Surg Clin North Am*. 1981;61(3):545–556.
57. Tilden SJ, Watkins S, Tong TK, Jeevanandam M. Measured energy expenditure in pediatric intensive care patients. *Am J Dis Child*. 1989; 143(4):490–492.
58. Phillips R, Ott L, Young B, Walsh J. Nutritional support and measured energy expenditure of the child and adolescent with head injury. *J Neurosurg*. 1987;67(6):846–851.
59. Chwals WJ, Bistrian BR. Predicted energy expenditure in critically ill children: Problems associated with increased variability. *Crit Care Med*. 2000;28(7):2655–2656.
60. Coss-Bu JA, Klish WJ, Walding D, et al. Energy metabolism, nitrogen balance, and substrate utilization in critically ill children. *Am J Clin Nutr*. 2001;74(5):664–669.
61. Ferrannini E. The theoretical bases of indirect calorimetry: A review. *Metabolism*. 1988;37(3):287–301.
62. Schoeller DA, van Santen E. Measurement of energy expenditure in humans by

doubly labeled water method. *J Appl Physiol*. 1982;53(4):955–959.
63. Schoeller DA, Hnilicka JM. Reliability of the doubly labeled water method for the measurement of total daily energy expenditure in free-living subjects. *J Nutr*. 1996;126(1):354.
64. Schoeller DA, Kushner RF, Jones PJ. Validation of doubly labeled water for measuring energy expenditure during parenteral nutrition. *Am J Clin Nutr*. 1986;44(2):291–298.
65. Jones PJ, Winthrop AL, Schoeller DA, et al. Validation of doubly labeled water for assessing energy expenditure in infants. *Pediatr Res*. 1987; 21(3):242–246.
66. Shew SB, Beckett PR, Keshen TH, et al. Validation of a [^{13}C]bicarbonate tracer technique to measure neonatal energy expenditure. *Pediatr Res*. 2000;47(6):787–791.
67. Jahoor F, Desai M, Herndon DN, Wolfe RR. Dynamics of the protein metabolic response to burn injury. *Metabolism*. 1988;37(4):330–337.
68. Weinstein MR, Oh W. Oxygen consumption in infants with bronchopulmonary dysplasia. *J Pediatr*. 1981;99(6):958–961.
69. Jones MO, Pierro A, Hammond P, Lloyd DA. The metabolic response to operative stress in infants. *J Pediatr Surg*. 1993;28(10):1258–1262.
70. Shew SB, Keshen TH, Glass NL, et al. Ligation of a patent ductus arteriosus under fentanyl anesthesia improves protein metabolism in premature neonates. *J Pediatr Surg*. 2000;35(9):1277–1281.
71. Pierro A, Carnielli V, Filler RM, et al. Partition of energy metabolism in the surgical newborn. *J Pediatr Surg*. 1991;26(5):581–586.
72. Goran MI, Kaskoun M, Johnson R. Determinants of resting energy expenditure in young children. *J Pediatr*. 1994;125(3):362–367.
73. Food and Nutrition Board, Commission on Life Sciences, National Research Council. Recommended Dietary Allowances. 10th ed. Washington, DC: National Academy Press; 1989.
74. Mehta NM, Compher C. A.S.P.E.N. Clinical Guidelines: Nutrition support of the critically ill child. *JPEN J Parenter Enteral Nutr*. 2009;33(3):260–276.
75. Kyle UG, Arriaza A, Esposito M, Coss-Bu JA. Is indirect calorimetry a necessity or a luxury in the pediatric intensive care unit? *JPEN J Parenter Enteral Nutr*. 2012;36(2):177–182.
76. Long CL, Spencer JL, Kinney JM, Geiger JW. Carbohydrate metabolism in man: Effect of elective operations and major injury. *J Appl Physiol*. 1971;31(1):110–116.
77. Jeevanandam M, Young DH, Schiller WR. Nutritional impact on the energy cost of fat fuel mobilization in polytrauma victims. *J Trauma*. 1990;30(2):147–154.
78. Jones MO, Pierro A, Garlick PJ, et al. Protein metabolism kinetics in neonates: Effect of intravenous carbohydrate and fat. *J Pediatr Surg*. 1995;30(3):458–462.
79. Srinivasan V, Spinella PC, Drott HR, et al. Association of timing, duration, and intensity of hyperglycemia with intensive care unit mortality in critically ill children. *Pediatr Crit Care Med*. 2004;5(4):329–336.
80. Preissig CM, Rigby MR. Pediatric critical illness hyperglycemia: Risk factors associated with development and severity of hyperglycemia in critically ill children. *J Pediatr*. 2009;155(5):734–739.
81. Ognibene KL, Vawdrey DK, Biagas KV. The association of age, illness severity, and glycemic status in a pediatric intensive care unit. *Pediatr Crit Care Med*. 2011;12(6):386–390.
82. Faustino EV, Apkon M. Persistent hyperglycemia in critically ill children. *J Pediatr*. 2005;146(1):30–34.

83. Turina M, Fry DE, Polk HC. Acute hyperglycemia and the innate immune system: Clinical, cellular, and molecular aspects. *Crit Care Med*. 2005;33(7):1624–1633.
84. Srinivasan V. Stress hyperglycemia in pediatric critical illness: The intensive care unit adds to the stress! *J Diabetes Sci Technol*. 2012;6(1):37–47.
85. van den Berghe G, Wouters P, Weekers F, et al. Intensive insulin therapy in critically ill patients. *N Engl J Med.* 2001;345(19):1359–1367.
86. Finfer S, Chittock DR, Su SY-S, et al. Intensive versus conventional glucose control in critically ill patients. *N Engl J Med*. 2009;360(13):1283–1297.
87. Vlasselaers D, Milants I, Desmet L, et al. Intensive insulin therapy for patients in paediatric intensive care: A prospective, randomised controlled study. *Lancet*. 2009;373(9663):547–556.
88. Agus MSD, Steil GM, Wypij D, et al. Tight glycemic control versus standard care after pediatric cardiac surgery. *N Engl J Med*. 2012; 367(13):1208–1219.
89. Nordenstrom J, Carpentier YA, Askanazi J, et al. Metabolic utilization of intravenous fat emulsion during total parenteral nutrition. *Ann Surg*. 1982;196(2):221–231.
90. Powis MR, Smith K, Rennie M, et al. Effect of major abdominal operations on energy and protein metabolism in infants and children. *J Pediatr Surg*. 1998;33(1):49–53.
91. Paulsrud JR, Pensler L, Whitten CF, et al. Essential fatty acid deficiency in infants induced by fat-free intravenous feeding. *Am J Clin Nutr*. 1972; 25(9):897–904.
92. Friedman Z, Danon A, Stahlman MT, Oates JA. Rapid onset of essential fatty acid deficiency in the newborn. *Pediatrics*. 1976;58(5):640–649.
93. Holman RT. The ratio of trienoic: Tetraenoic acids in tissue lipids as a measure of essential fatty acid requirement. *J Nutr*. 1960;70:405–410.
94. Aggett PJ, Haschke F, Heine W, et al. Comment on the content and composition of lipids in infant formulas. ESPGAN Committee on Nutrition. *Acta Paediatr Scand*. 1991;80(8–9):887–896.
95. Lee EJ, Simmer K, Gibson RA. Essential fatty acid deficiency in parenterally fed preterm infants. *J Paediatr Child Health*. 1993;29(1):51–55.
96. Van Aerde JE, Sauer PJ, Pencharz PB, et al. Metabolic consequences of increasing energy intake by adding lipid to parenteral nutrition in full-term infants. *Am J Clin Nutr*. 1994;59(3):659–662.
97. Cleary TG, Pickering LK. Mechanisms of intralipid effect on polymorphonuclear leukocytes. *J Clin Lab Immunol.* 1983;11(1):21–26.
98. Periera GR, Fox WW, Stanley CA, et al. Decreased oxygenation and hyperlipemia during intravenous fat infusions in premature infants. *Pediatrics*. 1980;66(1):26–30.
99. Freeman J, Goldmann DA, Smith NE, et al. Association of intravenous lipid emulsion and coagulase-negative staphylococcal bacteremia in neonatal intensive care units. *N Engl J Med*. 1990;323(5):301–308.
100. Trumbo P, Yates AA, Schlicker S, Poos M. Dietary reference intakes: vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. *J Am Diet Assoc*. 2001;101(3):294–301.
101. Forbes GB, Bruining GJ. Urinary creatinine excretion and lean body mass. *Am J Clin Nutr*. 1976;29(12):1359–1366.
102. Fomon SJ, Haschke F, Ziegler EE, Nelson SE. Body composition of reference children from birth to age 10 years. *Am J Clin Nutr*. 1982;35(5 Suppl):1169–1175.
103. Munro HN. Nutrition and muscle protein metabolism: Introduction. *Fed Proc*. 1978;37(9):2281–2282.

104. Munro HN. Fifth annual Jonathan E. Rhoads lecture. Metabolic integration of organs in health and disease. *JPEN J Parenter Enteral Nutr*. 1982;6(4):271–279.
105. Reichman B, Chessex P, Verellen G, et al. Dietary composition and macronutrient storage in preterm infants. *Pediatrics*. 1983;72(3):322–328.
106. Schulze KF, Stefanski M, Masterson J, et al. Energy expenditure, energy balance, and composition of weight gain in low birth weight infants fed diets of different protein and energy content. *J Pediatr*. 1987;110(5):753–759.
107. Kashyap S, Schulze KF, Forsyth M, et al. Growth, nutrient retention, and metabolic response in low birth weight infants fed varying intakes of protein and energy. *J Pediatr*. 1988;113(4):713–721.
108. Kashyap S. Enteral intake for very low birth weight infants: What should the composition be? *Semin Perinatol*. 2007;31(2):74–82.
109. Bechard LJ, Parrott JS, Mehta NM. Systematic review of the influence of energy and protein intake on protein balance in critically ill children. *J Pediatr*. 2012;161(2):333–339.
110. Kawagoe JY, Segre CA, Pereira CR, et al. Risk factors for nosocomial infections in critically ill newborns: A 5-year prospective cohort study. *Am J Infect Control*. 2001;29(2):109–114.
111. de Lucas C, Moreno M, Lopez-Herce J, et al. Transpyloric enteral nutrition reduces the complication rate and cost in the critically ill child. *J Pediatr Gastroenterol Nutr*. 2000;30(2):175–180.
112. Briassoulis G, Zavras N, Hatzis T. Malnutrition, nutritional indices, and early enteral feeding in critically ill children. *Nutrition*. 2001;17(7–8):548–557.
113. Briassoulis GC, Zavras NJ, Hatzis TD. Effectiveness and safety of a protocol for promotion of early intragastric feeding in critically ill children. *Pediatr Crit Care Med*. 2001;2(2):113–121.
114. Chellis MJ, Sanders SV, Webster H, et al. Early enteral feeding in the pediatric intensive care unit. *JPEN J Parenter Enteral Nutr*. 1996; 20(1):71–73.
115. Zaloga GP. Early enteral nutritional support improves outcome: Hypothesis or fact? *Crit Care Med*. 1999;27(2):259–261.
116. Sanchez C, Lopez-Herce J, Carrillo A, et al. Early transpyloric enteral nutrition in critically ill children. *Nutrition*. 2007;23(1):16–22.
117. Briassoulis G, Tsorva A, Zavras N, Hatzis T. Influence of an aggressive early enteral nutrition protocol on nitrogen balance in critically ill children. *J Nutr Biochem*. 2002;13(9):560.
118. de Groof F, Joosten KF, Janssen JA, et al. Acute stress response in children with meningococcal sepsis: Important differences in the growth hormone/insulin-like growth factor I axis between nonsurvivors and survivors. *J Clin Endocrinol Metab*. 2002;87(7):3118–3124.
119. Rogers EJ, Gilbertson HR, Heine RG, Henning R. Barriers to adequate nutrition in critically ill children. *Nutrition*. 2003;19(10):865–868.
120. Heyland DK, Cook DJ, Guyatt GH. Enteral nutrition in the critically ill patient: a critical review of the evidence. *Intensive Care Med*. 1993;19(8):435–442.
121. Atkinson S, Sieffert E, Bihari D. A prospective, randomized, double-blind, controlled clinical trial of enteral immunonutrition in the critically ill. Guy's Hospital Intensive Care Group. *Crit Care Med*. 1998;26(7):1164–1172.
122. Munshi IA, Steingrub JS, Wolpert L. Small bowel necrosis associated with early postoperative jejunal tube feeding in a trauma patient. *J Trauma*. 2000;49(1):163–165.
123. Agarwal KC, Khan MA, Falla A, Amato JJ. Cardiac perforation from central venous catheters: Survival after cardiac tamponade in an infant. *Pediatrics*.

1984;73(3):333–338.
124. Collier PE, Ryan JJ, Diamond DL. Cardiac tamponade from central venous catheters. Report of a case and review of the English literature. *Angiology*. 1984;35(9):595–600.
125. Ziegler M, Jakobowski D, Hoelzer D, et al. Route of pediatric parenteral nutrition: Proposed criteria revision. *J Pediatr Surg*. 1980;15(4): 472–476.
126. Marik PE, Zaloga GP. Immunonutrition in critically ill patients: A systematic review and analysis of the literature. *Intensive Care Med*. 2008;34(11):1980–1990.
127. Heyland DK, Samis A. Does immunonutrition in patients with sepsis do more harm than good? *Intensive Care Med*. 2003;29(5):669–671.
128. Heyland DK. Immunonutrition in the critically ill patient: Putting the cart before the horse? *Nutr Clin Pract*. 2002;17(5):267–272.
129. Heyland DK, Novak F. Immunonutrition in the critically ill patient: More harm than good? *JPEN J Parenter Enteral Nutr*. 2001;25 (2 Suppl):S51–55; discussion S55–56.
130. Tubman TR, Thompson SW, McGuire W. Glutamine supplementation to prevent morbidity and mortality in preterm infants. *Cochrane Database Syst Rev*. 2005(1):CD001457.
131. Poindexter BB, Thompson SW, McGuire W. Parenteral glutamine supplementation does not reduce the risk of mortality or late-onset sepsis in extremely low birth weight infants. *Pediatrics*. 2004;113(5):1209–1215.
132. Heyland D, Muscedere J, Wischmeyer PE, et al. A randomized trial of glutamine and antioxidants in critically ill patients. *N Engl J Med*. 2013;368(16):1489–1497.
133. Gura KM, Duggan CP, Collier SB, et al. Reversal of parenteral nutrition-associated liver disease in two infants with short bowel syndrome using parenteral fish oil: Implications for future management. *Pediatrics*. 2006;118(1):197–201.
134. Le HD, de Meijer VE, Robinson EM, et al. Parenteral fish-oil-based lipid emulsion improves fatty acid profiles and lipids in parenteral nutrition-dependent children. *Am J Clin Nutr*. 2011;94(3):749–758.
135. Reniers D, Rajakumar I, Ratko S, Atkison P. Use of parenteral fish oil to reverse cholestasis induced by parenteral nutrition in infants with intestinal failure: Single-centre case series. *Can J Hosp Pharm*. 2012;65(1):27–30.
136. Sant'Anna AMGA, Altamimi E, Clause R-F, et al. Implementation of a multidisciplinary team approach and fish oil emulsion administration in the management of infants with short bowel syndrome and parenteral nutrition-associated liver disease. *Can J Gastroenterol*. 2012;26(5):277–280.
137. Nehra D, Fallon EM, Carlson SJ, et al. Provision of a soy-based intravenous lipid emulsion at 1 g/kg/d does not prevent cholestasis in neonates. *JPEN J Parenter Enteral Nutr*. 2013;37(4):498–505.
138. Cober MP, Killu G, Brattain A, et al. Intravenous fat emulsions reduction for patients with parenteral nutrition-associated liver disease. *J Pediatr*. 2012;160(3):421–427.

Hyperlipidemia and Cardiovascular Disease

63

Sarah D. de Ferranti, MD, MPH

Cardiovascular disease (CVD) is a major cause of adult morbidity and mortality in the United States. Although rates of CVD mortality are decreasing, coronary heart disease (CHD) was the cause of death for more than 380,000 people in 2009, explaining 1 in 6 deaths.[1,2] Other atherosclerotic complications, including stroke and peripheral vascular disease, affect millions more.[2] Atherosclerotic CVD is multifactorial; dyslipidemia—high triglyceride (TG), high low-density lipoprotein cholesterol (LDL-C), and low high-density lipoprotein cholesterol (HDL-C)—is an important contributor. Elevated total cholesterol (TC) greater than 240 mg/dL was found in half of the Framingham Heart Study adult cohort who sustained a myocardial infarction;[3] interventions that reduced cholesterol by 8% reduced the risk of CHD by 19%.[4] Nutrition is an important determinant of dyslipidemia and has important effects on other CVD risk factors including hypertension and obesity. Given the ongoing childhood obesity epidemic,[5] pediatric and adolescent health-care providers can be assured that an important fraction of their clinical practice is at risk for diabetes and atherosclerotic vascular disease and that promoting improvements in nutrition and exercise is the first-line approach to treat—and ideally prevent—the cardiovascular risk factors in youth.

In this chapter, we describe how atherosclerotic vascular changes can begin in childhood based on evidence of preclinical disease by noninvasive testing, suggesting that early intervention is warranted on a population

basis and for individuals with identified risk factors. We briefly review lipid biochemistry and describe different pediatric hyperlipidemias, both primary and secondary. We discuss screening for lipid disorders, as recommended by the most recent guidelines related to pediatric lipid disorders, the National Heart, Lung, and Blood Institute (NHLBI) Expert Panel Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents (http://www.nhlbi.nih.gov/guidelines/cvd_ped/peds_guidelines_sum.pdf).[6] We review nutritional and pharmacologic treatments and lay out some practical approaches to treating children with hyperlipidemia including distinct dietary recommendations for elevated LDL-C and elevated TG in childhood.

ATHEROSCLEROSIS DURING CHILDHOOD

Atherosclerosis usually takes decades to develop. Symptoms and sequelae from atherosclerotic vascular disease are extremely rare in young persons and are limited to patients with profound genetic dyslipidemias. However, the disease pathophysiology itself and the nutritional habits that propagate the disease begin early. Autopsy studies of cohorts as early as the Korean War have shown atherosclerotic coronary artery disease in the second decade of life on gross examination[7] and by microscopy.[8,9] These pathology findings correlate with hyperlipidemia, as demonstrated in the Bogalusa cohort[10] and the Pathological Determinants of Atherosclerosis in Youth (PDAY) study,[11] among others.[12] In the PDAY study, fatty streak precursor lesions were present by age 15 years in the coronary arteries of children who died of noncardiac causes. Both the prevalence and the extent of fatty streaks and the fibrous plaques in the right coronary artery and in the thoracic aorta were directly related to non-HDL-C and decreased HDL-C, as measured at the time of death. A more recent autopsy study of US war veterans killed in combat suggests that the prevalence of atherosclerotic lesions has declined over the past several decades, although there is still an important atherosclerotic burden in young adults.[13,14]

PEDIATRIC DYSLPIDEMIA AND CARDIOVASCULAR DISEASE

Epidemiologic studies have shown that countries with higher rates of CHD

have higher pediatric population mean TC levels.[15] Cholesterol levels in the pediatric age group are important in part because they predict adult cholesterol levels. The Muscatine and Bogalusa cohorts showed cholesterol "tracks" reasonably well[16]; nearly half of Muscatine subjects with high cholesterol levels as children had high cholesterol levels as adults.[17] Childhood cholesterol levels may also be conceived as an indicator of lifetime cholesterol exposure.

There are data connecting cholesterol levels in young adults with future CHD. Johns Hopkins University medical students with high TC had a greater risk of CHD, mortality from CHD, and overall mortality over an average of 30 years of follow-up.[18] Similarly, the Princeton Lipid Research Clinic showed that childhood factors including abnormal cholesterol are a risk for early CVD.[19] These studies provide the most compelling link between measured cholesterol levels in young persons and subsequent risk of heart disease 30 or 40 years later. The Special Torku Coronary Risk Factor Intervention Project (STRIP) study showed infant boys whose parents were randomly assigned to receive counseling on low-saturated-fat diets had lower LDL-C and TC as adolescents, compared to a control group, by approximately 5%.[20] To date, there are no long-term studies demonstrating treatment for dyslipidemia during childhood results in a reduction in adult CVD events, due to the length of follow-up and the number of study participants needed to successfully answer these questions. Therefore, many of the recommendations we discuss here are based on inference, adult studies, and short-term interventions in the young.

HIGH-RISK PATIENTS

In adults, guidelines for primary and secondary prevention of CVD depend on accompanying cardiovascular risk factors. This concept has been applied to evaluating and treating lipid disorders in children,[6,21] although there are less data on which to base specific recommendations. Guidelines suggest when to initiate treatment and what target to use when treating lipid disorders; abnormal lipids should be modified in these high-risk groups. These include heart transplant recipients[22,23] and renal transplantation patients,[24,25] in whom the problem is related only in part to chronic transplant rejection. Children with previous Kawasaki disease who had or continue to have coronary aneurysms are another high-risk group.[26] Some types of congenital heart diseases are associated with an increased

risk of atherosclerosis even if repaired, such as coarctation.[27] Coronary manipulation as part of arterial switch operations for transposition of the great arteries may also make the coronaries more vulnerable.[28–30] Children with diabetes mellitus type 1 and type 2, both increasingly common in adolescence, are also at particular risk for CVD as young adults.[31] Children who have been treated for cancer during childhood are more vulnerable than the general population to future CVD and its precursors, including lipid disorders, hypertension, and central adiposity.[32,33] In assessing risk for individual children, solid organ transplantation and diabetes mellitus types 1 and 2 can reasonably be considered very high risk conditions in determining whether a child is a candidate for pharmacologic therapy and should lower the therapeutic cutpoint for treatment. For example, it is our practice to place all children on pravastatin after heart transplantation, regardless of cholesterol levels, due to the risk of coronary artery pathology and arteriopathy in this population.[34]

OBESITY, METABOLIC SYNDROME, AND TYPE 2 DIABETES MELLITUS

Obesity is associated with dyslipidemia, specifically hypertriglyceridemia and low HDL as well as a mild to moderate increase in LDL-C; other or additional diagnoses should be considered if the LDL is significantly elevated. In patients who are genetically prone to hypertriglyceridemia or have a family history of type 2 diabetes, trends toward “super-sized” fast-food meals, juice, and sedentary lifestyle are highly detrimental; overweight or obesity simply adds to this.

Metabolic syndrome is a term used to describe a collection of cardiovascular risk factors associated with a high rate of atherosclerotic disease and diabetes mellitus type 2. Childhood metabolic syndrome has been associated with adult metabolic syndrome.[19] There is controversy about pediatric metabolic syndrome as a concept, and many definitions have been proposed but none are generally agreed on.[35] Most proposed definitions, similar to the adult definitions,[36] include hypertension, disordered glucose metabolism, central obesity, low HDL, and high TG levels. Estimates of the prevalence of a pediatric metabolic syndrome in adolescents range between 4% and 12.7% of the population aged 13-19 years, depending on the population and the definition used.[37–38] Childhood obesity is associated with a greater burden of childhood metabolic risk factors and also associated with adult metabolic syndrome.[39] The main

clinical use of the concept of metabolic syndrome in childhood is to alert the clinician to the clustering of risk factors so that he or she can screen for other cardiovascular risk factors.

EMERGING CONCEPTS IN ATHEROSCLEROSIS: APPLICATIONS IN CHILDREN AND ADOLESCENTS

Abnormal cholesterol is an important CVD risk factor; however, half of the adults in the Framingham Heart Study who had myocardial infarctions had relatively normal cholesterol levels,[3] a finding supported by other studies.[40,41] This has prompted researchers to search for other contributions to atherosclerosis that might affect atherosclerosis accumulation, and eventually events.

Inflammation is an important physiologic process in atherosclerosis. It has been clinically assessed by measuring C-reactive protein (CRP). In adults categorized as moderate risk by the Framingham Risk Score, a CRP adds to CVD event risk prediction.[42,43] The relative risk of future CVD events is low in those with hsCRP levels less than 1 mg/L, average in those with hsCRP levels of 1.0–3.0 mg/L, and high in those with hsCRP greater than 3.0 mg/L.[44] No specific cardiovascular CRP cut point has been identified in youth because of the lack of long-term follow-up outcomes data, but it is likely that chronic inflammation in youth contributes to atherosclerosis. Offspring of first-degree relatives who had myocardial infarctions were found to have abnormally elevated CRP levels, even when controlling for body mass index.[45] Another study found that CRP was elevated in children with multiple cardiovascular risk factors.[46] At present, CRP is not recommended for clinical use in the assessment of the child for risk of future CVD,[47] although the test might arguably have some utility in individuals with high-risk family history and borderline lipid levels. It is worth bearing in mind that in clinical practice children with increased hsCRP levels may have an intercurrent illness (particularly if CRP > 10 mg/L) and/or excess adiposity. Other inflammatory biomarkers and cytokines are involved in the atherosclerotic process; however, none are presently used in clinical care.[47]

Perinatal exposures are another important feature related to cardiovascular risk in adulthood, often referred to as the fetal origins of adult disease hypothesis. For example, offspring of Dutch women who experienced famine during World War II had higher rates of obesity and

hyperlipidemia as adults.[48,49] A low birth weight has been associated with higher rates of type 2 diabetes, as described in a meta-analysis.[50] The relationship between birth weight and cholesterol levels is less clear. A twin study demonstrated that lower birth weight was associated with higher TC, and LDL levels.[51] The data on breast feeding and cholesterol levels later in life are mixed. A meta-analysis of 1532 British children and adults showed that breast-feeding was associated with higher TC and LDL in childhood but lower TC and LDL in adulthood.[52] However, being breast fed does seem to be protective against adult obesity.[53]

Lipoprotein (a), or Lp(a), is a variant LDL particle with a covalently bound protein portion, termed apolipoprotein (a). This particle is not measured on standard lipid profiles. High levels are thought to be atherogenic because the particle not only participates in atheromatous plaques (as does LDL) but also impairs fibrinolysis by preventing normal plasminogen activation,[54] and this particle, therefore, is likely involved in the thrombosis of CVD. Lp(a) is genetically determined, and high levels are associated with increased risk of coronary artery disease and stroke in a continuous fashion.[55] Normal levels are considered to be less than 30 mg/dL (or ~70 nmol/L), but higher levels are common in pediatric referral lipid clinic settings and the cutoff for increased cardiovascular risk may indeed be closer to 65 mg/dL.[51] Attention must be paid to the units among different assays, which measure either mass or molarity (mg/dL vs. nmol/L). Certainly, very high Lp(a) levels (more than 100 mg/dL) deserve attention and follow-up, particularly in the setting of a family history of early CVD events. Diet and exercise have no effect on Lp(a) levels. Pharmacologic doses of niacin lower Lp(a)[56]; however, there are very little clinical trial data in childhood,[57] and most adolescents find it difficult to tolerate this medication, even in the slow or extended release forms (adherence must be as near as possible to 100% to avoid tachyphylaxis). For this reason, and because niacin has not been shown to provide additional benefit in reducing CVD events in adults above and beyond standard medical therapy, including statins,[58] we generally do not recommend niacin to lower Lp(a) in pediatrics. However, high Lp(a) levels might be a reason to use lower statin treatment cut points in patients whose LDL values fall into the “gray zone” for pharmacotherapy, particularly if family history is a cause for concern.

LIPID BIOCHEMISTRY

Lipids, including cholesterol and other sterols, TGs, and free fatty acids, are essential for life. In the developed world, diet is typically a major contributor to total body lipid stores, although most fatty acids and TGs (three fatty acids esterified to glycerol) can also be endogenously synthesized from excess carbohydrates and proteins. The essential fatty acids including the basic precursors of polyunsaturated fatty acids (e.g., linoleic acid or linolenic acid) must come from the diet.

Fatty acids can be "saturated," with hydrogen at each carbon; "monounsaturated," with one double carbon bond; or "polyunsaturated," with two to six double bonds. Naturally occurring dietary fatty acids are in the "*cis*" configuration at each double bond. Polyunsaturated fats are prone to spoilage by oxygen, light, and heat; therefore, food processing, especially catalytic hydrogenation processing, which is used to avoid rancidity, produces nonnatural fatty acid chemistry, including a "*trans*" bond configuration. Readers will be familiar with "partially hydrogenated" oils from the list of ingredients in many processed foods, especially snack foods. In general, the larger the proportion of partially hydrogenated fats, the larger the proportion of undesirable *trans* conformation fatty acids. *Trans*-fatty acids not only act like saturated fats to increase LDL but also decrease HDL[59] and should be eliminated from, or at least minimized in, the diet.[60]

Cholesterol is partially absorbed from the diet and partially synthesized by the liver; 30%–50% of cholesterol consumed in the diet is actually absorbed (usually not much more than 300 mg/d). The only dietary source of absorbed cholesterol is animal products. Strict vegans synthesize 100% of their cholesterol needs, as vegetables contain no cholesterol. Dietary cholesterol is reesterified to fatty acids in plasma. Dietary TGs (typically dozens of grams daily) are degraded by lipoprotein lipase in the gut, absorbed, and reesterified to re-form circulating serum TGs. Dietary fat is distributed to body tissues, primarily the liver, in the form of chylomicrons. After absorption, cholesterol is esterified and carried in the serum on lipoprotein particles, as described here.

Cholesterol esters and neutral fats such as TGs are negligibly soluble in water and are therefore carried in the bloodstream by lipoproteins. Free fatty acids in plasma are carried mainly on albumin and are rapidly taken up by tissues for reesterification or for combustion as fuel. Lipoproteins are made up of hydrophobic lipids (TGs and cholesterol ester) on the inside and apolipoproteins, phospholipids, and unesterified cholesterol on the outside.

There are several types of lipoproteins, classified by density as

determined using plasma ultracentrifugation: chylomicrons, which float as a creamy supernatant; HDL; LDL; and very-low-density lipoprotein (VLDL):

- **Chylomicrons** are synthesized in the intestine from dietary TGs, cholesterol, and apolipoproteins synthesized in the liver. Chylomicrons have the lipoproteins apo A, apo C, and apo C-II. The chylomicron delivers the TG to muscle and adipose tissue, where it is modified by lipoprotein lipase, losing TGs, apo A, and apo C and gaining apo E. The chylomicron remnants are taken up by the liver via the apo E receptor and degraded.
- **HDL** is the densest and smallest lipoprotein. The major proteins (apoproteins) in HDL are apo A-I, apo A-II, apo C, and apo E. HDL is the primary lipoprotein responsible for reverse cholesterol transport, returning cholesterol from other tissues back to the liver. HDL also transports cholesterol from other lipoproteins, such as LDL, and cell membranes to VLDL and chylomicrons.
- **LDL** is the main reservoir of circulating cholesterol esters, supplying cell membranes and tissues. Each LDL particle carries an apolipoprotein B molecule, the protein responsible for LDL clearance by its cognate receptor. Cholesterol synthetic mechanisms are present in nearly all mammalian cell types, but exogenous sources are used prior to endogenous synthesis. Delivery to tissues is regulated by the number of LDL receptors, and a tight negative feedback system prevents cholestßerol synthesis and downregulates LDL receptors in normal humans when extracellular LDL is abundant. There is a small but measurable increased risk of atherosclerotic events in adults with small, dense LDL particles, above and beyond the effect of the LDL concentration,[61] which contain a relatively higher concentration of apo B. The effect of this is not known in pediatrics, and studies have shown some effect in young adults, although this may be less important in CVD events in young people.[62]
- **VLDL** is manufactured in the liver, particularly during fasting, as the main source of TGs when chylomicrons are absent. VLDL is secreted into the bloodstream and TGs are removed in the periphery by lipoprotein lipase. VLDL remnants are metabolized by the liver and serve as the precursor for LDL production.

MAJOR DYSLIPIDEMIAS

Normal lipid levels are age dependent; children have lower levels than adults. Normative pediatric values have been reported from National Health and Nutrition Examination Survey data.[63] Cut points for abnormal values are based on population distributions and are not determined by their relationship to health outcomes such as myocardial infarction, because events are vanishingly rare in childhood.[3] The NHLBI Guideline cutoffs for borderline and abnormal lipid values are shown in Table 63-1. Over two decades, there has been a small but important improvement in serum lipid levels in US youth, including a 5-mg/dL decrease in TC and 5-mg/dL decrease in LDL,[63] perhaps related to lower rates of smoking, less uses of *trans* fats, and a lower fat diet/higher glycemic diet.[64]

Dyslipidemia can be primary—monogenic or polygenic, and familial or sporadic—or secondary—related to medical conditions, medications, or lifestyle. The proportion of primary hyperlipidemia is higher in the pediatric population than in adults; however, secondary causes of hyperlipidemia should be considered because they may be modifiable or even treatable. The medical history and examination can help identify secondary causes of hyperlipidemia (Table 63-2) often with the help of simple laboratory testing.

TABLE 63-1 Low, Normal, Borderline, and High Lipid Values in Childhood

Lipid Parameter (mg/dL)	Low	Normal	Borderline	High
TC	—	<170	170–199	≥200
TGs (fasting)				
0–9 Years	—	<75	75–99	≥100
10–19 Years	—	<90	90–129	≥130
LDL (fasting)	—	<110	110–129	≥130
HDL	<40	>45	40–45	—
Non-HDL	—	<120	120–144	≥144

Acceptable, borderline, and abnormal cholesterol values according to the NHLBI pediatric guidelines are shown for TC, TG, HDL, and LDL.

Values are reported as serum; to convert to plasma values, divide by 1.03.

LDL and HDL values presented are for Caucasian subjects.

PRIMARY DYSLIPIDEMIAS

Primary hypercholesterolemias are usually dominantly inherited, with a

gene dosage effect. Heterozygous individuals have elevated lipids but are generally asymptomatic until the condition manifests as early stroke, angina, myocardial infarction, or sudden death in the fourth or fifth decade of life. However, homozygotes can manifest xanthomas, arcus cornea, and thickened tendons even before puberty and can have heart attacks or strokes in childhood or early adulthood. Although multiple genes affect lipid levels, several monogenic conditions have been distinguished and can be readily recognized if family history and laboratory data are available. A simplified guide for diagnosis based on lipid profile and family history is presented in Table 63-3. Among those with primary hyperlipidemias, dietary transgressions, lack of exercise, glucose intolerance, and obesity can worsen familial lipid disorders.

Familial hypercholesterolemia, though rare, is the genetic lipid disorder best understood at a biochemical level. The cause is a genetic defect in the LDL receptor gene. Heterozygotes show a decrease in the number of active receptors, and homozygotes have absence or near absence of functional receptors. Heterozygous deficiency has a prevalence of approximately 1 in 200–500 individuals (0.2%), but heterozygotes account for 5% of early myocardial infarction survivors.[65] These individuals usually have TC levels in the range of 200–500 mg/dL (5.2–13 mmol/L). They have a family history of high cholesterol and early coronary disease and require pharmacologic treatment. Premature atherosclerosis is seen in 30% of adults with heterozygous deficiency for familial hypercholesterolemia.[66]

Some populations have higher prevalence rates for familial hypercholesterolemia because of founder effects, such as French Canadians, Dutch Afrikaners in South Africa, and Lebanese Christians. Individuals with homozygous LDL receptor defects are rare, about one in a million individuals, and have very high TC levels of 500–1000 mg/dL (13–25.9 mmol/L). These are children who could have myocardial infarctions in the first or second decade of life and require not only pharmacologic treatment, which is usually ineffective even though it is sometimes used as an adjunct, but plasmapheresis or, if there is intractable coronary disease, consideration for liver transplantation. In the complete absence of LDL receptors (familial hypercholesterolemia patients homozygous for null mutations), statins are utterly ineffective at standard doses. LDL apheresis with devices such as Liposorber® columns is very effective in lowering LDL-C, although it carries a heavy burden for the child and family with respect to frequency and length of sessions (four to six hours per session every one to three weeks). Liver transplantation has

been used in some cases of progressive coronary disease,[67] or when apheresis is not available. Gene therapy approaches have been attempted,[68,69] but long-term benefit remains to be demonstrated.

Familial defective apolipoprotein B is clinically similar to familial hypercholesterolemia. Familial defective apolipoprotein B is attributable to decreased binding of LDL to its receptor because of a mutation in the apolipoprotein B protein itself rather than a receptor mutation. The condition is rare. The biochemical findings in untreated patients are indistinguishable from heterozygous familial hypercholesterolemia. However, response to pharmacologic therapy, including statins and other agents, tends to be poor, because the upregulation of hepatic LDL receptors, a key step in the mechanism of action for statins and bile-binding resins, will not increase clearance of the mutant particles from the blood.

TABLE 63-2 Medical, Physiologic, and Pharmacologic Causes of Secondary Hyperlipidemia

Endocrine and metabolic states
- Diabetes mellitus
  - Type I related to a large degree to poor glycemic control
  - Type II: particularly high TGs, low HDL
  - Insulin resistance states without overt diabetes (check for acanthosis nigricans)
- Pregnancy
- Hypothyroidism
- Hypopituitarism[a]
- Anorexia nervosa

Hepatic disease
- Hepatitis of any etiology
- Biliary atresia[b]
- Acute intermittent porphyria
- Glycogen storage diseaseb
- Genetic and acquired cholestatic disorders

Renal disease
- Nephrotic syndromeb
- Any renal inflammatory state
- Hemolytic uremic syndrome
- Idiopathic hypercalcemia

Lysosomal storage disease
- Cholesterol ester storage disease

- Niemann-Pick disease
- Other conditions, such as Tay-Sachs disease and Gaucher's disease

Recognizable genetic syndromes

- Klinefelter's syndrome
- Progeria (Hutchinson-Gilford disease)

Systemic inflammatory states

- Trivial inflammation (e.g., viral illnesses, simple urinary tract infection, and sinusitis); "healthy day" testing recommended
- Systemic lupus erythematosus: severe vascular risk if combined with primary hyperlipidemia
- Burns

Drugs and other agents

- Corticosteroids (systemic): primarily hypertriglyceridemia
- Thiazides
- β-Blockers
- Asparaginase
- Immunosuppressives: tacrolimus, cyclosporine, etc.
- Oral contraceptives: use with caution in those with severe hyperlipidemia
- Antiepileptic medications
- Ethanol: low-dose, regular consumption increases salutary HDL; in excess, hypertriglyceridemia

[a] If hypopituitarism is suspected, thyroid-stimulating hormone alone, as advocated for primary screening, is not sufficient to rule out central hypothyroidism.

[b] Elevations can be severe with xanthomas.

TABLE 63-3 Phenotype and Family History Commonly Found among Pediatric Lipid Clinic Patients

Common Phenotypes	Family History	Likely Diagnosis
Elevated LDL[a] and TC +/– low HDL (xanthoma)	Xanthomas/early heart disease[b]	Familial hypercholesterolemia (TC 500–1000 mg/dL found in homozygotes)
Elevated TGs + low HDL, often with obesity	Type 2 diabetes or similar lipid patterns	High risk for metabolic syndrome
Extremely elevated TGs and chylomicrons, pancreatitis	Elevated TGs, pancreatitis	Lipoprotein lipase deficiency, apoC2
Moderately elevated TGs and LDL	Elevated TGs, LDL, or both	Familial combined Hyperlipidemia

Low HDL only	Low HDL, early CHD	Familial hypoalphalipoproteinemia
High HDL (> 60 mg/dL), normal LDL	Often negative for early heart disease	Normal variant; 20% of children with elevated TC; no intervention

Cholesterol, HDL, and TG values were measured in a fasting state. Combination of phenotype with typical family history suggests diagnosis.

[a] LDL is calculated from fasting values using the Friedewald formula, in which LDL = TC – (HDL + TG/5), which is valid if the TG level is <400 mg/dL. When TG levels exceed 400 mg/dL, direct LDL is advised.

[b] Early heart disease is defined as myocardial infarction before age 55 in men and before age 60 in women.

There are several more recently described defects that phenotypically present in a manner similar to familial hypercholesterolemia. Proprotein convertase subtilisin-kexin type 9 (PCSK9) is a protein that promotes the degradation of the LDL receptor; gain of function mutations lead to elevated circulating LDL at levels similar to familial hypercholesterolemia, due to a lack of LDL uptake by the liver. PCSK9 promotes the degradation of LDL receptors; gain of function mutations lead to fewer LDL receptor and more circulating LDL. Autosomal recessive hypercholesterolemia is caused by the lack of a cofactor responsible for LDL binding to the LDL receptor, leading to less binding and more circulating LDL.

Familial hypertriglyceridemia is characterized by high TG levels, sometimes accompanied by low HDL. This is a very common profile, of polygenic origin, exacerbated by type 2 diabetes mellitus and obesity. Rare monogenic disorders cause chylomicronemia and profound hypertriglyceridemia. Homozygous deficiency of lipoprotein lipase or its cofactor, apolipoprotein C-II, causes isolated increase in chylomicrons, may present as recurrent pancreatitis, and is autosomal recessive. Affected individuals may have lipemia retinalis and a papular rash, but increased risk of early myocardial infarction has not been described.

Familial hypoalphalipoproteinemia exists; but in the general population, low HDL is a common trait, and low HDL is especially prevalent among young survivors of myocardial infarction. With rare exceptions, the exact genetic basis of low HDL is obscure.

Familial combined hyperlipidemia is the most common type of inherited hyperlipidemia. It is autosomal dominant and is found in 1 in 100 individuals. Of patients with early CHD, 10%–15% carry this diagnosis. The lipid abnormalities are moderate. About one-third of patients with familial combined hyperlipidemia have increased LDL, one-third have

increased TGs without altered LDL, and another third have increased TGs and increased LDL. The three types frequently coexist within a family. There are likely multiple genetic abnormalities that lead to this phenotype.

To date, genetic testing has not been used in clinical practice and treatment is based on phenotype; however, as the genetics of lipid metabolism are better understood it is likely that testing will be used in clinical practice, either related to therapeutic choices or screening of family members.

CHOLESTEROL SCREENING

Who to Screen: The Report of the NHLBI Expert Panel on Cardiovascular Risk Reduction in Children and Adolescents,[6] endorsed by the American Academy of Pediatrics, recommends selective screening of children with risk factors for atherosclerosis, including family history of hypercholesterolemia or early coronary artery disease (Figure 63-1 and Table 63-4), after age 2 years. Cholesterol screening is also recommended for children who have personal risk factors, such as obesity, hypertension, or smoking, and for those with high-risk medical histories, as described earlier. Additionally, and importantly, the NHLBI Guidelines recommended for the first time universal screening of all children once between the ages of 9 and 11 years and once again between the ages of 17 and 21 years in addition to selective screening. These recommendations have been very controversial,[70–72] and there are no published data on the extent to which these guidelines have been followed in general pediatric practices, although preliminary research results suggest that adoption is low.

Arguments for universal screening are as follows:

- Atherosclerosis begins during childhood and is associated with childhood cholesterol values.
- Cholesterol values track from childhood to adulthood, implying some degree of lifelong exposure.
- Hypercholesterolemia responds to lifestyle modification and pharmacotherapy, for example, there are available therapies.
- Family history–based screening identifies only approximately half of those with high cholesterol values. Often, the full family history is not available,[73] and very young children might have grandparents too young to have had early heart disease. Family history may be variably useful in screening in nonwhite

populations.[13,73,74]

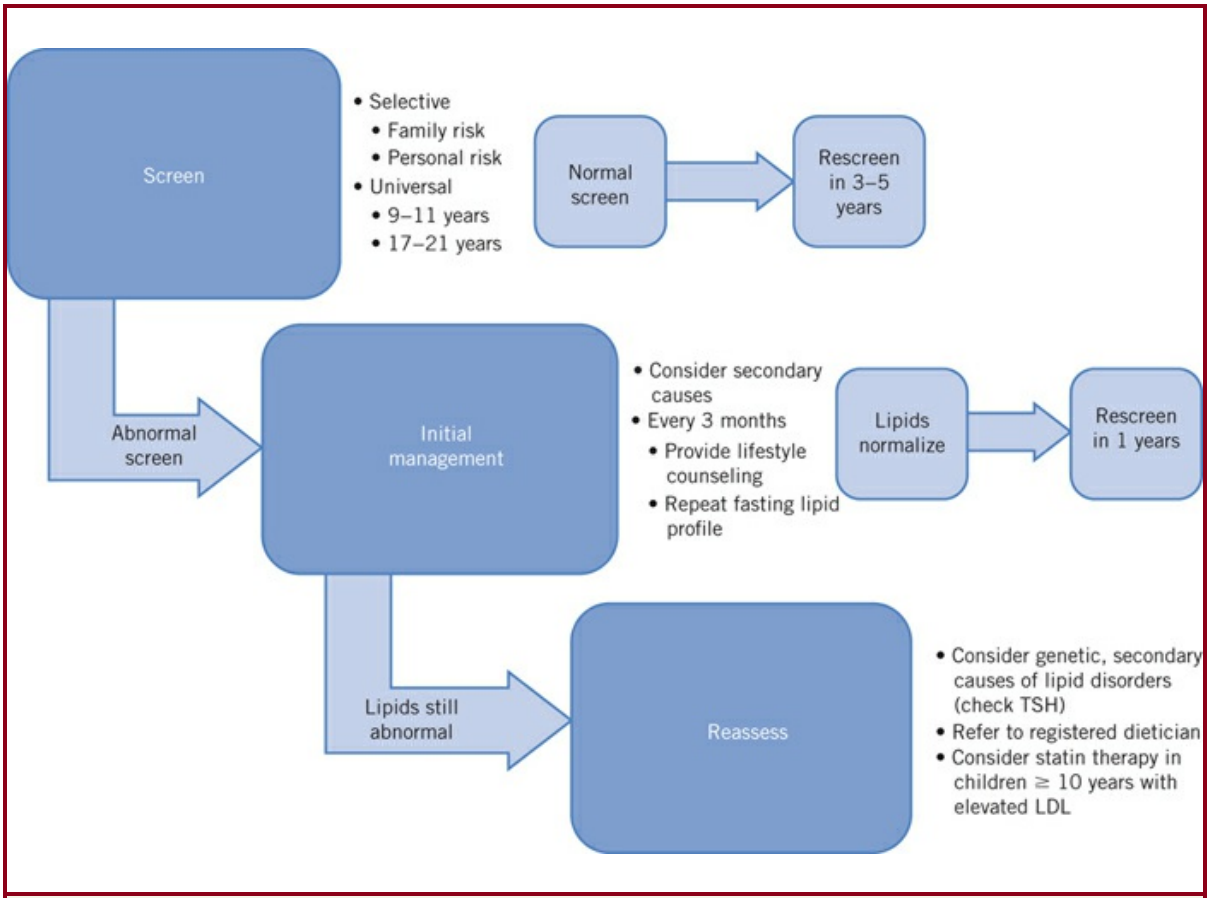


Figure 63-1 Selective screening approach based on our clinical practice and recommendations by the National Heart, Lung, and Blood Institute Expert Panel on Cardiovascular Risk Reduction in Children and Adolescents. (From Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents; National Heart, Lung, and Blood Institute. *Pediatrics*. 2011;128(Suppl 5):S213-S256.)[6]

Arguments against universal screening are as follows:

- The cost of screening some 60 million young persons is not known, nor is it clear how many additional patients would be identified by screening above and beyond a family history-based approach.
- Exact numbers are not known, but some argue that it would be rare to discover children by universal screening who would require medication before adulthood.
- All children should follow a heart-healthy diet and exercise

regularly, regardless of their cholesterol level.

- Labeling children as hypercholesterolemic unnecessarily could cause harm.

How to Screen: The 2011 NHLBI Guidelines recommend that selective screening should be done using a fasting lipid profile. The NHLBI-recommended method for universal screening is either a fasting lipid profile or nonfasting TC and HDL to calculate a non-HDL cholesterol, as is recommended for adult lipid screening.[75] Screening using only TC is not recommended due to high false-negative and false-positive results, based in part on differences in HDL.

NUTRITION AND CHOLESTEROL IN CHILDREN

Dietary habits among American children promote excess calories and CVD risk. Children and adolescents, like all Americans, eat out often, and this is associated with a higher calorie intake. According to national survey data, 34% of calories are now consumed outside the home.[76] Food eaten outside the home is higher in total and saturated fats and lower in calcium, iron, and fiber than food eaten at home.[77] Adolescents who eat fast food are more likely to have a poor-quality diet, one higher in saturated fat and sugar and lower in fruits and vegetables.[78] Fruit juice has been recognized as a major source of calories. High intake has been associated with both poor growth and also obesity in some[79] but not all studies[80]; reduced intake is one strategy for weight loss and TG reduction.

TABLE 63-4 Recommendations for Lipid Screening in Childhood According to the NHLBI Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents

	0–1.9 Years[a]	2–8 Years	9–11 Years	12–16 Years	17–21 Years
Screening recommendation	Do not screen	Screen if there is a family history of early CVD or high cholesterol, or if there is	Screen all children, once during this age range	Screen if there is a family history of early CVD or high cholesterol,	Screen all adolescents, once during this age range

		a high-risk condition		or if there is a high-risk condition	
Test	None	Fasting lipid profile	Fasting lipid profile or non-HDL[c]		Fasting lipid profile or non-HDL[c]
Treatment	None	Lifestyle modification[b]	Lifestyle modification for 6 months, statins for children ages 10 years and older	Lifestyle modification for 6 months, statins for children aged 10 years and older	Lifestyle modification for 6 months, statins for children ages 10 years and older

[a] Children younger than age 2 may be screened in rare situations of extreme family history or in the presence of xanthoma suggestive of homozygous familial hyperlipidemia. Treatment of any significant abnormalities at this young age should be supervised by an expert in pediatric lipid disorders. Metabolic disorders should be considered in the differential diagnosis.

[b] Children aged 8 and 9 years with severe lipid abnormalities that do not respond to lifestyle modification may be candidates for statin therapy under supervision of an expert in pediatric lipid disorders.

[c] Non-HDL is derived from subtracting HDL from TC. Results are valid in the fasting or nonfasting states, making it a convenient cholesterol screening test.

TREATMENT FOR DYSLIPIDEMIAS

Dietary treatment is the first line of intervention in childhood dyslipidemias. Diet modification is emphasized in the pediatric age range because pharmacologic agents have been tested primarily in adults and pediatric long-term safety data are limited. The NHLBI Guidelines recommend different nutritional approaches based on an individual's lipid abnormalities. It is recommended that all children keep fat intake to less than 30% of total calories, with a goal of saturated fat representing less than 10% of total calories and cholesterol intakes of less than 300 mg/day (CHILD 1).[6] The next step for children with elevated LDL is to restrict saturated fat intake to less than 7% of daily calories; added fiber and stanol esters can be used as adjunct nutritional therapy (CHILD 2 LDL). For children and adolescents with high TGs, the NHLBI Guidelines recommend the same restrictions on saturated, total fat, and cholesterol, and in addition decreased sugars in the form of simple carbohydrates and sugary beverages and increased fish to provide dietary ω-3 fatty acids. Calorie restriction and increased physical activity are recommended for obese and overweight children regardless of the lipid phenotype.[6]

In practice, low cholesterol and low saturated fat can be achieved by many children and adolescents. Relatively few US children now eat too much cholesterol, which is primarily found in shrimp, squid, and organ meats such as liver. Saturated fat is more prevalent in the diet; some suggestions that we provide to our patients for reducing dietary saturated fat are shown in Table 63-5. Professional nutritional counseling is very helpful for patients aiming for more significant changes in lipid values, such as might be achieved with the CHILD2 LDL diet. Dietary intervention trials in adults with stringent fat reductions show a TC reduction between 10% and 15%.[81] Several short- and medium-term studies demonstrate that diets low in saturated and total fat decrease TC and LDL cholesterol by approximately 10%.[82–84] Two key dietary counseling studies, the STRIP study[85–87] and the Dietary Intervention Study in Children (DISC) trial,[88] showed the safety of low-fat diets and the relatively small LDL-lowering effect in these large studies over time, 3–5 mg/dL, compared to control.

Some pediatric practitioners have raised concerns that diets designed to improve cholesterol will hamper pediatric growth and development; however, the DISC trial demonstrated that children counseled on a low-cholesterol, low-saturated-fat diet had the same growth and development than children on a diet close to the usual American intake.[88] The STRIP study also showed that dietary consumption of fat, saturated fat, and TC did not affect neurodevelopment, although higher protein intake was associated with better neurodevelopmental outcome in boys.[89] These studies demonstrate that some degree of saturated fat restriction is safe even in young children.

Several decades of dietary counseling focused on low-fat and low-saturated-fat approaches. However, dietary counseling in the current era is fairly individualized, focusing now not on eliminating fat from the diet so much as exchanging saturated fat for monounsaturated fat. This is in part because fat elimination can lead to substitution with refined sugars and carbohydrates that increase TG levels, whereas increased intake of monounsaturated fats can raise HDL levels and improve the cardiovascular risk profile. The Mediterranean diet, which focuses on mono- and polyunsaturated fats, has been associated with a reduced risk of cardiac mortality and morbidity in adults after their first myocardial infarction[90] and, more recently, has been associated with lower rates of CVD events in primary prevention populations.[91] There are limited trial data on the effect of the Mediterranean diet on lipids in children with hypercholesterolemia,

although one study did demonstrate a decline in TC, LDL, and carotid intimal-medial thickness, a measure of subclinical atherosclerosis,[92] and some practitioners recommend it to their patients and families.

TABLE 63-5 Simple Substitutions for a Healthy Diet

A. High LDL	
For	Substitute
Fatty meats	Fish, skinless chicken, lean beef, Vegetable proteins, pork tenderloin
Saturated fat (e.g., butter)	Monounsaturated fat
Partially hydrogenated oils (*trans*-fatty acids) in regular Margarines	Canola or olive oil, monounsaturated margarines
Regular peanut butter (partially hydrogenated)	"Natural" peanut butter
Ice cream	Frozen yogurt
Whole milk	Skim or 1% milk
Regular cheese	Low-fat cheese
Lunch meats	Low-fat equivalents
Chocolate (saturated oils)	Dark chocolate
Less healthful school lunch purchased	Lunch packed at home
B. High TGs	
For	Substitute
White rice	Brown Rice
Regular pasta	Whole grain pasta
White bread	Whole wheat bread
Instant oatmeal	Rolled oats or steel cut oats
White potatoes	Sweet potatoes
Low fiber cereals	High fiber cereals
Sugary beverages	Water or low calorie beverages

To modify TG and HDL levels, most recommend reducing carbohydrate intake as a percentage of total calories. A reduction in total calories leads to weight loss and an improvement in cardiovascular risk factors associated with the metabolic syndrome. Studies of low-carbohydrate diets in adults show greater initial weight loss in the short term (~6 months) compared to low-saturated-fat diets; as with all dietary interventions, results at one year are less impressive.[93,94] Low-glycemic-load diets are associated with lower rates of CVD in adults in

observational studies, and reduced CVD risk factors including lipid levels in short-term interventional trials.[95–97] Glycemic index is a property of carbohydrate-containing food that describes the rise of blood glucose occurring after a standardized amount of food is consumed. In general, most refined starchy foods eaten in the United States, such as white bread, prepared breakfast cereals, rice, and potato products, have a high glycemic index, whereas unprocessed grains, nonstarchy vegetables, fruit, and legumes tend to have a low glycemic index. Several studies in children have demonstrated greater weight loss in children on low-glycemic-index diets.[78,79] Weight loss in children has been shown to effectively decrease TC and TG and increase HDL[81] and is a goal of many nutritional interventions if the child is overweight. Furthermore, data are beginning to emerge about the effect of the low-glycemic-load approach to managing lipid disorders in children, and not all of them are favorable[98,99]; however, some trials do support the importance of the low-glycemic-load approach in weight and lipid management.[100–102]

Other types of interventions to improve lipid levels on a population basis have been explored, including community-based programs. Several studies of nutritional teaching in school resulted in a lower average TC in children at the intervention schools than in the comparison schools.[103,104] The DISC is the largest published experience with school-based interventions and also showed improved cholesterol.[88]

SPECIAL NUTRITIONAL INTERVENTIONS: FIBER, STANOLS, AND *ω* -3 FATTY ACIDS

Several specific nutritional tools have been used to modify lipid profiles in dyslipidemic children. Studies of children with familial hypercholesterolemia who received fiber supplementation on top of a low-cholesterol and low-saturated-fat diet had decreases in TC with preserved HDL.[105–107] Certain sterols, similar in structure to cholesterol but not absorbed by the small intestines, lower cholesterol by inhibiting the absorption of cholesterol, both dietary and in the form of bile acids. As part of a low-fat diet in adults, stanols decrease TC by 8%–10%, above and beyond the 7% reduction from the low-fat diet alone.[86] Studies in children confirm these findings.[108–110] In adult observational data, higher intakes of n-3 polyunsaturated fatty acids (*ω*-3 fatty acids) in the form of fish have been associated with lower risks of CVD mortality after a myocardial infarction[111]; however, an interventional trial of 1 g of *ω*-3

fatty acid supplement daily did not demonstrate lower event rates compared to olive oil placebo.[112] Of course, no such data are available for children. Fish are a rich source of ω-3 fatty acids and should be recommended. Flax oil and ground flaxseed are reasonable substitutes for children who will not eat fish. For children with high TGs, particularly those who do not consume many ω-3 fatty acids, supplements can be used, although data supporting their efficacy are not currently published; care should be taken with respect to docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) content as this varies substantially in over-the-counter preparations and is not standardized. In younger children with high TG levels unresponsive to lifestyle therapy, we have used a dose of 2000 mg/d of preparations containing approximately 700–800 mg of DHA+EPA per pill, over the counter or prescription, and 4000 mg/d of the high-dose ω-3 pills in older adolescents with high TG levels.

PHARMACOLOGIC INTERVENTION

Studies of adults with hypercholesterolemia demonstrate significant reduction in cardiovascular morbidity and mortality with cholesterol-lowering medications. The benefit is seen not only in severely affected patients or those who have existing heart disease[113] but also as primary prevention in patients with only moderate hypercholesterolemia and no previous CVD.[114,115] As in adults, lipid-lowering drugs effectively lower LDL in children.[6] There are no long-term trials evaluating the effect of lipid-lowering medication during childhood on CVD event rates. Studies are needed that consider long-term benefits, risks, and costs of statin therapy in children with FH, and other patient populations.

The NHLBI Guidelines make the following recommendations about lipid lowering in childhood: after approximately six months of nutritional and lifestyle changes, children aged 10 years or older with severe hyperlipidemia, defined as having LDL cholesterol greater than 190 mg/dL or greater than 160 mg/dL with two additional risk factors for CVD, are eligible for consideration for pharmacologic treatment. Children as young as 8 years of age who are severely affected may be treated with medications if supervised by a pediatric lipid specialist. Factors that are also considered in clinical practice include high beneficial HDL levels, high-risk medical conditions such as diabetes mellitus, a history of Kawasaki disease aneurysms, chronic inflammatory disorders such as lupus, severe family history, and so forth.

A number of pediatric trials have shown that lipid-lowering

medications act similarly in childhood to adulthood. Usual dosages are shown in Table 63-6. The oldest class of medications are bile acid–binding resins, including cholestyramine and colestipol, which bind to bile acids in the gut and prevent their reabsorption; this prompts the body to convert more cholesterol to bile acids, thereby lowering cholesterol by 10%–15%.[116,117] These medications may also decrease dietary cholesterol absorption from the gut and have been demonstrated to increase LDL receptor expression and HMGCoA reductase activity.[118] The bile acid-binding medications are not absorbed into the bloodstream, minimizing systemic effects; however, the powder forms have a gritty texture and the pills are large and can be difficult to swallow.[117] Both forms can cause constipation. For these reasons, compliance can be an issue. These medications also bind to lipophilic and acidic drugs, making them somewhat challenging to use in patients on multiple medications.

3-Hydroxy-3-methylglutaryl CoA reductase inhibitors, or statins, are the clear first-line agents for adults with elevated LDL cholesterol levels and have been used in selected, high-risk pediatric and adolescent patients for many years; statins are the recommended first-line therapy for children and adolescents with high LDL requiring pharmacotherapy.[6] Randomized controlled clinical trials of statins have shown good efficacy and safety in the short and medium terms, with an LDL lowering of 20%–50%.[119] These trials are too small and too short to definitively eliminate the possibility of long-term sequelae, the major but very rare adverse effect of rhabdomyolysis, or the theoretical modification of sex-steroid production. It should be noted that while these adverse effects cannot be excluded, there are no data to support them either; in fact, adults seem to experience benefits from statins over decades.

Gemfibrozil and fenofibrate are indicated for hypertriglyceridemia in adults but are used only for teenagers at risk for pancreatitis (TGs > 500–1000 mg/dL). Niacin can be an effective adjunct to lipid therapy in some adult settings, but flushing, abdominal discomfort, and risk of hepatotoxicity limit its use in children.[57]

Ezetimibe is a selective inhibitor of cholesterol absorption that was approved for use in adults by the US Food and Drug Administration in late 2002. It reduces LDL by approximately 20% with minimal side effects.[120] It is frequently used in adults as an adjunct to statins but has not been shown to decrease CVD events. Two pediatric trials of ezetimibe in children show 15%–30% decrease in LDL, with no significant adverse effects.[121] It is the primary medication recommended for sitosterolemia, a

rare genetic cause of early atherosclerotic disease associated with xanthomata and paradoxically only moderately elevated LDL levels.

OTHER THERAPIES FOR SEVERE HYPERCHOLESTEROLEMIA

For patients with severe hypercholesterolemia unresponsive to diet or pharmacologic intervention, plasmapheresis or LDL apheresis with a Liposorber column has been successfully used to lower TC, LDL, TGs, and lipoprotein (a). Case series of patients with homozygous familial hypercholesterolemia suggest a slowing down or even regression of atherosclerosis with the use of apheresis.[122] The technique has been used in children,[123] although the issue of vascular access is a difficult one. New pharmacotherapies for LDL lowering are now emerging for use in adults with resistant and/or severe hypercholesterolemia such as monoclonal antibodies PCSK9[124]; mipomerson is an antisense oligonucleotide complementary to the coding region for human apolipoprotein B (apo B) mRNA. These newer medications all have injection sight, and there are concerns about hepatic congestion.[125] There is little experience with these agents in childhood; however, their use might ultimately be considered when apheresis is not available. Liver transplantation has also been used in homozygous or compound heterozygous familial hypercholesterolemia, with good short-term success, albeit trading hypercholesterolemia for lifelong immunosuppression.

MULTIDISCIPLINARY CLINICAL APPROACH

Because atherosclerosis has a complex and multifactorial pathophysiology, pediatric lipid treatment programs that employ a multidisciplinary approach are a priori more likely to be successful. In our center, the approach is combined nutritional and physical activity counseling, along with risk assessment and lipid monitoring. We find that lifestyle habits can be more easily modified during childhood and that changes put into place in younger children may have an impact for years to come. This is the prime rationale for seeing children at young ages. Contrary to adult lipid treatment, pediatric lipid specialists begin with lifestyle modification and use pharmacotherapy in only the most severely affected patients. Figure 63-2 describes one such approach to patient management.

TABLE 63-6 Dosages of Medications Commonly Used in Hyperlipidemia

Lipid Lowering Bile Acid Binding	Dosage Forms	Typical Dosage Range	Comments
Cholestyramine powder	Generic powder is unflavored; Questran Light and LoCholest are flavored	Weight < 40 kg: start with 2 g bid, increase to 4 g bid if well tolerated Weight > 40 kg: start as above, to 6 g bid; max. adult dose, 24 g/d	Not systemically absorbed; side effects of bloating, constipation fairly common. Absorbs other medication including antibiotics; adherence is poor
Colesevelam	625 mg Tablets, 3750 mg/packet	1.875 gm bid or 3.75 g/d; take with liquids	Tablets are large; can be difficult for children to swallow
Colestipol	Powder or tablet (2 g)	5–30 g of powder div bid (supplied as 5 g packets or scoops) 2–16 g Tablet form, div bid	As above
Absorption Inhibitor[a]			
Ezetimibe (Zetia)	10 mg Tablets	10 mg po qd	Inadequate LDL control on statins, Statin intolerance, sitosterolemia
Statins[b]			
Lovastatin	10, 20, 40 mg Tablets	In children begin at lowest dose Increase gradually based on effect	
Pravastatin	10, 20, 40, 80 mg Tablets		
Simvastatin	5, 10, 20, 40, 80 mg Tablets	Doses above 40 mg/d are associated with increased risk for muscle toxicity	
Atorvastatin	10, 20, 40, 80 mg Tablets		
Rosuvastatin (Crestor)	5, 10, 20, 40 mg Tablets		No generic available for rosuvastatin

Fibrates			
Gemfibrozil	600 mg	600 mg twice per day	Use caution when combining with fibrates with a statin Significantly increased risk of muscle toxicity
Fenofibrate	(Depends on brand)	(Depends on brand)	May have less risk of muscle toxicity compared to gemfibrozil
ω-3 Fatty acids			
OTC	(Depends on brand)	~700 mg DHA + EPA/1000mg	For TG levels > 500 mg/dL (adult data)
	~700 mg DHA + EPA/1000 mg	900 mg of ethyl esters/1000 mg	Start with 2 g/d, measure effect
Lovaza		2–4 g/d	In ~4 weeks

Dosages are provided as a guide only and should be reviewed independently prior to prescribing.

[a] Side effects are minimal in adults and include gastrointestinal upset. No long-term studies in children.

[b] Statin therapy is contraindicated in pregnancy. Liver function tests should be monitored at baseline, after 6 weeks of therapy, and then intermittently if normal. Caution patients about rare rhabdomyolysis; creatine phosphokinase should be checked if patient has myalgias. Sleep disturbances and abdominal discomfort are infrequent side effects.

Clinic sessions may include group and individual nutritional education, review of all available laboratory results, and a session with a clinician. A three-day food recall provides a basis for individual nutritional counseling at the initial visit and in follow-up. Patient and family group education can include the basics of “good” and “bad” cholesterol, saturated and *trans*–fatty acids, protein, sugars, and fiber. The concept of glycemic index/load can be helpful in most cases of hypertriglyceridemia and obesity. Parents can be taught to read labels and to read between the lines on labels, and children can learn the concept of budgeting their fat and sugar intake so that, for example, birthday cake is not completely off limits but requires a lean period in compensation. It is useful to show children the actual amount of sugar in a can of soda or a serving of juice, or the appearance of fat clogging a tube that represents an artery. Methods of dieting for children can include using the “plate method” (Figure 63-3). We stress that diet does not mean weight loss for slender patients with genetic hyperlipidemia but that weight loss is paramount in patients with moderate to severe obesity, even when genetic hyperlipidemia is present. In general, emphasizing substitution of foods, as opposed to restricting or eliminating

food, is more positive and seems more effective. Table 63-5 presents some simple suggestions for dietary modifications in hyperlipidemia.

The physician or nurse practitioner portion of the visit should involve a review of family history, a review of general medical history, an assessment of physical activity and inactivity (screen time), and an evaluation of lipid values. Twelve-hour fasting lipid values are the most valuable for children referred to a specialist clinic, as it is important to have accurate TG and LDL levels, at least on two occasions. Because severely affected children in a pediatric lipid setting usually have severely affected parents, we have found that often the most pressing medical issue for the child is having his or her parents tested and if necessary treated for hyperlipidemia. It is important to ask about cigarette smoke exposure, encourage children not to smoke, and encourage parents not to smoke around their children because children exposed to cigarette smoking have lower HDL levels.[126,127] We stress the need to find physical activities that the child will enjoy.

We try to emphasize that for most children these issues are not an emergency but rather are long-term modifications that are important not for present health but for cardiovascular and metabolic risk 20–40 years in the future (a nearly unimaginable length of time for most children). This helps parents and children to view this as a long-term project and removes the pressures of enforcing an immediate turnaround. Having only appropriate food in the home is helpful in making choices easier. Lifestyle changes seem to be more effectively made if the entire family joins in the effort.[128] Often, the changes we suggest benefit multiple family members even if only one child in the family is referred. Follow-up is crucial for monitoring progress, medication compliance, and somatic growth, and for encouraging further efforts in lifestyle modification.

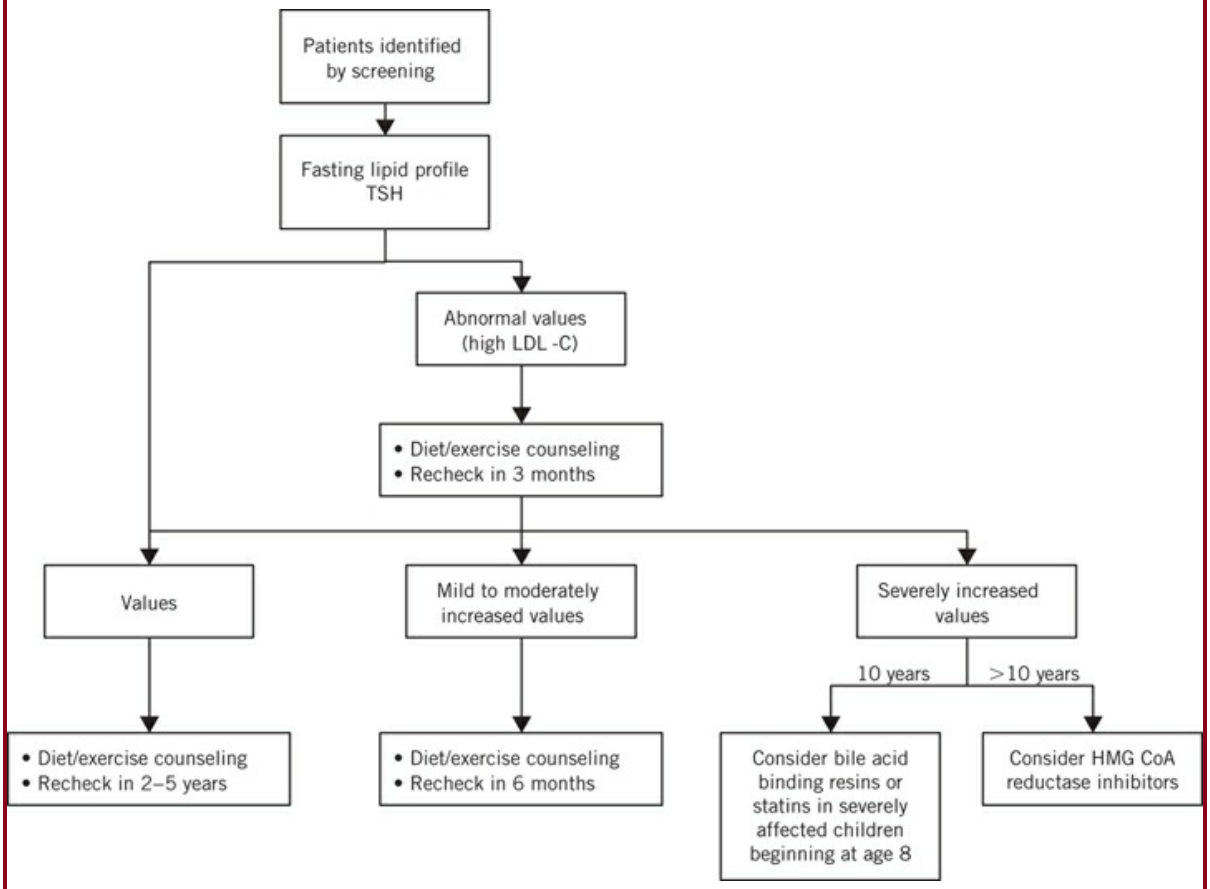


Figure 63-2 Proposed approach to cholesterol screening and initial treatment in pediatric patients. Thyroid-stimulating hormone (TSH) serves as an inexpensive screen for primary hypothyroidism.

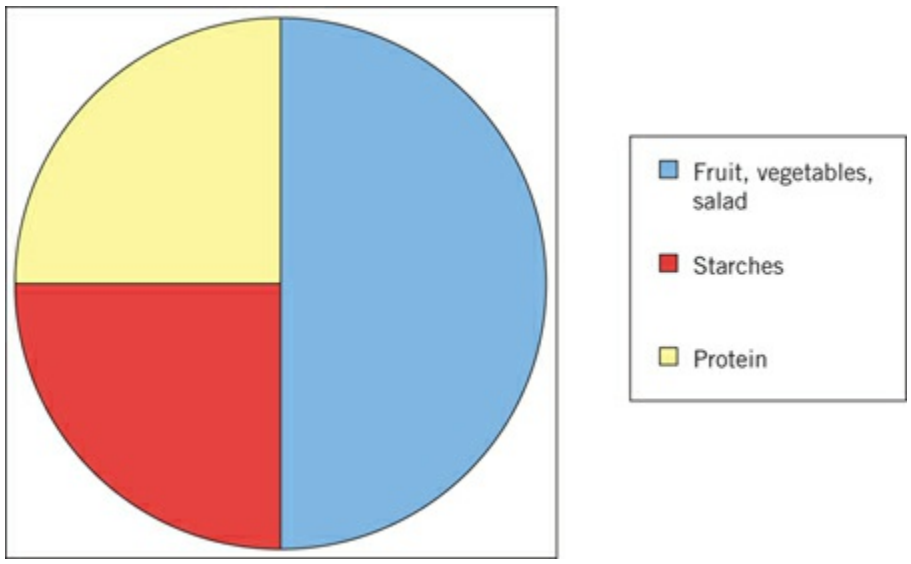


Figure 63-3 The plate method is a simple means for balancing the diet and reducing carbohydrate consumption without counting calories or grams of fat. One-half to one-third of the plate should contain fruits and vegetables, whereas one-quarter should contain starch and the final quarter should contain protein. The method is effective in patients with lifestyle-related hypertriglyceridemia and for obesity, if given with a few suggestions: (1) the entire family, not just the patient, should try the method for 4-6 weeks and then recheck lipids and weight; (2) if the patient wants second helpings, he or she must finish the entire plate (e.g., eat all the fruits and vegetables); and (3) "seconds" cannot be just starches, but it must again be balanced.

SUMMARY

Pediatric lipid disorders contribute to future cardiovascular risk, and as such it is important to address them directly. At the same time, the majority of pediatric hyperlipidemias are most safely, and perhaps most effectively, treated with a multidisciplinary lifestyle approach that includes plenty of physical activity, minimizing inactivity, nutritional counseling for an optimal diet, and adequate sleep in the setting of an individualized intervention for the child and the family. Care should be exercised to prevent the establishment of negative behavioral patterns because the health implications of dyslipidemias occur, for the most part, far in the future. Future research should focus on how lifestyle modification changes —including diet—can be best implemented; longer term evaluation of antihyperlipidemic medications in young patients; the utility of markers of preclinical disease either blood or vascular testing for evaluating the efficacy of interventions for atherosclerosis prevention; and better understanding provider, patient, and parent preferences around lipid screening and treatment that will impact implementation for lipid screening and treatment.

REFERENCES

1. Ford ES, Ajani UA, Croft JB, et al. Explaining the decrease in U.S. deaths from coronary disease, 1980-2000. *N Engl J Med.* 2007;356:2388–2398.
2. Go AS, Mozaffarian D, Roger VL, et al. Heart disease and stroke statistics—2013 update: a report from the American Heart Association. *Circulation*. 2013;127:e6–e245.

3. Kannel WB. Range of serum cholesterol values in the population developing coronary artery disease. *Am J Cardiol*. 1995;76:69C–77C.
4. The Lipid Research Clinics Coronary Primary Prevention Trial results. II. The relationship of reduction in incidence of coronary heart disease to cholesterol lowering. *JAMA*. 1984;251:365–374.
5. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in body mass index among US children and adolescents, 1999-2010. *JAMA*. 2012;307:483–490.
6. Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents; National Heart, Lung, and Blood Institute. *Pediatrics.* 2011;128(Suppl 5):S213–S256.
7. Holman RL. The natural history of atherosclerosis. *Am J Pathol*. 1958;34:209–235.
8. Stary HC. Evolution and progression of atherosclerotic lesions in coronary arteries of children and young adults. *Arteriosclerosis*. 1989;9: I19–I32.
9. Strong JP, Malcom GT, McMahan CA, et al. Prevalence and extent of atherosclerosis in adolescents and young adults: implications for prevention from the Pathobiological Determinants of Atherosclerosis in Youth Study. *JAMA*. 1999;281:727–735.
10. Berenson GS, Srinivasan SR, Bao W, et al. Association between multiple cardiovascular risk factors and atherosclerosis in children and young adults. The Bogalusa Heart Study. *N Engl J Med*. 1998;338:1650–1656.
11. McGill HC, Jr., McMahan CA. Determinants of atherosclerosis in the young. Pathobiological Determinants of Atherosclerosis in Youth (PDAY) Research Group. *Am J Cardiol*. 1998;82:30T–36T.
12. Solberg LA, Strong JP. Risk factors and atherosclerotic lesions. A review of autopsy studies. *Arteriosclerosis*. 1983;3:187–198.
13. Webber BJ, Seguin PG, Burnett DG, et al. Prevalence of and risk factors for autopsy-determined atherosclerosis among US service members, 2001–2011. *JAMA*. 2012;308:2577–2583.
14. Levy D. Combating the epidemic of heart disease. *JAMA*. 2012;308:2624–2625.
15. Knuiman JT, West CE, Katan MB, Hautvast JG. Total cholesterol and high density lipoprotein cholesterol levels in populations differing in fat and carbohydrate intake. *Arteriosclerosis*. 1987;7:612–619.
16. Webber LS, Srinivasan SR, Wattigney WA, Berenson GS. Tracking of serum lipids and lipoproteins from childhood to adulthood. The Bogalusa Heart Study. *Am J Epidemiol*. 1991;133:884–899.
17. Lauer RM, Lee J, Clarke WR. Factors affecting the relationship between childhood and adult cholesterol levels: the Muscatine Study. *Pediatrics*. 1988;82:309–318.
18. Klag MJ, Ford DE, Mead LA, et al. Serum cholesterol in young men and subsequent cardiovascular disease. *N Engl J Med*. 1993;328:313–318.
19. Morrison JA, Friedman LA, Gray-McGuire C. Metabolic syndrome in childhood predicts adult cardiovascular disease 25 years later: the Princeton Lipid Research Clinics Follow-up Study. *Pediatrics*. 2007;120:340–345.
20. Niinikoski H, Lagstrom H, Jokinen E, et al. Impact of repeated dietary counseling between infancy and 14 years of age on dietary intakes and serum lipids and lipoproteins. The STRIP Study. *Circulation*. 2007; Aug 28;116(9):1032–1040. Epub 2007 Aug 13.
21. Kavey RE, Allada V, Daniels SR, et al. Cardiovascular risk reduction in high-risk pediatric patients: a scientific statement from the American Heart Association

Expert Panel on Population and Prevention Science; the Councils on Cardiovascular Disease in the Young, Epidemiology and Prevention, Nutrition, Physical Activity and Metabolism, High Blood Pressure Research, Cardiovascular Nursing, and the Kidney in Heart Disease; and the Interdisciplinary Working Group on Quality of Care and Outcomes Research: endorsed by the American Academy of Pediatrics. *Circulation*. 2006;114:2710–2738.
22. Tuzcu EM, Kapadia SR, Tutar E, et al. High prevalence of coronary atherosclerosis in asymptomatic teenagers and young adults: evidence from intravascular ultrasound. *Circulation*. 2001;103:2705–2710.
23. Mulla NF, Johnston JK, Vander Dussen L, et al. Late rejection is a predictor of transplant coronary artery disease in children. *J Am Coll Cardiol*. 2001;37:243–250.
24. Pennisi AJ, Heuser ET, Mickey MR, et al. Hyperlipidemia in pediatric hemodialysis and renal transplant patients. Associated with coronary artery disease. *Am J Dis Child*. 1976;130:957–961.
25. Sharma AK, Myers TA, Hunninghake DB, et al. Hyperlipidemia in long-term survivors of pediatric renal transplantation. *Clin Transplant*. 1994;8:252–257.
26. Newburger JW, Takahashi M, Gerber MA, et al. Diagnosis, treatment, and long-term management of Kawasaki disease: a statement for health professionals from the Committee on Rheumatic Fever, Endocarditis, and Kawasaki Disease, Council on Cardiovascular Disease in the Young, American Heart Association. *Pediatrics*. 2004;114:1708–1733.
27. Warnes CA, Williams RG, Bashore TM, et al. ACC/AHA 2008 Guidelines for the Management of Adults with Congenital Heart Disease: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (writing committee to develop guidelines on the management of adults with congenital heart disease). *Circulation*. 2008;118:e714–e833.
28. Hauser M, Bengel FM, Kuhn A, et al. Myocardial perfusion and coronary flow reserve assessed by positron emission tomography in patients after Fontan-like operations. *Pediatr Cardiol*. 2003;24:386–392.
29. Bengel FM, Hauser M, Duvernoy CS, et al. Myocardial blood flow and coronary flow reserve late after anatomical correction of transposition of the great arteries. *J Am Coll Cardiol*. 1998;32:1955–1961.
30. Hauser M, Bengel FM, Kuhn A, et al. Myocardial blood flow and flow reserve after coronary reimplantation in patients after arterial switch and ross operation. *Circulation*. 2001;103:1875–1880.
31. Soedamah-Muthu SS, Fuller JH, Mulnier HE, et al. High risk of cardiovascular disease in patients with type 1 diabetes in the U.K.: a cohort study using the general practice research database. *Diabetes Care*. 2006;29:798–804.
32. Tukenova M, Guibout C, Oberlin O, et al. Role of cancer treatment in long-term overall and cardiovascular mortality after childhood cancer. *J Clin Oncol*. 2010;28:1308–1315.
33. Meacham LR, Chow EJ, Ness KK, et al. Cardiovascular risk factors in adult survivors of pediatric cancer—a report from the childhood cancer survivor study. *Cancer Epidemiol Biomarkers Prev*. 2010;19:170–181.
34. Mahle WT, Vincent RN, Berg AM, Kanter KR. Pravastatin therapy is associated with reduction in coronary allograft vasculopathy in pediatric heart transplantation. *J Heart Lung Transplant*. 2005;24:63–66.
35. Steinberger J, Daniels SR, Eckel RH, et al. Progress and challenges in metabolic syndrome in children and adolescents: a scientific statement from the American Heart Association Atherosclerosis, Hypertension, and Obesity in the Young

Committee of the Council on Cardiovascular Disease in the Young; Council on Cardiovascular Nursing; and Council on Nutrition, Physical Activity, and Metabolism. *Circulation*. 2009;119:628–647.
36. Ford ES, Giles WH, Dietz WH. Prevalence of the metabolic syndrome among US adults: findings from the third National Health and Nutrition Examination Survey. *JAMA*. 2002;287:356–359.
37. de Ferranti SD, Gauvreau K, Ludwig DS, et al. Prevalence of the metabolic syndrome in American adolescents. Findings from the Third National Health and Nutrition Examination Survey. *Circulation*. 2004; Oct 19;110(16):2494–2497. Epub 2004 Oct 11.
38. Cook S, Weitzman M, Auinger P, et al. Prevalence of a metabolic syndrome phenotype in adolescents: findings from the third National Health and Nutrition Examination Survey, 1988-1994. *Arch Pediatr Adolesc Med*. 2003;157:821–827.
39. Vanhala MJ, Vanhala PT, Keinanen-Kiukaanniemi SM, et al. Relative weight gain and obesity as a child predict metabolic syndrome as an adult. *Int J Obes Relat Metab Disord*. 1999;23:656–659.
40. Rubins HB, Robins SJ, Collins D, et al. Distribution of lipids in 8,500 men with coronary artery disease. Department of Veterans Affairs HDL Intervention Trial Study Group. *Am J Cardiol*. 1995;75:1196–1201.
41. EUROASPIRE. A European Society of Cardiology survey of secondary prevention of coronary heart disease: principal results. EUROASPIRE Study Group. European Action on Secondary Prevention through Intervention to Reduce Events. *Eur Heart J*. 1997;18:1569–1582.
42. Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. *N Engl J Med*. 2000;342:836–843.
43. Ridker PM, Cushman M, Stampfer MJ, et al. Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. *N Engl J Med*. 1997;336:973–979.
44. Pearson TA, Mensah GA, Alexander RW, et al. Markers of inflammation and cardiovascular disease: application to clinical and public health practice: a statement for healthcare professionals from the Centers for Disease Control and Prevention and the American Heart Association. *Circulation*. 2003;107:499–511.
45. Margaglione M, Cappucci G, Colaizzo D, et al. C-reactive protein in offspring is associated with the occurrence of myocardial infarction in first-degree relatives. *Arterioscler Thromb Vasc Biol*. 2000;20:198–203.
46. de Ferranti SD, Gauvreau K, Ludwig DS, et al. Inflammation and changes in metabolic syndrome abnormalities in US adolescents: findings from the 1988–1994 and 1999–2000 National Health and Nutrition Examination Surveys. *Clin Chem*. 2006; Jul;52(7):1325–1330. Epub 2006 May 4.
47. Balagopal PB, de Ferranti SD, Cook S, et al. Nontraditional risk factors and biomarkers for cardiovascular disease: mechanistic, research, and clinical considerations for youth: a scientific statement from the American Heart Association. *Circulation*. 2011;123:2749–2769.
48. Ravelli GP, Stein ZA, Susser MW. Obesity in young men after famine exposure in utero and early infancy. *N Engl J Med*. 1976;295:349–353.
49. Roseboom TJ, van der Meulen JH, Ravelli AC, et al. Effects of prenatal exposure to the Dutch famine on adult disease in later life: an overview. *Mol Cell Endocrinol*. 2001;185:93–98.
50. Whincup PH, Kaye SJ, Owen CG, et al. Birth weight and risk of type 2 diabetes: a systematic review. *JAMA*. 2008;300:2886–2897.

51. RG IJ, Stehouwer CD, Van Weissenbruch MM, et al. Evidence for genetic factors explaining the association between birth weight and low-density lipoprotein cholesterol and possible intrauterine factors influencing the association between birth weight and high-density lipoprotein cholesterol: analysis in twins. *J Clin Endocrinol Metab*. 2001;86:5479–5484.
52. Owen CG, Whincup PH, Odoki K, et al. Infant feeding and blood cholesterol: a study in adolescents and a systematic review. *Pediatrics*. 2002;110:597–608.
53. Parikh NI, Hwang SJ, Ingelsson E, et al. Breastfeeding in infancy and adult cardiovascular disease risk factors. *Am J Med*. 2009;122:656 e1–63 e1.
54. Nordestgaard BG, Chapman MJ, Ray K, et al. Lipoprotein(a) as a cardiovascular risk factor: current status. *Eur Heart J*. 2010;31:2844–2853.
55. Emerging Risk Factors C, Erqou S, Kaptoge S, et al. Lipoprotein(a) concentration and the risk of coronary heart disease, stroke, and nonvascular mortality. *JAMA*. 2009;302:412–423.
56. Guyton JR, Blazing MA, Hagar J, et al. Extended-release niacin vs gemfibrozil for the treatment of low levels of high-density lipoprotein cholesterol. Niaspan-Gemfibrozil Study Group. *Arch Intern Med*. 2000;160:1177–1184.
57. Colletti RB, Neufeld EJ, Roff NK, et al. Niacin treatment of hypercholesterolemia in children. *Pediatrics*. 1993;92:78–82.
58. Investigators A-H, Boden WE, Probstfield JL, et al. Niacin in patients with low HDL cholesterol levels receiving intensive statin therapy. *N Engl J Med*. 2011;365:2255–2267.
59. Mensink RP, Katan MB. Effect of dietary trans fatty acids on high-density and low-density lipoprotein cholesterol levels in healthy subjects. *N Engl J Med*. 1990;323:439–445.
60. U.S. Department of Agriculture and U.S. Department of Health and Human Services. Dietary Guidelines for Americans, 2010. 7th Edition, Washington, DC: U.S. Government Printing Office, December 2010. http://www.health.gov/dietaryguidelines/dga2010/DietaryGuidelines 2010.pdf.
61. Rosenson R, Lloyd-Jones D, Working group from the Adult Treatment Panel IIIotNCEP. Critical appraisal of revised cholesterol guidelines for the very high-risk patient. *Expert Rev Cardiovasc Ther*. 2005;3:173–178.
62. Goliasch G, Oravec S, Blessberger H, et al. Relative importance of different lipid risk factors for the development of myocardial infarction at a very young age (</=40 years of age). *Eur J Clin Invest*. 2012;42:631–636.
63. Kit BK, Carroll MD, Lacher DA, et al. Trends in serum lipids among US youths aged 6 to 19 years, 1988–2010. *JAMA*. 2012;308:591–600.
64. de Ferranti SD. Declining cholesterol levels in US youths: a reason for optimism. *JAMA*. Aug 8 2012;308(6):621–622. PMID: 22871874.
65. Austin MA, Hutter CM, Zimmern RL, Humphries SE. Genetic causes of monogenic heterozygous familial hypercholesterolemia: a HuGE prevalence review. *Am J Epidemiol*. 2004;160:407–420.
66. Vuorio AF, Turtola H, Piilahti KM, et al. Familial hypercholesterolemia in the Finnish north Karelia. A molecular, clinical, and genealogical study. *Arterioscler Thromb Vasc Biol*. 1997;17:3127–3138.
67. Malatack JJ. Liver transplantation as treatment for familial homozygous hypercholesterolemia: too early or too late. *Pediatr Transplant*. 2011;15:123–125.
68. Van Craeyveld E, Jacobs F, Gordts SC, De Geest B. Gene therapy for familial hypercholesterolemia. *Curr Pharm Des*. 2011;17:2575–2591.
69. Grossman M, Rader DJ, Muller DW, et al. A pilot study of ex vivo gene therapy for homozygous familial hypercholesterolaemia. *Nat Med*. 1995;1:1148–1154.

70. Psaty BM, Rivara FP. Universal screening and drug treatment of dyslipidemia in children and adolescents. *JAMA*. 2012;307:257–258.
71. Gillman MW, Daniels SR. Is universal pediatric lipid screening justified? *JAMA*. 2012;307:259–260.
72. de Ferranti SD, Daniels SR, Gillman M, et al. NHLBI Integrated Guidelines on Cardiovascular Disease Risk Reduction: can we clarify the controversy about cholesterol screening and treatment in childhood? *Clin Chem*. 2012;58:1626–1630.
73. Dennison BA, Jenkins PL, Pearson TA. Challenges to implementing the current pediatric cholesterol screening guidelines into practice. *Pediatrics*. 1994;94:296–302.
74. Rifai N, Neufeld E, Ahlstrom P, et al. Failure of current guidelines for cholesterol screening in urban African-American adolescents. *Pediatrics*. 1996;98:383–388.
75. Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Executive summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). *JAMA*. 2001;285:2486–2497.
76. Poti JM, Popkin BM. Trends in energy intake among US children by eating location and food source, 1977–2006. *J Am Diet Assoc*. 2011;111:1156–1164.
77. Guthrie JF, Lin BH, Frazao E. Role of food prepared away from home in the American diet, 1977–78 versus 1994–96: changes and consequences. *J Nutr Educ Behav*. 2002;34:140–150.
78. Bowman SA, Gortmaker SL, Ebbeling CB, et al. Effects of fast-food consumption on energy intake and diet quality among children in a national household survey. *Pediatrics*. 2004;113:112–118.
79. Dennison BA, Rockwell HL, Nichols MJ, Jenkins P. Children's growth parameters vary by type of fruit juice consumed. *J Am Coll Nutr*. 1999;18:346–352.
80. Skinner JD, Carruth BR, Moran J, 3rd, et al. Fruit juice intake is not related to children's growth. *Pediatrics*. 1999;103:58–64.
81. Nguyen TT. Cholesterol-lowering trials and coronary heart disease in the United States. In: Nguyen TT (ed.) *Postgraduate Medicine Special Report: New Developments in the Dietary Management of High Cholesterol*. Minneapolis, MN: McGraw-Hill Companies; 1998. pp. 15–26.
82. Polonsky SM, Bellet PS, Sprecher DL. Primary hyperlipidemia in a pediatric population: classification and effect of dietary treatment. *Pediatrics*. 1993;91:92–96.
83. Glueck CJ, Fallat RW, Mellies M, Tsang RC. Pediatric familial type II hyperlipoproteinemia: therapy with diet and colestipol resin. *Pediatrics*. 1976;57:68–74.
84. Kwiterovich PO, Jr., Bachorik PS, Franklin FA, Jr., et al. Effect of dietary treatment on the plasma levels of lipids, lipoprotein cholesterol and LDL B protein in children with type II hyperlipoproteinemia. *Prog Clin Biol Res*. 1985;188:123–137.
85. Lapinleimu H, Viikari J, Jokinen E, et al. Prospective randomised trial in 1062 infants of diet low in saturated fat and cholesterol. *Lancet*. 1995;345:471–476.
86. Niinikoski H, Lagstrom H, Jokinen E, et al. Impact of repeated dietary counseling between infancy and 14 years of age on dietary intakes and serum lipids and lipoproteins: the STRIP study. *Circulation*. 2007;116:1032–1040.
87. Niinikoski H, Pahkala K, Ala-Korpela M, et al. Effect of repeated dietary

counseling on serum lipoproteins from infancy to adulthood. *Pediatrics*. 2012;129:e704–e713.
88. Efficacy and safety of lowering dietary intake of fat and cholesterol in children with elevated low-density lipoprotein cholesterol. The Dietary Intervention Study in Children (DISC). The Writing Group for the DISC Collaborative Research Group. *JAMA*. 1995;273:1429–1435.
89. Rask-Nissila L, Jokinen E, Terho P, et al. Effects of diet on the neurologic development of children at 5 years of age: the STRIP project. *J Pediatr*. 2002;140:328–333.
90. de Lorgeril M, Salen P, Martin JL, et al. Mediterranean diet, traditional risk factors, and the rate of cardiovascular complications after myocardial infarction: final report of the Lyon Diet Heart Study. *Circulation*. 1999;99:779–785.
91. Estruch R, Ros E, Salas-Salvado J, et al. Primary prevention of cardiovascular disease with a Mediterranean diet. *N Engl J Med*. 2013;368: 1279–1290.
92. Giannini C, Diesse L, D'Adamo E, et al. Influence of the Mediterranean diet on carotid intima-media thickness in hypercholesterolaemic children: a 12-month intervention study. *Nutr Metab Cardiovasc Dis*. 2013; Jan;24(1):75–82. doi: 10.1016/j.numecd.2013.04.005. Epub 2013 Jun 25.
93. Yancy WS, Jr., Olsen MK, Guyton JR, et al. A low-carbohydrate, ketogenic diet versus a low-fat diet to treat obesity and hyperlipidemia: a randomized, controlled trial. *Ann Intern Med*. 2004;140:769–777.
94. Samaha FF, Iqbal N, Seshadri P, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. *N Engl J Med*. 2003;348:2074–2081.
95. Dumesnil JG, Turgeon J, Tremblay A, et al. Effect of a low-glycaemic index—low-fat—high protein diet on the atherogenic metabolic risk profile of abdominally obese men. *Br J Nutr*. 2001;86:557–568.
96. Bouche C, Rizkalla SW, Luo J, et al. Five-week, low-glycemic index diet decreases total fat mass and improves plasma lipid profile in moderately overweight nondiabetic men. *Diabetes Care*. 2002;25:822–828.
97. Pereira MA, Swain J, Goldfine AB, et al. Effects of a low-glycemic load diet on resting energy expenditure and heart disease risk factors during weight loss. *JAMA*. 2004;292:2482–2490.
98. Damsgaard CT, Papadaki A, Jensen SM, et al. Higher protein diets consumed ad libitum improve cardiovascular risk markers in children of overweight parents from eight European countries. *J Nutr*. 2013;143:810–817.
99. Iannuzzi A, Licenziati MR, Vacca M, et al. Comparison of two diets of varying glycemic index on carotid subclinical atherosclerosis in obese children. *Heart Vessels*. 2009;24:419–424.
100. Sondike SB, Copperman N, Jacobson MS. Effects of a low-carbohydrate diet on weight loss and cardiovascular risk factor in overweight adolescents. *J Pediatr*. 2003;142:253–258.
101. Lindquist CH, Gower BA, Goran MI. Role of dietary factors in ethnic differences in early risk of cardiovascular disease and type 2 diabetes. *Am J Clin Nutr*. 2000;71:725–732.
102. Fajcsak Z, Gabor A, Kovacs V, Martos E. The effects of 6-week low glycemic load diet based on low glycemic index foods in overweight/obese children—pilot study. *J Am Coll Nutr*. 2008;27:12–21.
103. Walter HJ, Hofman A, Vaughan RD, Wynder EL. Modification of risk factors for coronary heart disease. Five-year results of a school-based intervention trial. *N Engl J Med*. 1988;318:1093–1100.
104. Manios Y, Moschandreas J, Hatzis C, Kafatos A. Health and nutrition education

in primary schools of Crete: changes in chronic disease risk factors following a 6-year intervention programme. *Br J Nutr*. 2002;88:315–324.
105. Glassman M, Spark A, Berezin S, et al. Treatment of type IIa hyperlipidemia in childhood by a simplified American Heart Association diet and fiber supplementation. *Am J Dis Child*. 1990;144:973–976.
106. Zavoral JH, Hannan P, Fields DJ, et al. The hypolipidemic effect of locust bean gum food products in familial hypercholesterolemic adults and children. *Am J Clin Nutr*. 1983;38:285–294.
107. Davidson MH, Dugan LD, Burns JH, et al. A psyllium-enriched cereal for the treatment of hypercholesterolemia in children: a controlled, double-blind, crossover study. *Am J Clin Nutr*. 1996;63:96–102.
108. Tammi A, Ronnemaa T, Gylling H, et al. Plant stanol ester margarine lowers serum total and low-density lipoprotein cholesterol concentrations of healthy children: the STRIP project. Special Turku Coronary Risk Factors Intervention Project. *J Pediatr*. 2000;136:503–510.
109. Ketomaki AM, Gylling H, Antikainen M, et al. Red cell and plasma plant sterols are related during consumption of plant stanol and sterol ester spreads in children with hypercholesterolemia. *J Pediatr*. 2003;142:524–531.
110. Amundsen AL, Ose L, Nenseter MS, Ntanios FY. Plant sterol ester-enriched spread lowers plasma total and LDL cholesterol in children with familial hypercholesterolemia. *Am J Clin Nutr*. 2002;76:338–344.
111. Marchioli R, Barzi F, Bomba E, et al. Early protection against sudden death by n-3 polyunsaturated fatty acids after myocardial infarction: time-course analysis of the results of the Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico (GISSI)-Prevenzione. *Circulation*. 2002;105:1897–1903.
112. Risk and Prevention Study Collaborative G, Roncaglioni MC, Tombesi M, et al. n-3 Fatty acids in patients with multiple cardiovascular risk factors. *N Engl J Med*. 2013;368:1800–1808.
113. Heart Protection Study Collaborative G, Bulbulia R, Bowman L, et al. Effects on 11-year mortality and morbidity of lowering LDL cholesterol with simvastatin for about 5 years in 20,536 high-risk individuals: a randomised controlled trial. *Lancet*. 2011;378:2013–2020.
114. Shepherd J, Cobbe SM, Ford I, et al. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. West of Scotland Coronary Prevention Study Group. *N Engl J Med*. 1995;333:1301–1307.
115. Downs JR, Clearfield M, Weis S, et al. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. Air Force/Texas Coronary Atherosclerosis Prevention Study. *JAMA*. 1998;279:1615–1622.
116. Tonstad S, Knudtzon J, Sivertsen M, et al. Efficacy and safety of cholestyramine therapy in peripubertal and prepubertal children with familial hypercholesterolemia. *J Pediatr*. 1996;129:42–49.
117. McCrindle BW, O'Neill MB, Cullen-Dean G, Helden E. Acceptability and compliance with two forms of cholestyramine in the treatment of hypercholesterolemia in children: a randomized, crossover trial. *J Pediatr*. 1997;130:266–273.
118. Reihner E, Angelin B, Rudling M, et al. Regulation of hepatic cholesterol metabolism in humans: stimulatory effects of cholestyramine on HMG-CoA reductase activity and low density lipoprotein receptor expression in gallstone patients. *J Lipid Res*. 1990;31:2219–2226.
119. Vuorio A, Kuoppala J, Kovanen PT, et al. Statins for children with familial

hypercholesterolemia. *Cochrane Database Syst Rev*. 2010; Jul 7;(7):CD006401.
120. Gagne C, Bays HE, Weiss SR, et al. Efficacy and safety of ezetimibe added to ongoing statin therapy for treatment of patients with primary hypercholesterolemia. *Am J Cardiol*. 2002;90:1084–1091.
121. Clauss S, Wai KM, Kavey RE, Kuehl K. Ezetimibe treatment of pediatric patients with hypercholesterolemia. *J Pediatr*. 2009;154:869–872.
122. Schiel R, Bambauer R, Muller U. Low-density lipoprotein apheresis versus lipid lowering drugs in the treatment of severe hypercholesterolemia: four years' experience. *Artif Organs*. 1996;20:318–323.
123. Zwiener RJ, Uauy R, Petruska ML, Huet BA. Low-density lipoprotein apheresis as long-term treatment for children with homozygous familial hypercholesterolemia. *J Pediatr*. 1995;126:728–735.
124. McKenney JM, Koren MJ, Kereiakes DJ, et al. Safety and efficacy of a monoclonal antibody to proprotein convertase subtilisin/kexin type 9 serine protease, SAR236553/REGN727, in patients with primary hypercholesterolemia receiving ongoing stable atorvastatin therapy. *J Am Coll Cardiol*. 2012;59:2344–2353.
125. Akdim F, Stroes ES, Sijbrands EJ, et al. Efficacy and safety of mipomersen, an antisense inhibitor of apolipoprotein B, in hypercholesterolemic subjects receiving stable statin therapy. *J Am Coll Cardiol*. 2010;55:1611–1618.
126. Moskowitz WB, Schwartz PF, Schieken RM. Childhood passive smoking, race, and coronary artery disease risk: the MCV Twin Study. Medical College of Virginia. *Arch Pediatr Adolesc Med*. 1999;153: 446–453.
127. Neufeld EJ, Mietus-Snyder M, Beiser AS, et al. Passive cigarette smoking and reduced HDL cholesterol levels in children with high-risk lipid profiles. *Circulation*. 1997;96:1403–1407.
128. Porter JS, Bean MK, Gerke CK, Stern M. Psychosocial factors and perspectives on weight gain and barriers to weight loss among adolescents enrolled in obesity treatment. *J Clin Psychol Med Settings*. 2010;17:98–102.

Diet and Dental Disease

64

Rosalyn Sulyanto, DMD, MS and James H. Shaw, DMD, PhD

The position of the oral cavity at the entrance to the gastrointestinal tract requires that it perform functions that need diverse tissues and specialized structures. The foods and other items that children eat and drink vary widely in their particle size and texture, degree of hardness, pH, and other chemical stimuli, as well as wide ranges of temperature. These different characteristics produce stresses on the oral components that must be moderated before swallowing. In the mouth, food items must be subdivided by chewing and simultaneously diluted and buffered with saliva until the bolus is suitable to swallow and pass on to the esophagus and lower areas of the gastrointestinal tract. The amount and composition of saliva secreted on each occasion are in response to the whole experience of eating—from the initial seeing, smelling, and anticipating to the tasting, chewing, and swallowing.[1,2] Especially in young children, many undesirable materials are taken into the mouth, which contribute various kinds of microorganisms and/or other dangerous entities.

The temperature and humidity of the oral cavity, as well as the frequent presence of all required nutrients for cellular growth and multiplication in food and drink, result in the mouth being an ideal incubator for many types of microorganisms (oral biofilm) from birth onward. It has been demonstrated through evidence-based dentistry that dental caries is an infectious disease and that mutans streptococci are the primary etiologic bacterial agents in the biofilm in humans. The pathogenicity of these microorganisms is related to their adherence characteristics; acidic nature; and resistance to low pH levels. Early colonization by mutans streptococci is closely related to a high dental caries experience, although the mere presence of mutans streptococci is

not sufficient to develop dental caries. At present, there are three events for the development of carious lesions during early childhood: (1) primary infection and initial colonization by mutans streptococci; (2) increase of the mutans streptococcus population in the mouth associated with the frequent and long exposure to a cariogenic diet rich in carbohydrates; and (3) demineralization and development of caries.[76] An important source of the infant's oral microbiome is the primary caregiver.[3] There is evidence of matrilineal transmission of mutans streptococci in very early childhood. Strong correlations between salivary mutans streptococci counts in mothers and their children have been reported. Recent studies indicate that mutans streptococci can colonize the mouth of predentate infants and that horizontal transmission (e.g., between family members or children in a day care setting) as well as vertical transmission (between caregiver and child with the major reservoir from mother to infant) occurs.[4] Maternal salivary levels of mutans streptococci infection may influence infection rates of mutans streptococci in their children at ages 6–18 months and at 4–5 years. Salivary concentrations of 10^5 colony-forming units mutans streptococci per milliliter of maternal saliva were associated with a 52% infection rate in their children compared with only a 6% infection rate when the maternal saliva concentration was 10^3 or below.[5,6] The presence of caries in the mother and siblings increases risks for the child, but the most consistent predictor of risk of caries (dental decay) in children is past caries experience.[7] Nutrients and the foods that provide them have numerous relationships to the oral tissues and structures, and these relationships vary depending on the nature of the tissue or structure itself. Some relationships differ from those elsewhere in the body. For example, sugars, when metabolized throughout the body, are wholesome sources of energy for the cells' many and varied functions. However, in contrast to this vital systemic relationship, the sugars have an undesirable local effect in the mouth. On the surfaces of dental enamel, sugars support the metabolism of microorganisms in dental plaque with the production of acid.

In the life history of the soft tissues, a transition occurs at birth as a result of the new, stressful exposure of their outer surfaces to microorganisms and food, meanwhile still being nourished systemically. A much more striking transition occurs for the teeth, especially the enamel surfaces, at the time of their eruption into the oral cavity. Prior to eruption, the external surfaces of enamel are surrounded by the enamel cap, including the enamel-forming cells of ectodermal origin, the ameloblasts, and the inner surface of the dentin is covered by the layer of odontoblasts,

the dentin-forming cells, which are embryonically of mesodermal origin. Both enamel and dentin throughout their formation and mineralization are related fully to the systemic environment. At eruption, the enamel cap is ruptured and the outer surfaces of enamel become exposed to the oral milieu of saliva, microorganisms, and food. The odontoblastic layer remains intact in contact with the capillaries of the pulp and continues to metabolize slowly with the centripetal formation of secondary dentin at the expense of the connective tissue in the pulp. The latter process accelerates when toxic metabolites from a deep carious lesion in the dentin trigger a protective response with more rapid production of secondary dentin.

STRUCTURE OF ENAMEL

Enamel and dentin are characteristically different from other tissues in the body. When fully mature, enamel contains only about 1% organic material and 2% water. Its tiny organic component consists of proteins chemically similar to the keratins of skin and is dispersed throughout the inorganic crystals as a network of extremely delicate and intricate fibrils.[8] The rest of enamel is the complex calcium–phosphorus salt hydroxyapatite, which varies somewhat in its composition, depending on the concentrations of anions and cations in the extracellular fluids at the time it crystallizes in the embryonic organic matrix. The hydroxyapatite in enamel has more perfectly formed crystals than those in dentin and bone. After its eruption, enamel contains no cells and is cut off from systemic influences other than through saliva, which bathes it continuously throughout life. Elements in saliva exchange with those on the outer surface of the enamel, especially in the period shortly after eruption.[9] Owing to its acellular nature, enamel has no ability to heal or remodel, with the exception that remineralization of early (precaries) surface lesions with the incorporation of calcium and phosphorus from saliva can occur under suitable conditions.[10,11]

STRUCTURE OF DENTIN

Dentin contains about 20% protein of the collagen family comparable to bone. About 70% of dentin is hydroxyapatite. Although dentin continues to form centripetally at a slow rate, like enamel, it is unable to remodel or repair itself as it does not contain anything comparable to the haversian system of bone. Any developmental errors in the formation of enamel and dentin remain throughout life as chemical or histologic artifacts in the kymographic record.

NUTRITIONAL STUDIES ON THE ORAL TISSUES AND STRUCTURES

Soft Tissues

The soft tissues, mucosa, gingiva, tongue, and taste buds, are especially susceptible to local stresses. Their metabolic activity is high. Oral mucosal cells in rats, for example, are replaced in 3–5 days by rapid cell division in the basal layer and sloughing of the surface cells; in contrast, the turnover rate in cheek skin is about 10 days.[12] This high metabolic activity accounts for the rapid healing of injuries to soft tissues within the mouth. However, the high metabolic activity of oral tissues results in a high susceptibility to nutritional deficiencies as adequate nutrients must be available to support the rapid cell multiplication and replacement.

In the early decades of nutrition as a discipline, deficiencies were often recognized first as clinical entities because of manifestations in the oral cavity; scurvy (vitamin C) and pellagra (niacin) are the prime examples.[13] The ease of oral examination for abnormalities with minimal violation of the subject's privacy also contributed to early recognition of nutritional deficiencies that affected oral components.

The increased understanding of nutrition and the attention to preventive nutrition, including legally required fortification of foods, such as niacin for the prevention of pellagra, have led to almost total elimination of such nutritional deficiencies in children of the industrialized nations. However, in the developing nations of the third world, especially in times of famine, such deficiencies still occur.

Mineralized Tissues

In early experimental studies with rats, severe deficiencies of vitamin A caused abnormalities in the ameloblasts, which led to hypoplastic enamel.[14] Severe vitamin C deficiency in guinea pigs, which, unlike rats, require preformed vitamin C in their diets, produced a syndrome in which the odontoblasts produced much less dentin.[15] As a result, the erupting teeth were thin and fragile. Acute deficiencies in rats and dogs of vitamin D or calcium or grossly abnormal calcium-to-phosphorus ratios resulted in inadequate mineralization and hypoplasia of the teeth.[16,17]

Little evidence exists to suggest that moderate deficiencies of these nutrients are of clinical significance in children in industrialized countries, although we still see cases of vitamin D–resistant rickets and scurvy. In severely malnourished children in developing countries, a linear enamel

hypoplasia (natal line) has been described in the anterior teeth of the primary dentition (Figure 64-1), which is much less frequent or absent in better nourished children in the same communities.[18,19] The poorly mineralized ring around the teeth in this syndrome was being formed about the time the children were born and was highly susceptible to dental caries. All children at birth had low serum vitamin A levels.

Low protein intake during the reproductive cycle of white rat dams resulted in delayed eruption of the molars of their offspring. Their teeth were smaller and more susceptible to tooth decay than those of the offspring of loosely related females provided with adequate protein.[20] The increased caries susceptibility in the deprived rats may be attributable to structural abnormalities in the salivary glands produced by the protein deficiency.[21]

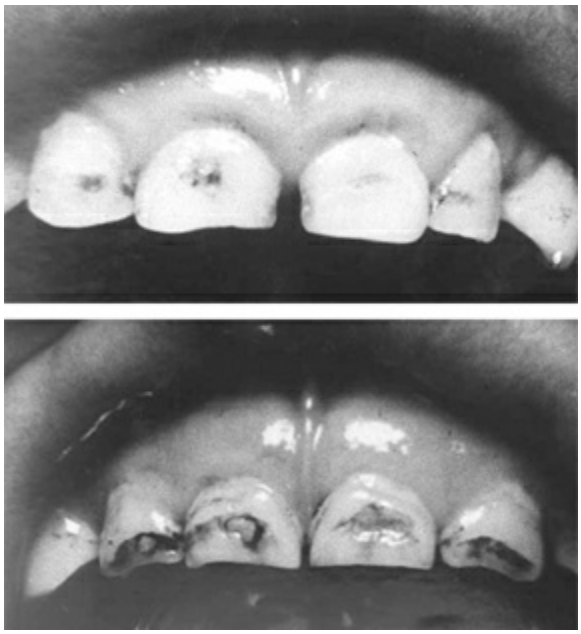


Figure 64-1 Photographs of linear hypoplasia of the anterior teeth of Guatemalan children. These hypoplastic areas are prone to develop dental caries. (Reproduced with permission from Sweeney and Guzman[18] and Sweeney et al.[19])

Alvarez[22] conducted two cross-sectional and one longitudinal study among Peruvian children in whom malnutrition in early life was prevalent. He studied the influence of malnutrition on the time of tooth eruption and of deciduous tooth exfoliation and the susceptibility of both deciduous and

permanent teeth to dental caries. As in the earlier studies with rodents, tooth eruption was delayed significantly among malnourished children. Because rodents have only a single dentition, exfoliation could not be studied in the laboratory. Among the malnourished Peruvian children, their deciduous teeth exfoliated appreciably later than those in better-nourished children in the same communities. When the length of time that the deciduous teeth were exposed to the oral environment was considered, instead of the traditional method of considering only the chronologic age of the children, the deciduous teeth of malnourished children were observed to have a higher incidence of caries than those of the better-nourished children. Even a single episode of moderate malnutrition before 1 year of age also resulted in a higher incidence of dental caries of the permanent incisors and first molars, which were being formed and were starting to mineralize during that interval. The increased incidence of dental caries among these children again paralleled the rodent studies conducted three decades earlier. Alvarez suggested that this observation may result from imperfect formation of dental enamel early in life.

Several studies, in animals and humans, have evaluated the effect of nutrition and nutrients on the growth and development of oral structures. The conclusion of many studies is that adequate nutrition is essential from the earliest periods of development. Further, a firm diet enhances proper growth and development compared with a soft diet. Specifically, the effect of diet consistency has been shown to affect growth of the mandible and maxilla and proper tooth eruption. Ongoing research on the harmful effects of nutrition on oral growth indicates that excess vitamin A can be a potent teratogen that may contribute to oral defects such as cleft palate. Protective effects of folic acid have been shown in animal studies but not in humans as yet, probably owing to the complexity of other factors also involved.[23]

Adequate amounts of all nutrients should be provided throughout the long periods of tooth formation in children to ensure their normal development, that is, from the last trimester of pregnancy through 16 years of age.

DISEASES OF THE ORAL CAVITY

The two principal diseases of the oral cavity are dental caries and periodontal disease (destruction of the tissues that support the teeth in the jaws). Although the latter is the chief cause of tooth loss in adults, juvenile periodontitis occurs infrequently in children, is much more localized than in adults, and may be associated with an immunologic defect in some

children. Both diseases are of multifactorial origin, are chronic in nature, and progress slowly and intermittently. Nutrition is not known to have a relationship to juvenile periodontitis except possibly through dietary influences on the dental plaque.

Etiology of Dental Caries

A carious lesion results from an interaction between the carbohydrates in food, especially the sugars, with cariogenic microorganisms in the dental plaque on tooth surfaces (dental biofilm) and the tooth surface in the presence of saliva; the gross anatomy of the tooth, the composition of the enamel and dentin, and the quantity and quality of saliva all vary in ways that influence the rate of progression of tooth decay. The most serious adverse influence on the ability of the teeth to be maintained in the oral cavity is any reduction in the flow of saliva owing to salivary disease, radiation, or drug use that modifies the ability of the salivary glands to function.[24,25]

Under certain circumstances, the acidic metabolic products of the dental biofilm's microorganisms on the tooth surfaces from sugars reduce the pH of plaque sufficiently to demineralize enamel and, later, the dentin progressively until a carious lesion is produced. The epidemiologist commonly speaks of this type of disease production as the relationship of the agent (microorganism[s]), host (the tooth and the person in which it functions), and environment (the components in the milieu around the tooth and the home and community in which the individual lives) (Figure 64-2).[26]

All three parameters must simultaneously be in a caries-producing mode for carious lesions to be initiated and progress to become visually detectable. Periodontal disease also results from the interaction of food, the microorganisms in the crevices between the teeth and the gingiva, and the tissues in the area.

Agent. The microorganisms producing carious lesions are able to colonize the smooth surfaces of the teeth and/or the pits and fissures of the biting surfaces.[27] These microbiota (known as dental biofilm) are able to produce sufficiently acidic environments (pH around 5) on the tooth surfaces when metabolizing actively to demineralize enamel and later dentin. They must be able to continue to metabolize actively at these low pH values as the lesion progresses deeper into the tooth. The primary sources of energy required by these organisms are the sugars. *Streptococcus mutans* is of special interest because it produces mutans, a

polysaccharide of glucose, which enables it to adhere to smooth surfaces and to build up colonies in dental plaque. The nature of plaque is such that the microbial metabolites are produced in close proximity to enamel with restricted access to buffering saliva. In the human mouth, plaque contains numerous species of microorganisms with acid-producing capabilities. The complex nature of the oral milieu makes it virtually impossible to fully apply Koch's principles for identification of a specific causative organism. Probably more than one species is involved in most situations, but especially as lesions progress more deeply into the tooth with the need to metabolize in a strongly acidic environment. The recent recognition that dental plaque is a bacterial biofilm and that it behaves quite differently from its individual constituents has led to research to control plaque formation by preventing bacterial adhesion to the tooth surface. Physical approaches include coating the enamel surface with a polymer, whereas biological approaches to prevent initial colonization include inhibition of plaque matrix formation and inhibition of the bacterial signaling mechanisms.[28]

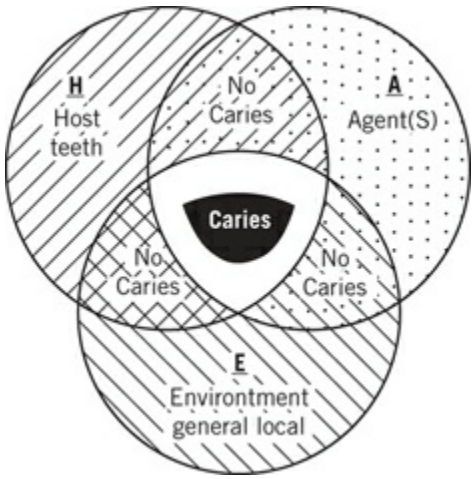


Figure 64-2 Diagrammatic illustration of the three major parameters simultaneously required for the initiation and progression of carious lesions. (Reproduced with permission from Keyes and Jordan.[26])

Host. Evidence of genetic determinants of caries resistance is limited in humans, yet a variety of subtle indications of genetic influences has been reported. Early suggestions that large differences in caries experience between national groups were genetically determined have been largely

discredited. The caries prevalence in industrialized nations tended to be routinely higher than in developing countries; however, as the dietary customs of the high-caries industrialized nations were adopted in developing countries with increased sugar consumption and more frequent eating, their low-caries experience has been increasing. Meanwhile, in the last 20 years, dental caries prevalence in the industrialized nations has been decreasing significantly.[29]

Comparison of the dental caries experience of twins indicated that identical twins had more similar caries experiences than fraternal twins.[30] This may not be a truly genetic influence as such environmental experiences as food preferences, frequency of eating, and tooth brushing techniques may be more similar for identical than for fraternal twins. Similarities in dental caries for twins reared separately were considered to be about 50% attributable to genetic influences.[31] Numerous trials have been conducted to develop animal strains of divergent caries resistance. Superficially, differences attributable to heredity seemed to be produced. However, when so-called resistant strains were heavily challenged early in life with cariogenic microorganisms, carious lesions developed readily, although not always as extensively as in the seemingly more susceptible strains.

Mandel[32] suggested three avenues through which genetic influences may be mediated: (1) salivary components that buffer plaque acids—elevating the pH and influencing the bacterial ecology of plaque, (2) immunologic and nonimmunologic constituents of oral fluids that have the capacity to alter the ability of cariogenic bacteria to attach to enamel, and (3) protein-bound lipids on the surfaces of teeth that may alter the permeability to acids produced in plaque.

Environment. The single most important variable in the oral environment is the frequency of consuming foods containing sugars that adhere to tooth surfaces. The absolute amount of sugars consumed is less important than the frequency of eating. In the 1930s and 1940s, numerous studies were conducted on the relation of frequency of eating to caries experience (Figure 64-3).[33] Almost invariably, the conclusion was that caries experience increased as frequency of food consumption, especially of between-meal snacks, increased. More recently, this type of trial has been less frequent and the above relationship has not been as readily demonstrable. Since the earlier studies, food habits have changed dramatically. At that time, most food was prepared and eaten at home, and the amount of sugar used was controlled by the homemaker. Between-meal snacks were usually candy and cookies. Today, relatively little food

is prepared at home; many items are commercially prepared, with the amount of sugar controlled industrially on the basis of the amount needed for "best" taste. In addition, a much higher percentage of meals are consumed away from the home. Snack consumption is much more frequent, and less dependence is placed on three main meals a day.

The amount of sugar available per capita in a country is a reasonable index of the prevalence of dental caries among its children (Figure 64-4).[34] The information supporting the pivotal role of sugars in the etiology of dental caries was considered by 1977 to be sufficiently conclusive that Horowitz stated that in human dental experimentation:

> *the control group should not be asked to engage in any practice that may have a harmful effect on the dentition or its surrounding tissues. For example, subjects in a control group should not be given or asked to use chewing gum sweetened with sucrose, nor should they be asked to use sucrose rinses or other agents to produce decalcification of tooth enamel or initiate carious lesions, unless the investigators are certain that they can completely "reverse" the defects.*[35]

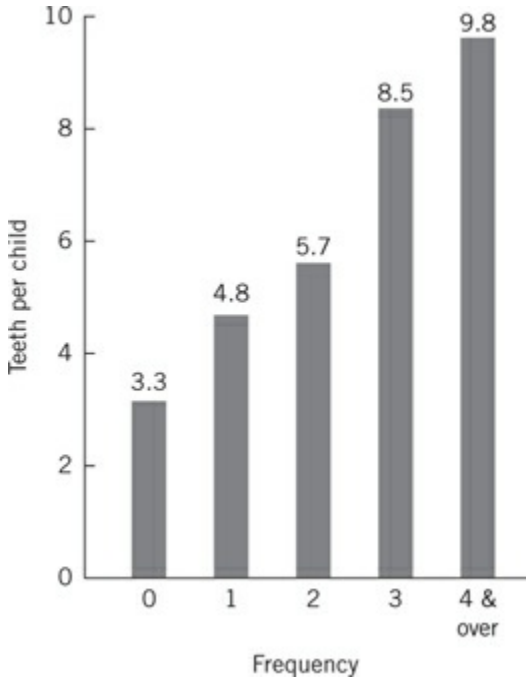


Figure 64-3 This bar graph compares the number of decayed, missing, and filled deciduous teeth with the frequency of between-meal eating in preschool children in Tennessee.

*Includes extracted primary molars. (Reproduced with permission from Weis and Trithart.[33])

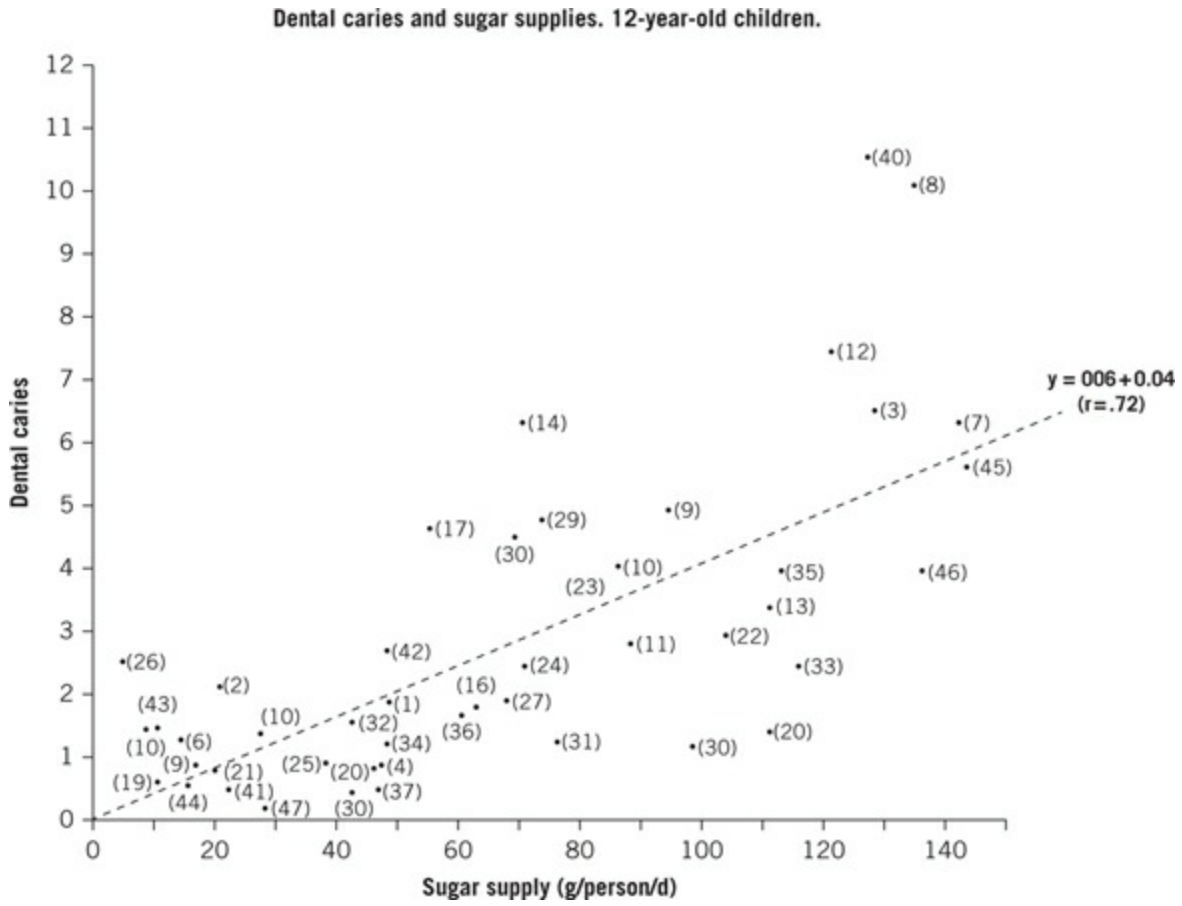


Figure 64-4 This graph represents a comparison of the number of decayed, missing, and filled permanent teeth with the sugar supply in grams per person per day for 47 countries. (Reproduced with permission from Sreebny.[34])

Mandel[32] stated that little disagreement exists that differences in amount, form, and frequency of readily fermentable carbohydrates can affect the rate of caries attack, but considerable variation between children exists when intake appears to be generally comparable.

Foods are complex mixtures of many chemicals. As yet, it has not been possible to devise a series of animal trials and evaluations in the human oral cavity that dependably estimate the caries-producing value of a specific food.[36] Some food items that contain relatively little sugar but substantial amounts of starch are more capable of producing caries lesions

than expected; others containing high amounts of sugar are less capable of supporting the caries process than expected.

A special case of rampant dental caries limited to the maxillary anterior deciduous teeth and posterior molars has become known as early childhood caries. This problem occurs in young children who are put to bed at night or for rests during the day with a bottle of formula or sweetened juice. When they go to sleep, salivary secretion decreases drastically, and the fermentable carbohydrates in the milk or juice are not diluted or washed away but are readily available to the microorganisms on tooth surfaces.[37] It is believed that the tongue affords protection to the mandibular anterior teeth so that they are pathognomonically not affected. Excessive on-demand breastfeeding is also capable of causing the same problem. The sugar, lactose, in human milk is clearly able to support the caries process.

Pediatricians and pediatric dentists on a preventive basis need to urge mothers of young infants to follow feeding practices in which excessive exposure to sweetened fluids and breast milk does not occur. When early childhood caries is already present, appropriate dietary corrections need to be made by less frequent feeding and the use of fewer sweet snacks of all kinds. Appropriate professional dental care is needed, probably including topical fluoride applications, such as a fluoride varnish, coupled with strong oral hygiene procedures at home.[38] The latter will include the parent's brushing of the infant's teeth with fluoridated toothpaste. Care is needed so that excess fluoride is not swallowed.

The importance of maintaining the deciduous teeth is not to be underestimated. They are needed as "space maintainers" as the jaws grow in order that there will be adequate room for eruption of the permanent teeth. Increased likelihood of orthodontic problems accompanies the early loss of deciduous teeth. High levels of untreated caries in the deciduous teeth provide reservoirs of cariogenic microorganisms from which the permanent teeth become quickly infected when they erupt into the oral cavity.

Reduced Ability of Polyols to Support Caries. Measurement of the pH of dental plaque in vivo with tiny, sensitive electrodes has become a convenient means of evaluating how various solutions and foods in the oral cavity affect the metabolism in the dental plaque in proximity to the tooth surface. In 1944, Stephan reported on a sharp depression in the pH of dental plaque after a glucose rinse was swished around the mouth.[37]

This depression from the resting value with a pH around 6.6–7.0 (the range observed in saliva) to 5.0 or less within minutes after the start of the

glucose rinse lasted for a few minutes and then slowly returned to the resting value. Plots of results from this kind of evaluation have become known as Stephen curves (Figure 64-5).[39,40] The consensus from the Stephen curve concerning the relation of plaque pH to caries production is shown in Table 64-1.

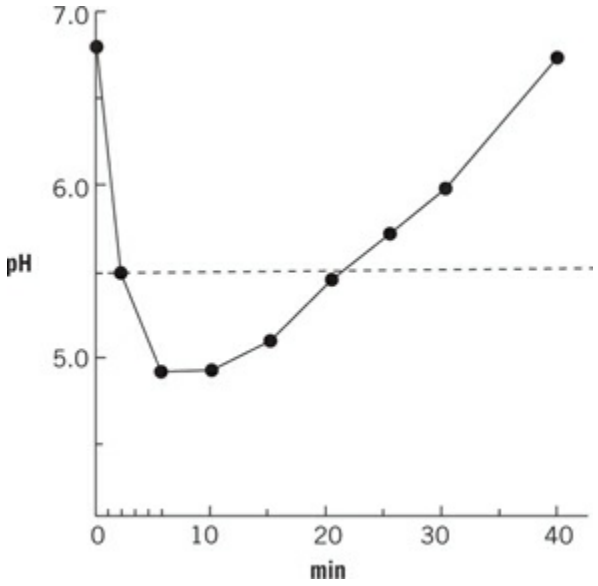


Figure 64-5 The typical Stephen curve showing the precipitous decrease in pH measured in vivo in dental plaque after a glucose rinse and the slower return to the resting pH. (Adapted from Stephan[39] and Edgar.[40])

Both the equipment and the procedures for intraoral tests have been refined and are used widely today. For example, certain varieties of cheese eaten shortly after a sugar rinse or a sweet food prevent the rapid pH drop in dental plaque (Figure 64-6).[40] Likewise, chewing a gum sweetened with a polyol, sorbitol, or xylitol does not result in a pH decrease, whereas chewing a gum sweetened with sucrose causes the typical caries-producing decrease (Figure 64-7).

Oral microorganisms have greatly limited abilities to metabolize the polyols, xylitol (5 carbon), sorbitol, and mannitol.[41,42] The effect of sugar substitutes on changes in caries rates has been evaluated in several observational studies as well as clinical trials, with results consistently demonstrating a protective effect of xylitol on caries incidence. Sorbitol was also shown to decrease caries rates compared with controls; however, the reductions in caries rates were greatest when xylitol was the sugar

substitute. Some limitations of these studies include a lack of radiographs in caries diagnosis, high loss to follow-up, potential confounding, and bias owing to the nature of long-term community intervention studies. The criteria for causality—consistency, strength association, biologic plausibility, temporal sequence, and dose–response relationship—should be considered. First, these studies are remarkably consistent both in terms of the magnitude of the effect observed as well as the consistent demonstration that xylitol was superior to sorbitol in decreasing the risk of dental caries. Second, the relative risks observed are considered strong (0.19–0.4). It is biologically plausible that xylitol can reduce dental caries because the pH of plaque is lowered with xylitol compared with sucrose. Fourth, a dose–response trend was observed in the two studies that evaluated different concentrations of xylitol, with the greatest effect observed in the subjects using the strongest xylitol preparations. The evidence to date suggests a strong caries-protective effect of xylitol.[43]

TABLE 64-1 Relation of Plaque pH to Caries Production

	Potential for Caries
Plaque p^H	**Initiation and development**
Above 6.0	Safe area; low solubility of enamel; calcium and phosphorous adequate to saturate environment
6.0–5.5	Doubtful or very slow progression
Below 5.5	Danger area; high solubility of enamel; calcium and phosphorous inadequate to saturate environment

Sources: Adapted from Stephan[39] and Edgar.[40]

In vivo pH testing has been used extensively in Switzerland, not only in basic research but also in the estimation of the potential of a specific product to cause dental caries when used excessively. When a product has been evaluated by approved methods for in vivo testing and is found not to cause an excessive decrease in pH, the Swiss Health Authority grants permission to identify the product as “zahnfreundlich” (friendly or safe for the teeth) and imprint a distinctive logo on the label (Figure 64-8).[44] This procedure has not been adopted elsewhere because of the inability to find a consensus that this procedure is adequate for the intended purpose.

FLUORIDES AND HUMAN HEALTH

Because fluoride has relationships to dental caries through both the host and the environment, a separate section for fluoride discussion is appropriate. Fluoride ion from any salt of the highly reactive halogen fluorine occurs ubiquitously in water supplies, soils, and foods. Fluoride is also present in all body fluids and tissues, even when the amount of fluoride ingested is low. Probably no nutrient demonstrates more clearly the physiologic spectrum from the influence of an insufficient intake, through normal to toxic levels of ingestion.

TOXICITY OF EXCESS FLUORIDE: MOTTLED ENAMEL

Toxic manifestations of excess fluoride ingestion were observed before recognition of its benefits, unlike other nutrients, for which the effects of deficiencies were described first. In 1901, Eager,[45] a physician in the U.S. Public Health Service stationed in Naples, Italy, described an esthetically disfiguring problem of dental enamel in otherwise healthy adults. Their enamel was variously mottled with brown stains; he attributed the problem to local geologic conditions. In striking contrast, he recognized that the enamel of teeth in young children in the area was normal, which he postulated might be attributable to a recent change known to have occurred in the water supply.

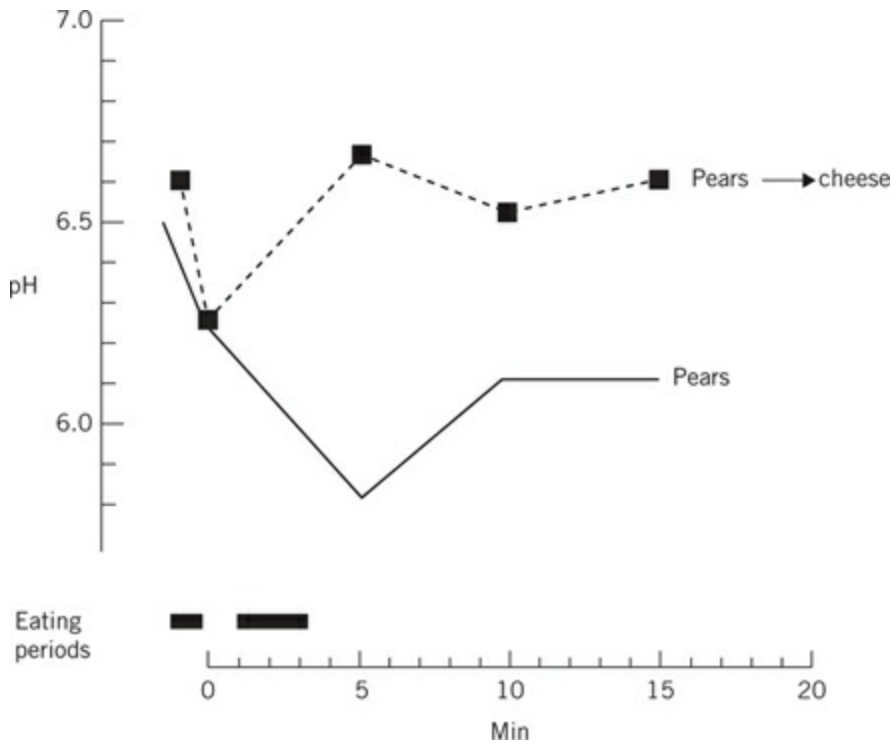


Figure 64-6 An example of the sharp pH decrease after eating a portion of sweetened pears and the moderating influence of eating cheese immediately after the same portion of pears. (Reproduced with permission from Edgar.[40])

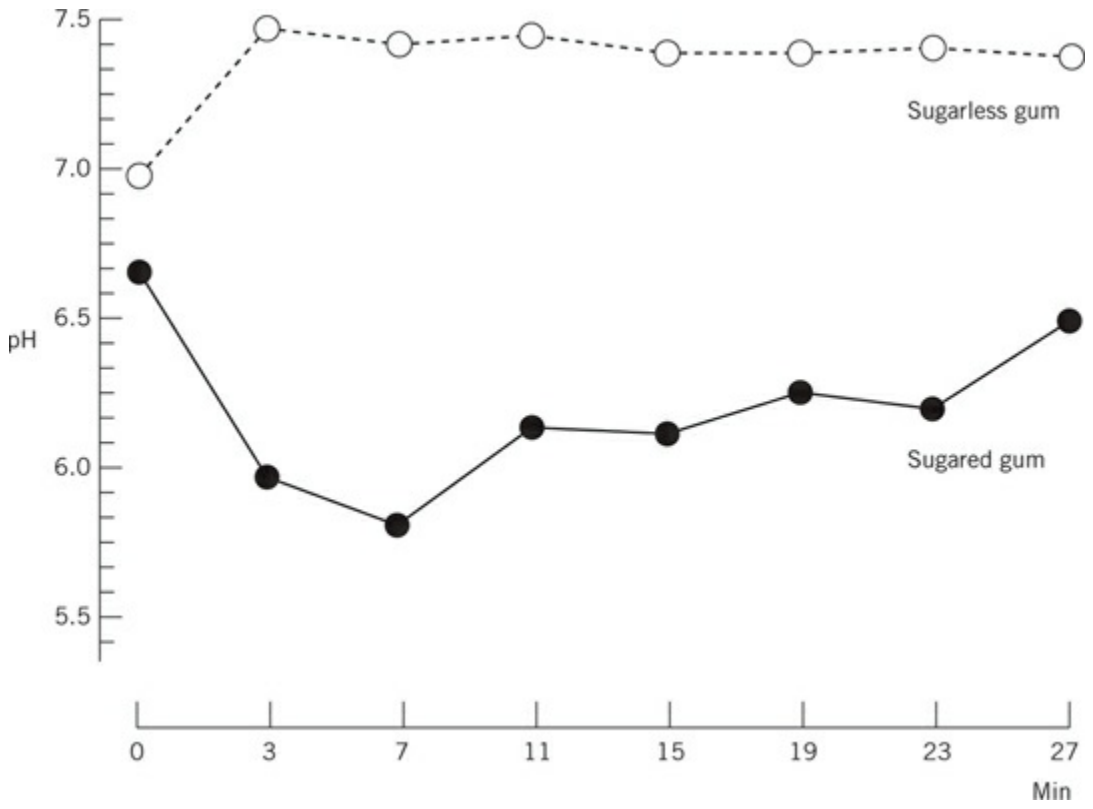


Figure 64-7 A comparison of the Stephen curve after chewing sucrose-sweetened gum with the pH curve after chewing gum sweetened with a noncariogenic polyol. (Reproduced with permission from Edgar.[40])

Subsequently, descriptions of mottled enamel were reported from numerous parts of the world in humans as well as in domestic animals.[44] Evidence became increasingly clear in humans that this esthetically disfiguring problem was caused by some agent in community or home water supplies. In animals, the problem sometimes was caused by their water supplies and in other situations was related to rock phosphate supplements added to their feeds as a source of calcium and phosphorus.

By 1931, reports from several widely separated laboratories established that the mottled enamel in humans and experimental animals was attributable to excess ingestion of fluoride from drinking water that contained more than 1.5 ppm fluoride, that is, 1.5 mg/L of water.[46–48] At this point, the popular name, mottled enamel, was superseded by the scientific descriptor, chronic endemic dental fluorosis.

This abnormality was determined to result from some as yet undefined altered relationship(s) in the deposition of crystals of hydroxyapatite in the

organic matrix formed by ameloblasts, the enamel-forming cell of ectodermal origin. This developmental interrelationship is apparently the most sensitive process to excess fluoride in the extracellular fluids of any in the body. Higher intake (20 mg) of fluoride per day in industrial exposure causes abnormal calcification in the spine and, after 20 years, serious crippling fluorosis.[49]

CLASSIFICATION SYSTEM FOR MOTTLED ENAMEL

Dean developed a classification system to describe the various visual levels of enamel pathology and a weighting system to provide a community fluorosis index.[50] He used the terms: (1) questionable—slight aberrations from the normal glossy translucency of enamel, which the examiner could not be certain was pathologic (weight = 0.5); (2) very mild—small, dull, paper-white areas scattered irregularly over the tooth surface but covering less than 25% of its area (1.0); (3) mild—more extensive white opacities but less than 50% of the area involved (2.0); (4) moderate—all surfaces affected with marked wear on the chewing surface accompanied by brown stains (3.0); and (5) severe—all surfaces affected with marked hypoplasia, discrete or confluent pitting, and widespread brown stains.



Figure 64-8 The “safe for teeth” logo approved for use on the label of an approved product in Switzerland. (Reproduced with permission from Guggenheim.[44])

The community fluorosis index was calculated by combining the weight for each child with the frequency of occurrence of each grade in the child population. A community fluorosis index over 0.5 was considered undesirable, indicating the need to reduce the water fluoride level.

As shown in Figure 64-9, mottled enamel does not become a problem until in excess of 1.5 ppm fluoride in drinking water was ingested during

tooth development.[49] However, water concentrations beyond this amount caused increasingly severe fluorosis and increasing frequency. As a public health measure, excessively high fluoride amounts must be reduced or a new lower fluoride water supply found. It must be emphasized that mottled enamel cannot result from consuming excessively high amounts of fluoride after the teeth are formed because this abnormality is completely of developmental origin.

DENTAL BENEFITS OF NATURALLY AVAILABLE FLUORIDES

One of the earliest observers of enamel fluorosis in this country, McKay,[52] in Colorado Springs, where the water supply was later shown to contain 2.0 ppm fluoride, reported that teeth with mottled enamel did not seem to have an increased susceptibility to dental caries despite the surface irregularities. In the late 1920s and 1930s, several small studies suggested that fluorosis appeared to be associated with reduced dental caries experience.

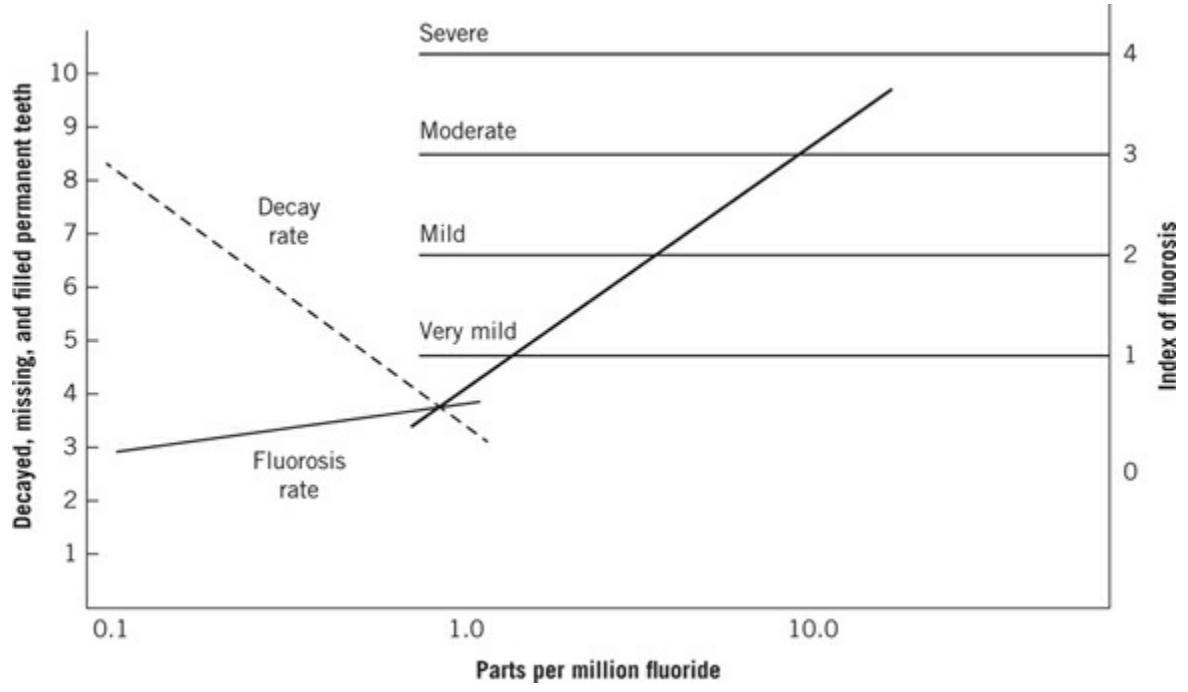


Figure 64-9 Relation between decayed, missing, and filled permanent teeth (broken line), severity of dental fluorosis (solid line), and fluoride concentration in the public water supply, with

the latter on a logarithmic scale. (Reproduced with permission from Hodge and Smith.[51])

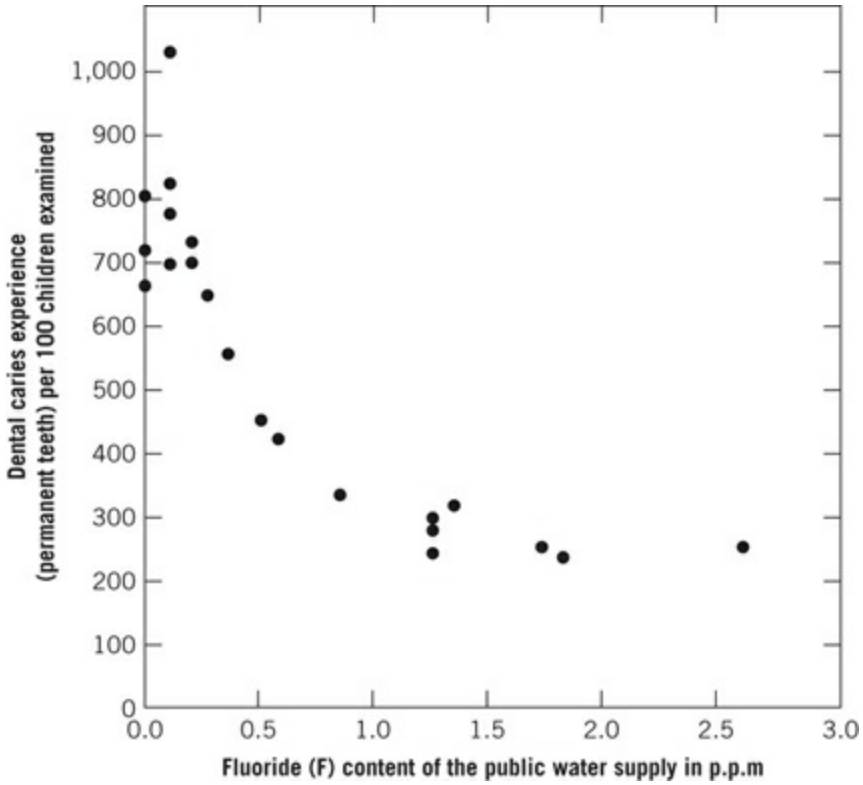


Figure 64-10 Relationship between the fluoride content naturally available in public water supplies and the number of decayed, missing, and filled permanent teeth per 100 children 12–14 years old. (Reproduced with permission from Dean et al.[53,54])

A combination of the need to know the permissible upper limit of fluoride in drinking water plus the increasing concern about the generally high incidence of dental caries among American children led to many extensive epidemiologic surveys among children. Numerous increasingly large surveys in the 1930s and 1940s indicated the strong correlation between fluoride ingestion during tooth development and tooth decay. For example, Dean et al.[53,54] reported on data for 7257 12- to 14-year-old children in 21 cities in 4 states. Figure 64-10 indicates the convincing relationship between fluorides during development and dental caries experience. They also considered other factors that might be related—composition of diet, hours of sunshine, and hardness of the water—and

concluded that none could explain the naturally available fluoride–dental caries relationship.

DENTAL BENEFITS OF WATER FLUORIDATION

By 1945, the dental benefits of fluoride in communal water supplies had been demonstrated to the satisfaction of almost all investigators and many health-related professional organizations. The possibility of pathology elsewhere in the human body or of increased susceptibility to other diseases had been thoroughly studied, with negative results with one exception, the crippling fluorosis described above, which has never been observed at the optimal level for caries reduction. Hence, in 1945, three clinical trials were initiated to adjust the fluoride concentration of community water supplies to an optimal level, with the expectation that 10 years would be needed for demonstration of the benefits. Grand Rapids, Michigan, was selected as one trial community, with Muskegon, Michigan, to serve as the unfluoridated control city and Aurora, Illinois, as a natural fluoride city. A second study was initiated in Newburgh, New York, with nearby Kingston as the unfluoridated control. The third community was Brantford, Ontario, with Sarnia, Ontario, as the unfluoridated control and Stratford, a nearby community with an optimal fluoride level naturally present. The data soon demonstrated that the results from water fluoridated in the distribution system duplicated those observed where fluoride was naturally present, with caries reductions from 50% to 60% in permanent teeth (Figure 64-11).[54–58]

The benefits in teeth developed during the study began to show up sufficiently rapidly that Muskegon dropped out as the control community for Grand Rapids before the 10-year test period ended and initiated its own water fluoridation program. In addition, some unexpected reductions in caries initiation were observed among the teenage population in teeth formed before fluoridation was begun; this observation suggested that an oral environmental influence was occurring in addition to the expected systemic influence on developing teeth. As in communities with naturally available fluoride, the greatest benefit was for the smooth surfaces of the anterior teeth and the least for the carious lesions that develop in the developmental pits and fissures on the grinding surfaces of the posterior teeth. No evidence of esthetically disfiguring mottling was observed in these trials.

Fluoridation of water supplies has been widely adopted in the United States as a valuable and inexpensive public health measure. The

Fluoridation Census in 1992 reported that 134.7 million individuals on communal water supplies in 8572 communities received water fluoridated in the water processing system; in addition, 10.0 million individuals lived in 1924 communities where the water contained at least 0.7 ppm fluoride naturally.[59] The total percentage of the population benefiting from fluoride in the water in the United States was 62.1.

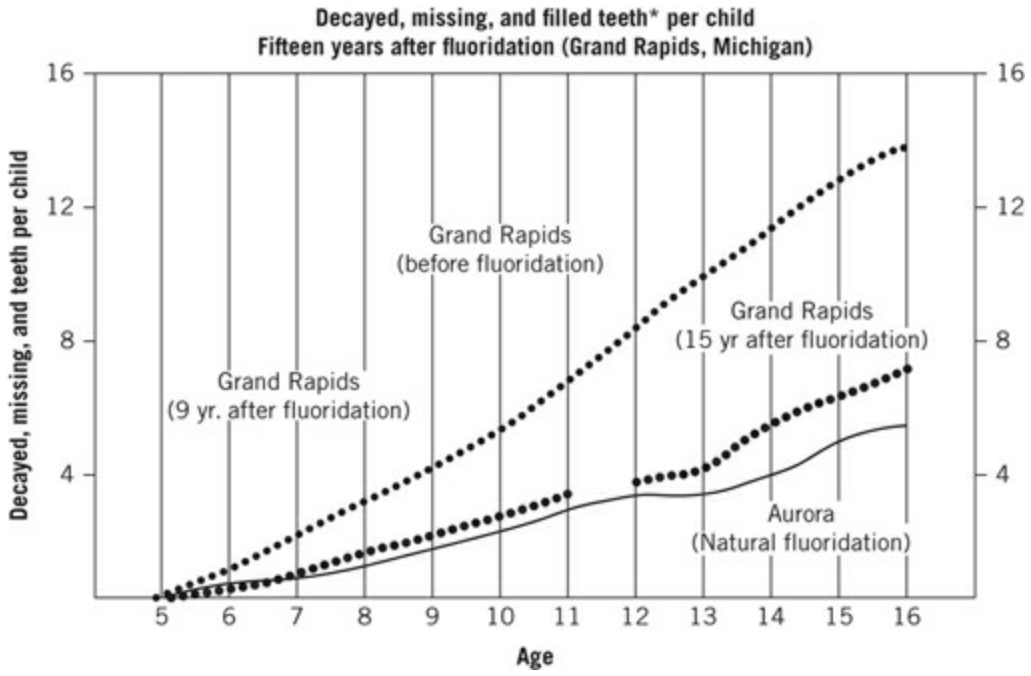


Figure 64-11 The benefits of fluoridation of the communal water supply in Grand Rapids, Michigan, are shown for the deciduous teeth after 9 years and for the permanent teeth after 15 years. The inclusion of the data for Aurora, Illinois, where the water naturally contains an adequate comparison of how similar the benefits are between natural fluoride and fluoride introduced at the water treatment facility. (Reproduced with permission from Arnold et al.,[55,56] Ast et al.,[57] and Hutton et al.[58])

METABOLISM OF FLUORIDES

The varying evidence of mottled enamel within the child population of any community in both natural fluoride areas and areas where fluoridation is practiced has been the subject of much interest since the first evidence of the fluoride relationships. Some variation undoubtedly occurs in the amount of water consumed by individual children; the amounts of canned

fish with bones and the use of tea, the only foods with substantial amounts of fluoride, constitute additional variables.

Fluorides from soluble salts are readily absorbed from the gastrointestinal tract.[60] In the presence of high concentrations of cations that produce much less soluble salts of fluoride, its absorption is reduced. When much less soluble sources of fluoride are ingested, absorption of fluoride is also reduced.

The kidneys are the major route for fluoride removal from the body, which is characterized by variable degrees of tubular resorption. A range in resorption has been reported in humans; in four studies, the average resorption of fluoride after filtration into the tubules varied from 23% to 38%.[61] In general, fluoride clearance tended to increase with increased urinary flow rate. Urinary pH also influenced fluoride excretion. In rats with a urinary pH less than 5.6 induced by administration of ammonium chloride, urinary excretion was less than 5% of the amount filtered; alkaline pH as high as 8.0, induced by administration of sodium bicarbonate, resulted in fluoride excretion approaching 70%. Various factors can chronically alter the acid–base status and affect fluoride relationships. The composition of the diet is normally the major determinant of acid–base balance and urinary pH. In addition, various metabolic and respiratory disorders, level of physical activity, and altitude of residence contribute to acid–base status.

The combination of slightly different levels of fluoride ingestion by children with the above factors, which modify excretion in the urine, is probably adequate to explain the different degrees of mottled enamel in children within the same community.

DENTAL BENEFITS OF FLUORIDE TABLETS, TOOTHPASTES, AND ORAL RINSES

Efforts to extend the systemic benefits of fluoride to populations for whom waterborne fluoride was not available led to the development of fluoride dietary supplements. Three major factors have combined to change the way in which fluoride supplements are used in contemporary practice: (1) a shift in our understanding of fluoride’s mode of action from systemic to topical; (2) an increased exposure among children to other sources of fluoride; and (3) concern for the risk of fluorosis in permanent teeth. With those factors in mind it is recommended that prior to prescribing fluoride supplements, assay the child’s primary drinking water supply for fluoride

content. In addition, practitioners should consider other sources of fluoride exposure for their patients and do a caries risk assessment.[62] The levels recommended by the American Academy of Pediatric Dentistry for different ages and levels of fluoride in the water supply are shown in Table 64-2.[63]

Because young children are prone to swallow toothpaste, use of fluoridated toothpaste (usually containing 1000 ppm fluoride, i.e., 1 mg F per gram) contributes to the level of fluoride ingested. Therefore, it must be used sparingly; the size of a pea on the brush is adequate and must be applied by the child's caregiver. As a safety factor, a maximum of 120 mg of fluoride should be prescribed at one time. This amount would be a certainly lethal dose only for those children weighing less than 8 kg and would be a probably toxic dose for children weighing 24 kg or less.[64]

The overall effectiveness of fluoride supplements has been estimated to provide 20%–30% reduction in dental disease. The level of evidence for the effectiveness of fluoridated dentifrices in the permanent dentition is high and sufficient to warrant a strong recommendation for their use to reduce the incidence of caries. Unfortunately, there is no evidence regarding their effectiveness in the primary dentition, but logically it should follow that they would be as effective in younger patients.

TABLE 64-2 Dietary Fluoride Concentration in the Water Supply

	Fluoride Concentration in the Water Supply		
Age of Child	**Less Than 0.3 ppm F**	**0.3–0.6 ppm F**	**More Than 0.6 ppm F**
Birth–6 months	0	0	0
6 months–3 years	0.25 mg	0	0
3–6 years	0.50 mg	0.25 mg	0
6 years up to atleast 16 years	1.0 mg	0.50 mg	0

Source: Adapted from Whitford.[61]

Fluoridated mouthrinses (0.05 sodium fluoride) and self-applied gels (stannous fluoride) should be reserved for use with children judged to be at moderate or high risk for dental caries, including children with orthodontic appliances or prosthetic appliances, children with special health care needs, and those with reduced salivary flow. Fluoride mouthrinses should

be recommended only for children who have demonstrated mastery of their swallowing reflex.[62]

In children with special health care needs who are especially at risk for dental caries (children with gastroesophageal reflux disease) who cannot control their swallow reflex, it is recommended that the caregiver dip the brush in the mouthrinse before bedtime or use a supplemental gel.

Fluoride varnish recently received Food and Drug Administration approval in 1994 for use as a desensitizing agent and as a cavity liner. It has been used off-label as a caries preventive agent. Fluoride varnish is easily applied to the teeth, and it sets relatively quickly in contact with moisture. A typical application requires 0.2–0.5 mL, resulting in a total fluoride ion application of approximately 5–11 mg. The Cochrane Database for Systematic Reviews found a 46% pooled prevented fraction for permanent teeth and 33% for primary teeth. The authors concluded that fluoride varnish provides a substantial caries-inhibiting effect in both dentitions.[65]

The extent to which the various uses of fluorides contribute to the dental health of the United States is demonstrated by a statement by Harold Varmus, Nobel laureate and director of the National Institutes of Health, at a Senate Appropriations Subcommittee Hearing on March 17, 1994.[66] In response to Senator Harkin's question, "Does medical research contribute to long-term cost savings?" Varmus stated that his favorite example of the benefits of research concerned the use of fluorides to prevent dental caries, which alone saves the taxpayer several billion dollars per year.

MECHANISMS OF ACTION OF FLUORIDES

When the dental benefits of fluoride were first observed, systemic mechanisms of action were sought. The degree of crystallinity of hydroxyapatite in enamel increased as its fluoride concentration increased. Acid solubility of hydroxyapatite decreased as its fluoride concentration increased. Enamel surfaces had particularly high fluoride concentrations.[67]

Saliva contains about 0.019 ppm fluoride (1 μmol/L), about one-fiftieth of the concentration in communal water fluoridated at 1 ppm.[68] Even this low concentration appears to be important in the maintenance of the integrity of the tooth surface and in the remineralization of precarious lesions in the enamel. The mechanisms of action of fluoride in caries inhibition include the following: (1) the enhancement of remineralization during the repeated cycles of demineralization/remineralization in the early

stages of the caries process, (2) the inhibition of glycolysis by which sugar is metabolized by bacteria to produce acid, and (3) while a preeruptive systemic effect cannot be ruled out, no direct evidence of such an effect has been demonstrated.[69]

TREATMENT OF CARIOUS LESIONS

Because a visually detectable carious lesion is rarely self-limiting (as in most bacterial infections), it continues to progress until the pulp is infected, with the probable loss of the tooth, unless specific action is taken to interrupt the progress of the lesion. Decayed enamel and dentin must be removed and replaced with an inert material that is able to resist the forces of mastication and the influences of the oral milieu.

Significant bactericidal benefits can be achieved by the use of fluoride-containing products preparations, such as stannous fluoride gels in young children. Bacteristatic benefits can be seen in high fluoride (5000 ppm) containing toothpastes for children over 6 years of age.

Xylitol is a low-calorie, naturally occurring, and diabetic safe sugar that is not metabolized by cariogenic bacteria. Incorporating xylitol into the diet inhibits pH changes and reduces bacterial attachment to teeth. Xylitol currently is available in many forms (gums, mints, chewable tablets).

Use of xylitol gums by primary caregivers has been shown to decrease oral bacterial load and interrupt vertical transmission. Xylitol products also stimulate salivary flow and aid in the symptomatic treatment of xerostomia sometimes associated with chronic childhood disease.

Although there are no definitive clinical studies to support the dosage in tooth wipes containing xylitol, they may be used to compliment, but not completely replace, regular mechanical removal of plaque through brushing. It is important to note that many sugarless gums containing xylitol also contain phenylalanine and are contraindicated for phenylketonurics.[75]

Pit and fissure sealants have been demonstrated to be effective in the primary prevention of caries, and their effectiveness remains strong as long as the sealants are maintained.[6] Sealants are intended to protect caries-susceptible tooth surfaces, which are benefited least by fluoride. They should be placed as soon as possible after the tooth erupts and isolation to prevent moisture contamination during placement can be achieved.

Identification of early carious lesions and treatment with preventive

resin restorations allow for decreased dental bioload while preserving tooth structure. This very conservative treatment allows for the removal of just the small pit and fissure lesion and subsequent restoration with esthetic composite resin to which a sealant can be applied. By removing only the carious enamel and dentin, the preventive resin technique provides a restoration with a minimum of tooth reduction while ensuring the prevention of future caries in other pits and fissures through sealant placement.[70]

Amalgam is the least expensive restorative material to purchase and insert. The new high copper amalgams are expected to last over 30 years. Questions have been raised in a few publications about the safety of amalgams because of their gradual release of mercury vapor. The media picked up this concern and exaggerated the problem with regard to both its severity and its frequency. Berglund[71] estimated that 1.7 μg of mercury vapor was inhaled from 12 amalgam fillings in a 24-hour period. He noted that the World Health Organization threshold limit value for mercury vapor in the workplace is 100 times greater than this amount.

Osborne[72] stated that allergies to amalgam has been reported in the literature infrequently but that it is speculative as to which component(s) of amalgam (mercury, silver, tin, or copper) is causative. Owing to their frequent handling of amalgam, dentists and their staffs are exposed to more mercury than any patient. Even though the profession urges great care in amalgam handling and ventilation of dental offices, guidelines that are followed by most dentists, dentists still accumulate a greater body burden of mercury than do nondentists. However, dentists have 21/2 years longer life expectancies than the population in general, including physicians. Dentists and their staffs do not have elevated levels of spontaneous abortions, and their children do not have a higher incidence of birth defects.[73]

SUMMARY AND RECOMMENDATIONS

Typical of all areas of the body, the mouth and each of its components require a nutritionally well-balanced diet containing adequate amounts of all essential nutrients throughout development and maintenance.

Attention must be drawn to one special requirement for the adequate development and maintenance of the teeth throughout their entire life histories, namely, a source of fluoride. The easiest and least expensive way to provide fluoride is through a communal water supply containing 1.0 ppm fluoride that is present naturally or is added in the local water

treatment plant. If, for any reason, water fluoridation is not possible, a proprietary source of fluoride in tablets or drops should be used. However, it is imperative that the diet should not be supplemented if the water already contains 0.6 ppm or more fluoride; an undesirable level of mottling is likely to occur with fluoride from two sources.

Söderling demonstrated that habitual use of xylitol chewing gum by mothers was associated with a statistically significant reduction in the probability of vertical transmission of mutans streptococci at age 2 years. Saliva sharing activities, such as tasting food before feeding and shared toothbrushes also contributes to the horizontal and vertical transmission. Clearly, efforts made to reduce the risk for early vertical and horizontal transmission of mutans streptococci translate into a decreased risk for dental caries in the child.[74]

Consumption of sticky snacks containing sugars should be kept to a minimum and be replaced whenever possible by such noncariogenic snacks as fresh fruits and vegetables, cheese, or nuts. Candy and gum sweetened with a xylitol are good substitutes for products sweetened with sugar.

Good oral hygiene habits need to be followed at least once a day using a small amount of fluoridated toothpaste. Use of a fluoride oral rinse or varnish is appropriate, especially when the child has a high degree of susceptibility and when there is a problem of reduced manual dexterity.

Visits to a dentist regularly are essential to detect and treat early lesions. Usually, every 6 months is sufficient, but when the child is especially susceptible, more frequent visits are important, as suggested by the dentist. Preventive procedures, such as topical fluoride applications to the tooth surfaces and the use of sealants to flow into and fill the susceptible pits and fissures on the occlusal surfaces of the posterior teeth, are to be encouraged.

Children who have special health care needs or have had radiation of the head or neck or chemotherapy may have a reduced salivary flow, which is probably the most serious deleterious influence on caries initiation and progression. Special attention to frequent and careful oral hygiene and frequent use of oral fluoride rinses are imperative.

REFERENCES

1. Baum BJ. Principles of saliva secretion. In: Malamud D, Tabak L, editors. *Saliva as a Diagnostic Fluid*. *Ann N Y Acad Sci*. 1993;694:17–23.
2. Turner RJ. Mechanisms of fluid secretion by salivary glands. In: Malamud D, Tabak L, editors. *Saliva as a Diagnostic Fluid*. *Ann N Y Acad Sci*. 1993;694:24–

35.
3. Berkowitz RJ, Jordan HV. Similarity of bacteriocins of Streptococcus mutans from mother and infant. *Arch Oral Biol*. 1975;20:725–730.
4. Berkowitz RJ. Mutans streptococci: acquisition and transmission. *Pediatr Dent*. 2006;28:106–109.
5. Berkowitz RJ, Turner J, Green P. Maternal salivary levels of Streptococcus mutans and primary oral infection in infants. *Arch Oral Biol.* 1981;26:147–149.
6. Kohler B, Bratthall D. Intrafamilial levels of Streptococcus mutans and some aspects of the bacterial transmission. *Scand J Dent Res*. 1978;86:35–42.
7. National Institutes of Health. Diagnosis and management of dental caries throughout life. NIH Consensus Statement March 26–28, 2001;18(1):1–30.
8. Scott DB, Ussing MJ, Sognnaes RF, et al. Electron microscopy of mature dental enamel. *J Dent Res*. 1952;31:74–84.
9. Sognnaes RF, Shaw JH. Salivary and pulpal contributions to the radiophosphorus uptake in enamel and dentin. *J Am Dent Assoc*. 1952;44:489–505.
10. Silverstone LM, Hicks MJ, Featherstone MJ. Dynamic factors affecting lesion initiation and progression in human dental enamel. Part I. The dynamic nature of dental caries. *Quintessence Int*. 1988;19:683–711.
11. Silverstone LM, Hicks MJ, Featherstone MJ. Dynamic factors affecting lesion initiation and progression in human dental enamel. Part II. Surface morphology of sound enamel and caries-like lesions of enamel. *Quintessence Int*. 1988;19:773–785.
12. Cutright DE, Bauer H. Cell renewal in the oral mucosa and skin of the rat. I. Turnover time. *Oral Surg*. 1967;23:249–259.
13. Jolliffe N. The clinical signs. In: Jolliffe N, editor. *Clinical Nutrition*, 2nd ed. New York: Hoeber Medical Books; 1962. pp. 28–87.
14. Wolbach SB, Howe PR. Tissue changes following deprivation of fat-soluble A vitamin. *J Exp Med*. 1925;42:753–757.
15. Wolbach SB, Howe PR. Effect of scorbutic state upon the production of intercellular substances. *Proc Soc Exp Biol Med*. 1925;22:400–402.
16. Mellanby M. An experimental study of the influence of diet on teeth formation. *Lancet*. 1918;ii:767–770.
17. Weinman JP, Schour I. Experimental studies in calcification. I. The effect of a rachitogenic diet on the dental tissues of the white rat. *Am J Pathol*. 1945;21:821–831.
18. Sweeney EA, Guzman M. Oral conditions in children from three highland villages in Guatemala. *Arch Oral Biol*. 1966;11:687–698.
19. Sweeney EA, Cabrera J, Urrutia J, et al. Factors associated with linear hypoplasia of human deciduous incisors. *J Dent Res*. 1969;48:1275–1279.
20. Shaw JH, Griffiths D. Dental abnormalities in rats attributable to protein deficiency during reproduction. *J Nutr*. 1963;80:123–141.
21. Menaker L, Navia JM. Effect of undernutrition during the perinatal period on caries development in the rat. III. Effects of undernutrition on biochemical parameters in the developing submandibular salivary gland. *J Dent Res*. 1973;52:688–691.
22. Alvarez JO. Nutrition, tooth development, and dental caries. *Am J Clin Nutr*. 1995;61:410–416.
23. Hayes C. Nutrition and growth and development of oral tissues and structures. In: Palmer C, editor. *Diet and Nutrition in Oral Health*. Upper Saddle River, NJ: Prentice Hall; 2002.
24. Papas AS, Joshi A, MacDonald SL, et al. Caries prevalence in xerostomic

individuals. *J Can Dent Assoc*. 1993;59:171–179.
25. Sreebny LM. Salivary flow and dental caries. In: Guggenheim B, editor. *Cariology Today*. Basel, Switzerland: S. Karger; 1984. pp. 56–69.
26. Keyes PH, Jordan HV. Factors influencing the initiation, transmission, and inhibition of dental caries. In: Sognnaes RF, editor. *Mechanisms of Hard Tissue Destruction*. Washington, DC: American Association for the Advancement of Science; 1963. pp. 261–83.
27. Gibbons RJ, van Houte J. Bacteriology of dental caries. In: Shaw JH, et al., editors. *Textbook of Oral Biology*. Philadelphia, PA: WB Saunders; 1978. pp. 975–91.
28. Cummins D. The impact of research and development on the prevention of oral diseases in children and adolescents: an industy perspective. *Pediatr Dent*. 2006;28:118–127.
29. Glass RL, editor. The first international conference on the declining prevalence of dental caries. *J Dent Res*. 1982;61:1301–1383.
30. Kent RL Jr, Moorrees CFA. Associations in interproximal caries prevalence from a longitudinal twin study [abstract]. *J Dent Res*. 1979;58:225.
31. Conroy JP, Messer LB, Boraas JC, et al. Dental caries and treatment characteristics in human twins reared apart. *Arch Oral Biol*. 1993;38:937–943.
32. Mandel ID. Nature versus nurture in dental caries. *J Am Dent Assoc*. 1994;125:1345–1351.
33. Weis RL, Trithart AH. Between-meal eating habits and dental caries experience in preschool children. *Am J Public Health*. 1960;50:1097–1104.
34. Sreebny LM. Sugar availability, sugar consumption and dental caries. *Community Dent Oral Epidemiol*. 1982;10:1–7.
35. Horowitz HS. Ethical considerations in human experimentation. *J Dent Res*. 1977;56:154–159.
36. Hefferren JJ, editor. Scientific consensus conference on methods for the assessment of the cariogenic potential of foods. *J Dent Res*. 1986;65:1473–1543.
37. Dilley GJ, Dilley DH, Machen JB. Prolonged nursing habit: a profile of patients and their families. *ASDC J Dent Child*. 1980;47:102–108.
38. Weinstein P, Domoto P, Koday M, et al. Results of a promising open trial to prevent baby bottle tooth decay: a fluoride varnish study. *J Dent Child*. 1994;61:338–341.
39. Stephan RM. Intra-oral hydrogen-ion concentrations associated with dental caries activity. *J Dent Res*. 1944;23:257–266.
40. Edgar WM. Plaque pH assessments related to food cariogenicity. In: Hefferren JJ, Koehler HM, editors. *Foods, Nutrition and Dental Health*. Volume 1. Park Forest South, IL: Pathotox; 1981. pp. 137–147.
41. Scheinin A, Makinen KK, Tammisalo E, et al. Turku sugar studies. XVIII. Incidence of dental caries in relation to 1-year consumption of xylitol chewing gum. *Acta Odontol Scand*. 1975;33:269–278.
42. Scheinin A, Makinen KK, Ylitalo K. Turku sugar studies. V. Final report on the effect of sucrose, fructose and xylitol diets on the caries incidence in man. *Acta Odontol Scand*. 1975;33:67–104.
43. Hayes C. The effect of non-cariogenic sweeteners on the prevention of dental caries: a review of the evidence. *J Dent Educ*. 2001;65:1112–1115.
44. Guggenheim B. Preface. In: Guggenheim B, editor. *Cariology Today*. Basel, Switzerland: Karger; 1984. p. x-ii.
45. Eager JM. Denti di Chiaie (Chiaie teeth). *US Public Health Rep*. 1901;16:2576–2577.

46. Smith MC, Lantz EM, Smith HV. *The Cause of Mottled Enamel, a Defect of Human Teeth*. Tempe, AZ: University of Arizona Agricultural Experimentation Station; 1931. Bulletin No. 32.
47. Churchill HV. Occurrence of fluorides in some waters of the United States. *Ind Eng Chem*. 1931;23:996.
48. Velu H. Le darmous (ou dermes). Fluorose spontanee des zones phosphate. *Arch Inst Pasteur Alger*. 1932;1:41–118.
49. Roholm K. *Fluorine Intoxication. A Clinical-Hygienic Study with a Review of the Literature and Some Experimental Investigations*. London: HK Lewis and Co; 1937.
50. Dean HT. Chronic endemic dental fluorosis (mottled enamel). In: Gordon SM, editor. *Dental Science and Dental Art*. Philadelphia, PA: Lea & Febiger; 1938. pp. 387–414.
51. Hodge HC, Smith FA. Some public health aspects of water fluoridation. In: Shaw JH, editor. *Fluoridation as a Public Health Measure*. Washington, DC: American Association for the Advancement of Science; 1954. p. 80.
52. McKay FS (in collaboration with Black GV). Investigations of mottled teeth: an endemic developmental imperfection of the enamel of the teeth, heretofore unknown in the literature of dentistry. *Dent Cosmos*. 1916;58:627–644, 781–92, 894–904.
53. Dean HT, Jay P, Arnold FA Jr, et al. Domestic water and dental caries. II. A study of 2,832 children aged 12–14 years of eight suburban Chicago communities, including L. acidophilus studies of 1,761 children. *US Public Health Rep.* 1941;56:761–769.
54. Dean HT, Arnold FA Jr, Elvove E. Domestic water and dental caries. V. Additional studies of relation of fluoride domestic waters to dental caries experience in 4,425 children, aged 12 to 14 years, of 13 cities in 4 states. *US Public Health Rep*. 1942;57:1155–1179.
55. Arnold FA Jr, Dean HT, Jay P, et al. Effect of water fluoridation of public water supplies on dental caries prevalence, tenth year of the Grand Rapids-Muskegon study. *US Public Health Rep*. 1956;71:652–658.
56. Arnold FA Jr, Likins RC, Russell AL, et al. Fifteenth year of the Grand Rapids fluoridation study. *J Am Dent Assoc*. 1962;65:780–785.
57. Ast DB, Smith DJ, Wachs B, et al. Newburgh-Kingston caries-fluorine study: Combined clinical and roentgenographic dental findings after ten years of fluoride experience. *J Am Dent Assoc*. 1956;52:314–325.
58. Hutton WL, Linscott BW, Williams B. Final report of local studies on water fluoridation in Brantford. *Can J Public Health*. 1956;47:89–92.
59. US Department of Health and Human Services. *Fluoridation Census, 1992*. Atlanta, GA: US Department of Health and Human Services, Division of Oral Health, National Centers for Disease and Prevention; September 1993.
60. Whitford GM. Overview of fluoride metabolism. In: Whitford GM, editor. *The Metabolism and Toxicity of Fluoride*. Basel, Switzerland: Karger; 1989. pp. 1–30.
61. Whitford GM. Renal handling of fluoride. In: Whitford GM, editor. *The Metabolism and Toxicity of Fluoride*. Basel, Switzerland: Karger; 1989. pp. 51–66.
62. Adair S. Evidence-based use of fluoridein comtemporary pediatric dental practice. *Pediatr Dent*. 2006;28:133–141.
63. Clark DC. Appropriate use of fluorides in the 1990s. *J Can Dent Assoc*. 1993;59:272–279.
64. Whitford GM. Fluoride in dental products: Safety considerations. *J Dent Res*.

1987;66:1056–1060.
65. Marinho VCC, Higgins JPT, Logan S, Sheiham A. Fluoride varnishes for preventing dental caries in children and adolescents. *Cochrane Database Syst Rev*. 2002;(3):CD002279.
66. Varmus H. 1995 Senate Appropriations Subcommittee Hearing, March 17, 1994.
67. Dawes C. Fluorides: mechanisms of action and recommendations for use. *J Can Dent Assoc*. 1989;55:721–723.
68. Dawes C, Weatherell JA. Kinetics of fluoride in the oral fluids. *J Dent Res*. 1990;69:638–644.
69. Fejerskov O. Changing paradigms in concepts on dental caries: consequences for oral health care. *Carfies Res.* 2004;38:182–191.
70. Meiers JC, Jensen ME. Management of the questionable carious fissure: invasive vs. noninvasive techniques. *J Am Dent Assoc*. 1984;108:64.
71. Berglund A. Estimation by a 24-hour long study of the daily dose of intra-oral mercury vapor inhaled after release from dental amalgam. *J Dent Res*. 1990;69:1646–1651.
72. Osborne JW. Dental amalgams and mercury vapor release. In: Effects and Side Effects of Dental Restorative Materials. An NIH Technology Assessment Conference, National Institute of Dental Research, Bethesda, MD, August 26–28, 1991. *Adv Dent Res*. 1992;6:135–138.
73. Jones DW. The enigma of amalgam in dentistry. *J Can Dent Assoc*. 1993;59:155–166.
74. Söderling E, Isokangas P, Pienihakkinen K, et al. Influence of maternal xylitol consumption on acquisition of mutans streptococci by infants. *J Dent Res*. 2000;79:882–887.
75. Mouri A, Fernandez J, Daronch M, et al. Nutrition and oral health considerations in children with special health care needs: implications for oral health care providers. *Pediatr Dent*. 2010;32:333–342.
76. Gudiño S, Rojas N, Castro C, et al. Colonization of mutans Streptococci in Costa Rican children from a high-risk population. *J Dent Child*. 2007;74:36–40.

Adolescent Eating Disorders

65

Kendrin R. Sonneville, RD, ScD and Alison E. Field, ScD

DEFINITIONS

There is confusion about the prevalence of eating disorders (EDs), with the general public believing they are common and many clinicians believing they are relatively rare. One reason for this discrepancy in perception is that the Diagnostic and Statistical Manual for Mental Disorder (DSM) versions IIIR and IV only recognized two EDs, anorexia nervosa and bulimia nervosa, which combined affect <5% of women, but many more people meet some, but not all, criteria of those disorders. In both treatment-seeking and general population samples, the majority of eating-disordered individuals do not meet the DSM cut-offs and are therefore categorized as having an eating disorder not otherwise specified (EDNOS). This is a large heterogeneous group in terms of their symptoms and treatment needs.[1–4] In the DSM-5, this group will be slightly smaller and less heterogeneous and will be renamed to other specified feeding and eating disorders (OSFEDs).

Anorexia nervosa (AN) is considered the most serious ED. It is characterized by having a very low weight, a distorted body perception, an intense fear of gaining weight despite being underweight, amenorrhea (females only), and usually a refusal to recognize the seriousness of their low weight. In adults, the low weight is the result of a large weight loss, but in children and adolescents who are expected to be growing, the low weight may present as failure to make adequate gains in weight. In the DSM-IIIR (1987–1993) and DSM IV (1994–2013), it was suggested that a

weight ≤85% of expected weight for age and gender would meet the weight criterion. Although 85% of expected weight was given as an example in the DSM-IV, it is widely used as an accepted cut-off. There have been many criticisms of the diagnostic criteria for anorexia nervosa. First, the weight cut-off is arbitrary. A nontrivial number of treatment-seeking cases do not meet the stringent weight criterion and therefore considered to have partial or atypical AN, a subtype of EDNOS. Moreover, since people lose weight, or for children fail to make adequate gains in weight, because of the eating-disordered cognitions and behaviors, individuals with shorter duration of illness may not (yet) have achieved the requisite weight criteria to meet the diagnostic criteria. To remedy this problem, the DSM-5 will have a less stringent weight criterion. It will require that an individual, relative to age-, gender-, and activity-level-specific needs, has limited his or her calorie intake sufficiently to result in a significantly low body weight. This change in diagnostic criteria will have several consequences. First, it will reduce the number of OSFED cases because some will now be classified as having AN. Second, it will capture a wider spectrum of anorexia cases for whom treatment may differ by severity of illness and duration of insufficient intake.

Another limitation of the diagnostic criteria for AN and other EDs is that they are not developmentally appropriate for children. Children and some adolescents have limited verbal skills and reasoning ability. Until children transition from concrete to abstract reasoning, concepts, such as denial of the seriousness of low body weight, are beyond their cognitive level. Therefore, some ED cases may fail to meet full diagnostic criteria because they do not appear to have the adult cognitive features of the disorder or they have difficulties understanding the criteria and reporting their feelings.[5] In addition, since the prefrontal cortex, the region of the brain involved in complex decision making, such as future consequences of a behavior and planning for achieving goals, does not finish maturing until young adulthood, it has been suggested that the cognitive criterion related to evaluation being heavily influenced by weight and shape and those related to denying the consequences of their behaviors and weight should not be require for a diagnosis in a child.[6]

Bulimia nervosa (BN) is the other recognized ED in DSM-IV. It is characterized by frequently engaging in overeating episodes during which the person feels out of control and they cannot stop eating. In addition, the person engages frequently in inappropriate compensatory behaviors to counteract the large caloric intake during the binges. The compensatory behaviors include self-induced vomiting, use of laxatives, diuretics,

enemas, and fasting. DSM-IV required that individuals engaged in both binge eating and the compensatory behaviors at least two times per week. In DSM-5, the frequency criteria will be reduced to at least once per week. Although AN and BN are the recognized disorders, more adolescents and young adults engage in only binge eating or purging. Those who only binge eat, but do not engage in compensatory behaviors, are considered to have binge eating disorder (BED). In DSM-IV, this has been a subgroup of EDNOS; in DSM-5, BED will be a third recognized ED. In contrast, individuals with purging disorder (PD) who purge but do not binge eat, will remain part of OSFED.

In recognition that AN and BN do not capture some of food avoidant behavior in children Avoidant Restrictive Food Intake Disorder (ARFID) will be another officially recognized condition in DSM-5. ARFID is more common among children, but DSM-5 will not restrict the diagnosis to children. People with ARFID will refuse to eat many foods due to characteristics of the food.

INCIDENCE AND PREVALENCE

The peak age of onset of AN and BN is during adolescence.[7,8] Some have reported a bimodal age of onset during adolescence,[7] others have reported only one mean or median age of onset. However, the exact distribution of age of onset has been difficult to study since it is a rare disorder and this requires a very large sample size to ascertain. In the National Survey Replication Survey sample of 9282 adults, median age of recalled onset of AN was 18 years, with an interquartile range of 16–22 years[9]; whereas, among the 21,425 adults in the European Study of the Epidemiology of Mental Disorders (ESEMeD) project, all cases of anorexia developed before 20 years of age.[7]

Among adolescent and young adult women in the United States, approximately 0.1%–1% have AN and 1%–3% have BN.[9,10] The prevalence of BED is higher; in population-based samples, the prevalence has ranged from 2.5% to 4.1% among females[11–14] and slightly lower among males.[9,15] Purging disorder has been much less studied, but among the 9000 girls in the Growing Up Today Study, 4.1% of the girls developed PD.[11] A slightly lower rate (3.4%) was observed in a smaller sample followed for 8 years.[13] The rates of EDs are thought to be much lower in males. Swanson et al. observed, among 10,123 adolescents, that BN and BED were three times more common among females than males,

but the prevalence of partial criteria BED was similar among females and males.[15] Among the 5527 adolescent and young adult males in the Growing Up Today Study, only 0.8% had partial or full criteria BN or PD, many more (2.9%) had partial or full criteria BED. Since the adolescence and young adulthood are the periods of highest incidence for BED, rates are lower in younger samples. Among adults, BED is the most common ED, and the lifetime prevalence of partial or full BED is similar for males (3.9%) and females (4.1%).[9]

CLINICAL ASSESSMENT

Although many ED screening questionnaires are available, the SCOFF questionnaire (Table 65-1) is a popular, brief screening tool among health professionals, which demonstrates good validity compared to clinical interview.[16] Once an D has been diagnosed, a careful history and physical examination can reveal much information about the chronicity of an adolescent's illness, abnormal eating behaviors, and potential strategies for management. A clinician can start the history-taking session by asking the parent and the adolescent individually why he or she is being seen. This information can be most helpful in revealing hidden agendas or different perceptions of parents and their teenagers. Confidentiality should be established immediately, with the explicitly stated caveat that the provider will break confidentiality if the situation is life-threatening or extremely dangerous. In addition to the physical exam conducted by a medical provider, a teen should be assessed by a mental health provider and should meet with a registered dietitian for a thorough nutrition assessment. The nutrition assessment will have different components depending on the stage of illness and the treatment setting.[17] A full nutrition assessment reveals current dietary intake, present eating patterns, history related to foods, nutrient deficiencies, supplement use, risk of refeeding syndrome, beliefs about food, binge purge patterns, and physical activity patterns.[17]

TABLE 65-1 The "SCOFF" Questions[16]

S	Do you make yourself SICK (vomit) because you feel uncomfortably full?
C	Do you worry that you have lost CONTROL over how much you eat?
O	Have you recently lost more than ONE stone (15 pounds) in a three-month period?
F	Do you believe yourself to be FAT when others say you are thin?
F	Would you say that FOOD dominates your life?

Letting patients tell their story provides an in-depth and personal picture of the ED. Ideally, the parent and the adolescent each should be given time alone with the provider to promote a sense of individualized caring and listening to each one's needs. In general, the younger a child, the more the registered dietitian (RD) will rely on parental report.[17] Parents will provide information about their child such as eating history, typical food intake, weight history, temperament, and behavior changes.[17] In children and adolescents, it is important to find out about family meals, who buys and prepares food and who will be providing care to the young person with an ED.[17] Weight history and lifetime growth charts can provide information for estimation of goal weight, while inquiring about the frequency of weight monitoring, desired weight, and body checking can help assess fear of weight gain and body image concerns.[17] Information about typical meal patterns and food rules and rituals such as counting calories, focusing on numbers, rigid food patterns, dietary extremes, or missing food groups should be collected during the nutrition assessment.[17] A review of purging behaviors should be conducted and should involve asking about amount, frequency, and type, including self-induced vomiting, laxative use, use of enemas, suppositories, diuretics, and diet pills.[17] Physical activity routines including type of activity, sessions per week, intensity, and duration of exercise should be assessed in addition to information about compulsive exercise.[17] Macro and micronutrient deficiencies are evaluated by dietary intake, diet history, physical signs and symptoms, and laboratory values.[17] Initial laboratory assessment typically includes complete blood count, comprehensive serum metabolic profile, electrolytes, liver function, and thyroid function texts.[17] Reduced sodium levels may result from water loading, water intoxication, and excessive fluid loss due to vomiting, diarrhea, sweating, while hypernatremia may be seen with dehydration and insufficient water intake.[17] The most common cause of low potassium is gastrointestinal loss due to purging behaviors, while dehydration from water withholding may elevate potassium.[17] Low bicarbonate levels are seen with laxative use, while elevated bicarbonate level is seen with vomiting.[17] Although most laboratory results will be normal in patients with EDs, normal laboratory results do not exclude serious illness or medial instability.

MEDICAL FINDINGS AND COMPLICATIONS

No organ system is spared the effects of EDs, weight-control behavior, and

malnutrition.[18] As such, the medical complications (Table 65-2) can be serious or even life-threatening, thereby warranting a comprehensive history and a comprehensive physical examination.[18] Physical exam findings in some adolescents with EDs are shown in Table 65-3. Physiological signs of EDs include marked weight changes, absence of expected weight gain, or growth delay in children or adolescents, low bone mineral density, and weakness, fatigue, or lethargy.[17] Cardiovascular signs and symptoms include low pulse; low blood pressure; orthostasis with blood pressure and/or pulse changes; and poor peripheral perfusion characterized by cold extremities, delayed capillary refill, and acrocyanosis.[17,19] Gastrointestinal complaints include abdominal pain, postprandial fullness, and bloating related to delayed gastric emptying, constipation, and gastroesophageal reflux.[17,19] Endocrine dysfunction may include hypothyroidism, hypercortisolism, and disturbances of the hypothalamic–pituitary–adrenal axis, which may result in hypogonadism, loss of menstrual cycle, or delayed menarche.[17,19] Hair and nail changes are seen in EDs and include dry skin, brittle nails, hair loss, and lanugo (white downy hair).[17,19] Neuropsychiatric symptoms include poor concentration, memory loss, insomnia, depression, anxiety, and over concern with weigh/shape.[17]

Medical stabilization and nutritional rehabilitation are the most important determinants of short-term outcomes and are essential for correcting cognitive deficits to allow for effective mental health interventions.[19] Most of the medical complications of EDs resolve with refeeding and/or resolution of purging; however, there is increasing concern that some complications, particularly growth retardation, structural brain changes, and low bone density, may become irreversible over time.[17]

REFEEDING SYNDOME

Refeeding syndrome is a potentially life-threatening cluster of metabolic events that occur when feeding is initiated in the starved catabolic individual. Patients at risk for refeeding syndrome include those with AN, any child with prolonged under-nutrition over weeks and months, and individuals with a rapid and significant weight loss prior to hospital admission regardless of current weight. The starved catabolic individual has low body stores of phosphate, thiamin, and potassium, which are needed to synthesize metabolic intermediates, particularly of carbohydrate

metabolism. In uncomplicated starvation, however, laboratory values may be normal and then change once feeding begins.[17] The start of aggressive refeeding shifts serum electrolytes into the intracellular compartments, which drops serum levels. Biochemical findings in refeeding syndrome include hypophosphatemia, hypomagnesemia, hypokalemia, glucose intolerance, fluid overload, and thiamin deficiency. To avoid complications of refeeding syndrome, feed very conservatively at the beginning of refeeding.[17] Initiating replacement therapy for electrolytes and fluids prior to refeeding and monitoring electrolytes (K, P, Ca, Mg) frequently at the beginning of refeeding is also recommended.[17]

TABLE 65-2 Medical Complications and Consequences of EDs

General	Dehydration Hypokalemia Hypomagnesemia Hyponatremia Irreversible cardiomyopathy and myositis (ipecac toxicity) Amenorrhea and menstrual irregularities Low bone mineral density; osteoporosis Cognitive deficits Mood symptoms Obsessive/compulsive symptoms Suicide
Caloric restriction and weight loss-related	Inability to maintain body temperature Prolonged corrected QT interval or increased QT dispersion (uncommon but may predispose patient to sudden death) Dysrhythmias (including supraventricular beats and ventricular tachycardia, with or without exercise) Other electrocardiographic abnormalities Mitral valve prolapse Pericardial effusions Delayed gastric emptying and impaired gastrointestinal tract motility Constipation Bloating; postprandial fullness Hypoglycemia Hypercholesterolemia Abnormal liver function test results Sterile pyuria Anemia, leukopenia; thrombocytopenia Sick-euthyroid syndrome Growth retardation Cortical atrophy
Vomiting-related	Hypochloremic metabolic alkalosis (vomiting)

	Esophagitis Gastroesophageal reflux Dental erosions Mallory–Weiss tears Esophageal or gastric rupture (rare) Aspiration pneumonia (rare)
Laxative-related	Hyperchloremic metabolic acidosis (laxative abuse) Hyperuricemia Hypocalcemia Fluid retention (may gain up to 10 lb in 24 h) with laxative withdrawal
Binge eating-related	Weight-related hypertension Abnormal lipid profile Diabetes
Refeeding-related	Diaphoresis and night sweats Polyuria and nocturia Peripheral edema Refeeding syndrome

Source: Adapted from references 18 and 20.

TABLE 65-3 Physical Exam Findings Seen in Some Patients with EDs

Sinus bradycardia; other cardiac arrhythmias
Orthostatic changes in pulse (>20 beats per min) or blood pressure (>10 mm Hg)
Hypothermia
Cachexia; facial wasting
Cardiac murmur (one-third with mitral valve prolapse)
Dull, thinning scalp hair
Sialoadenitis (parotitis most frequently reported)
Angular stomatitis; palatal scratches; oral ulcerations; dental enamel erosions
Dry, sallow skin; lanugo
Bruising/abrasions over the spine related to excessive exercise
Delayed or interrupted pubertal development
Atrophic breasts; atrophic vaginitis (postpubertal)
Russell sign (callous on knuckles from self-induced emesis)
Cold extremities; acrocyanosis; poor perfusion
Carotenemia (orange discoloration of the skin, particularly palms and soles)
Edema of the extremities
Flat or anxious affect
Parotid gland swelling

Source: Adapted from references 18 and 20.

Treatment

The goals of ED treatment are numerous (Table 65-4). As such, ED treatment requires a collaborative approach by a multidisciplinary team of mental health, nutrition, and medical specialists.[17] Best treatment practices

in general for AN and BN include early detection, early and aggressive intervention, and early return to physical stability. Normalization of attitudes and eating behaviors usually follows physical restoration, but this aspect of recovery can take years and clinicians vary in terms of what they define as full recovery from EDs.[17]

Various levels of care for EDs are available including outpatient, intensive outpatient, partial hospitalization, inpatient hospitalization, and residential treatment, although most adolescents with EDs will be managed in an outpatient setting. Family-based treatment (FBT), often referred to as the "Maudsley method," is emerging as the outpatient treatment of choice for adolescent AN and BN. FBT for adolescent AN and BN is a short-term treatment designed to mobilize parents in assisting their ill child reverse his/her state of starvation (AN), eliminate eating-disordered behaviors such as eating and purging episodes (BN), reestablish healthy eating patterns, and ultimately achieve remission.[20] FBT consists of three phases, the first of which involves parents taking charge of their ill child's eating. Control over eating is returned to the adolescent in the second phase and the final phase addresses issues of family structure and normal adolescent development.[20] FBT is gaining scientific acceptance in light of its demonstrated efficacy to date.[20] A small number of studies have demonstrated that FBT for AN is effective for weight restoration thereby preventing hospitalization in the short term[21–23] and that gains are maintained four to five years after treatment ends.[24–26] FBT for BN has been shown to be superior to supportive psychotherapy for binge/purge abstinence in one study.[27] While outpatient treatment for BED among adolescent remains understudied, both cognitive behavioral therapy and interpersonal psychotherapy (IPT) appear to be promising psychological treatments for BED.[28]

Hospitalization of an adolescent with an ED is necessary in the presence of severe malnutrition, physiologic instability, severe mental health disturbance, or failure of outpatient treatment.[18] Medical decisions regarding stability in AN cases are based on pulse, blood pressure, body temperature, orthostatic changes, electrolyte imbalance, refusal to eat, lack of progress in the outpatient setting, percent of weight lost and percent body fat and cardiac arrhythmias.[17] With BN, different criteria for medical stability are taken into consideration and include syncope, serum electrolyte concentrations, esophageal tears, hematemesis, body temperature, suicide risk, cardiac arrhythmias, and also failure to progress in outpatient treatment.[17]

Long-term Consequences

Health effects of EDs are often immediate, but some may persist well beyond ED remission (Table 65-2). While little is known about the long-term consequences of BED and PD since they have been less studied, more is known about the myriad of health consequences of AN. The peak age of onset of AN corresponds to a time of intense physical, psychosocial, and cognitive development in adolescence and overlaps with the pubertal growth spurt during which adolescents acquire up to 50% of adult weight, more than 20% of adult height, and 50% of adult skeletal mass.[29] Adolescence is also a critical window for bone development, approximately 40%–60% of the maximum adult bone mineral density is acquired between 11 and 18 years of age.[30,31] The severe caloric restriction among people with AN causes increases in bone resorption and decreases in bone formation. Not only do adolescents with AN have low estrogen levels but also they have high cortisol, both of which adversely impact bone mineral density. As a result, adolescents with AN are at increased risk of osteopenia, which is defined as being two standard deviations below the mean for bone mineral density, and the loss in density may not be completely reversible. This loss makes them up to seven times more likely to sustain a fracture.[32] Adolescent females who recover by mid-adolescents may partially recover for the loss,[33] but adolescent-onset cases who recover after peak bone mineral density has been achieved, appear to have a permanent decrease in bone mineral density.[33,34] However, there are cases that onset in adulthood have less bone loss than those with onset during adolescence, a critical window for bone development.

Young female athletes who have amenorrhea, engage in eating-disordered behaviors or have an ED, and have osteopenia is a group at particularly high risk of fracture. This constellation of symptoms is known as the female athlete triad. Although physical activity can promote bone formation, when activity volume becomes very high, weight loss and oligomenorehea or amenorrhea may develop. Thus, at high levels, activity is associated with a hypoestrogenic state, hypothalamic dysfunction, and osteopenia and fractures, particularly stress fractures. Moreover, among children and young adolescents, high levels of activity and AN can both interrupt or delay puberty, and thus decrease bone density and cause stunting. Although some studies have found that girls with BN or subthreshold EDs are at increased risk of low bone mass density, the results have not been consistent.

TABLE 65-4 Goals of ED Treatment	
Treatment goal	Treatment components
1. Restore patients to a healthy body weight, if necessary 2. Eliminate disordered behaviors including restricting, bingeing, purging, and compulsive exercise 3. Improve self-esteem and body image 4. Correct dysfunctional thoughts and beliefs related to food and weight 5. Learn new coping skills 6. Develop healthy, flexible eating habits 7. Stabilize accompanying symptoms and medical conditions of the ED	Outpatient treatment (individualized meal planning and nutrition counseling + medical monitoring + psychotherapy) or family-based treatment or non-outpatient supervised refeeding (partial hospitalization, inpatient hospitalization, or residential program) Multidisciplinary outpatient treatment (psychotherapy + nutrition counseling + medical monitoring ± family therapy) Medical monitoring

Although estrogen may have an independent effect on bone mass in adolescents, hormonal replacement therapy alone is inadequate without weight gain to overcome the detrimental effect of low weight for height on bone density.[35] Weight gain and resumption of menses appears to be the best means of increasing peak bone mass in teens with EDs.[35]

CONCLUSION

EDs, including AN, BN, and BED, and related subthreshold conditions are complex illnesses that often begin in adolescence. EDs are accompanied by myriad complications and consequences that affect almost every organ system of the body. ED treatment should include a collaborative, multidisciplinary team and is most effective when initiated early in the course of illness. Because EDs are prevalent and difficult to treat, effort should be dedicated to prevention and early detection.

REFERENCES

1. Eddy KT, Celio Doyle A, Hoste RR, et al. Eating disorder not otherwise specified in adolescents. *J Am Acad Child Adolesc Psychiatry.* 2008;47(2):156–164.
2. le Grange D, Binford RB, Peterson CB, et al. DSM-IV threshold versus subthreshold bulimia nervosa. *Int J Eat Disord.* 2006;39(6):462–467.
3. Patton GC, Coffey C, Carlin JB, et al. Prognosis of adolescent partial syndromes of eating disorder. *Br J Psychiatry.* 2008;192(4):294–299.
4. Field AE, Javaras KM, Aneja P, et al. Family, peer, and media predictors of becoming eating disordered. *Arch PediatrAdolesc Med.* 2008;162(6):574–579.
5. Bravender T, Bryant-Waugh R, Herzog D, et al. Classification of child and adolescent eating disturbances. Workgroup for Classification of Eating Disorders in Children and Adolescents (WCEDCA). *Int J Eat Disord.* 2007;40 Suppl:S117–122.
6. Bravender T, Bryant-Waugh R, Herzog D, et al. Classification of child and adolescent eating disturbances. *Int J Eat Disord.* 2007;40(S3):S117–S122.
7. Preti A, Girolamo Gd, Vilagut G, et al. The epidemiology of eating disorders in six European countries: Results of the ESEMeD-WMH project. *J Psychiatr Res.* 2009;43(14):1125–1132.
8. Currin L, Schmidt U, Treasure J, Jick H. Time trends in eating disorder incidence. *Br J Psychiatry.* 2005;186(2):132–135.
9. Hudson JI, Hiripi E, Pope Jr HG, Kessler RC. The prevalence and correlates of eating disorders in the National Comorbidity Survey Replication. *Biol Psychiatry.* 2007;61(3):348–358.
10. No Authors. Practice guideline for the treatment of patients with eating disorders (revision). American Psychiatric Association Work Group on Eating Disorders. *Am J Psychiatry.* 2000;157(1 Suppl):1–39.
11. Field AE, Sonneville KR, Micali N, et al. Prospective association of common eating disorders and adverse outcomes. *Pediatrics.* 2012;130(2):e289–295.
12. Kessler RC, Berglund PA, Chiu WT, et al. The prevalence and correlates of binge eating disorder in the World Health Organization World Mental Health Surveys. *Biol Psychiatry.* 2013;73(9):904–914.
13. Stice E, Marti CN, Rohde P. Prevalence, incidence, impairment, and course of the proposed DSM-5 eating disorder diagnoses in an 8-year prospective community study of young women. *J Abnorm Psychology.* 2013;122(2):445–457.
14. Striegel-Moore RH, Dohm FA, Kraemer HC, et al. Health services use in women with a history of bulimia nervosa or binge eating disorder. *Int J Eat Disord.* 2005;37(1):11–18.
15. Swanson SA, Crow SJ, Le Grange D, et al. Prevalence and correlates of eating disorders in adolescents: Results from the national comorbidity survey replication adolescent supplement. *Arch Gen Psychiatry.* 2011; 68(7):714–723.
16. Hill LS, Reid F, Morgan JF, Lacey JH. SCOFF, the development of an eating disorder screening questionnaire. *Int J Eat Disord.* 2010;43(4):344–351.
17. Ozier A, Henry B. Practice paper of the American Dietetic Association: Nutrition intervention in the treatment of eating disorders. *J Am Diet Assoc.* 2011;111(8):1236–1241.
18. Golden NH, Katzman DK, Kreipe RE, et al. Eating disorders in adolescents: Position paper of the Society for Adolescent Medicine. *J Adolesc Health.* 2003;33(6):496–503.
19. Rosen DS, American Academy of Pediatrics Committee on Adolescence. Identification and management of eating disorders in children and adolescents. *Pediatrics.* 2010;126(6):1240–1253.

20. Loeb K, le Grange D. Family-based treatment for adolescent eating disorders: Current status, new applications and future directions. *Int J Child Adolesc Health.* 2009;1:243–254.
21. Lock J, Agras W, Bryson S, Kraemer H. A comparison of short-and long-term family therapy for adolescent anorexia nervosa. *J Am Acad Child Adolesc Psychiatry*. 2005;44:632–639.
22. Eisler I, Dare C, Hodes M, et al. Family therapy for adolescent anorexia nervosa: The results of a controlled comparison of two family interventions. *J Child Psychol Psychiatry.* 2000;41(6):727–736.
23. Le Grange D, Eisler I, Dare C, Russell G. Evaluation of family treatments in adolescent anorexia nervosa: A pilot study. *Int J Eat Disord.* 1992;12:347–357.
24. Eisler I, Dare C, Russell G, et al. Family and individual therapy in anorexia nervosa: A 5-year follow-up. *Arch Gen Psychiatry.* 1997;54:1025–1030.
25. Eisler I, Simic M, Russell G, Dare C. A randomised controlled treatment trial of two forms of family therapy in adolescent anorexia nervosa: A five-year follow up. *J Child Psychol Psychiatry.* 2007;48(6):552–560.
26. Lock J, Couturier J, Agras W. Comparison of long-term outcomes in adolescents with anorexia nervosa treated with family therapy. *J Am Acad Child Adolesc Psychiatry*. 2006;45:666–672.
27. Le Grange D, Crosby R, Rathouz P, Leventhal B. A randomized controlled comparison of family-based treatment and supportive psychotherapy for adolescent bulimia nervosa. *Arch Gen Psychiatry.* 2007;64:1049–1056.
28. Wilfley DE, Kolko RP, Kass AE. Cognitive-behavioral therapy for weight management and eating disorders in children and adolescents. *Child Adolesc Psychiatr Clin N Am.* 2011;20(2):271–285.
29. Stang J, Story M, eds. *Guidelines for adolescent nutrition services*. Minneapolis, MN: Center for Leadership, Education and Training in Maternal and Child Nutrition, Division of Epidemiology and Community Health, School of Public Health, University of Minnesota; 2005.
30. Bonjour J-P, Theintz G, Buchs B, et al. Critical years and stages of puberty for spinal and femoral bone mass accumulation during adolescence. *J Clin Endocrinol Metab.* 1991;73(3):555–563.
31. Theintz G, Buchs B, Rizzoli R, et al. Longitudinal monitoring of bone mass accumulation in healthy adolescents: Evidence for a marked reduction after 16 years of age at the levels of lumbar spine and femoral neck in female subjects. *J Clin Endocrinol Metab.* 1992;75(4):1060–1065.
32. Rigotti N, Neer R, Skates S, et al. The clinical course of osteoporosis in anorexia nervosa: A longitudinal study of cortical bone mass. *JAMA.* 1991;265(9):1133–1138.
33. Misra M, Prabhakaran R, Miller KK, et al. Prognostic indicators of changes in bone density measures in adolescent girls with anorexia nervosa-II. *J Clin Endocrinol Metab.* 2008;93(4):1292–1297.
34. Bachrach LK, Katzman DK, Litt IF, et al. Recovery from osteopenia in adolescent girls with anorexia nervosa. *J Clin Endocrinol Metab.* 1991;72(3):602–606.
35. Gordon CM, Nelson LM. Amenorrhea and bone health in adolescents and young women. *Curr Opin Obstet Gynecol.* 2003;15(5):377–384.

Adolescence: Advancing Bone Health

66

Keith J. Loud, MDCM, MSc and Catherine M. Gordon, MD, MSc

Adolescence is a critical period for lifetime bone health. Evidence has accumulated that an individual's peak bone mass (PBM), which is the strongest predictor of his or her future risk of osteoporosis, is reached by early adulthood. At least half of all adults' mineralized calcium is laid down during the adolescent years, with the most rapid bone accretion occurring in late childhood and early adolescence.[1,2] Any condition that impairs this process may create a deficit in bone mass that will have permanent ramifications for future risk of osteoporosis, which afflicts over 10 million Americans[3] and results in more than $22 billion in direct medical expenditures in 2008.[4]

Many disease states, each related to nutritional deficits, can inhibit bone accretion during adolescence, including rickets, eating disorders, the "female athlete triad," inflammatory bowel disease (IBD), celiac disease, cystic fibrosis, cancer, and other multisystem or inflammatory disorders. In this chapter, these conditions are discussed in detail. Adolescent behavior itself may also prevent maximal PBM attainment, as the dietary habits of many teenagers do not lead to optimization of calcium and vitamin D intake.

Normal Bone Physiology and Development

Bones are the fundamental units of the skeletal system, providing an adaptable framework that supports and protects the major organ systems of the body while allowing for locomotion. Because of its role in calcium and

phosphate homeostasis, bone can also be considered an organ system. The skeletal system is the reservoir for the calcium used in essential mechanisms ranging from the regulation of cardiac electrical activity, contraction of muscles, and varied inter- and intracellular messaging systems. Each bone, housing lymphoid and myeloid precursor cells within the marrow, is also an organ unto itself. Finally, these organs are composed of a unique tissue substance that is called "bone."[5]

Types of Bone

Bone as a tissue takes on two forms in normal health: cortical (dense) and trabecular (cancellous or spongy) bone.[5] Cortical bone, which makes up 80% skeletal bone mass, is found in the diaphysis (shaft) of long bones such as the femur, as well as the "walls" of almost all other bones, including the vertebrae. It provides strength against tensile, torsional, and shear forces. The remaining 20% bone mass consists of trabecular bone, found in the metaphyses (ends) of long bones and the entire bodies of the vertebrae. It is well suited to resist compressive forces. Because of its high surface area-to-volume ratio, trabecular bone occupies greater than 20% volume of the skeletal system and has approximately eight times the metabolic activity of cortical bone.[5] Because of its higher metabolic availability, trabecular bone is more significantly affected by disease states.

Histology: The Building Blocks of Bone

Regardless of its structure or ultrastructure, on a microscopic level bone consists of thin layers, or lamellae, of a mineralized protein matrix. This mature lamellar bone is 70% (by dry weight) mineral, predominantly calcium and phosphate in the form of hydroxyapatite crystals [$Ca_{10}(PO_4)_6(OH)_2$] and a less pure, carbonate-rich analogue called bone apatite, which is more soluble and thus more available for metabolic activity.[5] The remaining 30% mass is an organic substance called osteoid, primarily protein in the form of type I collagen. Cellular elements, which produce all of the metabolic activity of bone, make up only 2% of the osteoid.

Osteoblasts are bone-forming cells derived from fixed-tissue osteoprogenitor cells.[6] Osteoblasts synthesize osteoid on the surface of already mature lamellar bone, periosteum, or endosteum. They then regulate the process of mineralization of the collagen matrix through the actions of bone-specific alkaline phosphatase (BSAP), which is different

than the alkaline phosphatase produced in the liver and can be distinguished from it by specific assays.

Once the osteoid matrix has mineralized, it is considered bone. The osteoblasts, which are synchronized in such a way as to create the parallel lamellae, become trapped inside small caverns called lacunae and mature into osteocytes that communicate via long processes with each other to direct mineral homeostasis.

Osteoclasts are multinucleated cells from the marrow monocyte–macrophage cell line.[7] By producing lysosomal enzymes such as acid phosphatase, osteoclasts break down the mineralized bone matrix, mobilizing calcium and phosphorus for participation in homeostasis, in a process called bone resorption.

Growth and Remodeling: The Bone Cycle

Bone is a dynamic tissue. Growth in long bones occurs at the physes (growth plates) and apophyses (secondary centers of ossification) by a process known as enchondral ossification in which osteoblasts lay down bone on a cartilaginous framework that is ultimately resorbed by osteoclasts. Even after skeletal maturity, osteoblasts respond to structural stresses in bone by laying down new matrix. Osteoclasts concomitantly resorb areas of bone under less stress. Thus, bone is constantly remodeling. Newly identified factors, which comprise the Wnt/Lrp5 signaling and BMP pathways, among others, are responsible for the "cross-talk" that occurs between osteoblasts and osteoclasts.[8] During times of growth, bone formation by osteoblasts is greater than resorption by osteoclasts. With disuse, osteoclasts outpace osteoblasts and bone mass decreases. Otherwise, osteoblasts and osteoclasts are tightly "coupled" to keep formation and resorption equally balanced in what is known as "the bone cycle."[6,7]

Importance of Adolescence

Bones grow at different rates throughout the skeleton. The extremities (appendicular skeleton) have largely completed growth by the time of the pubertal growth spurt (peak height velocity [PHV]). Increases in estrogen and testosterone during puberty result in growth of the trunk and spine (axial skeleton). These increases in vertebral bone size are accompanied by dramatic increases in bone mineral density (BMD) in cross-sectional[9,10] and longitudinal studies.[11] This trabecular bone reaches its peak BMD by age 20–30 years, but steadily declines thereafter.[12] Alarmingly, one animal

model showed that calcium restriction in adolescence resulted in a decreased bone volume throughout life, even after restoration to recommended calcium intake in young adulthood.[13] Adolescence is an essential time for significant bone accretion. Over half of adult bone calcium is laid down during the teenage years.[1] Maximal rates of bone mineral accretion follow PHV by 6–12 months.[14] As a consequence, at the time of PHV (Tanner pubertal stage II–III in girls, Tanner III–IV in boys), teenagers have reached approximately 90% of their adult height but they have acquired only 60% of their adult total body mineral content, resulting in relatively less mineralized bone.[14] In North America, these pubertal stages are reached by age 11–14 in girls and 13–17 in boys. Classic studies by Thientz et al.[2] are consistent, showing that girls' BMD may plateau by age 16 (or 2 years post menarche) and boys' by age 20. This is a much earlier window of opportunity than was previously suspected. In fact, 95%–100% PBM may be attained by the end of adolescence.[11,15]

Maximizing Bone Health

Nonnutritional Factors

Intrinsic Factors: Genetics

Intrinsic factors dominate the determination of bone density. Males have a higher bone mass than females at nearly all ages (Figure 66-1).[11,16] African–American children demonstrate an approximately 10% higher BMD by dual-energy X-ray absorptiometry (DXA) than do children of other ethnicities,[11,16] even after correcting for bone size.[15] A large cohort study in the United States showed that these differences contribute to a doubling in fracture risk among children of Caucasian European descent when compared with their peers.[17] An interesting finding among African–American and Caucasian girls was that their spinal BMD was similar until puberty, at which point the African–American girls had an increase of 34% while the Caucasian girls improved only 11%.[18] Family history of osteoporosis in postmenopausal women predicts lower BMD for their daughters,[19] and both elderly men and women have an increased risk of osteoporosis when other family members have been affected.[20] In all, 60%–80% variance in PBM is attributed to heritable factors.[12] Polymorphisms in the genes encoding receptors for vitamin D, estrogen, type I collagen,[21] insulin-like growth factor I (IGF-I), transforming growth

factor beta (TGF-β), and interleukin 6 (IL-6) are under ongoing study,[22] but none have been able, singly or in combination, to account for more than a small percentage of this variance in PBM.

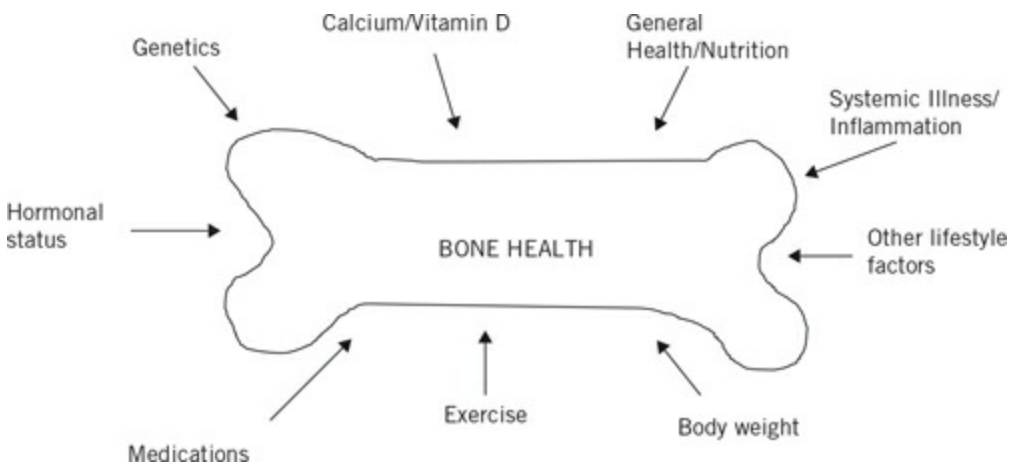


Figure 66-1 Factors affecting bone health. (Reproduced with permission from Loud KJ and Gordon CM.[194])

Hormonal Status

A balanced hormonal milieu is essential to attain and maintain normal bone formation. Early menarche and regular menses are strong predictors of increased bone mass in women,[23] demonstrating the importance of estrogen.[24] Among adolescent girls, an estimate of estrogen exposure was positively correlated with spine BMD.[12] Estrogen deficiency may also slow growth in axial bone size[25–27] when it occurs during adolescence, a time when truncal growth typically accelerates.

Androgens exert independent and positive effects on PBM.[28] Although men with a history of delayed puberty and documented low serum androgen levels have been shown to have decreased BMD, a meta-analysis of randomized, placebo-controlled clinical trials of testosterone use in men showed only a modest improvement in lumbar spine BMD and inconclusive results at the femoral neck.[29] Nonetheless, androgen replacement therapy to teenage boys may provide skeletal benefits during the formative adolescent years.[30] Patients with androgen insensitivity have a mutation in the androgen receptor, with consequent impaired androgen action, such that there is resistance to even elevated androgen levels. Previous reports have shown that affected individuals have decreased bone

mineralization.[31–33] Even after administration of estrogen replacement therapy in these individuals, BMD did not significantly increase, again highlighting the importance of androgens in bone physiology.[31] Women with androgen excess also have been shown to have a higher BMD.[34,35]

Children and adults with childhood-onset growth hormone (GH) deficiency have lower BMD than controls.[36,37] Although patients on adequate replacement doses of GH have increased BMD compared to GH-deficient patients,[38] those adolescents who discontinued the medication at the completion of linear growth demonstrated decreased spine and hip BMD in long-term follow-up, suggesting that bone mineral accretion continues beyond completion of linear growth.[39] The actions of GH are likely mediated by IGF-I, an anabolic factor that stimulates osteoblast function directly.

Exercise

Patients with impaired activity due to central nervous system or neuromuscular disease develop disuse osteoporosis.[40] It is well established that individuals engaged more intensely in weight-bearing activities have higher measured BMD than less active controls.[41] Even among women with amenorrhea and adolescents with anorexia nervosa (AN), those who are more athletic have higher bone density.[42,43] Unfortunately, swimmers do not consistently reap this benefit, as their activity is largely weightless, highlighting the importance of weight-bearing activities.[44] The increases in BMD are site specific; for example, runners and ballet dancers have higher proximal femur (hip) BMD than controls, whereas gymnasts have higher measured radial (arm) BMD.[45] Elite men and women rowers have also been shown to have a higher spinal bone density.[46] Although not all intervention studies have replicated these findings, and the specifics of optimal frequency, intensity, and duration remain elusive, most clinical trials of physical activity demonstrate a positive association between exercise and the bone health of children, adolescents, and adults.[47,48]

Chronic Illness and Systemic Inflammation

Many of the medical conditions associated with impaired bone density are listed in Table 66-1. Some of these diagnoses are associated with malnutrition, with concomitant decreases in BMI and absorption of important nutrients that are described in detail later in this chapter, in the sections on celiac disease, inflammatory bowel disease, and cystic fibrosis. Many are additionally marked by systemic inflammation, which is

increasingly known to result independently in decreased BMD. For example, the magnitude of glucocorticoid use is the single greatest predictor of low bone mass in patients with both ulcerative colitis (UC) and Crohn's disease (CD).[49] Glucocorticoids directly decrease bone formation by inhibiting osteoblast activity. Indirectly, these agents also decrease the secretion of estrogen and testosterone, which are necessary for bone accretion, as well as decrease the absorption of calcium in the gut and reabsorption of calcium in the kidneys.

These adverse effects of glucocorticoids are a concern for other patients with systemic steroid–dependent diseases such as idiopathic juvenile arthritis (IJA; formerly juvenile rheumatic arthritis), childhood leukemia, collagen vascular and connective tissue diseases, and solid organ transplant recipients. One study has shown a dose-related loss of BMD at the hip in premenopausal adult women using inhaled glucocorticoids for treatment of their asthma.[50] Although there is a similar concern for children and adolescents, it is largely speculative. Reassuring evidence comes from a study of steroid-responsive nephrotic syndrome in children and adolescents, which showed that decreased BMD is not correlated with steroid dosage but with underlying disease severity, for which steroid use is merely a marker. Body mass was also shown to be protective of BMD in that study.[51]

Other autoimmune-mediated diseases, allergic conditions, and even chronic infection have been associated with decreased regional and total body bone density, further implicating systemic inflammation as a cause of bone loss.[52] The proinflammatory cytokines IL-1α, IL-1β, IL-6, IL-11, IL-17, tumor necrosis factor (TNF)-α and TNF-β, TGF-α, EGF, and prostaglandin E_2 all activate osteoclastic resorption of bone.[52] TNF-α and IL-1β also decrease osteoblastic bone formation while stimulating the osteoblasts to secrete IL-6, further activating the osteoclasts.[52]

Basic experiments have continued to elucidate the final common pathways of the signaling cascade that modulates the coupling of the bone remodeling cycle. Of particular interest is the osteoblast surface ligand called the receptor activator of nuclear factor kappa B ligand, or RANKL. When binding to the osteoclast precursor, RANK is predominant and osteoclast differentiation and maturation is favored, leading to increased bone resorption. Osteoprotegerin (OPG) blocks the RANK–RANKL interaction, thus shifting the balance in favor of bone formation.[6] The observation that RANKL also regulates early lymphocyte development and some T-cell interactions[6] further links bone remodeling with the

activity of the immune system.

Nutritional Influences

General Fitness and Macronutrients

In addition to participating in weight-bearing exercise, individuals must maintain appropriate weight. At most weights, there is a direct relationship between body mass index (BMI) and BMD, with underweight individuals at increased risk for lower BMD.[53] Adolescent girls with low weight due to AN have decreased BMD, as is discussed below. Excessive BMI may also have a deleterious effect on BMD. Work by Goulding et al. has suggested that overweight boys and girls are fracture prone,[54] and have bone mass and bone area that are increased for their age, but not appropriately relative to their total body weight.[55] An anthropometric study of normal weight, overweight, and obese female adolescents demonstrated no difference in measures of bone mineral content and density when controlling for lean body mass, suggesting a deleterious effect of increased adiposity on the skeletal development of overweight children and adolescents.[55,56] Clinical investigations linking visceral, rather than subcutaneous, adipose tissue to low bone mass in obese adolescent girls have begun to implicate adipokines (adiponectin, leptin) and proinflammatory cytokines (as mentioned above) in the pathophysiology.[57] These adipokines also appear to be at play in the mediation of bone loss in amenorrheic women athletes and adolescent girls with AN.[58]

TABLE 66-1 Conditions Predisposing to Osteoporosis in Children and Adolescents

Chronic illnesses mechanisms = malnutrition, pro-inflammatory and antiresorptive cytokines, endocrinopathies, medication effects, decreased activity	Inflammatory bowel disease (IBD) (Crohn's disease, ulcerative colitis) Celiac disease Asthma Chronic renal failure Cystic fibrosis (CF) Connective tissue diseases Cancer Idiopathic juvenile arthritis (IJA) Other chronic illnesses
Organ transplantation mechanisms = malnutrition,	Bone marrow

cytokines, immunosuppression, endocrinopathies, medication effects, decreased activity	Heart Lung Kidney Pancreas Liver
Endocrine abnormalities	Genetic sex hormone deficiency Turner syndrome Acquired sex hormone deficiency Anorexia nervosa Exercise-induced amenorrhea Primary ovarian insufficiency Irradiation, chemotherapy Other endocrine abnormalities Cushing syndrome Hyperparathyroidism Hyperthyroidism Growth hormone deficiency Hyperprolactinemia Diabetes mellitus (type 1 and 2)
Medications	Glucocorticoids Anticonvulsants Cyclosporine A Lithium GnRH agonists Heparin Methotrexate
Family history	Osteoporosis Frequent fractures
Female athlete triad definition = interrelations of energy availability, menstrual function, and bone density	Any female athlete, but particularly those in activities that emphasize lean physique
Prolonged immobilization	Cerebral palsy and other neurologic conditions Postoperative immobilization

The macronutrients carbohydrate, fat, and protein are essential building blocks for growth and attainment of appropriate BMI to maximize BMD. These nutrients also provide energy for the muscle contractions that mechanically load bone, thereby stimulating bone formation. In addition, lipids are substrates for the synthesis of prostaglandins, which are cytokines that may play an essential role in regulating bone metabolism.[59]

Proteins are particularly important as the fundamental elements of muscle tissue, as well as the predominant components of osteoid, the unmineralized precursor of bone. In one study, protein intake and bone mass gains were positively correlated in children, possibly mediated by IGF-I production.[60] Although small, poorly controlled additional studies[61] cite the importance of increased macronutrient intake to bone health, most larger scale research focuses on the effects of calcium and vitamin D.

Other nutrients with putative, but less well studied, influences on bone density include magnesium, phosphorus, copper, manganese, zinc, and vitamins C and K (see Appendix II for Recommended Daily Allowances [RDA] of these nutrients). Approximately 60% body's magnesium and 85% phosphorus is in bone. Magnesium may decrease the brittleness of bone by modulating the size of hydroxyapatite crystals. The other minerals and vitamins are important cofactors in the synthesis, folding, and cross-linking of the proteins found in bone, including collagen, elastin, and osteocalcin.[59] A small study of British female endurance athletes showed a significant positive correlation between lumbar spine BMD and dietary magnesium and zinc intake.[62]

Calcium

As described above, bone is a calcified tissue that derives its compressive strength from its calcium phosphate crystal structure, and in turn serves as the body's vast reservoir of calcium. Enormous gains are made in total skeletal calcium over childhood and adolescent development, from approximately 25 g at birth to 900 g (women) to 1200 g (men) at skeletal maturity.[14] It follows that calcium is an essential nutrient for skeletal as well as general health. Dairy products are an important source of dietary calcium, providing approximately 75% calcium in a typical North American diet.[59,63] Epidemiologic data suggest that osteoporosis is more prevalent in regions where dairy intake is low[64] and that milk consumption in childhood and adolescence correlates with postmenopausal BMD.[65]

Although dietary calcium intake would therefore be a seemingly obvious factor promoting increased BMD, the literature is inconsistent. In growing children, the influence of calcium availability on gains in bone density has been demonstrated. Dietary calcium is one of the most significant predictors of bone mass in cross-sectional studies of children and adolescents.[66–68] Controlled supplementation trials in healthy children and adolescents,[69–73] including twin studies,[74,75] have generally shown increased gains in BMD in at least some subgroups of those supplemented

with calcium, milk extract, or dairy products.[76] However, the response to calcium varied among studies with respect to skeletal site, pretreatment calcium consumption, pubertal stage, and genetic factors.[77] There is also uncertainty in how well the gains are maintained after supplementation is discontinued, as only one of the studies demonstrated persistence of the increase more than one year.[78] Similarly, women consuming greater than 1000 mg calcium daily made no significant gains in their BMD if they did not concomitantly increase their activity patterns,[79] suggesting an interesting synergy between calcium intake and exercise that represents an area of active research. This is an area of much scrutiny,[77] but expert panels have recently reaffirmed the recommended daily intake of 1300 mg elemental calcium for adolescents (see Table 66-2).[63,80]

There has been recent controversy regarding potential adverse cardiovascular side effects of calcium supplementation in the elderly that has raised questions about its safety among youth. Specifically, reports have suggested that daily calcium supplementation of 500 mg or above is associated with increased risk of stroke and myocardial infarction, while others refute this claim.[81–83] Of note, no data currently exist to substantiate concerns about adverse side effects in children and adolescents, but the adult data have generated concern among patients and their families. Promoting adequate dietary calcium remains the primary recommendation with use of supplemental calcium reserved for known lactose intolerance or in the setting of documented vitamin D deficiency.

Vitamin D

The active form of vitamin D, 1,25-dihydroxyvitamin D [1,25(OH)2D], is necessary for normal mineralization of bone. In addition to being absorbed through the gut, Vitamin D is also synthesized endogenously in the skin under the influence of ultraviolet sunlight. It is converted to 25-hydroxyvitamin D [25(OH)D] in the liver and then to 1,25(OH)2D in the kidney. Activated 1,25(OH)2D facilitates absorption of calcium and phosphate from the duodenum and jejunum, as well as reabsorption of calcium from the renal tubules. At high levels in the serum, it can stimulate osteoclasts to resorb bone, but this action is mediated via receptors on osteoblasts.[84]

The effects of calcium and vitamin D are difficult to separate. When calcium intake is sufficient, non-vitamin D-dependent passive absorption predominates. However, in the face of low calcium intake, vitamin D-dependent active transport becomes the major route for calcium

absorption. Thus, the effect of vitamin D deficiency on calcium bioavailability is critical when it coexists in the setting of low dietary calcium intake.[85,86] Fortunately, such a deficit can be easily treated, and the incidence of fractures possibly decreased,[87] with combined vitamin D and calcium supplementation.[88]

Vitamin D deficiency has been clearly linked to fractures in postmenopausal and osteoporotic women, and is a common problem across the age spectrum.[84,89–93] Plasma 25(OH)D levels, the most sensitive clinical index of vitamin D status, are positively correlated with BMD in middle-aged and elderly women.[90] Seasonal variation of vitamin D metabolites, with high summer/fall and low winter values, has been established in adults and is generally more pronounced in northern latitudes, where there is reduced cutaneous production of vitamin D for up to six months of the year.[91–97] Some,[98–101] but not all,[102] studies have indicated a similar seasonal difference in BMD. In postmenopausal women with low intakes of vitamin D, BMD significantly decreases in winter, which can be prevented by vitamin D supplementation.[101]

In a study of hospitalized adults, Thomas et al. reported that 57% men and women screened were vitamin D-deficient on serum testing.[103] Research in Scandinavia has shown that the dietary intake of vitamin D in children, even when combined with vitamin stores from the summer months, is not sufficient to sustain healthy vitamin D levels during the winter.[104,105] The prevalence of vitamin D insufficiency among otherwise healthy American adolescents has been demonstrated to be as high as 42%, with prevalence highest among African–American teens and during the winter months.[89] Other studies have replicated the findings of a high prevalence of vitamin D deficiency among teenagers.[106,107] A concern is that the occurrence of vitamin D deficiency during this important developmental period results in an increased bone remodeling rate, which could impair bone density achievement during skeletal growth.[108]

TABLE 66-2 Dietary Reference Intakes/Recommended Daily Allowance of Calcium and Vitamin D

	Calcium			Vitamin D		
Life Stage Group	Estimated Average Requirement (mg/d)	Recommended Dietary Allowance (mg/d)	Upper Level Intake (mg/d)	Estimated Average Requirement (IU/d)	Recommended Dietary Allowance (IU/d)	Upper Level Intake (IU/d)
Infants 0–6 months	*	*	1000	**	**	1000
Infants 6–12 months	*	*	1500	**	**	1500
1–3 years old	500	700	2500	400	600	2500
4–8 years old	800	1000	2500	400	600	3000
9–13 years old	1100	1300	3000	400	600	4000
14–18 years old	1100	1300	3000	400	600	4000
19–30 years old	800	1000	2500	400	600	4000
31–50 years old	800	1000	2500	400	600	4000
51- to 70-year-old males	800	1000	2000	400	600	4000
51- to 70-year-old females	1000	1200	2000	400	600	4000
>70 years old	1000	1200	2000	400	800	4000
14–18 years old, pregnant/lactating	1100	1300	3000	400	600	4000
19–50 years old, pregnant/lactating	800	1000	2500	400	600	4000

*For infants, adequate intake is 200 mg/d for 0–6 months of age and 260 mg/d for 6–12 months of age. **For infants, adequate intake is 400 IU/d for 0–6 months of age and 400 IU/d for 6–12 months of age.

Source: Adapted from Institute of Medicine.[80]

Dietary vitamin D intake of some of the patients identified as deficient in the Thomas study exceeded the RDA and many had no apparent risk factors for vitamin D deficiency.[103] To address this issue, investigations are ongoing into the mechanisms of vitamin D action. One such area is the role of the vitamin D receptor (VDR) gene in calcium absorption and bone mass accretion. One study noted a significantly lower spinal BMD in girls with a homozygous recessive (BB) genotype at one restriction site when compared to heterozygous (Bb) or dominant (bb) genotypes.[109] Thus, alleles for this receptor may influence an individual's response to calcium intake and other environmental factors.

Finally, there is an ongoing debate regarding both the appropriate vitamin D supplementation dose for children and adolescents as well as the optimal serum 25(OH)D concentration needed to promote bone health, among other health outcomes. Vitamin D dosing depends on whether the young person is found to have vitamin D deficiency (25(OH)D < 20 ng/mL) or insufficiency (25(OH)D < 21–29 ng/mL).[80,110] In 2010, the Institute of Medicine (IOM) of the US National Academy of Science published recommendations aimed at the healthy population that established a new RDA of 600 IU daily for the pediatric age group (see

Table 66-2).[80] In 2011, the Endocrine Society, an international organization of pediatric and adult endocrinologists, published similar guidelines that were targeted at individuals with chronic diseases associated with bone loss, and/or who are at high risk for vitamin D deficiency.[110] In these patients, vitamin D supplementation doses as high as 2000 IU daily are recommended. Either vitamin D_2 or vitamin D_3 is effective in raising blood levels to the desired range, although recent research suggests that vitamin D_3 is more potent and supports cortical bone mass and strength better than vitamin D_2.[110,111]

DISTURBANCES IN ADOLESCENT NUTRITION WITH IMPLICATIONS FOR BONE HEALTH

General Nutrition

It should be remembered that rates of gastrointestinal calcium absorption and urinary excretion significantly influence the bioavailability of calcium. Foods high in oxalic acid such as spinach, beans, sweet potatoes, and rhubarb, which are not typically major components of the average teenager's diet, may inhibit calcium absorption.[59] Otherwise, calcium absorption is relatively high and excretion relatively low in adolescents when compared with adults. The typical adolescent's high-salt diet may negate this advantage; each gram of urinary sodium excretion obligates a loss of 26 mg of urinary calcium.[112]

Dairy intake

A US survey in the late 1990s showed that although adolescents overwhelmingly (>90%) believed that calcium is healthy and important for bones, few (19%) were aware of the RDA for calcium and even fewer (10%) knew how to meet this goal with dairy products.[113] It is not surprising, therefore, that less than 16% adolescent girls in the United States met the RDA of calcium of 1300 mg/d.[114] In 2006 the mean calcium intake for girls aged 9–13 years was approximately 988 mg/d, or 76% of DRI; this value dropped to approximately 918 mg/d, or 71% of DRI in 14–18 year-old girls.[115] For vitamin D, girls aged 9–13 years consumed 7.7 μg/d (308 IU) but declined to 5.0 μg/d (200 IU) by age 14–18.[115] Boys in the United States fared slightly better, with mean daily intakes of 1093 mg calcium (84% DRI) and 7.5 μg (300 IU) vitamin D in

ages 9–13 and 1296 mg calcium (99.6% DRI) and 6.9 μg (276 IU) vitamin D in ages 14–18,[115] probably due to better dairy consumption.

Carbonated Beverages and Tobacco

Wyshak has shown that consumption of carbonated beverages, particularly cola (phosphoric acid containing) drinks,[116] in teenaged girls may be associated with increased risk of bone fractures.[117] The association was not found to hold for teenaged boys.[117] Similarly, a large observational study in Northern Ireland linked lower BMD with increased carbonated soda beverage consumption, but again only in 12-to-15-year-old girls.[118] The pathophysiology is not completely understood, and is a source of significant debate.[119] Cola drinks, in particular, are implicated because they create a high phosphoric acid load that both chelates calcium and may increase parathyroid hormone (PTH) secretion, but others argue that this is a less important factor than the simple replacement of milk by carbonated beverages.[119] Between 1970 and 1997 in the United States, per capita consumption of carbonated beverages increased by 118% while consumption of milk declined by 23%.[120]

Finally, tobacco, by decreasing osteoblast function,[121] and excessive alcohol use increase the risk of lower BMD in young women. A study of these factors in younger adolescents showed that the effects of alcohol use and smoking were synergistic, and magnified the negative associations between teen anxiety, depression, and BMD.[122]

BONE DISEASES

Osteomalacia

Rickets, or vitamin D-deficient osteomalacia, is a classic example of the detrimental effect of malnutrition on bone and continues to be a problematic condition for growing children in North America and worldwide. As previously mentioned, evidence has accumulated that vitamin D deficiency may be prevalent among American adolescents and young adults, particularly those who live in northern latitudes.[89,106,107]

Low levels of 1,25(OH)2D lead to low levels of serum calcium, which stimulates PTH secretion. Under the stimulus of PTH, osteoclasts increase the rate of bone resorption to maintain calcium homeostasis. Osteoblasts respond to the increased osteoclast activity by increasing osteoid formation. Because of the relative lack of calcium and phosphate, this

osteoid is poorly mineralized. As a consequence, vitamin D deficiency impacts negatively not only on trabecular bone, which is more hormonally responsive and more commonly affected by such deficiencies, but also on cortical bone.

In addition to poor dietary intake, vitamin D deficiency can be caused by inadequate exposure to sunlight. Anticonvulsants cause increased hepatic breakdown of vitamin D. Patients with renal disease may have decreased activation of 25(OH)D to 1,25(OH)2D, in addition to increased tubular losses of calcium and phosphate. Use of aluminum-containing antacids impairs phosphate absorption. Other gastrointestinal conditions also impair absorption of vitamin D, calcium, and phosphate, as described later.

Clinical manifestations of rickets in skeletally immature children include flaring of the metaphyses and growth retardation. In skeletally mature adolescents and adults, findings are more subtle, including bone pain, susceptibility to fracture, and pseudo-fracture. Serum levels of 25(OH)D are a reliable measure of total vitamin D stores. However, the desirable level for 25(OH)D, including what constitutes the threshold stimulus for rises in PTH, remains a subject of debate.[85,105] A recent study by Hill et al. showed that the 25OHD inflection point for maximal PTH suppression is more difficult to define in children and adolescents versus adults. Thus, this endpoint is difficult to use as an outcome for defining optimal vitamin D status in a young population.[123]

Prevention of vitamin D deficiency and rickets includes ensuring adequate dietary intake of vitamin D, with empiric supplementation now recommended by the IOM for healthy neonates (400 IU daily) and children age 1 year and above (600 IU daily).[80] For children with a documented low bone mass or previous history of vitamin D deficiency, oral supplementation doses of 1000–2000 IU daily or 50,000 IU once weekly may be appropriate.[110] There are data suggesting an increased efficacy of vitamin D_3 (animal-derived) over vitamin D_2[124,125]; this preparation is generally preferred, if readily available. Parenteral dosing of 50,000–300,000 IU provided as vitamin D_2 intramuscularly can also be used, but must be undertaken cautiously, with assurance of adequate calcium intake and appropriate monitoring of vitamin D and PTH levels. However, parenteral preparations are presently difficult to obtain due to limited production.

Osteoporosis

Definitions

In contrast to osteomalacia, which refers to a condition in which the bone matrix is poorly mineralized regardless of the actual mass of bone tissue produced, osteoporosis is defined as a systemic skeletal disease characterized by low bone mass and micro-architectural deterioration of bone tissue, with a consequent increase in bone fragility and susceptibility to fractures.[126] A related term, osteopenia, is used to describe a significant reduction in bone mass for age and sex, but only in postmenopausal women and older adult men.[127] The term osteopenia should be avoided in a child, as this terminology generates confusion.[128] In each of these conditions, the osteoid is mineralized normally to form calcified bone, but there is less of it and its structural arrangement is abnormal.

Diagnosis

Adult osteoporosis is defined by the World Health Organization (WHO) as a BMD below 2.5 standard deviations from the mean peak BMD attained for sex (i.e., T-score < –2.5) as measured by DXA. Osteopenia in adults is the term reserved for BMD with T-score between –1.0 and –2.5.[127] Using these definitions, DXA scanning has become the standard modality for diagnosing osteoporosis in postmenopausal women and older adult men. In adolescents and younger adults, these WHO definitions do not apply; the International Society for Clinical Densitometry (ISCD), in a 2007 position statement, specifies that osteoporosis in these age groups is a clinical diagnosis, requiring evidence of skeletal fragility in addition to decreased BMD.[127,128] Appropriate DXA use and terminology in the pediatric population is discussed below.

Nutritional Disease States Associated with Impaired Bone Health

Anorexia Nervosa

Patients with AN are at risk for profound deficits in BMD, which are often irreversible, even with a return to appropriate weight. Patients with chronic AN have a sevenfold increased incidence of fractures and are at increased risk for developing early osteoporosis.[129–132] Because the incidence of anorexia increases during the years of attainment of PBM, the lifelong impact on skeletal health in maturity is particularly concerning. The mechanisms of this bone loss are certainly related to decreased dietary intake of calcium, vitamin D, and other macronutrients. Recent investigations have focused on the role of paradoxically increased fat in

the bone marrow of these patients.[133,134] One of the first investigations of male adolescents with AN confirmed the decreases in BMD seen in women, and suggests some of the same mechanisms are at play.[135]

Women with AN also develop hypothalamic amenorrhea that results in low circulating levels of estrogen.[136] It is hypothesized that estrogen helps regulate bone mass by impairing osteoblast-mediated bone resorption. Oral contraceptive (OCP) use by young women with AN was shown in a retrospective cross-sectional study to be associated with a higher BMD.[137] In a small pilot study of women with AN and hypothalamic amenorrhea, ERT resulted in increases in BMD.[138] A recent trial of transdermal estrogen showed gains in bone density in the young adolescents studied, but below what was expected for age.[139] Unfortunately, ERT alone has not consistently been shown in other controlled studies to be able to reverse the bone loss in patients with AN, with the possible exception of those most profoundly affected (below 70% of ideal body weight).[140] Ultimately, nutritional rehabilitation and weight restoration are necessary to return menstrual function, circulating estrogen levels, bone formation, and BMD to normalcy.[141] It may be frustrating to the patients that it often requires a higher body weight to resume menses than the level at which they ceased.[136] Even prior to attainment of menstrual regularity, however, bone density increases with weight gain in adolescent girls with AN.[142]

It has been suggested that bone formation is more profoundly decreased than resorption is increased in patients with AN.[131] This "uncoupling" of the normal bone cycle has led investigators to address both sides of the formation–resorption dyad.[43,143–145] Serum levels of IGF-I have been found to be decreased in patients with AN.[145] Randomized controlled trials of IGF-I administration showed significant increases in bone formation in patients with AN, with greatest improvement in those who were also given OCPs,[146] arguing for a synergistic effect between ERT and IGF-I. Unfortunately, use of this agent in children and adolescents may be limited because of its availability only as an intravenous or subcutaneous preparation. Longitudinal data are also needed to determine long-term efficacy.

Nutritional Supplements: Dehydroepiandrosterone

Similar to IGF-I, levels of the adrenal steroid dehydroepiandrosterone (DHEA)[147] can be low in patients with AN. DHEA is positively correlated with BMD in adults[148] and parallels the pattern of bone accretion in that

secretion of DHEA peaks during the same adolescent years as bone mass acquisition accelerates.[149,150] DHEA is thought to increase levels of IGF-I, thereby increasing bone formation by stimulating osteoblasts, and also decrease mediators of osteoclast-mediated resorption.

Studies on the administration of oral DHEA, which is currently sold as a nutritional supplement, in adolescent girls with AN have shown that this agent is well tolerated.[151] The adolescents who were given DHEA showed significant decreases in concentration of NTx, a marker of bone resorption, and increases in osteocalcin and BSAP, markers of formation.[151] In a randomized, double-blind clinical trial, the effect of DHEA on BMD in these patients was the same as that of ERT; both agents resulted in maintenance of BMD, but no significant gains, after controlling for weight gain.[151] However, bone turnover markers again revealed increased bone formation after DHEA, and a more significant suppression of bone resorption after ERT.[151] A subsequent randomized placebo-controlled trial showed an arrest of bone loss in older adolescents who received the combined therapy.[152] Because DHEA is unregulated by the FDA, it is potentially unsafe, depending on the purity of the preparation and dosing. For all of these reasons, DHEA cannot yet be recommended as a therapy for patients with bone loss secondary to AN.

The Female Athlete

Athletic participation has become increasingly common among adolescent girls since the enactment of Title IX of the US Educational Amendments Act in 1972, which required women's intercollegiate athletic offerings to be proportional to their representation in the student body. As noted previously, an active lifestyle has the potential for improved skeletal health. Unfortunately, some female athletes may develop patterns of unhealthy eating, irregular menstrual cycles, and an ensuing state of low estrogen levels, which can undo the beneficial effects of exercise on bone density.[136] In 1992, 20 years after Title IX, the American College of Sports Medicine (ACSM) coined the term "female athlete triad" to define this negative inter-relatedness of disordered eating, amenorrhea, and osteoporosis in certain female athletes.[153] Since most athletic women with menstrual irregularities do not meet criteria for osteoporosis nor frank eating disorders, the prevalence of the entire female athlete triad has been difficult to establish, although commonly cited references estimated that 3.4%–66% female athletes have experienced amenorrhea,[154,155] compared with only 2%–5% in the general population, and 15%–62% female college

athletes demonstrated disordered eating, compared with a 5%–10% prevalence of eating disorders in the general population.[156] A more complete epidemiologic characterization of athletic female adolescents conducted in the United States showed that while approximately one in five demonstrated any one of the components, only 1.2% met criteria for the full triad.[157]

Moreover, many active women with irregular menses consume adequate calories, leading some clinicians to use the term “athletic amenorrhea,” believing it is caused by the stress of exercise. Loucks et al. have disputed this concept, showing that low energy availability, defined as a failure to match food intake directly to exercise energy expenditure, not stress of exercise, alters luteinizing hormone (LH) pulse frequency and amplitude.[158] These disturbances in LH pulsatility can cause amenorrhea or more subtle menstrual dysfunction, including irregular, anovulatory, and short luteal phase cycles, all of which can cause a relative estrogen deficiency and increased bone resorption. Work with elite gymnasts and runners has shown that even within day, hourly energy deficits cause changes in body composition, with greater amplitude and frequency variations in energy balance correlating with increased body fat percentage.[159] The authors suggest that these wide swings also predispose to menstrual dysfunction.[159]

An alternative but complementary hypothesis is that cognitive dietary restraint, defined as the intention to limit food intake to control body weight, regardless of actual food limitation, is associated with higher levels of cortisol, which in turn impairs bone mass accretion by directly decreasing osteoblast activity and indirectly increasing osteoclast activity. In studies by McLean et al., exercising women with high dietary restraint measures by questionnaire had similar BMI and menstrual cycle length compared to those with low restraint, but higher urinary cortisol excretion[160] and lower total body bone mineral content.[161] In a group of younger female endurance athletes, elevated dietary restraint was similarly associated with decreased BMC and lumbar spine BMD without affecting menstrual regularity.[162] In any event, it is important to counsel adolescent female athletes to maintain positive energy balance throughout the day with frequent appropriate pre-, post- and between-exercise meals. Loucks’ work, in particular, suggests that a daily energy availability of approximately 30 kcal/kg in lean body mass may be required to protect against a catabolic state.[158]

Recognizing the spectrum of behaviors exhibited by female athletes, in

2007 ACSM refined the definition of the triad to encompass all possible relationships among energy availability, menstrual function, and bone health. This shift has removed the pathologic stigma of the female athlete triad, allowing it to include those active girls and women who achieve optimum bone health by maintaining adequate dietary intake to preserve normal menstrual cycles.[163]

CELIAC DISEASE

Celiac disease is a disorder of nutrient absorption in the small intestine, which affects as many as 1 in 250 people of European descent. In susceptible individuals, an immunologic reaction to gluten (a protein found in wheat, barley, and rye) leads to a chronic enteropathy of the proximal intestine. See Chapter 54: Celiac Disease for details.

Low BMD is a common complication of celiac disease, caused by general malnutrition secondary to anorexia (reduced dietary intake) and impaired absorption of nutrients, especially calcium and vitamin D,[164] among other etiologies. Vitamin D deficiency can be challenging to treat due to malabsorption in the setting of active, untreated celiac disease.

A gluten-free diet (GFD) is the mainstay of treatment for celiac disease, decreasing intestinal manifestations and improving BMD in adults and, in short-term trials, children. A long-term longitudinal study in children and adolescents showed that spinal BMD in patients with celiac disease was significantly lower than in controls at the time of diagnosis. After long-term GFD treatment (four years), patients had regained BMC and BMD to levels similar to controls, with significant “catch up” gains made in the first year of GFD treatment.[164] Provision of adequate calcium to these patients is critically important for bone mass.[165]

INFLAMMATORY BOWEL DISEASE

Similar to patients with celiac disease, adolescents with IBD are at risk for low BMD.[166,167] Patients with CD have a higher incidence of low bone mass than those with ulcerative colitis (UC),[168] likely related to the more widespread distribution of intestinal inflammation in the former versus the latter diagnosis. In addition to malabsorption of calcium, phosphate, and vitamin D, these patients have generalized malnutrition. Recent studies highlight the high prevalence of vitamin D deficiency among these patients, and the challenges encountered in trying to replete levels.[169,170]

They may also have delayed puberty, leading to inadequate levels of estrogen and/or testosterone to promote adequate bone accretion. The negative role of inflammation and proinflammatory/resorptive cytokines IL-1β, IL-6, and TNF-α on bone turnover and ultimately, bone density can be seen in pediatric patients with CD.[171,172]

Patients with IBD have relatively reduced lean body mass as a percentage of body composition, leading some to describe this deleterious combination of cytokines and malnutrition as "inflammatory cachexia."[173] Finally, extensive mucosal disease or surgical resection of the small bowel, particularly the terminal ileum, seen in CD further impairs calcium and vitamin D absorption. See also Chapter 53: Inflammatory Bowel Disease for more detail.

CYSTIC FIBROSIS

Patients with cystic fibrosis (CF) are at increased risk for significant bone disease. A review of the literature from 1988 to 1998 showed relative BMD decreases for patients with CF, with measured Z-scores ranging from –0.31 to –2.14 in studies with varying age populations and densitometry methods. This decrease in BMD translates into measurable increases in fracture rates and kyphosis (compression of the thoracic vertebrae) in CF patients aged 6 years and older.[174]

As with other chronic diseases, the pathophysiology of bone loss in these patients is felt to be multifactorial, with malabsorption of calcium and vitamin D, general malnutrition, inflammatory cytokines, and systemic corticosteroid use all playing roles. Vitamin D is a fat-soluble vitamin that relies on pancreatic enzymes for optimal absorption and reabsorption via enterohepatic circulation. Patients with CF have been shown to have a high prevalence of vitamin D deficiency, even after exogenous supplementation.[174] In addition, CF patients have decreased levels of IGF-I[175] and growth impairment, with height deficits as great as one standard deviation when compared with controls,[174] which impairs maximal bone mineral accretion and may warrant GH replacement. Pubertal delay and hypogonadism ensue, even in adulthood, leading to deficits in the estrogen and testosterone necessary to optimize the bone cycle. Low concentrations of IGF-I have also been directly correlated with decreased bone density.[176] An additional consideration in CF can be a lack of exercise tolerance in patients because of their respiratory disease. Since bones require mechanical loading for remodeling, lack of weight-bearing activity further

hinders normal bone mineral acquisition. See also Chapter 56: Exocrine Pancreatic Disease Including Cystic Fibrosis.

EVALUATION AND MANAGEMENT

Diagnostic Tools

Dual energy x-ray absorptiometry

DXA utilizes X-rays of two differing photon energies passing through the body to a sensitive detector. The beams are attenuated differentially depending on the tissues through which they pass. A computer then calculates body composition, BMC, and BMD based on the attenuation. DXA scanners report BMC in grams of mineralized calcium and BMD in grams per area of bone region in centimeters squared (g/cm^2). Advantages of DXA scanning include relatively rapid scanning times (approximately five minutes for each scan), low levels of radiation exposure (1/10th the radiation of a chest X-ray), and precision in the range of 1%. For all of these reasons, DXA can be used in children, with reliable normative databases consistently available, with some including children as young as 5 years.[12,128]

Limitations of DXA for Adolescents

The major limitation of DXA is that it utilizes a 2-dimensional technique to quantify a 3-dimensional structure. Density is, by definition, a measure of mass per unit volume. The BMD reported by DXA scanners is areal, or per unit area in gram per square centimeter. The significance of this calculation is more than academic, as the mechanical properties of bone are dependent on its architecture and geometry, as well as on its composition. Good mathematical models have been developed to calculate the approximate volumetric BMD—known as bone mineral apparent density (BMAD) in gram per cubic centimeter—although their prognostic value is not well established.[177]

These limitations of DXA are more significant in adolescents.[128] First, not all DXA scanners are equipped with the proper software to evaluate pediatric body composition and, therefore, BMD. Even comparing a single adolescent with him or herself on a single machine with reliable pediatric software is subject to flaws. Presumably, one could measure serial DXA scans over time to assess the effect of a disease state or treatment on BMD. Unfortunately, the growth of the child complicates interpretation. To illustrate, consider quantifying the density of a tree trunk by measuring the

shadow cast by the tree in the sun. After one year of growth, the tree trunk casts a wider (and perhaps darker) shadow, but the trunk itself may be no denser than it was previously. Using DXA, the "shadow" or areal BMD (aBMD) of this tree would be reported as greater than it was the previous year. The same phenomenon occurs when measuring a growing child's bones. The opposite phenomenon, that of a growing child's measured aBMD remaining constant, should therefore appear ominous to the clinician. Unfortunately, limitations on the actual scores reported for BMD do not allow the clinician to quantify the degree of risk. Fortunately, bone grows stronger as it increases in diameter, even at the same BMD.[178]

As mentioned, interpreting even reliable values for BMD is challenging. One standard deviation score, the T-score, which compares an individual adult's BMD to the mean peak BMD (or PBM) attained by young adults of the same sex, provides a valid estimate of the risk of fracture for postmenopausal adult women. A second standard deviation score, the Z-score, compares an individual's BMD to the mean BMD (in standard deviation units) for his or her age and sex, which is useful to grade patients with respect to their peers. The Z-score has limited prognostic significance, although a large prospective population-based cohort has begun to quantify a significantly increased risk of fracture associated with decreased BMD Z-scores starting at age 10.[179] Reliable pediatric databases report both T- and Z- scores, but the T- scores are not generally informative since most children and adolescents have not reached their peak young adult BMD and therefore may prove confusing to the reader of the report. For this reason, the ISCD has recommended that T-scores not appear on printouts or reports from DXA scans of children and adolescents younger than age 20 years.[128] In the same position statement, ISCD recommends the terms "low bone density for chronologic age" or "below the expected range for age" for Z-scores lower than –2.0 and discourages use of the term osteopenia entirely in this age group.[128]

Furthermore, as bone mass accretion is strongly influenced by Tanner stage, which does not directly correlate with chronologic age, comparing BMDs based on age alone (as in Z-scores) may be problematic. Finally, most databases include predominantly or exclusively Caucasian populations, which make the interpretation of BMD data more difficult for patients of different ethnic groups.[128]

Other Imaging Techniques

Quantitative computed tomography (QCT) is a technique of bone

densitometry, which is rapidly becoming the gold standard in research protocols because it can generate direct measures of bone volume, and thus a more accurate volumetric BMD. Pediatric algorithms are available, but the increased time required and significantly increased radiation doses limit its clinical use in this age group. Peripheral QCT (pQCT) is becoming increasingly used as a research tool in pediatric patients as it affords a measure of volumetric BMD in assessment of the appendicular skeleton, and provides insight into bone geometry and strength.[12,180] A final appeal of this methodology is that it involves a minimal radiation dose. Another technology, quantitative ultrasound (QUS), showed promise as it has no associated radiation exposure and demonstrates reasonable precision, but has proven difficult to validate in comparison with DXA and QCT.[181,182]

Serum markers

Bone turnover markers are used infrequently in the assessment of metabolic bone disease, primarily for research purposes, to gauge the relative contributions of bone formation versus bone resorption in the bone remodeling cycle.[183] Some endocrinologists also use them to monitor response to medical treatment. Serum alkaline phosphatase levels, particularly BSAP, reflect osteoblast function, as does serum osteocalcin, an osteoblast-specific product. Serum tartrate-resistant acid phosphatase (TRAP) is an osteoclast equivalent for BSAP, whereas collagen type I cross-linked *N*-telopeptides (NTx), which, as breakdown products of collagen, reflects osteoclast activity and can be measured in the urine. Adult studies have shown these markers to be inferior to bone densitometry for diagnosing osteoporosis and monitoring skeletal health, and pediatric reference ranges are more limited (compared to adults), as growth, puberty, and diurnal cycling cause wide variation in these values.[183]

Diagnostic Considerations

The conditions described above are only a few of those which predispose children and adolescents to osteoporosis (Table 66-1). The wide variety of conditions that impair skeletal health necessitates a high index of suspicion and heightened level of concern for bone health on the part of the pediatric health care or nutrition practitioner. The limitations of DXA scanning in pediatric populations detailed above have delayed its widespread use. Therefore, there is little evidence on which to make general

recommendations as to when and how frequently specific groups of children should have bone density measurement by DXA. The Cystic Fibrosis Foundation (CFF) first convened an expert panel in 2002 to develop practice guidelines for the management of bone disease in CF, which has served as a model for providers of other patient groups.[184] These guidelines include a screening and treatment protocol calling for a baseline DXA scan at the time of diagnosis of patients age 8 years and older. It may be reasonable to apply this recommendation and others from the CFF consensus statement, to patients with celiac disease, IBD, intestinal failure, and IJA, among other diagnoses. Standard practice in the authors' clinics is to perform DXA scans on girls and young women with AN after six months of hypothalamic amenorrhea. Bone density measurement is also recommended for patients with multiple fractures, regardless of etiology, pathologic or atraumatic fractures, and repeated long-bone stress fractures. A higher level of suspicion should also be raised in any patient with a strong family history of osteoporosis or with long-term use of systemic corticosteroids. DXA scans should then be repeated over time, but not more frequently than annually in clinical practice, as the changes noted over a shorter period are unlikely to be significant.[128]

Patients found to have a subnormal BMD on DXA scanning should have a screening evaluation of calcium homeostasis in addition to any clinically indicated laboratory assessments of their underlying medical conditions. A panel including serum calcium, phosphorus, magnesium, 25(OH)D, PTH, thyroid-stimulating hormone (TSH), and urinary spot calcium-to-creatinine ratio (normal < 0.2) would be sufficient. At present, it is generally agreed that a teenager with a BMD Z-score less than −2.0 warrants referral to a provider (e.g., endocrinologist) experienced in calcium metabolism and skeletal health. Consideration should be given to early referrals for patients with a BMD Z-score less than −1.0 if the adolescent has sustained recurrent fractures or a strong family history of documented osteoporotic fracture.

Management of Bone Diseases

Management of bone disease in patients with chronic illnesses described otherwise consists primarily of optimizing nutritional status, particularly with respect to calcium and vitamin D, and ensuring a normal hormonal milieu, with exogenous replacement of estrogen, testosterone, and growth hormone, as warranted. GFDs have already been mentioned for patients with celiac disease. Systemic glucocorticoids should be used judiciously,

but should not be withheld if that would jeopardize management of the underlying disease process. As with many decisions in clinical medicine, potential risks and benefits must be balanced for each individual case.

Novel Therapies

The uses of DHEA, IGF-I, and transdermal estrogen as new treatment modalities in patients with AN have already been discussed. The positive effects on BMD may make them attractive candidates for use in other diseases, particularly IGF-I for CF. However, persistent long-term improvements as well as solutions to the limitations mentioned above (nonstandard preparations for DHEA, parenteral administration for IGF-I) will need to be demonstrated before widespread use can be recommended.

Another class of agents being explored in several clinical trials of patients with AN[185] and some of the other abovementioned subgroups are the bisphosphonates. These agents are irreversible inhibitors of bone resorption, which become incorporated permanently into the mineralized bone matrix. These investigations have been prompted by the successful use of the bisphosphonate pamidronate in children with osteogenesis imperfecta (OI),[186] which represents a life-changing therapy for these children, as well as widespread experience with the bisphosphonate alendronate in postmenopausal women with osteoporosis. The CFF consensus statement includes the bisphosphonate alendronate as a therapeutic option in OI screening and treatment protocol.[184] However, concerns still exist around use of bisphosphonates in pediatrics, especially with respect to possible inhibition of primary remodeling of growing bone, increased risk of osteomalacia, long half-life in bone (>10 years), and unknown effects on the growing fetus. In addition, despite increases in BMD, there is concern that bone matrix, intercalated with bisphosphonate, may not provide normal mechanical strength. A case report of osteopetrosis (excessively dense and brittle bone) induced in an adolescent treated with very high-dose bisphosphonates highlights the caution with which these agents should be used.[187] Therefore, although there is promise to translate the established benefits of bisphosphonates in the elderly and younger patients with specific diagnoses to a wider pediatric population, these agents should currently be used extremely cautiously and/or in the setting of a controlled research study.

Finally, early recognition of the conditions that predispose to low bone density and early identification of patients at risk are essential to the prevention of bone loss in adolescents with chronic illnesses, which may be the most effective way to manage bone disease in these patients.

Prevention

In addition to managing the skeletal complications of chronic illnesses, one of the most important goals of adolescent nutrition is the potential prevention of osteoporosis by maximizing skeletal health in otherwise healthy teenagers. It is essential to ascertain family history of osteoporosis, especially early-onset osteoporosis, or frequent fractures. Dietary sources of calcium and vitamin D should be assessed and inadequacies addressed. Recommended age-appropriate daily calcium and vitamin D intake guidelines developed in the United States by the IOM[80] and endorsed by the American Academy of Pediatrics (AAP) have been highlighted in this chapter (see Table 66-2). Metabolic studies of calcium retention in adolescent girls[188,189] showed such large variation, however, that it might be necessary to individualize calcium requirements based on post-menarcheal age. Others have argued that sex and physical activity should also modify recommendations for optimal calcium intake.[190]

The success shown by a single supplementation trial in maintaining increases in BMD several years after discontinuation of concentrated milk protein as the primary calcium source may suggest the superiority of milk over other sources,[78] but further study is necessary to confirm this finding. Adolescent girls may need to be reassured that consuming low-fat dairy products instead of calcium supplements will not increase their weight or percent body fat. Indeed, evidence from one large study often cited in the lay press suggests that regular consumption of milk may be protective against weight gain.[191]

The AAP once again reaffirmed the importance of dairy products for bone health in a position statement that recommends the increasingly widely available "partially digested" forms of milk, cheese, and yogurt as the primary source of calcium and vitamin D for patients with lactose intolerance due to relative lactase deficiency.[192] Pretreatment with lactase enzyme replacement tablets were also recommended. For those patients with the most severe forms of lactose intolerance or those who will not consume adequate dairy products, there are other dietary sources of calcium (see Table 66-3), along with calcium supplements as a suitable alternative. Despite concerns to the contrary, chronic consumption of high levels of calcium has not been shown to impair absorption of such essential nutrients as iron,[193] magnesium, and zinc, as long as the intake is held below 2500 mg daily.[59]

TABLE 66-3 Examples of Dietary Sources of Calcium

Food	Serving Size	Calcium Content (mg)	No. of Servings to Equal Calcium Content in 1 Cup of Low-Fat Milk
Dairy foods			
Low-fat (1%) milk	1 cup (244 g)	264	—
Yogurt, nonfat, fruit variety	6 oz (170 g)	258	1.0
Frozen yogurt, vanilla, soft serve	½ cup (72 g)	103	2.6
Cheese	1 1-oz slice (28 g)	202	1.3
Cheese, pasteurized, processed	1 ¾-oz slice (21 g)	144	1.8
Nondairy foods			
Salmon, sockeye canned, drained, with bones	3 oz (85 g)	203	1.3
Tofu, firm, prepared with calcium sulfate and magnesium chloride	½ cup (126 g)	204	1.3
Broccoli, cooked	1 cup, chopped (156 g)	62	4.3
Collards, cooked, boiled, drained	1 cup, chopped (190 g)	266	1.0
Baked beans, canned	1 cup (253 g)	127	2.1
Foods fortified with calcium			
Calcium-fortified orange juice	1 cup (240 mL)	300	0.9
Selected fortified breakfast cereals	¾–1 cup (30 g)	100	2.6
Calcium-fortified soy milk	1 cup (240 mL)	200–500	0.5–1.3

Conclusions

The growing body of literature highlighted in this chapter should alert

clinicians in all disciplines to the adverse effects of chronic illnesses and inadequate nutrition on the skeletal health of adolescents. Most clinicians should find the solutions less obscure, however, as the promotion of skeletal health is ultimately no more than the promotion of good general health, including recommending balanced general nutrition, moderate weight-bearing exercise, and avoidance of alcohol and tobacco for all adolescents. The challenge lies in helping teenagers modify their behaviors to meet these goals.

REFERENCES

1. Bonjour JP, Thientz G, Buchs B, et al. Critical years and stages of puberty for spinal and femoral bone mass accumulation during adolescence. *J Clin Endocrinol Metab.* 1991;73:555–563.
2. Thientz G, Buchs G, Rizzoli R, et al. Longitudinal monitoring of bone mass accumulation in healthy adolescents: evidence for a marked reduction after 16 years of age at the levels of lumbar spine and femoral neck in female subjects. *J Clin Endocrinol Metab.* 1992;75:1060–1065.
3. National Osteoporosis Foundation. Learn about osteoporosis: Bone health basics [online report]. http://www.nof.org/learn/basics. Accessed January 7, 2013.
4. Blume SW, Curtis JR. Medical costs of osteoporosis in the elderly Medicare population. *Osteoporos Int.* 2011;22:1835–1844.
5. Robey PG, Boskey AL. The composition of bone. In: Rosen CJ, Compston JE, Lian JB, eds. *Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism*. 7th ed. Washington, DC: American Society for Bone and Mineral Research, 2008:32–38.
6. Krause C, de Gorter DJJ, Karperien M, ten Dijke P. Signal transduction cascades controlling osteoblast differentiation. In: Rosen CJ, Compston JE, Lian JB, eds. *Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism*. 7th ed. Washington, DC: American Society for Bone and Mineral Research, 2008:10–16.
7. Ross FP. Osteoclast biology and bone resorption. In: Rosen CJ, Compston JE, Lian JB, eds. *Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism*. 7th ed. Washington, DC: American Society for Bone and Mineral Research, 2008:16–22.
8. Monroe DG, McGee-Lawrence ME, Oursler MJ, Westendorf JJ. Update on Wnt signaling in bone cell biology and bone disease. *Gene.* 2012;492:1–18.
9. Southard RN, Morris JD, Mahan JD, et al. Bone mass in healthy children: measurement with quantitative DXA. *Radiology.* 1991;179:735–738.
10. McCormick DP, Ponder SW, Fawcett HD, Palmer JL. Spinal bone mineral density in 335 normal and obese children and adolescents: evidence for ethnic and sex differences. *J Bone Miner Res.* 1991;6:507–513.
11. Zemel BS, Kalkwarf HJ, Gilsanz V, et al. Revised reference curves for bone mineral content and areal bone mineral density according to age and sex for Black and non-Black children: results of the bone mineral density in childhood study. *J Clin Endocrinol Metab*. 2011;96(10):3160–3169.
12. Ma NS, Gordon CM. Pediatric osteoporosis: where are we now? *J Pediatr.* 2012;161(6):983–990.

13. Peterson CA, Eurell JAC, Erdman JW. Alterations in calcium intake on peak bone mass in the female rat. *J Bone Miner Res.* 1995;10:81–95.
14. Bailey DA, McKay HA, Mirwald RL, et al. A six-year longitudinal study of the relationship of physical activity to bone mineral accrural in growing children: the University of Saskatchewan Bone Mineral Accrural Study. *J Bone Miner Res.* 1999;14:1672–9.
15. Bachrach LK, Hastie T, Marcus R, et al. Bone mineral acquisition in healthy Asian, Hispanic, Black, and Caucasian youth: a longitudinal study. *J Clin Endocrinol Metab.* 1999;84:4702–4712.
16. Kalkwarf HJ, Zemel BS, Gilsanz V, et al. The bone mineral density in childhood study: bone mineral content and density according to age, sex, and race. *J Clin Endocrinol Metab*. 2007;92(6):2087–2099.
17. Wren TA, Shepherd JA, Kalkwarf HJ, et al. Racial disparity in fracture risk between white and nonwhite children in the United States. *J Pediatr.* 2012;161:1035–1040.
18. Gilsanz V, Roe TF, Mora S, et al. Changes in vertebral bone density in black girls and white girls during childhood and puberty. *New Engl J Med.* 1991;325:1597–1600.
19. Seeman E, Hopper JL, Bach LA, et al. Reduced bone mass in daughters of women with osteoporosis. *N Engl J Med.* 1989;320:554–558.
20. Pocock NA, Eisman JA, Hopper JL, et al. Genetic determinants of bone mass in adults. *J Clin Invest.* 1987;80:706–710.
21. Brown MA, Haughton MA, Grant SF, et al. Genetic control of bone density and turnover: role of the collagen 1alpha1, estrogen receptor, and vitamin D receptor genes. *J Bone Miner Res.* 2001;16:758–764.
22. Richards JB, Zheng HF, Spector TD. Genetics of osteoporosis from genome-wide association studies: advances and challenges. *Nat Rev Genet*. 2012;13(8):576–588.
23. Gerdhem P, Obrant KJ. Bone mineral density in old age: the influence of age at menarche and menopause. *J Bone Miner Metab.* 2004;22(4):372–375.
24. Riggs BL, Khosla S, Melton LJ. Sex steroids and the construction and conservation of the adult skeleton. *Endocr Rev.* 2002;23:279–302.
25. Bass S, Delmas PD, Pearce G, et al. The differing tempo of growth in bone size, mass and density in girls is region-specific. *J Clin Invest.* 1999;104:795–804.
26. Cameron N, Tanner JM, Whitehouse RH. A longitudinal analysis of the growth of limb segments in adolescence. *Ann Hum Biol.* 1982;9:211–220.
27. Seeman E, Karlsson MK, Duan Y. On exposure to anorexia nervosa, the temporal variation in axial and appendicular skeletal development predisposes to site-specific deficits in bone size and density: a cross-sectional study. *J Bone Miner Res.* 2000;15:2259–2265.
28. Buchanan JR, Myers C, Lloyd T, et al. Determinants of peak trabecular density in women: the role of androgens, estrogen and exercise. *J Bone Miner Res.* 1988;3:673–680.
29. Tracz MJ, Sideras K, Bolona ER, et al. Testosterone use in men and its effects on bone health. A systematic review and meta-analysis of randomized placebo-controlled trials. *J Clin Endocrinol Metab.* 2006;91:2011–2016.
30. Rogol AD. Pubertal androgen therapy in boys. *Pediatr Endocrinol Rev.* 2005;2(3):383–390.
31. Munoz-Torres M, Jodar E, Quesada M, Escobar-Jiminez F. Bone mass in androgen-insensitivity syndrome: response to hormonal replacement therapy. *Calcif Tissue Int.* 1995;57:94–96.
32. Marcus R, Leary D, Schneider DL, et al. The contribution of testosterone to

skeletal development and maintenance: lessons from the androgen insensitivity syndrome. *J Clin Endocrinol Metab.* 2000;85:1032–1037.
33. Bertelloni S, Baroncelli GI, Federico G, et al. Altered bone mineral density in patients with complete androgen insensitivity syndrome. *Horm Res.* 1998;50:309–314.
34. Buchanan JR, Hospodar P, Myers C, et al. Effect of excess endogenous androgens on bone density in young women. *J Clin Endocrinol Metab.* 1988;67;937–943.
35. Good C, Tulchinsky M, Mauger D, et al. Bone mineral density and body composition in lean women with polycystic ovary syndrome. *Fertil Steril.* 1999;72:21–25.
36. Shore RM, Chesney RW, Mazess RB, et al. Bone mineral status in growth hormone deficiency. *J Pediatr.* 1980;96:393–396.
37. Boot AM. Body composition and bone mineral density in adolescents with partial growth hormone deficiency. *J Clin Endocrinol Metab.* 2003;88:5099–5100.
38. Boot AM, Engels MA, Boerma GJ, et al. Changes in bone mineral density, body composition, and lipid metabolism during growth hormone (GH) treatment in children with GH deficiency. *J Clin Endocrinol Metab.* 1997;82:2423–2428.
39. Baroncelli GL, Bertelloni S, Sodini F, Saggese G. Acquisition of bone mass in normal individuals and in patients with growth hormone deficiency. *J Pediatr Endocrinol Metab.* 2003;16(suppl 2):327–335.
40. Ferretti JL. On new opportunities for absorptiometry. *J Clin Densitom.* 1998;1:41–53.
41. Kohrt WM, Bloomfield SA, Little KD, et al. American College of Sports Medicine Position Stand: physical activity and bone health. *Med Sci Sports Exerc.* 2004;36:1985–1996.
42. Marcus R, Cann C, Madvig P, et al. Menstrual function and bone mass in elite women distance runners: endocrine and metabolic features. *Ann Intern Med.* 1985;102:158–163.
43. Gordon CM, Goodman E, Emans SJ, et al. Physiologic regulators of bone turnover in young women with anorexia nervosa. *J Pediatr.* 2002;141:64–70.
44. Taafe DR, Snow-Harter C, Connolly DA, et al. Differential effects of swimming versus weight-bearing activity on bone mineral status of eumenorrheic athletes. *J Bone Miner Res.* 1995;10:586–593.
45. Duncan CS, Blimkie CJ, Cowell CT, et al. Bone mineral density in adolescent female athletes: relationship to exercise type and muscle strength. *Med Sci Sports Exerc.* 2002;34:286–294.
46. Lariviere JA, Robinson TL, Snow CM. Spine bone mineral density increases in experienced but not novice collegiate female rowers. *Med Sci Sports Exerc*. 2003;35:1740–1744.
47. Hind K, Burrows M. Weight-bearing exercise and bone mineral accrual in children and adolescents: a review of controlled trials. *Bone.* 2007;40:14–27.
48. Strong WB, Malina RM, Blimkie CJR, et al. Evidence based physical activity for school-age youth. *J Pediatr.* 2005;146:732–737.
49. Berstein CN, Seeger LL, Sayre JW, et al. Decreased bone density in inflammatory bowel disease is related to corticosteroid use and not disease diagnosis. *J Bone Miner Res.* 1995;10:250–256.
50. Israel E, Banerjee TR, LeBoff MS, et al. Effects of inhaled glucocorticoids on bone density in premenopausal women. *New Engl J Med.* 2001;345:941–947.
51. Leonard MB, Feldman HI, Shults J. Long-term, high-dose glucocorticoids and bone mineral content in childhood glucocorticoid-sensitive nephrotic syndrome. *N Engl J Med.* 2004;351:868–875.

52. Bernstein CN, Leslie WD. The pathophysiology of bone disease in gastrointestinal disease. *Eur J Gastroenterol Hepatol.* 2003;15:857–864.
53. Blum M, Harris SS, Must A, et al. Weight and body mass index at menarche are associated with premenopausal bone mass. *Osteoporos Int.* 2001;12:588–594.
54. Goulding A, Grant AM, Williams SM. Bone and body composition of children and adolescents with repeated forearm fractures. *J Bone Miner Res.* 2005;20:2090–2096.
55. Goulding A, Taylor RW, Jones IE, et al. Overweight and obese children have low bone mass and area for their weight. *Int J Obes Relat Metab Disord.* 2000;24:627–632.
56. El Hage R, Moussa E, Jacob C. Bone mineral content and density in obese, overweight, and normal-weighted sedentary adolescent girls. *J Adol Health.* 2010;47:591–595.
57. Russell M, Mendes N, et al. Visceral fat is a negative predictor of bone density measures in obese adolescent girls. *J Clin Endocrinol Metab.* 2010;95:1247–1255.
58. Russell M, Misra M. Influence of ghrelin and adipocytokines on bone mineral density in adolescent female athletes with amenorrhea and eumenorrheic athletes. *Med Sport Sci.* 2010;55:103–113.
59. Weaver CM, Peacock M, Johnston CC. Adolescent nutrition in the prevention of postmenopausal osteoporosis. *J Clin Endo Metab.* 1999;84:1839–1843.
60. Cadogan J, Blumsehn A, Barker M, Eastell R. A longitudinal study of bone gain in pubertal girls: anthropometric and biochemical correlates. *J Bone Miner Res.* 1998;13:1602–1612.
61. Marwaha RK, Puri S, Tandon N, et al. Effects of sports training & nutrition on bone mineral density in young Indian healthy females. *Indian J Med Res.* 2011;134:307–313.
62. Burrows M, Nevill AM, Bird S, et al. Physiological factors associated with low bone mineral density in female endurance runners. *Br J Sports Med.* 2003;37:67–71.
63. Greer FR, Krebs NF, the Committee on Nutrition, American Academy of Pediatrics. Optimizing bone health and calcium intakes of infants, children and adolescents. *Pediatrics.* 2006;117:578–585.
64. Heaney, RP. Calcium in the prevention and treatment of osteoporosis. *J Intern Med.* 1992;231:169–180.
65. Kalkwarf HJ, Khoury JC, Lamphear BP. Milk intake during childhood and adolescence, adult bone density, and osteoporotic fractures in US women. *Am J Clin Nut.* 2003;77:257–265.
66. Baoshe A, Matkovic V. Relation of nutrition, body composition, and physical activity to skeletal development: a cross-sectional study in preadolescent females. *J Am Coll Nutr.* 1998;17:136–147.
67. Ruiz JC, Mandel C, Garabedian M. Influence of spontaneous calcium intake and physical exercise on the vertebral and femoral bone mineral density of children and adolescents. *J Bone Miner Res.* 1995;10:675–682.
68. Boot AM, de Ridder MA, Pols HA, et al. Bone mineral density in children and adolescents: relation to puberty, calcium intake, and physical activity. *J Clin Endocrinol Metab.* 1997;82:57–62.
69. Gunnes M, Lehmann EH. Physical activity and dietary constituents as predictors of forearm cortical and trabecular bone gain in healthy children and adolescents: a prospective study. *Acta Pediatr.* 1996;85:19–25.
70. Lloyd T, Andon MB, Rollings N, et al. Calcium supplementation and bone mineral density in adolescent girls. *JAMA.* 1993;270:841–844.

71. Chan GM, Hoffman K, McMurry M. Effects of dairy products on bone and body composition in pubertal girls. *J Pediatr.* 1995;126:551–556.
72. Cadogan J, Eastell R, Jones N, Barker ME. Milk intake and bone mineral acquisition in adolescent girls: randomized, controlled intervention trial. *BMJ.* 1997;315:1255–1260.
73. Lee WT, Leung SS, Leung DM, et al. A randomized double-blind controlled calcium supplementation trial, and bone and height acquisition in children. *Br J Nutr.* 1995;74:125–139.
74. Johnston CC, Miller JZ, Slemenda CW, et al. Calcium supplementation and increases in bone mineral density in children. *N Engl J Med.* 1992;327:82–87.
75. Nowson CA, Green RM, Hopper JL, et al. A co-twin study of the effect of calcium supplementation on bone density during adolescence. *Osteoporos Int.* 1997;7:219–225.
76. Bonjour JP, Carrie AL, Ferrari S, et al. Calcium-enriched foods and bone mass growth in prepubertal girls: a randomized, double-blind, placebo-controlled trial. *J Clin Invest.* 1997;99:1287–1294.
77. Lanou AJ, Berkow SE, Barnard ND. Calcium, dairy products and bone health in children and young adults: a reevaluation of the evidence. *Pediatrics.* 2005;115:736–743.
78. Bonjour JP, Chevalley T, Ammann P, et al. Gain in bone mineral mass in prepubertal girls 3.5 years after discontinuation of calcium supplementation: a follow-up study. *Lancet.* 2001;358:1208–1212.
79. Specker BL. Evidence for an interaction between calcium intake and physical activity on changes in bone mineral density. *J Bone Miner Res.* 1996;11:1539–1544.
80. Institute of Medicine. *Dietary Reference Intakes for Calcium and Vitamin D.* Washington, DC: The National Academies Press; 2011.
81. Bolland MJ, Barber PA, et al. Vascular events in healthy older women receiving calcium supplementation: randomised controlled trial. *BMJ.* 2008;336(7638):262–266.
82. Bolland MJ, Avenell A, Baron JA, et al. Effects of calcium supplements on risks of myocardial infarction and cardiovascular events: meta-analysis. *BMJ.* 2010;341:c3691.
83. Reid IR, Mason B, Horne A, et al. Randomized controlled trial of calcium in healthy older women. *Am J Med.* 2006;119:777–785.
84. Holick MF. Vitamin D deficiency. *N Engl J Med.* 2007;357:266–281.
85. Lips P. Vitamin D deficiency and osteoporosis: the role of vitamin D deficiency and treatment with vitamin D analogues in the prevention of osteoporosis-related fractures. *Eur J Clin Invest.* 1996;26:436–442.
86. Krall EA, Dawson-Hughes B. Relation of fractional 47-calcium retention to season and rates of bone loss in healthy postmenopausal women. *J Bone Miner Res.* 1991;6:1323–1329.
87. Chapuy MC, Arlot ME, DuBoef F, et al. Vitamin D3 and calcium to prevent hip fractures in elderly women. *N Engl J Med.* 1992;327:1637–1642.
88. Dawson-Hughes B, Harris SS, Krall EA, Dallal GE. Effect of calcium and vitamin D supplementation on bone density in men and women 65 years of age and older. *N Engl J Med.* 1997;337:670–676.
89. Gordon CM, DePeter KC, Feldman HA, et al. Prevalence of vitamin D deficiency among healthy adolescents. *Arch Pediatr Adolesc Med.* 2004;158:531–537.
90. Parfit AM, Gallagher JC, Heaney RP, et al. Vitamin D and bone health in the elderly. *Am J Clin Nutr.* 1982;32:1014–1031.

91. Webb AR, Kline L, Holick MF. Influence of season and latitude on the cutaneous synthesis of vitamin D3: exposure to winter sunlight in Boston and Edmonton will not promote vitamin D3 synthesis in human skin. *J Clin Endocrinol Metab.* 1988;67:373–378.
92. Krall EA, Sahyoun N, Tannenbaum S, et al. Effect of vitamin D intake on seasonal variation in parathyroid hormones secretion in postmenopausal women. *N Engl J Med.* 1989;321:1777–1783.
93. Ala-Houhala M, Parviainen MT, Pyykko K, Visakorpi JC. Serum 25-hydroxyvitamin D levels in Finnish children aged 2-17 years. *Acta Pediatr Scand.* 1984;73:232–236.
94. Juttman JR, Visser TJ, Buurman C, et al. Seasonal fluctuations in serum concentrations of vitamin D metabolites in normal patients. *BMJ*. 1981;282:1349–1352.
95. Landin-Wilhelmsen K, Wilhelmsern L, Wilske J, et al. Sunlight increases serum 25-OH vitamin D concentration whereas 25(OH)D3 is unaffected: results from a general population study in Goteborg, Sweden. *Europ J Clin Nutr*. 1995;49:400–407.
96. Poskitt EME, Cole TJ, Lawson DEM. Diet, sunlight, and 25-hydroxyvitamin D in healthy children and adults. *BMJ*. 1979;1:221–223.
97. Vik T, Try K, Srtomme JH. The vitamin D status of man at 70 degrees north. *Scan J Clin Lab Invest.* 1980;80:227–232.
98. Aitken JM, Anderson JB, Horton PW. Seasonal variations in bone mineral content after the menopause. *Nature.* 1973;241:59–60.
99. Krolner B. Seasonal variation of lumbar spine bone mineral content in normal women. *Calcif Tissue Int.* 1983;35:145–147.
100. Vanderschueren D, Gevers G, Dequeucker J, et al. Seasonal variation in bone metabolism in young healthy subjects. *Calcif Tissue Int.* 1991;49:84–89.
101. Dawson-Hughes B, Dallal GE, Krall EA, et al. Effect of vitamin D supplementation on wintertime and overall bone loss in healthy postmenopausal women. *Ann Int Med.* 1991;115:505–512.
102. Overgaard K, Johansen NL, Christiansen C. Lack of seasonal variation in bone mass and biochemical estimates of bone turnover. *Bone.* 1988;9:285–288.
103. Thomas MK, Lloyd-Jones DM, Thadhani RI, et al. Hypovitaminosis D in medical inpatients. *N Engl J Med.* 1998;338:777–783.
104. Guillemant J, Allemandou A, Cabrol S, et al. Vitamin D status in the adolescent: seasonal variations and effects of winter supplementation with vitamin D3. *Arch Pediatr*. 1998;5:1211–1215.
105. Docio S, Riancho JA, Perez A, et al. Seasonal deficiency of vitamin D in children: a potential target for osteoporosis-preventing strategies? *J Bone Miner Res.* 1998;13:554–558.
106. Harkness LS, Cromer BA. Vitamin D deficiency in adolescent females. *J Adolesc Health.* 2005;37:75.
107. Sullivan SS, Rosen CJ, Halteman WA. Adolescent girls in Maine are at risk for vitamin D insufficiency. *J Am Diet Assoc*. 2005;105(6):971–974.
108. Slemenda CW, Peacock M, Hui S, et al. Reduced rates of skeletal remodeling are associated with increased bone mineral density during the development of peak bone mass. *J Bone Miner Res.* 1997;12:676–682.
109. Ferrari SL, Rizzoli R, Slosman DO, Bonjour JP. Do dietary calcium and age explain the controversy surrounding the relationship between bone mineral density and vitamin D receptor gene polymorphisms? *J Bone Miner Res.* 1998;13:363–370.

110. Holick MF, Binkley NC, Bischoff-Ferrari HA, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab.* 2011;96:1911–1930.
111. Sayers A, Fraser WD, Lawlor DA, Tobias JH. 25-Hydroxyvitamin-D(3) levels are positively related to subsequent cortical bone development in childhood: findings from a large prospective cohort study. *Osteoporos Int.* 2012;23:2117–2128.
112. Massey LK, Whiting SJ. Dietary salt, urinary calcium, and bone loss. *J Bone Miner Res.* 1996;11:731–736.
113. Harel Z, Riggs S, Menzies G, et al. Adolescents and calcium: what they do and do not know and how much they consume. *J Adol Health.* 1998;22:225–228.
114. Albertson AM, Tobelmann RC, Marquart L. Estimated dietary calcium intake and food sources for adolescent females: 1980-1992. *J Adolesc Health.* 1997;20:20–26.
115. Bailey RL, Dodd KW, et al. Estimation of total usual calcium and vitamin D intakes in the United States. *J Nutrition.* 2010;140:817–822.
116. Wyshak G, Frisch RE. Carbonated beverages, dietary calcium, the dietary calcium/phosphorous ration, and bone fractures in girls and boys. *J Adol Health.* 1994;15:210–215.
117. Wyshak G. Teenaged girls, carbonated beverage consumption, and bone fractures. *Arch Pediatr Adolesc Med.* 2000;154:610–613.
118. McGartland C, Robson PJ, Murray L, et al. Carbonated soft drink consumption and bone mineral density in adolescence: the Northern Ireland Young Hearts project. *J Bone Miner Res*. 2003;18:1563–1569.
119. Amato D, Araujo F, Paniaqua R. The soda debate fizzes on. *J Bone Miner Res*. 2004;19:521.
120. Putnam JJ, Allhouse JE. *Food Consumption, Prices and Expenditures, 1970-1997*. Washington, DC: Food and Economics Division, Economic Research Service, US Department of Agriculture; 1999. Statistical Bulletin 965.
121. Need AG, Kemp A, Giles N, et al. Relationships between intestinal calcium absorption, serum vitamin D metabolites and smoking in postmenopausal women. *Osteoporos Int.* 2002;13:83–88.
122. Dorn LD, Pabst S, et al. Bone mass, depressive, and anxiety symptoms in adolescent girls: variation by smoking and alcohol use. *J Adol Health*. 2011;49:498–504.
123. Hill KM, McCabe GP, McCabe LD, et al. An inflection point of serum 25-hydroxyvitamin D for maximal suppression of parathyroid hormone is not evident from multi-site pooled data in children and adolescents. *J Nutr.* 2010;140:1983–1988.
124. Armas LA, Hollis BW, Heaney RP. Vitamin D2 is much less effective than vitamin D3 in humans. *J Clin Endocrinol Metab.* 2004;89(11):5387–5391.
125. Houghton LA, Vieth R. The case against ergocalciferol (vitamin D2) as a vitamin supplement. *Am J Clin Nutr*. 2006;84(4):694–697.
126. Consensus Development Conference. Diagnosis, prophylaxis, and treatment of osteoporosis. *Am J Med.* 1993;90:646–650.
127. Leib ES, Writing Group for the ISCD Position Development Conference. Diagnosis of osteoporosis in men, premenopausal women, and children. *J Clin Densitom*. 2004;7:17–26.
128. Gordon CM, Bachrach LK, Carpenter TO, et al. Dual energy X-ray absorptiometry interpretation and reporting in children and adolescents: the 2007 ISCD Pediatric Official Positions. *J Clin Densitom.* 2008;11:43–58.
129. Bachrach LK, Guido D, Katzman D, et al. Decreased bone density in adolescent

girls with anorexia nervosa. *Pediatrics.* 1990;86:440–7.
130. Rigotti NA, Neer RM, Skates SJ, et al. The clinical course of osteoporosis in anorexia nervosa. *JAMA.* 1991;265:1133–1138.
131. Soyka LA, Grinspoon S, Levitsky L, et al. The effects of anorexia nervosa on bone metabolism in female adolescents. *J Clin Endocrinol Metab.* 1999;84:4489–4496.
132. Castro J, Lazaro L, Pons F, et al. Predictors of bone mineral density reduction in adolescents with anorexia nervosa. *J Am Acad Child Adolesc Psychiatry.* 2000;39:1365–1370.
133. Bredella MA, Fazeli PK, Miller K, et al. Increased bone marrow fat in anorexia nervosa. *J Clin Endocrinol Metab.* 2009;94:2129–2136.
134. Ecklund K, Vajapeyam S, Feldman HA, et al. Bone marrow changes in adolescent girls with anorexia nervosa. *J Bone Miner Res.* 2010;25:298–304.
135. Misra M, Katzman DK, Cord J, et al. Bone metabolism in adolescent boys with anorexia nervosa. *J Clin Endocrinol Metab.* 2008;93:3029–3036.
136. Gordon CM. Functional hypothalamic amenorrhea. *N Engl J Med.* 2010;363:365–371.
137. Seeman E, Szmuckler GI, Formica C, et al. Osteoporosis in anorexia nervosa: the influence of peak bone density, bone loss, oral contraceptive use and exercise. *J Bone Miner Res.* 1992;7:1267–1274.
138. Hergenroeder AC, Smith EO, Shypailo R, et al. Bone mineral changes in young women with hypothalamic amenorrhea treated with oral contraceptives, medroxy-progesterone, or placebo over 12 months. *Am J Ob Gyn.* 1997;176:1017–1025.
139. Misra M, Katzman D, Miller KK, et al. Physiologic estrogen replacement increases bone density in adolescent girls with anorexia nervosa. *J Bone Miner Res.* 2011;26:2430–2438.
140. Klibanski A, Biller BM, Schoenfeld DA, et al. The effects of estrogen administration on trabecular bone loss in young women with anorexia nervosa. *J Clin Endocrinol Metab.* 1995;80:898–904.
141. Misra M, Prabhakaran R, Miller KK, et al. Prognostic indicators of changes in bone density measures in adolescent girls with anorexia nervosa. *J Clin Endocrinol Metab.* 2008;93:1292–1297.
142. Bachrach LK, Katzman DK, Litt IF, et al. Recovery from osteopenia in adolescent girls with anorexia nervosa. *J Clin Endocrinol Metab.* 1991;72:602–606.
143. Stefanis N, MacIntosh C, Abaha HD, et al. Dissociation of bone turnover in anorexia nervosa. *Ann Clin Biochem.* 1998;35:709–716.
144. Caillot-Augusseau A, Lafage-Prous MH, Margaillan P, et al. Weight gain reverses bone turnover and restores circadian variation of bone resorption in anorexic patients. *Clin Endocrinol (Oxf).* 2000;52:113–121.
145. Grinspoon S, Miller K, Coyle C, et al. Severity of osteopenia in estrogen-deficient women with anorexia nervosa and hypothalamic amenorrhea. *J Clin Endocrinol Metab.* 1999;84:2049–2055.
146. Grinspoon S, Thomas L, Miller K, et al. Effects of recombinant human IGF-I and oral contraceptive administration on bone density in anorexia nervosa. *J Clin Endocrinol Metab.* 2002;87:2883–2891.
147. Nicholas V, Prwett A, Bettica P, et al. Age-related decreases in IGF-I and TGF-beta in femoral cortical bone from both men and women: implications for bone loss with aging. *J Clin Endocrinol Metab.* 1994;78:1011–1016.
148. Haden ST, Glowacki J, Hurwitz S, et al. Effects of age and serum dehydroepiandrosterone sulfate, IGF-I and IL-6 levels in women. *Calcif Tissue Int.* 2000;66:414–418.

149. Meikle AW, Daynes RA, Aranco BA. Adrenal androgen secretion and biologic effects. *Endocri Metab Clin North Am.* 1991;20:381–400.
150. Labrie F, Diamond R, Cusan L, et al. Marked decline in serum concentrations of adrenal C19 sex steroid precursors and conjugated androgen metabolites during aging. *J Clin Endocrinol Metab*. 1997;82:2396–2402.
151. Gordon CM, Grace E, Eman S, et al. Effects of oral DHEA on bone density in young women with anorexia nervosa: a randomized trial. *J Clin Endocrinol Metab.* 2002;87:4935–4941.
152. DiVasta AD, Feldman HA, Giancaterino C, et al. The effect of gonadal and adrenal steroid therapy on skeletal health in adolescents and young women with anorexia nervosa. *Metabolism.* 2012;61:1010–1020.
153. Otis CL, Drinkwater B, Johnson M, et al. American College of Sports Medicine Position Stand: The Female Athlete Triad. *Med Sci Sports Exerc.* 1997;29:i–ix.
154. Feicht CB, Johnson TS, Martin BJ, et al. Secondary amenorrhea in athletes. *Lancet.* 1978;2:1145–1146.
155. Shangold M, Rebar RW, Wentz AC, et al. Evaluation and management of menstrual dysfunction in athletes. *JAMA.* 1990;263:1665–1669.
156. Warren BJ, Stanton AL, Blessing DL. Disordered eating patterns in competitive female athletes. *Int J Eat Disord.* 1990;9:565.
157. Nichols JF, Rauh MJ, Lawson MJ, et al. Prevalence of the female athlete triad syndrome among high school athletes. *Arch Pediatr Adolesc Med.* 2006;160:137–142.
158. Loucks AB, Verdun M, Heath EM. Low energy availability, not stress of exercise, alters LH pulsatility in exercising women. *J Appl Physiol.* 1998;84:37–46.
159. Deutz RC, Bernadot D, Martin DE, Cody MM. Relationship between energy deficits and body composition in elite female gymnasts and runners. *Med Sci Sports Exerc.* 2000;32:659–668.
160. McLean JA, Barr SI, Prior JC. Cognitive dietary restraint is associated with higher urinary cortisol excretion in healthy premenopausal women. *Am J Clin Nutr.* 2001;73:7–12.
161. McLean JA, Barr SI, Prior JC. Dietary restraint, exercise, and bone density in young women: are they related? *Med Sci Sports Exerc.* 2001;33:1292–1296.
162. Barrack MT, Rauh MJ, Barkai HS, et al. Dietary restraint and low bone mass in female adolescent endurance runners. *Am J Clin Nutr*. 2008;87:36–43.
163. Nattiv A, et al. American College of Sports Medicine position stand: the female athlete triad. *Med Sci Sports Exerc.* 2007;39:1867–1882.
164. Mora S, Barera G, Beccio S, et al. A prospective, longitudinal study of the long-term effect of treatment on bone density in children with celiac disease. *J Pediatr.* 2001;139:516–521.
165. Pazianas M, Butcher GP, Subhani JM, et al. Calcium absorption and bone mineral density in celiacs after long term treatment with gluten-free diet and adequate calcium intake. *Osteoporos Int.* 2005;16:56–63.
166. Burnham JM, Shults J, Semeao E, et al. Whole body BMC in pediatric Crohn disease: independent effects of altered growth, maturation, and body composition. *J Bone Miner Res.* 2004;19:1961–1968.
167. Sylvester FA. IBD and skeletal health: children are not small adults! *Inflamm Bowel Dis.* 2005;11:1020–1023.
168. Ulivieri FM, Piodi LP, Taioli E, et al. Bone mineral density and body composition in ulcerative colitis: a six-year follow-up. *Osteoporos Int.* 2001;12:343–348.
169. Pappa HM, Mitchell PD, Jiang H, et al. Treatment of vitamin D insufficiency in children and adolescents with inflammatory bowel disease: a randomized clinical

trial comparing three regimens. *J Clin Endocrinol Metab.* 2012;97:2134–2142.
170. Pappa HM, Langereis EJ, Grand RJ, Gordon CM. Prevalence and risk factors for hypovitaminosis D in young patients with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr.* 2011;53:361–364.
171. Pappa H, Thayu M, Sylvester F, et al. Skeletal health of children and adolescents with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr.* 2011;53:11–25.
172. Sylvester FA, Wyzga N, Hyams JS, et al. Natural history of bone metabolism and bone mineral density in children with inflammatory bowel disease. *Inflamm Bowel Dis*. 2007;13:42–50.
173. Burnham JM, Shults J, Semeao E, et al. Body-composition alterations consistent with cachexia in children and young adults with Crohn disease. *Am J Clin Nutr.* 2005;82:413–420.
174. Ott SM, Aitken ML. Osteoporosis in patients with cystic fibrosis. *Clin Chest Med.* 1998;19:555–567.
175. Arumugam R, LeBlanc A, Seilheimer DK, Hardin DS. Serum leptin and IGF-I levels in cystic fibrosis. *Endocr Res.* 1998;24:247–257.
176. Gordon CM, Binello E, LeBoff MS, et al. Relationship between insulin-like growth factor I, dehydroepiandrosterone sulfate and proresorptive cytokines and bone density in cystic fibrosis. *Osteoporos Int.* 2006;17:783–790.
177. Carter DR. Bouxsein ML, Marcus R. New approaches for interpreting projected bone densitometry data. *J Bone Miner Res.* 1992;7:137–145.
178. S Judex. *Exercise and Prevention of Osteoporosis Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism*. 7th ed. Washington, DC: American Society for Bone and Mineral Research; 2008:227–231.
179. Clark EM, Ness AR, Bishop NJ, Tobias JH. Association between bone mass and fractures in children: a prospective cohort study. *J Bone Mineral Res.* 2006;21:1489–1495.
180. Rauch F, Schonau E. Peripheral quantitative computed tomography of the distal radius in young subjects new reference data and interpretation of results. *J Musculoskelet Neuronal Interact.* 2005;5:99–126.
181. Specker BL, Schoenau E. Quantitative bone analysis in children: current methods and recommendations. *J Pediatr.* 2005;146:726–731.
182. Pettinato AA, Loud KJ, Bristol SK, et al. Effects of nutrition, puberty, and gender on bone ultrasound measurements in adolescents and young adults. *J Adol Health.* 2006;39:828–834.
183. Camacho P, Kleerekoper M. Biochemical markers of bone turnover. In: Favus MJ, ed. *Primer on the Metabolic Bone Diseases and Disorders of Mineral Metabolism*. 6th ed. Durham, NC: American Society for Bone and Mineral Research; 2006:36–42.
184. Aris RM, Merkel PA, Bachrach LK, et al. Consensus statement: guide to bone health and disease in cystic fibrosis. *J Clin Endocrinol Metab.* 2005;90:1888–1896.
185. Golden NH, Iglesias EA, Jacobson MS, et al. Alendronate for the treatment of osteopenia in anorexia nervosa: a randomized, double-blind, placebo-controlled trial. *J Clin Endocrinol Metab.* 2005;90:3179–3185.
186. Glorieux F, Bishop N, Poltoon H, et al. Cyclic administration of pamidronate in children with severe osteogenesis imperfecta. *N Engl J Med.* 1998;339:947–952.
187. Whyte MP, Wenkert D, Clements KL, et al. Bisphosphonate-induced osteopetrosis. *N Engl J Med.* 2003;349(5):457–463.
188. Jackman LA, Millane SS, Martin BR, et al. Calcium retention in relation to calcium intake and postmenarcheal age in adolescent females. *Am J Clin Nutr.*

1997;66:327–333.
189. Weaver CM, Peacock M, Martin BR, et al. Calcium retention estimated from indicators of skeletal status in adolescent girls and young women. *Am J Clin Nutr.* 1996;64:67–70.
190. Anderson JJ. Calcium requirements during adolescence to maximize bone health. *J Am Coll Nutr.* 2001;20(suppl 2):186S–191S.
191. Pereira MA, Jacobs DR Jr, Van Horn L, et al. Dairy consumption, obesity, and the insulin resistance syndrome in young adults: the CARDIA Study. *JAMA.* 2002;287(16):2081–2089.
192. Heyman MB, Committee on Nutrition. Lactose intolerance in infants, children, and adolescents. *Pediatrics.* 2006;118;1279–1286.
193. Abrams SA, Griffin IJ, Davila P, Liang L. Calcium fortification of breakfast cereal enhances calcium absorption in children without affecting iron absorption. *J Pediatr.* 2001;139:522–526.
194. Loud KJ, Gordon CM. Bone and nutrition in health and disease. *Int Semin Pediatr Gastroenterol Nutr.* 2002;11(2):1–7.

Enteral Nutrition and Formulas

67

Zoë McCallum, MBBS, FRACP and Julie E. Bines, MBBS, FRACP, MD

Enteral nutrition refers to both the delivery of nutrients via a feeding tube directly into the stomach or intestine and the provision of oral nutritional supplementation. The concept of enteral nutrition support is not new. In ancient Egypt, and later in Greece, feeds were introduced into the rectum, and in the nineteenth century, rudimentary tubes were used to infuse basic foods such as broths, eggs, milk, and even alcohol into the esophagus and stomach.[1] Since the 1980s enteral nutrition has been increasingly recognized as a safe, effective, and cost-efficient method of providing nutrition to patients with a functional gut. The last three decades have seen advances in the formulation of nutritionally complete and disease-specific formulas as well as improvements in feeding access devices.[2] Patients who had previously received parenteral nutrition can now be successfully managed with enteral nutrition alone or in combination with parenteral nutrition. Home enteral nutrition support is an important adjunct to the management of infants and children with chronic disease or feeding problems.

Nutrition support is required by patients who are unable to consume orally any, or adequate, fluid and nutrients for survival or for normal growth and development in children (Figure 67-1). Enteral nutrition support has a number of advantages over parenteral nutrition therapy. Enteral nutrition mimics the normal gastrointestinal (GI) response following the ingestion of a meal, with the exception of the oral phase. The presence of nutrients within the intestinal lumen provides stimulation of the gallbladder and normal GI function, helping to maintain balance within

the complex intraluminal environment. Luminal nutrients stimulate GI neuroendocrine function, effecting motility and digestion through the secretion of digestive enzymes and GI hormones. Intestinal functional and structural changes occur through local and systemic interaction of nutrients and neuroendocrine peptides, cytokines, and hormones.[3] The list of these mediators is extensive and includes gastrin, enteroglucagon, peptide YY, interleukin-3, 11, or 15, epidermal growth factor, growth hormone, insulin-like growth factors-I and -II, and glucagon-like peptide-1 and -2.[3] Monosaccharides and fatty acids influence the secretion of enteroglucagon and peptide YY and via these mediators effect mucosal growth and decrease intestinal transit time.[4] Carbohydrate, protein, zinc, magnesium, potassium, or manganese deficiency can modify the effect of growth hormone and insulin-like growth factor II.[5–7] Luminal nutrients assist in the maintenance of gut mucosal mass, including the gut-associated lymphoid tissue (GALT).[8] The GALT consists of the lamina propria, intraepithelial lymphocytes, immunoglobulin A (IgA), Peyer's patches, and mesenteric nodes and is responsible for processing antigens presented to the gut mucosa.[8] During periods of "bowel rest," such as occur with intravenous feeding and starvation, there is a reduction in gut mass and the function of the GALT is suppressed.[9] This has been associated with a reduction in IgA secretion and increased gut permeability resulting in increased bacterial adherence to the intestinal wall, cellular injury, and bacterial penetration, translocation with adverse systemic host responses.[10–12] Oral or enteral feeding may reduce the potential risk of bacterial translocation, except when disturbances in intestinal permeability are related to an underlying disease process (e.g., short bowel syndrome) or the chemical composition of the enterally provided substance (e.g., blue food dye).[11–15]

The GI tract is a delicate ecosystem with the balance determined and maintained through the interplay between nutrients, bacteria, and the intestinal defense system (luminal, mucosal, and submucosal immune system). Luminal nutrients play an important role in the development and function of the GI ecosystem through the modulation of the resident bacterial flora. The contribution of prebiotics to development of normal intestinal flora and the intestinal immune system is now recognized.[16,17] The interaction between specific bacteria and toll-like receptors located on intestinal enterocytes and lymphoid cell from as early as birth influences the development of physiologic intestinal immune response.[16,17]

Enteral nutrition therapy is easier and safer to administer than is

parenteral nutrition. Not only are the risks of intravenous access avoided but there is also a wider margin for error with most metabolic complications.[18,19] Due to advances in formulas, enteral feeding tubes and delivery systems, development of protocols for management, specialized Nutrition Support Teams, and better education of parents and caregivers, the administration of enteral nutrition has been associated with improved clinical outcome and safety profiles.[18,19] There are also a relative cost advantage of providing nutrition support via the enteral route compared to parenteral nutrition therapy. Enteral nutrition support is approximately two- to four-fold cheaper than parenteral nutrition on an inpatient or outpatient basis.[20–23] Based on U.S. Medicare charges, the annual cost of providing enteral nutrition per patient is approximately USD$9605 ± $9327 compared with USD$55,193 ± $30,596 for parenteral solutions.[20] In addition, the frequency and cost of hospitalizations are higher for patients supported on parenteral nutrition therapy compared with enteral nutrition therapy.[23] However, despite these relative advantages, the delivery of safe and effective enteral nutrition therapy may still present challenges for families and caregivers in terms of time, technical expertise, and cost.[21]

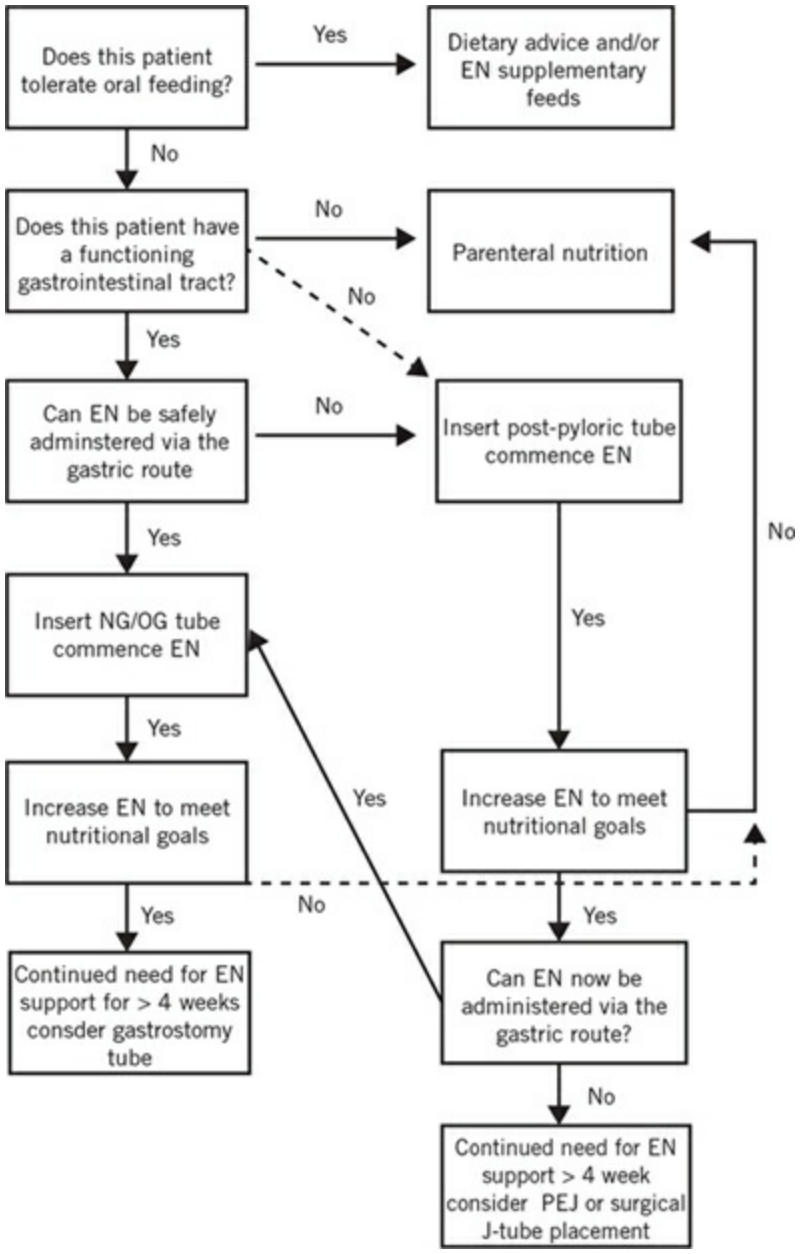


Figure 67-1 Decision flowchart for the provision of enteral nutrition support.

INDICATIONS FOR ENTERAL NUTRITION

Enteral nutrition should be considered for any patient with a functional GI tract who requires nutritional support (Figure 67-2). Enteral feeding may be required if adequate oral nutrient intake cannot be provided in children with growth failure, weight faltering, or weight deficit. See Chapter 47,

"Failure to Thrive." This may be defined as a child with a weight or weight for height less than third percentile below the mean for sex and age and/or crossing of two growth curves on the weight, height or weight/height percentile charts, no weight gain, or weight loss during the last 2–3 months and/or triceps skinfold measurements less than the fifth percentile. The requirement for enteral nutrition support may be temporary in the presence of acute illness or inadequate quantity or quality of nutrition provided. Enteral nutrition support may be an important adjunct in the long-term management of children with chronic diseases associated with increased energy needs that are difficult to achieve via the oral route such as may occur in cystic fibrosis[24,25] or in congenital heart disease (Figure 67-2).[26] Children with severe neurological dysfunction may have difficulty with the oral and esophageal phase of feeding and feeding may be slow and entail risks of aspiration. Tube feeding can provide welcome respite for families and caregivers who previously may have spent over 6 hours a day assisting with oral feeding.[27] Children with cognitive, psychiatric, and behavioral disorders that interfere with oral feeding may develop nutritional deficiency and enteral tube feeding may provide a safe and reliable route for the delivery of essential nutrients.

Disorders of the GI tract that are characterized by abnormal digestion and absorption of oral diet are an important indication for enteral nutrition support. Patients with short bowel syndrome, secretory diarrhea, or dysmotility syndromes resulting in excessive GI losses, frequently will have improved absorption and a reduction in losses with small volume continuous enteral feeds of a specialized formula.[28,29] See Chapter 61, "Intestinal Failure." Because of their composition, these formulas are often unpalatable and require tube administration to obtain adequate volumes of administration. Most patients receiving parenteral nutrition will also receive some enteral nutrition to sustain gut mucosa integrity.[28] Enteral nutrition usually provides an important transition stage as the patient progresses from parenteral nutrition to oral diet.[29]

Advances in understanding of the role of nutrients in the modification of inflammation and immunity has led to the development of disease-specific formulas as a primary treatment modality, such as in Crohn's disease[30,31] (see Chapter 52, "Inflammatory Bowel Disease"). Enteral nutrition has been advocated as a primary treatment for conditions associated with a metabolic disturbance, such as the use of gastrostomy tube feeding to infuse a ketogenic diet in children with epilepsy.[32] Although in most clinical scenarios enteral nutrition support has a

therapeutic intent, it is also used to prevent the development of malnutrition, such as can occur during stem cell transplantation.[33]

CONTRAINDICATIONS TO ENTERAL NUTRITION

With developments in options for GI access, delivery systems, and enteral formulas, the list of absolute contraindications for enteral nutrition therapy has been reduced significantly. Contraindications include GI ischemia, including the initial phase of necrotizing enterocolitis and toxic megacolon, severe intractable vomiting or diarrhea, diffuse peritonitis, and mechanical intestinal obstruction. Extreme care should be taken when administering enteral nutrition in patients in whom the GI blood flow could be compromised, such as during treatment with hypothermia, low cardiac output, multiorgan failure, compression syndromes, or infusions of specific drugs.[34] However, enteral nutrition has been shown to protect the splanchnic oxygen balance during intraoperative duodenal feedings in severely burned patients,[35] and in some forms of GI bleeding enteral nutrition may protect the gut mucosa and reduce further bleeding.[36] Controversy remains regarding the appropriate form and timing of nutritional support for the patient with circulatory shock.[37]

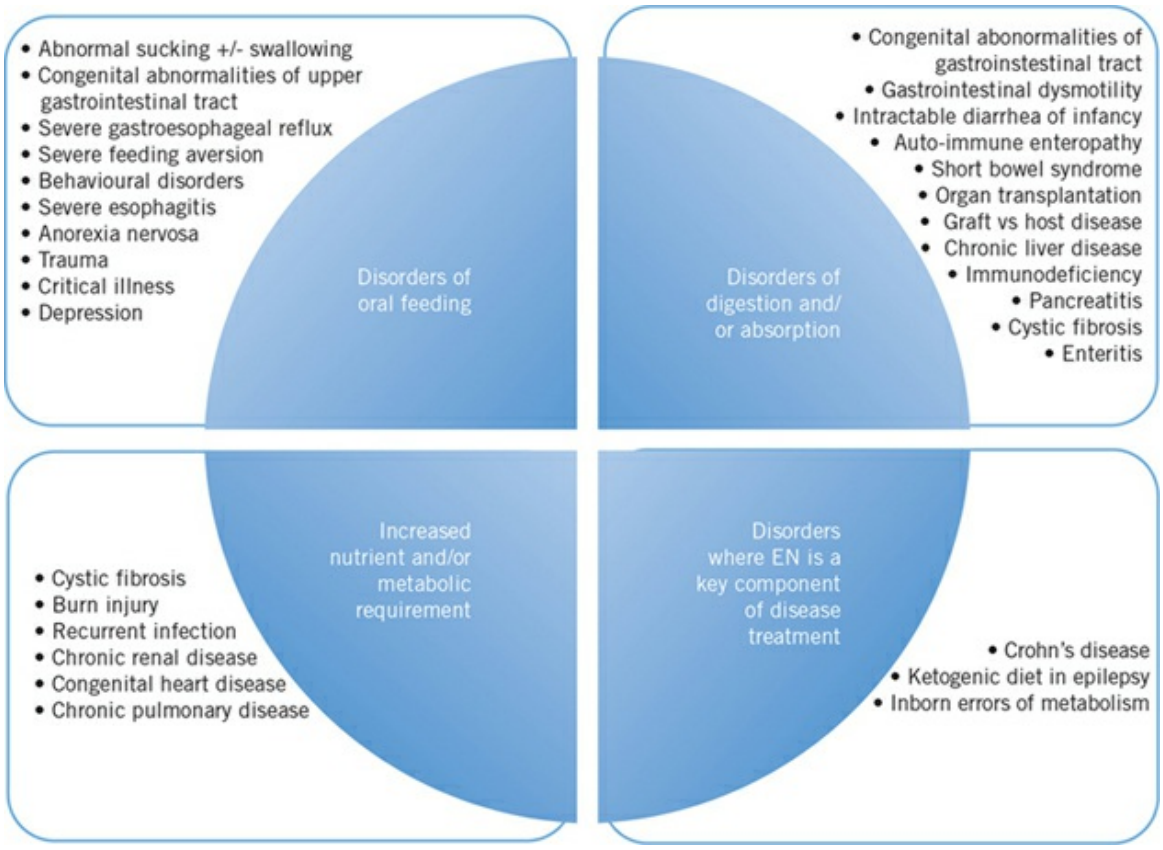


Figure 67-2 Indications for enteral nutrition in infants, children, and adolescents.

ENTERAL FORMULAS

In the early days of enteral nutrition, a mixture of blenderized diets and milk products was administered through a large-bore feeding tube. This approach was associated with nutritional imbalance, micronutrient deficiencies, feeding intolerance, and tube blockage. Today, there is a wide range of enteral nutrition products suitable for use in infants, children, and adolescents (see Chapter "Enteral Products" in Appendix III). Formulas have been designed for different age groups and for children with certain diseases.

Composition of Formulas

The majority of patients with a normal GI tract will tolerate the gastric administration of a polymeric formula. Polymeric formulas are based on intact protein or polypeptides usually derived from cow's milk or soybean.

The nitrogen to non-nitrogen calorie ratio approximates 1 to 150. Carbohydrates are sourced from different starches, including corn and tapioca. Maltodextrin and hydrolyzed cornstarch, glucose-derived saccharides, and corn syrup are commonly used. Formulas can have different lactose contents. Fats are usually present as polyunsaturated fatty acids from corn, safflower, sunflower, or soybean oil or from animal fat. The content of medium-chain triglycerides (MCTs) is increased in formulas developed for the treatment of patients with malabsorption. Some enteral nutrition formulas contain soluble fiber. The soluble fiber is added primarily to normalize GI transit, but after it is converted to short-chain fatty acids it provides an additional source of calories and can exert trophic effects on the colonic mucosa. In all complete enteral formulas, electrolytes, vitamins, and trace elements are added to provide the Dietary Reference Intakes of micronutrients and minerals at the target volume. At standard dilution, the caloric content of infant formula is usually 0.67 kcal/mL, and of standard enteral formula, 1 kcal/mL. Concentrated enteral nutrition formulas are also available (1.5 and 2 kcal/mL). The osmolality of enteral formulas can range widely depending on the nutrient composition and caloric density (~200–750 mOsm/L).

Age Considerations

Due to the difference in normal macro- and micronutrient requirements at different stages of development, enteral formulas have been developed to meet the recommended needs of specific age groups.

Infant Formulas

Human breast milk should be considered as the first option for enteral nutrition support for infants requiring a feeding tube. Human milk has a number of key nutritional, immunological and neuroendocrine advantages over commercial infant formulas (see Chapters 30 and 31), but if breast milk is not available, commercial infant formulas will be required. Standard infant cow's milk based protein formulas have been shown to support normal growth in full-term infants.[38] The serum amino acid profiles of infants fed casein predominant cow's milk formulas are different to that of breast-fed infants and have been linked to metabolic disturbances in some infants.[39] The amino acid profile of infants fed whey protein predominant cow's milk formulas more closely resembles human milk than cow's milk, but is associated with a higher percentage of branched chain amino acids and threonine compared with human milk.

Whether this has any clinical significance is not known.[40,41] Nucleotides are normally present in human breast milk and have been added to some cow's milk–based formulas. The rational for this addition is based on reports of decreased diarrheal disease in infants receiving formulas supplemented with nucleotides.[42] Most standard cow's milk based formulas are also supplemented with iron to prevent iron deficiency, particularly in preterm and small-for-gestational-age infants. The global standard for the iron present in infant formula ranges from 0.3 mg to 1.3 mg/100 kcal.[43] The renal solute load of standard cow's milk based formulas is higher than human milk and may pose a problem for preterm infants or infants with renal impairment. Plant oils are used in cow's milk based infant formulas to replace animal fats. Improved fat and calcium absorption has been observed in infants receiving formulas with a greater proportion of triglycerides with palmitic acid in the beta position of the glycerol backbone, similar to human milk.[44] Arachidonic acid (AA) and docosahexaenoic acid (DHA) are important structural elements of cell membranes in the retina and brain. Preterm infants fed formula supplemented with AA and DHA have been observed to have improved visual acuity and cognitive development.[45,46] However, this observation has not been consistently reported in term infants.[47,48] Lactose-containing cow's milk–based formulas are contraindicated for infants with galactosemia and lactose intolerance.

To meet the nutritional needs to support the rapid growth expected in preterm low-birth-weight and extremely low-birth infants who also have immature intestinal and renal systems, specialized preterm infant formulas are required. These formulas are energy rich, usually with a higher proportion of MCTs to enhance fat absorption. A benefit of these formulas is enhanced calcium absorption and nitrogen retention.[49,50] The calcium and phosphorus concentrations are also increased in preterm infant formulas to support rapid bone growth.

Soy protein formulas are used in the management of infants with cow's milk protein intolerance. Although soy protein is a high-quality protein, soy formulas are supplemented with L-methionine, taurine, and/or carnitine to meet the needs of human infants. The carbohydrate source is sucrose, corn syrup and/or maltodextrin and therefore soy formulas can also be used to treat infants with lactose intolerance. Zinc and iron are supplemented due to their decreased bioavailability in the presence of phytic acid and polysaccharides.[51] High plasma concentrations of isoflavones have been reported in infants receiving soy formulas although

the clinical significance of this finding is unclear.[52] Aluminum is present as a contaminant in soy formulas, therefore soy formulas should be avoided in infants with renal disease. Soy protein formulas are not recommended in infants with fructose intolerance or in preterm infants weighing less than 1800 g due to concerns regarding aluminum toxicity, and lower weight gain and bone mineralization observed in some infants.[53]

Formulas for Infants >6 Months of Age and Children

Follow-up formulas are aimed at supplementing the weaning diet for children greater than 6 months of age. These formulas have adjustments in the nutrient concentration compared to standard cow's milk based or soy protein infant formulas. It is not intended that these formulas interfere with the normal transition to solid food.

As it is expected that healthy children will have transitioned to solid food and complimentary liquids by 1 year of age, formulas for children over 1 year are aimed at providing complete or supplementary nutrition for children with acute or chronic illness. These formulas have a higher caloric density and protein concentration compared to standard infant formulas and micronutrient content reflects age-based recommendations. In patients with underlying GI disease or those requiring jejunal feeding, an oligomeric formula might be indicated. The protein in oligomeric formulas has been hydrolyzed to peptides or a combination of peptides and amino acids. The carbohydrate complexity varies among formulas, although most oligomeric formulas are lactose free. The potential advantages of oligomeric formulas over formulas containing chemically derived amino acids is the lower osmolality and faster intestinal uptake of amino acids derived from peptides.[54] Elemental or monomeric formulas contain digested macronutrients, including monosaccharides, MCT, and amino acids, with an essential to nonessential amino acid ratio reflecting high biologic protein values. Lactose and gluten are absent and residues are low. The unpalatability and high osmotic load of simple sugars and amino acids generally limit the use of elemental formulas to tube feeding when feeding intolerance occurs with other types of formula (e.g., severe malabsorption or short bowel syndrome). Modular feeds are built from individual nutrient components aimed at providing a specific nutritional requirement as a supplement to a diet or formula or to provide a complete formula to meet the specific needs of an individual. Indincations include conditions requiring a change in the calorie to protein ratio in a patient with burns or trauma or in increased caloric density in a patient who is severely fluid restricted.

Disease-Specific Formulas

Advances in the understanding of the role of specific nutrients and their effects on metabolism have led to modification of enteral nutrition formulas for the treatment of an expanding range of diseases. Immunonutrition is the enrichment of formulae with single components or a combination of key substrates that might play a crucial role in metabolism in sepsis, inflammation, tissue healing, and growth.[55] The aim of the disease-specific formulas is to provide therapeutic benefits in addition to the maintenance of general nutritional status. Glutamine-enriched formula has been advocated for the prevention and treatment of intestinal mucosa injury associated with chemotherapy and critical illness.[56,57] Glutamine is an essential amino acid in critical illness and is an important source of energy for the intestine. Glutamine supplemented formula was associated with a decrease in the frequency of sepsis and bacteremia in adult trauma patients and those supplemented with 0.4 gm of glutamine/kg/d had better enteral nutrition tolerance.[58] The American Society for Parenteral and Enteral Nutrition,[59] the European Society for Parenteral and Enteral Nutrition (ESPEN), and Canadian guidelines recommend the use of enteral glutamine in adult patients with burn injury and trauma.[60] Formulas supplemented with arginine, glutamine, ribonucleic acid nucleotides, MCTs, and/or omega-3 fatty acids have been also been advocated as enhancing the immune system of critically ill patients.[56,57] However, whether the benefit of glutamine or immune-enhancing formulas in malnourished and critically ill children outweighs the additional cost of these formulas remains unresolved.[61,62]

Formulas have been tailored for patients with inborn errors of metabolism. These formulas aim to prevent toxicity in patients unable to metabolize a specific amino acid, such as phenylalanine in phenylketouria, or as part of the management of acute metabolic instability during infection. Formulas have been modified for patients with severe pulmonary disease to provide 40%–50% of energy from fat in an effort to limit CO_2 production from the metabolism of carbohydrates. Formulas for patients with renal disease have a lower protein and sodium chloride content. Formulas for burns patient have a low fat and high carbohydrate content. Patients with chronic liver failure receiving formulas with a high concentration of branched amino acids had improved clinical outcome and quality of life.[63] However, evidence for improved clinical and economic outcomes of many of the disease-specific formulas, compared with standard formulas, is still required to justify recommendations of their

routine use.[56,58]

ROUTES OF ADMINISTRATION

During enteral nutrition therapy, nutrients are directly delivered via a tube into the stomach, duodenum, or jejunum. The tube is inserted either through the nose or mouth for short-term enteral nutrition (less than 4 weeks) or through a surgically or endoscopically created stoma for long-term enteral nutrition (more than 4 weeks). The choice of the location and the route of administration will depend on the patient's underlying medical or surgical condition, including GI anatomy and function, the indication and duration of enteral nutrition therapy, and psychosocial factors. Additional factors, such as local technical expertise, tube availability, and cost, will also influence the route and type of device selected. As a general principle, tubes that deliver nutrients into the stomach are the preferred choice. Gastric tubes are easier to insert and allow a physiological digestive process with bolus or continuous feeding regimens. However, in the presence of gastric outlet dysfunction, severe gastroesophageal reflux, or gastric paralysis, transpyloric access may be indicated.

Gastric Access Feeding Tubes

Nonsurgical or nonendoscopic placement of a feeding tube through the mouth or nose is the most common method of establishing GI access in infants and children due to the ease of placement and cost. The nasal access is usually preferred, except in preterm infants or in patients with nasopharyngeal abnormalities or obstruction as may occur following trauma or with congenital malformations. The tube is generally of small diameter (5–12 Fr) and is well suited to nutritional support of short or intermediate duration or intermittent nutritional therapy. However, the small luminal diameter renders these tubes susceptible to blockage particularly when medications or nutrient supplements are infused. To minimize the risk of blockage it is recommended that the tube is flushed with water after each feed and each infusion of medication. There is a wide range of enteric feeding tubes available for use in children. Current feeding tubes are made from flexible silicone, polyurethane, or elastomer and may require a stylet to assist placement. Despite the increased flexibility, they have a long lifespan and may incorporate specialized features. Weighted tips are designed to allow gravity to assist with small bowel placement and to prevent retrograde displacement. However, with the possible exception of patients requiring mechanical ventilation, the weighted tubes have not

been shown to have significant benefit over nonweighted tubes.[64] With training, nasogastric tubes can be safely inserted by allied health staff, caregivers, family members, and even the patients themselves.

Placement of a feeding tube using surgical, radiological, or endoscopic techniques is recommended for long-term enteral nutrition therapy or gastric decompression. There are a number of important advantages of these tubes over nasogastric or nasoenteric tubes in children. The gastrostomy tube can be maintained in position for a longer period because it is fixed against the anterior abdominal wall. The tube does not interfere with breathing and avoids potential complications of chronic nasal discharge, sinusitis, and developmental abnormalities of the nose. In addition, for some children, repeated insertion of a nasogastric tube is associated with psychological trauma and feeding aversion. Gastrostomy tubes are usually large-bore tubes (14–24 Fr) to deliver high feeding volumes and medications with minimal risk of occlusion. Most gastrostomy tubes are made of a biocompatible material such as silicone or polyurethane and are anchored in place with either parallel bumpers or a mushroom or balloon at the gastric site with a retention disk at the skin. This allows the tube to be secured to the stomach wall without sutures. A wide range of gastrostomy tubes is available for use in children. Most devices have an antireflux valve to prevent the release of gastric contents when the tube is accessed. Gastrostomy device can be easily disguised under clothing and the feeding tube is connected only at the time of infusion as a result it is particularly popular with older children and adolescents (Figure 67-3).

The percutaneous endoscopic gastrostomy (PEG) technique has revolutionized the placement of enteric feeding tubes in children.[65] This relatively simple and fast procedure can be performed during esophagogastroduodenoscopy in an endoscopy suite with the use of conscious sedation and local anesthesia or general anesthesia with a success rate of up to 96% even in small infants (<3.5 kg).[66] Although several techniques (Ponsky pull, Sachs-Vine push, Russel introducer) have been developed, all have in common the basic principle that the endoscope locates the site of tube placement from within the stomach while transillumination of the light from the endoscope through the abdominal wall identifies the site of skin incision. Radiological placement of gastrostomy tubes has also been shown to be safe and cost-effective in pediatric patients.[67]

The external bolster should be routinely checked for secure position to avoid tube migration and to prevent injury to the abdominal wall due to

compression between the internal and external bolsters. The stomal site requires regular cleaning to avoid chemical injuries from the gastric secretions and infection. Stomal granuloma and skin irritation or infection need prompt attention with referral to a stomal therapist if necessary, to prevent further complications. Eventually gastrostomy tubes will degrade over months to years due to physical or chemical trauma from yeast contamination or medications. Balloon-type skin level gastrostomy devices can also suffer from balloon rupture so it is general practice to replace these every 6 months as needed.[68]

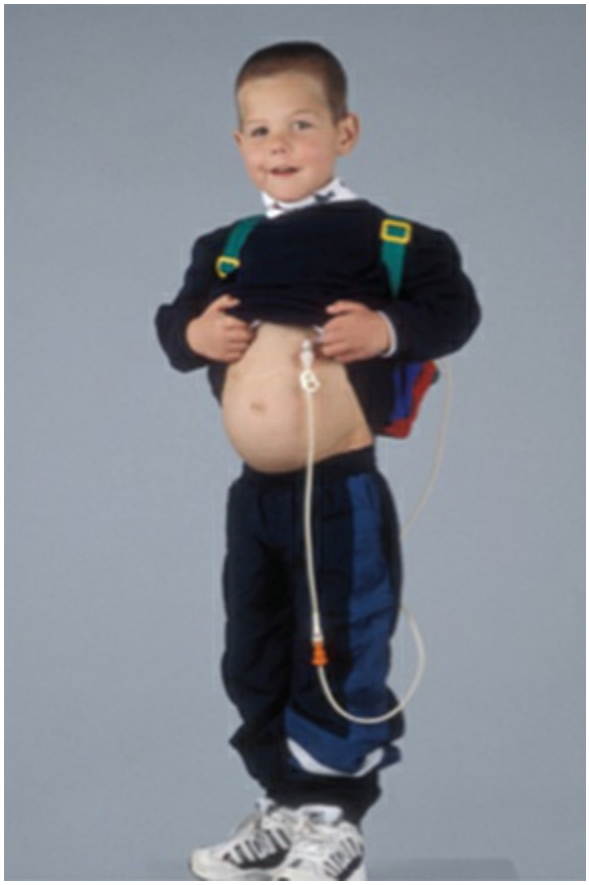


Figure 67-3 Child receiving continuous enteral nutrition via a gastrostomy using a portable enteral feeding pump in a backpack.

Postpyloric Feeding Devices

Advances in tube technology and the techniques for placement of nasoenteric tubes in children have provided an opportunity for postpyloric feeding in children in whom gastric feeding is difficult or contraindicated

(Figure 67-1). As a result, there has been renewed enthusiasm for the use of postpyloric feeding in the critically ill. Compared with intragastric feeding, postpyloric feeding in the critically ill is associated with a higher caloric intake and avoids complications associated with parenteral nutrition therapy. In a study of critically ill children requiring mechanical ventilation, the risk of aspiration was similar in patients receiving either postpyloric or intragastric feeding was less in the intragastric fed group compared to the post-pyloric fed group. The percentage of daily caloric goal achieved was less in the gastric group compared to the small bowel group.[69] Nasoenteric tubes can be placed blindly or under fluoroscopic or endoscopic guidance.[70,71] The percutaneous endoscopic technique can also be used to place an enteric tube using either a direct approach (direct percutaneous endoscopic jejunostomy) or by the creation of a PEG and placement of a specialized double-lumen tube (percutaneous endoscopic gastrostomy jejunostomy).[72] Among the range of surgical techniques described for jejunostomy tube placement, the needle–catheter jejunostomy is the most common. Using this technique, a large-bore needle is tunneled through the seromuscular layers of the jejunum distal to the ligament of Treitz. Enteric feeding tubes are generally smaller in diameter (9–12 Fr) than gastrostomy tubes and tend to have a shorter lifespan (3–6 months). Dislodgement of gastrojejunal tubes is not uncommon and may be a limiting factor for the use of this type of tube in patients requiring long-term home enteral nutrition.[19,73]

MODES OF DELIVERY

The mode of delivery of enteral nutrition will depend on the route of administration (gastric, duodenal, or jejunal), characteristics of the feeding tube (small- versus large-bore catheter), the desired feeding pattern (bolus, intermittent, cyclic, or continuous), and the cost and availability of equipment (by gravity or syringe, or by infusion pump). Bolus tube infusion usually mimics the normal meal pattern based on age. Intermittent or cyclic feedings are delivered at a specified rate over 1 or more hours, with 4–8 h/d of gut rest allowing freedom from tubes and pumps for a period of the day. Continuous feeding delivers a constant-rate infusion, usually by an infusion pump, over the entire day or a prolonged period. The infusion pump allows accurate nutrient delivery. Some pumps are specifically designed for outdoor activities and may be carried in a small backpack improving mobility for children on long-term enteral nutrition therapy (Figure 67-3).

Gastric feeding is preferred because it is considered more physiologic, allows bolus feeds through large-bore tubes, and is generally cheaper and easier to administer.[34] Many patients intolerant of bolus feeds can be successfully fed intragastrically using an intermittent or continuous feeding regimen. Energy intake goals are more successfully achieved in brain trauma patients receiving postpyloric feeds compared with those receiving intragastric feeds.[74] Postpyloric feeding is usually delivered as an intermittent or continuous infusion because rapid-rate infusion of nutrients is often limited by abdominal discomfort, diarrhea, or dumping syndrome. There are specific physiologic and metabolic considerations associated with continuous-rate feeding. During continuous intragastric feeding, gastric emptying increases parallel to the rate of infusion if the infusion rate is maintained at less than 3 kcal/min.[75] Increased caloric density, fat, and osmolarity of the formula can delay gastric emptying. The protein composition also influences gastric emptying. Whey-based enteral formulas enter the duodenum faster than casein-based enteral formulas.[76] In animals, the absorptive capacity of the proximal small intestine is unchanged during continuous enteral nutrition, but the enzymatic and functional capacity of the distal small intestine and colon is reduced.[75] The continuous delivery of nutrients into the small intestine affects glycemic control by modifying the typical fluctuating pattern of insulin and glucagon production.[77,78] The reduction in steatosis during continuous enteral nutrition compared with parenteral nutrition with the same carbohydrate intake is consistent with this observation. In addition, energy expenditure due to the thermic effect of feeding is lower in patients receiving continuous enteral nutrition than in patients receiving the same quantity of nutrients delivered by bolus.[74,77]

ENTERAL NUTRITION IN SPECIFIC DISEASE STATES

Critically Ill Children

Enteral feeding has now eclipsed parenteral nutrition in its role as primary nutrition support of the critically ill. Enteral nutrition not only reduces the risk of sepsis and metabolic complications associated with parenteral nutrition therapy but also prevents mucosal atrophy, reduced enzymatic activity, and dysmotility[79] associated with starvation.[80] The lack of bowel sounds on auscultation or lack of passage of flatus or stool is no longer

absolute contraindications for the initiation of enteral nutrition. In a recent multicenter cohort study of 31 pediatric intensive care units (ICUs) across eight countries, 30% of patients had severe malnutrition on admission.[80] Mortality was lower in those children who received enteral nutrition (rather than parenteral nutrition) and children receiving a higher percentage of their energy goal energy via the enteral route had lower 60-day mortality.[80] ICUs that implemented enteral nutrition protocols had a lower prevalence of acquired infections.[80] Despite potential complications such as diarrhea and aspiration, early enteral nutrition has been shown to decrease infectious complications and decrease mortality and infections when compared with parenteral nutrition.[81] Early enteral nutrition (<48 hours) in adult with severe traumatic brain injury has been shown to improve survival and the Glascow Coma Score on day 7 and was associated with an improved clinical outcome 1 month postinjury.[82] Gastric feeding is problematic in some patients and up to 50% of adult patients with traumatic brain injury exhibit intolerance to enteral nutrition due to impaired GI motility, reduced absorption, hypoalbuminemia, and chronic diarrhea. In a meta-analysis of critically ill adult patients, enteral nutrition reduced the risk of potentially life-threatening infections and noninfection-related events when compared with parenteral nutrition, as well as reducing length of stay, time in the ICU, and length of nutritional treatment.[83] Savings from reduced adverse event risks averaged nearly $1500 per patient and savings from reduced length of stay amounted to nearly $2500 per patient. The authors concluded that shifting 10% of parenterally treated adult patients in the United States to enteral nutrition would save $35 million annually due to reduced adverse events and another $57 million due to shorter hospital length of stay.[84] Adequate nutrition is crucial in children after surgery for congenital heart disease and can be challenging in the setting of greater demands and poor oral intake. Use of feeding protocols and a home surveillance system for hypoplastic left heart syndrome improves outcomes.[84] Children who have undergone cardiac surgery that is complicated by chylothorax require a fat-free diet or a formula that has a high proportion of medium-chain fatty acids.[85] Although myriad of barriers may exist to prevent achieving complete nutrition support in critically ill children, it is generally recommended to commence enteral nutrition within 48 hours of admission to the ICU.[86] See Chapter 64, “The Critically Ill Child.”

Gastrointestinal Diseases

Short bowel syndrome is an important cause of intestinal failure in childhood and usually as a consequence of surgical resection of the small intestine. Parenteral nutrition is essential for survival and to support growth while awaiting intestinal adaptation and improved enteral tolerance to occur. Although enteral feeding may be limited in the initial phase of recovery, if possible small amounts of enteral nutrition should be infused to support mucosal integrity and enhance the adaptive response. Enteral feeding will also promote normal motility and gall bladder emptying.[28] Human milk rich in nucleotides, IgA, glutamine, and growth factors is first choice but if unavailable oligomeric feeds with a high MCT content may be tolerated. The route of enteral feeding, either oral or via tube and continuous versus bolus, remains a matter of debate in terms of efficiency and feasibility.[28,29] A slow and constant infusion rate may promote absorption as a result of increased mucosal contact time however may also alter the normal intestinal motility pattern.[28,29,87] See Chapter 63, "Intestinal Failure."

Exclusive enteral nutrition has resulted in induction of remission of Crohn's disease, especially in children.[30] There is also emerging evidence regarding the role of ongoing supplementary enteral nutrition in the maintenance of remission from Crohn's disease in children and adolescents.[31] This is postulated to be the result of benefits of improved general nutrition as well as intestinal specific effects resulting in enhanced mucosal healing.[32] There is now a substantial body of evidence that enteral nutrition significantly reduces infectious complications, surgical interventions, and mortality in adults with severe acute pancreatitis compared to that observed in patients treated with parenteral nutrition.[88,89] This has yet to be studied rigorously in pediatric pancreatitis.[90] There is evidence, albeit relatively weak, to support the provision of minimal enteral nutrition (preferably with human milk) within the first 2 days of life, in the prevention of necrotizing enterocolitis in high-risk neonates.[91] After a diagnosis of necrotizing enterocolitis it has been recommended that feeds be withheld. Early introduction of enteral nutrition may have the potential for benefit by limiting the duration of parenteral nutrition therapy and the risk of catheter-related sepsis as well as reducing the risk of postnecrotizing enterocolitis intestinal stricture and length of hospitalization.[92]

MANAGEMENT OF ENTERAL ACCESS

Principles of Management

Monitoring patients receiving enteral nutrition will minimize the risk of developing complications and ensure optimal nutrition. The use of protocols for the management of enteral nutrition support has been associated with improved outcomes.[93] Enteral nutrition support monitoring includes nutritional, anthropometric, clinical, and laboratory measures. Frequency of laboratory monitoring of electrolytes, blood glucose, liver function, renal function, and serum lipids will depend on the clinical and nutritional status of the patient.[94] Nasally inserted tubes require daily check for nasal erosion, tube fixation, and tube integrity (Table 67-1). Gastrostomy and postpyloric tubes require daily stoma site and tube position check, weekly tube rotation, and balloon water volume check. Other anthropometric measures such as body mass index, mid-arm circumference, and triceps skinfold thickness may be performed as required to monitor growth and nutritional progress.

TABLE 67-1 Bedside Evaluation Tests for Tube Placement in Children

Aspirate	Color and Appearance	pH value	Pepsin (μg/mL)	Trypsin (μg/mL)	Bilirubin (mg/dL)
Gastric secretions	Yellow-gray or white-tan; cloudy	< 6	> 100 (> 20)[a]	< 30 (< 50)[a]	< 5
Intestinal secretions	Green	> 6	< 100 (< 20)[a]	> 30 (> 50)[a]	> 5
Respiratory secretions	Yellow-gray; mucoid	6–8	< 100 (< 20)[a]	< 30 (< 50)[a]	< 5

[a]Trypsin and pepsin ranges specific for children

Complications

Despite the potential benefits of enteral nutrition, complications can occur. Fortunately, life-threatening events are rare. However, problems related to the tube, the method of delivery, or the composition of the formula can seriously interfere with achieving nutritional goals. To minimize complications, prior consideration should be given to the patient's medical and physical condition, including any metabolic or electrolyte abnormalities and previous dietary tolerance.

Gastrointestinal Complications

Intestinal discomfort, bloating, cramping, diarrhea, nausea, and vomiting can occur during enteral feeding. Whenever symptoms occur, the position and integrity of the tube should be confirmed. In some cases, these symptoms relate to the high osmolality of the formula or the rate of infusion and alterations to the formula composition or infusion rate will be sufficient to improve feeding tolerance. The assessment of a sample of stool from a patient with diarrhea can assist in directing further investigations as required (bacterial culture, absorptive status by microscopy, analyzing pH, and reducing substances). The use of concomitant medications should be reviewed for possible drug–nutrient interactions or GI side effects. Bacterial contamination of the feed or the tubing can result in diarrhea or vomiting.[93] Therefore, the method of formula preparation and storage and the technique of hanging and administering should be reviewed. Samples of the feed, the tubing, and the feeding reservoirs are necessary to confirm this diagnosis.

Patients with a primary GI disorder could be at risk of bacterial overgrowth owing to disturbances in gut motility. Bacterial culture of an intestinal biopsy or a lactulose breath hydrogen test will assist in establishing the presence of bacterial overgrowth of the small intestine. Constipation associated with enteral nutrition is uncommon and when it occurs is usually associated with insufficient fluid or fiber intake, intestinal dysmotility or obstruction, or medications. Gastroesophageal reflux can be exacerbated by intragastric feeding. The nasogastric tube can stent open the lower esophageal sphincter, and irritation owing to tube trauma can contribute to lower sphincter incompetence. The placement of a gastrostomy tube will plicate the stomach against the anterior abdominal wall, distorting the normal gastric anatomy, with a potential impact on gastric function.

Pulmonary Complications

Aspiration of gastric contents due to incorrect tube placement or tube dislodgment into the airway can be fatal. Children at high risk include those with chronic neurologic disease, depressed conscious state, intestinal dysmotility, severe gastroesophageal reflux, and those requiring mechanical ventilation. In addition to checking tube position, tracheal aspirates can be measured for glucose content or fat-laden macrophages. The addition of blue dye to the formula to assess tube position and aspiration patients should be avoided because it has been associated with fatalities.[15]

Infectious Complications

Irritation due to the mechanical trauma or exposure to gastric or intestinal secretions render the skin and mucosa susceptible to infection. A well-fitting, well-maintained device specifically designed for enteral nutrition use will limit this complication. Contamination of the formula and the delivery set can occur as a result of preparation or administration of the feed.[94] Bacterial contamination occurred in 35%–50% of enteral feeding bags at the end of the infusions in a pediatric hospital.[95] Feeding devices also harbor biofilm growth of various bacteria and fungi in the outer and inner surface with potential risk of local and systemic infections.[96] Long-term nasogastric and nasoenteric tubes can be associated with chronic nasal discharge, otitis media, and sinusitis.

Mechanical Complications

Mechanical complications related to enteral tube feeding are common. However, these can generally be successfully treated and do not involve the same risks as a central venous catheter in critically ill children. Tube occlusion can occur as a result of problems related to the tube (length, caliber, characteristics), the infusion (formula, drugs), pumps and clamps, the method and rate of delivery, and the level of tube care (method, frequency). Infusions that contain a highly viscous formula or crushed or powdered drugs can be associated with tube occlusion. If flushing the tube with warm water is unsuccessful in clearing the blockage, a number of other options are available. The instillation of pancreatic enzyme supplements is sometimes successful in clearing a blockage. The use of meat tenderizer is not recommended because of its high sodium content and the risk of tube breakage during infusion. Cytology brushes, guide wires, neonatal tubes inserted into larger tubes, and specially designed catheters have been developed to treat tube occlusion. Key to the prevention of tube occlusion is careful monitoring and repeated flushing of the tube. Mixing of drugs and formula should be avoided. Irritation can occur anywhere along the interface between the tube and the skin or mucosa, resulting in inflammation, ulceration, or even perforation of any structure present along the tube lining (e.g., laceration or bleeding of nasal or oral cavity, perforation or laceration of the gut). Granulation tissue formation at the stoma site can occur as a result of chronic irritation. The migration of tubes with an internal balloon or retention device distally along the GI tract can cause bowel obstruction. Migration of a gastrostomy tube along the fistula tract can cause pain and inflammation around the site or the buried bumper syndrome. Complications such as dislodgment,

ampullary obstruction, and jaundice can occur with jejunal tubes.

Metabolic Complications

Metabolic complications associated with enteral nutrition are uncommon. Patients with chronic malnutrition or cardiac, hepatic, or renal impairment require careful monitoring of fluid and electrolyte status to prevent imbalance. Patients commencing enteral nutrition who have had a period of prolonged fasting, inadequate nutritional intake, or significant weight loss (>10% body weight) should be monitored for the metabolic features of the refeeding syndrome.[97] Regular monitoring of fluid status, serum sodium, potassium, phosphate, and magnesium levels is required until stability has been achieved with feeding advancement. Serum glucose levels, which are usually stable during continuous enteral nutrition therapy, can increase with overfeeding or as part of a stress response during critical illness. Hypoglycemia during intermittent feeding can be prevented with a progressive slowing of the rate of infusion in the period prior to disconnection. Dumping syndrome is reported in patients during enteral feeding in association with a rapid infusion of a formula with a high nutrient density and is treated with modifications in the rate of delivery or to the formula (such as adding uncooked cornstarch).

Drug–Nutrient Interactions

Administration of medications through an enteral feeding tube, either in combination with formula or alone, is problematic because only a few drugs have been tested and approved for tube delivery. Characteristics of the composition of the tube can influence the binding of drug to the tube wall.[98] Interactions between a drug and a nutrient can result in an undesired side effect of the drug or feeding intolerance. Drug–nutrient interactions can result in changes in medication bioavailability, distribution, metabolism, or excretion. Common drug–nutrient interactions are described with substances containing calcium, zinc, and iron or when acidic and neutral liquid medications are combined with casein or soy protein. Impaired absorption of phenytoin is well documented in the literature and occurs as a result of pharmacokinetic incompatibility.[98] Long-chain fatty acids can enhance the absorption of lipid-soluble drugs but hasten the degradation of other medications (e.g., carbamazepine). Liquid preparations of medications are preferred by children and for enteral administration. However, liquid drug preparations tend to have a high osmolality (1000 and 11,000 mOsm/kg) and may cause feeding intolerance when infused into the jejunum. The osmotic properties of these

liquids are usually attributable to the presence of sorbitol, propylene glycol, or polyethylene glycol. Although included among the inactive ingredients on drug labels, the amount is often not quantified. Osmotic diarrhea and delayed gastric emptying can occur if the cumulative dose is more than 20 g (range: 7.5–30 g).

HOME ENTERAL NUTRITION

Over the past three decades, advances in tube design, methods of delivery, and formulas have made enteral nutrition therapy safer, cheaper, and easier to administer for a wide range of disorders in childhood. With attention to appropriate patient selection, education, and providing adequate technical support, enteral nutrition therapy can be safely and effectively provided in the home.[99] It was estimated that in 1992, 152,000 patients of all ages received home enteral nutrition in the United States.[7] In Britain, home enteral nutrition therapy has been increasing rapidly—at a rate of up to 20% per year.[100] Of patients receiving home enteral nutrition therapy in Britain, 40% are children, compared with 5%–20% reported in three U.S. cohort studies.

Best-practice guidelines for the administration of safe and effective home enteral nutrition therapy have been developed by national nutrition support organizations. These guidelines take into consideration patient selection, assessment, monitoring, and the development of the enteral nutrition plan, including methods of implementation, documentation, and protocols for termination of therapy. Training patients, their families or caregivers in safe and effective home enteral nutrition therapy is ideally performed by a multidisciplinary team, including the gastroenterologist or surgeon, dietitian, stomal therapist, or nurse specialist.[98] An essential component of a successful home enteral nutrition service is availability of health professionals to provide support to the patient or caregivers at home to address concerns and direct appropriate intervention when necessary. A mechanism should be available to manage after-hours tube malfunction. To facilitate this process, all patients should carry a card with the type and size of tube and the date of last insertion clearly listed. Written documentation of routine tube care, as well as the recommended steps to take in the event of tube malfunction, is invaluable to the patient and caregiver at home. Regular review by the multidisciplinary home enteral nutrition team is cost-effective and improves quality of care.[101] At regular outpatient appointments, all aspects of the administration of enteral nutrition can be assessed. The tube is examined for size, function, and

integrity and can be changed if necessary. The stoma site is examined. The method of delivery is reviewed, along with protocols for formula preparation. The intake of formula is assessed with reference to the nutritional goals. The patient is examined for growth, weight gain, and nutritional status. Laboratory markers of nutritional status can be obtained as indicated.

CONCLUSION

Advances in enteral formulas, tube placement techniques, tube design, and delivery systems have enabled the safe and effective provision of enteral nutrition support for a broad range of disease indications in children. The successful provision of enteral nutrition support has limited the use of parenteral nutrition therapy, resulting in important benefits in terms of safety, manageability, and cost. The management of nutritional problems in patients with chronic diseases, such as neurologic disorders, Crohn's disease, cystic fibrosis, and short bowel syndrome can be enhanced by home enteral nutrition therapy. The development of disease-specific formulas provides new therapeutic options. Further improvements in technology enabling light, simple, feeding pumps and modifications to tube design continue to assist in the administration of enteral nutrition to children.

REFERENCES

1. Harkness L. The history of enteral nutrition therapy: from raw eggs and nasal tubes to purified amino acids and early post-operative jejunal delivery. *J Am Diet Assoc*. 2002;102:399–404.
2. Phillips MS, Ponsky JL. Overview of enteral and parenteral feeding access techniques: principles and practice. *Surg Clin North Am.* 2011; 91:897–911.
3. Pereira PM, Bines JE. New growth factor therapies aimed at improving intestinal adaptation in short bowel syndrome. *J Gastroentrol Hepatol*. 2006;21(6):932–940.
4. Drozdowski L, Thomson AB. Intestinal hormones and growth factors: effects on the small intestine. *World J Gastroenterol*. 2009;28;15(4):385–406.
5. Aponte GW, Fink AS, Meyer JH, et al. Regional distribution and release of peptide YY with fatty acids of different chain lengths. *Am J Physiol*. 1985;249:745–750.
6. Thissen JP, Ketelslegers JM, Underwood LE. Nutritional regulation of the insulin-like growth factors. *Endocr Rev*. 1994;15:80–101.
7. Ninh NX, Thissen JP, Collette L, et al. Zinc supplementation increases growth and circulating insulin-like growth factor I (IGF-1) in growth- retarded Vietnamese children. *Am J Clin Nutr*. 1996;63:514–519.
8. Li J, Kudsk KA, Gocinski B, et al. Effects of parenteral and enteral nutrition on gut-associated lymphoid tissue. *J Trauma*. 1995;39:44–51.

9. Kudsk KA. Importance of enteral feeding in maintaining gut integrity. *JPEN J Parenter Enteral Nutr*. 2001;25:S2–S8.
10. Alverdy JC, Aoys E, Moss GS. Total parenteral nutrition promotes bacterial translocation from the gut. *Surgery*. 1988;104:185–190.
11. Kudsk KA, Li J, Renegar KB. Loss of upper respiratory tract immunity with parenteral feeding. *Ann Surg*. 1996;223:629–638.
12. MacFie JC. Current status of bacterial translocation as a cause of surgical sepsis. *Br Med Bull*. 2004;71:1–11.
13. Alverdy JC, Aoys E, Moss GS. Effect of commercially available chemically defined liquid diets on the intestinal microflora and bacterial translocation from the gut. *JPEN J Parenter Enteral Nutr*. 1990;14:1–6.
14. Weber TR. Enteral feeding increases sepsis in infants with short bowel syndrome. *J Pediatr Surg*. 1995;30:1086–1089.
15. Maloney JP, Bhargava R, Ryan TA, et al. FD&C Blue No. 1 food dye is a mitochondrial poison that can be absorbed from enteral tube feedings in sepsis: a report of 3 deaths linked to systemic absorption. *JPEN J Parenter Enteral Nutr*. 2001;25:S25.
16. Quigley EM. Prebiotics and probiotics: modifying and mining the microbiota. *Pharmacol Res*. 2010;61(3):213–218.
17. Forchielli ML, Walker WA. The role of gut-associated lymphoid tissues and mucosal defence. *Br J Nutr*. 2005;93(suppl 1):S41–S48.
18. Shojania KG, Duncan BW, McDonald KM, et al. Making health care safer: a critical analysis of patient safety practices. *Evid Rep Technol Assess*. 2001;43:1–668.
19. Al-Zubeidi D, Demir H, Bishop WP, Rahhal RM. Gastrojejunal feeding tube use by pediatric gastroenterologists: 10-year experience in a pediatric academic center. *J Pediatr Gastroenterol Nutr*. 2012;56:523–527.
20. de Lucas C, Moreno M, Lopez-Herce J, et al. Transpyloric enteral nutrition reduces the complication rate and cost in the critically ill child. *J Pediatr Gastroenterol Nutr*. 2000;30:175–180.
21. Heyman MB, Harmatz P, Acree M, et al. Economic and psychologic costs for maternal caregivers of gastrostomy-dependent children. *J Pediatr*. 2004;145:511–516.
22. Abunnaja S, Cuviello A, Sanchez JA. Enteral and parenteral nutrition in the perioperative period: state of the art. *Nutrients*. 2013;5(2):608–623.
23. Reddy P, Malone M. Cost and outcome analysis of home parenteral and enteral nutrition. *JPEN J Parenter Enteral Nutr*. 1998;22:302–310.
24. Conway S, Morton A, Wolfe S. Enteral tube feeding for cystic fibrosis. *Cochrane Database Syst Rev*. 2012;(12):CD001198.
25. Bradley GM, Carson KA, Leonard AR, et al. Nutritional outcomes following gastrostomy in children with cystic fibrosis. *Pediatr Pulmonol*. 2012;47(8):743–748.
26. Medoff-Cooper B, Ravishankar C. Nutrition and growth in congenital heart disease: a challenge in children. *Curr Opin Cardiol*. 2013;28(2):122–129.
27. Mahant S, Friedman JN, Connolly B, et al. Tube feeding and quality of life in children with severe neurological impairment. *Arch Dis Child*. 2009;94(9):668–673.
28. Goulet O, Olieman J, Ksiazyk J, et al. Neonatal short bowel syndrome as a model of intestinal failure: physiological background for enteral feeding. *Clin Nutr*. 2012;32(2):162–171.
29. Olieman JF, Renning C, Ijsselstijn, et al. Enteral nutrition in children with short

bowel syndrome: current evidence and recommendations for the clinician. *J Am Diet Assoc*. 2010;110:420–426.
30. Lambert B, Lemberg DA, Leach ST, Day AS. Longer-term outcomes of nutritional management of Crohn's disease in children. *Dig Dis Sci*. 2012;57(8):2171–2177.
31. Yamamoto T, Nakahigashi M, Umegae S, Matsumoto K. Enteral nutrition for the maintenance of remission in Crohn's disease: a systematic review. *Eur J Gastroenterol Hepatol*. 2010;22(1):1–8.
32. Hosaid SA, La Vega-Talbott M, Solomon GE. Ketogenic diet in pediatric epilepsy patients with gastrostomy feeling. *Pediatr Neurol*. 2005;32:81–83.
33. Bicakli DH, Yilmaz MC, Aksoylar S, et al. Enteral nutrition is feasible in pediatric stem cell transplantation patients. *Pediatr Blood Cancer*. 2012;59(7):1327–1329.
34. A.S.P.E.N. American Society for Parenteral and Enteral Nutrition Board of Directors. Clinical guidelines for the use of parenteral and enteral nutrition in adult and pediatric patients, 2009. *JPEN J Parenter Enteral Nutr*. 2009;33(3):255–259.
35. Andel D, Kamolz LP, Donner A, et al. Impact of intraoperative duodenal feeling on the oxygen balance of the splanchnic region in severely burned patients. *Burns* 2005;31:302–305.
36. McClave SA, Chang WK. When to feed the patient with gastrointestinal bleeding. *Nutr Clin Pract*. 2005;20(5):544–550.
37. Cresci G, Cue J. The patient with circulatory shock: to feed or not to feed? *Nutr Clin Pract*. 2008;23(5):501–509.
38. Fomon SJ. Comparative study of adequacy of protein from human milk and cow's milk in promoting nitrogen retention by normal full term infants. *Pediatrics*. 1960;26:51.
39. Raiha NCR, Heinonen K, Rassin DK, Gaull GE. Milk protein quantity and quality in low-birth-weight infants. I: metabolic responses and effects on growth. *Pediatrics*. 1976;57:659–684.
40. Janas LM, Picciano MF, Hatch TF. Indices of protein metabolism in term infants fed human milk, whey-predominant formula, or cow's milk formula. *Pediatrics*. 1985;75:775–784.
41. Jarvenpaa AL, Rassin DK, Raiha NC, Gaull GE. Milk protein quantity and quality in the term infant. II: effects on acidic and neutral amino acids. *Pediatrics*. 1982;70:221–230.
42. Lama More RA, Gil-Alberdi Gonzalez B. Effect of nucleotides as dietary supplement on diarrhea in healthy infants. *An Esp Pediatr*. 1998;48:371–375.
43. Koletzko B, Baker S, Cleghorn G, et al. Medical Position Paper. Global standard for the composition of infant formula: recommendations of an ESPGHAN coordinated international expert group. *J Pediatr Gastroenterol Nutr*. 2005;41:584–599.
44. Carnielli VP, Luijendijk IH, Van Goudovever JB, et al. Structural position and amount of palmitic acid in infant formulas: effects on fat, fatty acid, and mineral balance. *J Pediatr Gastroenterol Nutr*. 1996;23:553–560.
45. O'Connor DL, Hall R, Adamkin D, et al. Ross preterm lipid study. Growth and development in preterm infants fed long-chain polyunsaturated fatty acids: a prospective, randomized controlled trial. *Pediatrics*. 2001;108:359–371.
46. Groh-Wargo S, Jacobs J, Auestad N, et al. Body composition in preterm infants who are fed long-chain polyunsaturated fatty acids: a prospective, randomized, controlled trial. *Pediatr Res*. 2005;57:712–718.
47. Auestad N, Halter R, Hall RT, et al. Growth and development in term infants fed

long-chain polyunsaturated fatty acids: a double-masked, randomized, parallel, prospective, multivariate study. *Pediatrics*. 2001;108:372–381.
48. Hoffman DR, Birch EE, Birch DG, et al. Impact of early dietary intake and blood lipid composition of long-chain polyunsaturated fatty acids on later visual development. *J Pediatr Gastroenterol Nutr*. 2000;31:540–553.
49. Tantibhedyangkul P, Hashim SA. Medium-chain triglyceride feeding in premature infants: effects on calcium and magnesium absorption. *Pediatrics*. 1978;61:537–545.
50. Tantibhedyangkul P, Hashim SA. Medium-chain triglyceride feeding in premature infants: effects on fat and nitrogen absorption. *Pediatrics*. 1975;55:359–370.
51. Lonnerdal B. Trace element absorption in infants as a foundation to setting upper limits for trace elements in infant formulas. *J Nutr*. 1989;119:1839–1844.
52. Setchell KD, Zimmer-Nechemias L, Cai J, Heubi JE. Exposure of infants to phyto-oestrogens from soy-based infant formula. *Lancet*. 1997;350:23–27.
53. Committee on Nutrition. American Academy of Pediatrics. Soy protein-based formulas: recommendations for use in infant feeding (RE9806). *Pediatrics*. 1998;101:148–153.
54. Mathews DM, Adibi SA. Peptide absorption. *Gastroenterology*. 1976;71:151–161.
55. Koletzko B. Progress of enteral feeding practice over time: moving from energy supply to patient- and disease-adapted formulations. In: Koletzko B, Koletzko S, Ruemmele F eds. *Drivers of Innovation in Pediatric Nutrition*. Nestle Nutrition Institute. Basel, Switzerland: Karger; 2009:41–54.
56. Heyland DK, Novak F, Drover JW, et al. Should immunonutrition become routine in critically ill patients? A systematic review of the evidence. *JAMA*. 2001;286:944–953.
57. Kenler AS, Swails, WS, Driscoll DF, et al. Early enteral feeding in postsurgical cancer patients: fish oil structured lipid-based polymeric formula versus a standard polymeric formula. *Ann Surg*. 1996;223:316–333.
58. Buchman AL. Glutamine: commercially essential or conditionally essential? A critical appraisal of the human data. *Am J Clin Nutr*. 2001;74:25–32.
59. McClave SA, Martindale RG, Vanek VW, et al. A.S.P.E.N. Board of Directors; American College of Critical Care Medicine; Society if Critical Care Medicine. Guidelines for the provision and ssessment of nutrition support therapy in the adult critically ill patient: Society of Critical Care Medicine (SCCM) and American Society for Parenteral and Enteral Nutrition (A.S.P.E.N.). *JPEN J Parenter Enteral Nutr*. 2009;33(3):277–316.
60. Bailey N, Clark M, Nordlund M, Shelton M, Farver K. New paradigm in nutrition support: using evidence to drive practice. *Crit Care Nurs Q*. 2012;35:255–267.
61. Briassoulis G, Filippou O, Hatzi E, et al. Early enteral administration of immunonutrition in critically ill children: results of a blinded randomized controlled trial. *Nutrition*. 2005;21:799–807.
62. Lima AAM, Brito LFB, Ribeiro HB, et al. Intestinal barrier function and weight gain in malnourished children taking glutamine supplemented enteral formula. *J Ped Gastroent Nutr*. 2005;40:28–35.
63. Muto Y, Sato S, Watanabe A, et al. Long-Term Survival Study Group Effects of oral branched-chain amino acid granules on event-free survival in patients with liver cirrhosis. *Clin Gastroenterol Hepatol*. 2005;3(7):705–713.
64. Lord LM, Weser-Maimone A, Pulhamus M, et al. Comparison of weighted versus unweighted enteral feeding tubes for efficacy of transpyloric intubation. *JPEN J Parenter Enteral Nutr*. 1993;17:271–273.

65. Ellett MLC, Croffie JMB, Cohen MD, Perkins SM. Gastric tube placement in young children. *Clin Nurs Res*. 2005;14:238–252.
66. Wilson L, Oliva-Hemker M. Percutaneous endoscopic gastrostomy in small medically complex infants. *Endoscopy*. 2001;33:433–436.
67. Barron MA, Duncan DS, Green GJ, et al. Efficacy and safety of radiologically placed gastrostomy tubes in paediatric haematology/oncology patients. *Med Pediatr Oncol*. 2000;34:177–182.
68. Michaud L, Guimber D, Blain-Stregloff AS, et al. Longevity of balloon-stabilized skin-level gastrostomy device. *J Pediatr Gastroenterol Nutr*. 2004;38(4):426–429.
69. Meert KL, Daphtary KM, Metheny NA. Gastric vs small-bowel feeding in critically ill children receiving mechanical ventilation. *Chest*. 2004;126:872–878.
70. Chellis MJ, Sanders SV, Dean JM, et al. Bedside transpyloric tube placement in the pediatric intenstive care unit. *JPEN J Parenter Enteral Nutr*. 1996;20:88–90.
71. Guitierrez ED, Balfe DM. Fluoroscopically guided nasoenteric feeding tube placement: results of a 1-year study. *Radiology*. 1991;178:759–762.
72. Shike M, Latkany L, Gerdes H, Bloch AS. Direct percutaneous endoscopic jejunostomies for enteral feeding. *Gastrointest Endosc*. 1996;44:536–540.
73. Fortunato JE, Darbari A Mitchell SE, et al. The limitations of gastro- jejunal (G-J) feeding tubes in children: a 9-year pediatric hospital database analysis. *Am J Gastroenterol*. 2005;100:186–189.
74. Graham TW, Zadrozny DB, Harrington T. The benefits of early jejunal hyperalimentation in the head-injured patient. *Neurosurgery*. 1989;25:729–735.
75. Goulet O. Paeditric enteral feeding. *Int Semin Paediatr Gastroenterol Nutr.* 1998;7:1–8.
76. Khoshoo V, Brown S. Gastric emptying of two whey-based formulas of different energy density and its clinical implication in children with volume intolerance. *Eur J Clin Nutr*. 2002;56:656–658.
77. Heymsfield S, Casper K, Grossman GD. Bioenergetic and metabolic response to continuous versus intermittent feeding. *Metabolism*. 1987;36:570–575.
78. Pinchofsky-Devin GD, Kaminsky MV. Visceral protein increase associated with interrupt versus continuous enteral hyper-alimentation. *JPEN J Parenter Enteral Nutr*. 1985;9:474–476.
79. Fruhwald S, Holzer P, Metzler H. Intestinal motility disturbances in intensive care patients pathogenesis and clinical impact. *Intensive Care Med*. 2007;33(1):36–44.
80. Mehta NM, Bechard LJ, Cahill N, et al. Nutritional practices and their relationship to clinical outcomes in critically ill children—an international multicenter cohort study. *Crit Care Med*. 2012;40:2204–2211.
81. Gerlach AT, Murphy C. An update on nutrition support in the critically ill [Review]. *J Pharm Pract*. 2011;24(1):70–77.
82. Chiang YH, Chai DP, Chu SF, et al. Early enteral nutrition and clinical outcomes of severe traumatic brain injury patients in acute stage: a multi-center cohort study. *J Neurotrauma*. 2012;29(1):75–80.
83. Cangelosi MJ, Auerbach HR, Cohen JT. A clinical and economic evaluation of enteral nutrition. *Curr Med Res Opin*. 2011;27(2):413–422.
84. Cabrera AG, Prodhan P, Bhutta AT. Nutritional challenges and outcomes after surgery for congenital heart disease. *Curr Opin Cardiol*. 2010;25(2):88–94.
85. Blewer ES, Zurn, Arnold R, et al. Chylothorax after surgery on congenital heart disease in newborns and infants -risk factors and efficacy of MCT-diet. *J Cardiothorac Surg*. 2010;5:127.
86. de Aguilar-Nascimento JE, BIcudo-Salomao A, Portari-Filho PE. Optimal timing for the initiation of enteral and parenteral nutrition in critical medical and surgical

conditions. *Nutrition*. 2012;28(9):840–843.
87. Braegger C, Decsi T, Dias JA, et al. ESPGHAN Committee on Nutrition: Practical Approach to paediatric enteral nutrition: a comment by the ESPGHAN committee on nutrition. *J Pediatr Gastroenterol Nutr*. 2010;51:110–122.
88. McClave SA. Nutritional support in acute pancreatitis. Failure to utilize the gastrointestinal tract leads to loss of gut integrity and a gut-lung axis of inflammation that generates a secondary systemic inflammatory response syndrome response, further worsening patient outcome. *World Rev Nutr Diet*. 2013;105:160–168.
89. Yi F, Ge L, Zhao J, et al. Meta-analysis: total parenteral nutrition versus total enteral nutrition in predicted severe acute pancreatitis. *Intern Med*. 2012;51(6):523–530.
90. Mekitarian FE, Carvalho WB, Silva FD. Acute pancreatitis in pediatrics: a systematic review of the literature. *J Pediatr*. 2012;88(2):101–114.
91. Fallon EM, Nehra D, Potemkin AK, et al. A.S.P.E.N. clinical guidelines: nutrition support of neonatal patients at risk for necrotizing enterocolitis [Review]. *JPEN J Parenter Enteral Nutr*. 2012;36(5):506–523.
92. Brotschi B, Baenziger O, Frey B, et al. Early enteral feeding in conservatively managed stage II necrotizing enterocolitis is associated with a reduced risk of catheter-related sepsis. *J Perinatat Med*. 2009;37(6):701–705.
93. Bankhead R, Boullata J, Brantley S, et al. Enteral nutrition administration. In: A.S.P.E.N. Enteral Nutrition Practice Recommendations. *JPEN J Parenter Enteral Nutr*. 2009;33:122–167.
94. Bott L, Husson MO, Guimber D, et al. Contamination of gastrostomy feeding systems in children in a home-based enteral nutrition program. *J Pediatr Gastroenterol Nutr*. 2001;33:266–270.
95. Roy S, Rigal M, Doit C, et al. Bacterial contamination of enteral nutrition in a pediatric hospital. *J Hosp Infect*. 2005;59:311–316.
96. Dautle MP, Wilkinson TR, Gauderer MWL. Isolation and identification of biofilm microorganisms from silicone gastrostomy devices. *J Pediatr Surg*. 2003;38:216–220.
97. Byrnes MC, Stangenes J. Refeeding in the ICU: an adult and pediatric problem. *Curr Opin Clin Nutr Metab Care* 2011;14(2):186–192.
98. Thomson FC, Naysmith MR, Lindsay A. Managing drug therapy in patients receiving enteral and parenteral nutrition. *Hosp Pharm*. 2000;7:155–164.
99. Daveluy W, Guimber D, Mention K, et al. Home enteral nutrition in children: an 11-year experience with 416 patients. *Clin Nutr*. 2005;24:48–54.
100. Elia M. Home enteral nutrition: general aspects and a comparison between the United States and Britain. *Nutrition*. 1994;10:115–123.
101. Scott F, Beech R, Smedley F, et al. Prospective, randomized, controlled, single-blind trial of the costs and consequences of systemic nutrition team follow-up over 12 months after percutaneous endoscopic gastrostomy. *Nutrition*. 2005;21:1071–1077.

Parenteral Nutrition

68

Corina Hartman, MD, Arie Riskin, MD, and Raanan Shamir, MD

INTRODUCTION

Providing nutritional support to patients unable to eat has been a dream and a challenge for many centuries before the advent of modern parenteral nutrition (PN). The ability to provide intravenous alimentation has been made possible only through the advances in our understanding of anatomy and physiology, biochemistry and metabolism, microbiology and mostly, the tremendous technological advances that occurred in the 20th century.[1] To these should be added the landmark research in several areas including, but not limited to, the development of accurate methods for analysis of body fluids and body composition, useful for nutritional assessment; the identification of the essential amino acids and their requirements for humans; the use of intravenously infused hydrolyzed protein and emulsified fats for nutritional support. In the 1930s, another major milestone was reached by Cuthbertson's landmark demonstrations of "catabolic response to injury" that revolutionized the approaches to management of severely ill patients.[2,3]

The era of modern PN has been initiated by the pioneer work of several researchers. In 1924, Robert Matas, a New Orleans surgeon, was the first to use a continuous drip infusion of glucose in humans.[4] Progress in protein metabolism and essential amino acids requirements made by William Rose, enabled Robert Elman to provide parenteral replacement of protein by injection of an enzymatic hydrolysate of casein.[5] Using an admixture of glucose, amino acids, and electrolytes, Elman showed improvement in nitrogen balance, serum protein concentration, and clinical status of 35 malnourished patients.[6] Concomitantly, the first intravenous protein hydrolysate solution in Europe was developed by Arvid Wretlind

in 1944.[7] Wretlind and Oscar Schuberth were also the designers of the first readily available fat emulsion from soybean oil with egg yolk phospholipids as an emulsifying agent.[8] Although frequently used in Europe, their use in United States has been banned for several years due to serious adverse effects reported in the previous years with cottonseed oil infusions. In 1968, a new milestone in the history of PN was marked by Stanley Dudrick who used a catheter placed in the superior vena cava (SVC) to deliver a solution of concentrated glucose, along with all other essential nutrients (although, lacking lipid emulsions), in a child with small bowel and colonic atresia. During the 44 days of intravenous nutrition, until "running out" of venous access, the child achieved normal growth and development.[9]

Currently, provision of nutrients parenterally enables short- and long-term growth in infants and children suffering from intestinal failure (IF) or immaturity. Improved knowledge of nutrient requirements and methods of nutrient delivery, and better understanding of prevention and management of complications, have enabled safer and more efficient delivery of PN in hospitals and at home. In 2005, the European Society of Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) and the European Society for Parenteral and Enteral Nutrition (ESPEN) published exhaustive guidelines on PN in children. That document represents the consensus statement of a multidisciplinary working party of professionals, knowledgeable in the management of children treated with PN, and represents so far the most exhaustive collection of data and guiding principles on PN use in children.[10]

INDICATIONS

PN is generally indicated in infants and children who are unable to tolerate adequate oral or enteral feedings to sustain their nutritional requirements. This commonly includes (a) premature infants, due mainly to immaturity of their gastrointestinal tracts, (b) infants or children in whom a period of prolonged fasting is anticipated, that is, necrotizing enterocolitis (NEC), pancreatitis, gastrointestinal graft-versus-host disease, or postoperatively, and (c) pediatric patients with temporary or permanent IF from any reason, that is, congenital diarrheal syndromes, motility disorders, congenital malformations, short bowel syndrome, and so forth. Given the risks and the costs associated with its use, PN should be avoided whenever possible by the appropriate use of oral or enteral nutrition.

When should nutritional support be started? The maximum tolerable

period of fasting depends on the patient's age, nutritional status, and underlying medical conditions. Energy substrate utilization relies on substrate availability during periods of fasting, but depends on the "neurohormonal" milieu and inflammatory/injury mediators elicited during stress. During fasting, most organs derive their energy from the oxidation of fatty acids (FAs) or ketone bodies produced from FAs in the liver. The central nervous system, nevertheless, is an obligate glucose user under ordinary conditions. In the absence of carbohydrate (CHO) intake or significant stored CHO, the only source of glucose is gluconeogenesis from amino acids, most of which are supplied by the breakdown of body proteins. However, there are no reserves of visceral proteins and each body protein has a specific function. Therefore, utilization of proteins for energy purposes results in an obligatory functional insult. In adults, this demand for glucose results in the catabolism of about 75 g of protein (which is about 375 g muscle tissue) a day during fasting.[11]

As starvation progresses, several adaptations take place which aim to conserve lean body mass. The most important of these are (a) gradual decrease in metabolic rate, and (b) progressive rise in serum concentration of ketone bodies. At high concentrations, ketone bodies cross the blood–brain barrier and can be utilized as an energy source by the central nervous system.[12] The adaptation to starvation is limited in sick and fasting patients. Illness and injury increase metabolic energy expenditure and the hormonal milieu of stress favor glycogenolysis and gluconeogenesis.[13] The rise in serum insulin, which accompanies the elevated blood glucose inhibits the increase in serum ketone body concentration that would be used by the brain as primary fuel. Thus, production of glucose and concomitant loss of protein continue at a high rate in sick and starving hospitalized patients.[14]

How can these observations be applied to children? There are very few studies on the physiology of starvation in children, but some reasonable estimates can be made. Protein makes up about 10% (dry weight) of total body weight.[15] In infants, about half the total protein is in muscle and half is found in other tissues. Taking into consideration body composition, basal metabolic rate (BMR), and brain metabolic rate for patients of various ages, one can roughly estimate the number of days in which one-third of body protein will be catabolized in an individual who is totally fasted. This ranges from less than four days in small premature infant to 10–15 days in older children and adolescents, which clearly illustrates the different time frames for nutritional intervention in infants and young children. Furthermore, each day without nutrition support will have

significantly different consequences in premature infants compared to full-term infants, toddlers, children, and adolescents.

Hospitalized patients are rarely totally starved as most intravenous fluids contain glucose, although the same principles hold for "semistarvation." In 1947, Gamble was the first to demonstrate the protein-sparing effect of CHO when he found that nitrogen excretion was considerably reduced in fasted subjects who were given 100–150 g of glucose.[16] This amount of glucose supplies a major part of the energy requirements of the central nervous system, thereby reducing, although not eliminating, the need for gluconeogenesis. Provision of sufficient glucose (100–150 g/d in adults) to meet the metabolic requirements of the brain has been shown to reduce nitrogen losses by half.[17] The nitrogen-sparing effect of CHO may be even greater in children as the brain consumes a much higher proportion of total metabolic expenditure, but this has not been specifically studied. The addition of proteins, vitamins, and minerals to the intravenous infusion will further slow the rate of catabolism of endogenous body protein. However, it has been repeatedly shown, that patients are in negative nitrogen balance unless their complete protein and caloric requirements are met.[18] In practice, if the child is taking nothing by mouth for more than a few days, proteins, vitamins, and minerals should be added on the third to seventh day, depending on age, clinical status, and disease. The exceptions are the very low-birth-weight infants (VLBW) as discussed below. The number of days must be reduced considerably for children who have already lost weight prior to hospitalization or who have increased metabolic requirements.

As part of nutritional care, PN should be provided only after thorough consideration/trial of other means of nutritional support, that is, enteral nutrition. Planning for PN should include a clear statement of PN support goal and patient's further evaluation and follow-up including (a) nutritional requirements, (b) baseline metabolic parameters, (c) nutritional goal (maintain nutritional status, improving nutritional status), (d) anticipated PN duration, (e) venous access including route and type of catheter used for delivery, and (f) monitoring for efficacy and complications of therapy. Despite the obvious benefits of PN, numerous complications (metabolic, infectious, and mechanical) may arise as a result of PN use. Consequently, the multiple risks of PN should be weighed against the benefits of nutrition support prior to the initiation of therapy in each individual patient.

PARENTERAL NUTRITION IN PRETERM INFANTS—GENERAL CONSIDERATIONS

The survival of preterm infants has increased significantly over the last few decades, especially for VLBW infants weighing less than 1500 g. The goal of nutritional support for the VLBW infant is to achieve a postnatal growth rate that approximates that of a normal fetus at the same gestational age. Nutrition aims to mimic intrauterine growth and body composition, and equally important to obtain functional outcome comparable to infants born at term.[19–21]

Human growth is unique due to special emphasis on the protection of brain development. The period between 20 and 32 weeks after conception is one of rapid brain growth and development. Illness, infection, or undernutrition during this critical time period may compromise neurodevelopment and affect later outcome. The clinical consequences may include motor problems, principally cerebral palsy, visual and hearing impairments, learning difficulties, and psychological, behavioral, and social problems.[22] Thus, the importance of providing early nutrition to preterm infants is related not only to short-term outcomes, but has implications on long-term development as well.[23] Nutritional strategy in preterm infants should be directed to achieve the best possible outcomes in terms of body growth and composition and neurodevelopment.[24,25]

The current focus of nutritional science has shifted from meeting nutritional needs to the biological effects of nutrition on short term and lifetime health. The term “programming” was coined to emphasize the long-term effects of nutritional and environmental factors especially for preterm infants. Programming relates to a stimulus or insult during a critical or sensitive period of development that can have long-term effects. In that context, there is ample evidence that nutrition in early life has a major impact on long-term health and development, notably on growth, cognitive function, bone mineralization, and cardiovascular health.[26] Nutritional insults at a vulnerable period of brain development are associated with effects on brain size, cell number, and function. Although slow growth below the intrauterine rate might have some benefit for later cardiovascular outcome, it carries the risks of undernutrition and its adverse consequences, and has profound adverse effects on later cognition.[27] Thus, in contrast to hypothetical reservations regarding catch-up growth in full-term infants, the current accepted concept is to provide the best optimal nutrition possible and try to achieve fetal accretion rates in premature infants and disregard the potential long-term disadvantages of

catch-up growth. The immediate postnatal period is specifically important, as the old practice of providing intravenous fluids with very little nutritional value has been shown to have negative long-term outcomes on growth and neurodevelopment.[21]

To minimize the harmful effects of near starvation associated with the interruption of fetal nutrient supply at and after delivery, the approach to nutritional support of preterm infants has been switched to "early aggressive nutrition." According to this approach full nutrient supply should be delivered to the preterm infant as soon as possible to prevent the catabolic state associated with delivery and separation from intrauterine nutrient supply as well as the morbidities associated with premature delivery (e.g., respiratory distress syndrome).[20,28–30] Delivering adequate amounts of nutrients to premature infants is challenging at all times because the infant's immature gastrointestinal tract is initially unable to accept enteral feedings, necessitating the use of PN. This is especially true for VLBW sick preterm infants who are usually dependent on PN during their first weeks of life. Not surprisingly, neonates represent today one of the largest age group of patients receiving PN.[31]

Protein intake is the major driving force of weight gain and brain development in VLBW infants and deficits and growth deficits start already in the first week of life.[32,33] Early amino acids administration improves outcome by providing substrate for tissue growth (especially the brain), and for protective glycoproteins, such as glutathione.[34,35] Early PN, started as soon as possible after delivery, is mandatory for VLBW premature infants in order to provide the required high protein intake and to prevent negative nitrogen balance. These tiny preterm infants can tolerate up to 3.5–4.0 g/Kg/d within the first days of life with only relatively mild transient increase in serum urea nitrogen and no significant acidosis.[35,36] As the minimum protein intake needed to prevent tissue breakdown is 2.0–2.5 g/kg/d, probably the best practice is to start with at least 2.0–3.0 g/kg/d amino acids as soon as possible following birth, and advance to full protein intake of 3.5–4.0 g/kg/d of amino acids within 3–4 days.[37] Early high amino acids' intake requires higher energy intake.[37] This implies that CHO–amino acids PN admixture solutions are not sufficient, and addition of lipids is needed very early in life, probably from the first day, to provide the extra energy.[38] Lipids provide not only energy for growth, but also essential fatty acids (EFAs), that is, long chain polyunsaturated fatty acids (LC-PUFA), especially docosahexaenoic acid (DHA), which are particularly important for brain and retinal development

of premature infants.

Early PN in premature infants should provide (a) adequate protein intake and supply of essential amino acids to encourage nitrogen retention that is essential for growth and brain development; (b) adequate calories for energy expenditure, protein synthesis, and growth; (c) CHO to prevent hypoglycemia and provide energy supply; (d) lipids to meet the caloric needs and supply of essential FAs; (e) essential nutrients including electrolytes, minerals (especially calcium and phosphate), trace elements (TEs), and vitamins.[39] Nutrition support in VLBW infants should prevent growth failure and improve extrauterine growth,[40] decrease morbidity and mortality, and support better short as well as long-term outcomes,[23] especially neurodevelopmental outcomes.[41]

ENERGY REQUIREMENTS

The fundamental cornerstones of nutritional support are prevention of weight loss in the short term and promotion of growth in the long term. Providing the energy needs is fundamental both in health and disease. Positive energy balance is associated with energy storage and tissue deposition, weight gain, and adequate linear growth. Negative energy balance is associated with depletion of tissue and thus weight loss, and if prolonged, leads to poor linear growth. Although protein is a potential energy substrate, it should be utilized only for tissue synthesis (accretion or renewal) and not as an energy source. The recommendations for energy needs in children usually include the protein contribution to caloric intake as well as fat and CHO, and as such are referred to in this chapter. Ideally, the distribution of calories should be 45%–55% CHO, 10%–15% proteins, and 35%–40% fats.[42]

Total daily energy expenditure (TEE) is the sum of the BMR, diet-induced thermogenesis (DIT), the energy spent on physical activity, and losses in urine and stools (obligatory losses). BMR, the largest (60%–70%) component of TEE, represents the energy needed to maintain essential body functions and tissue and cellular integrity. BMR can be considered as the energy expended by a fasting individual at rest in a thermoneutral environment, and is similar to measured resting energy expenditure (REE). The energy expended for physical activity is the second largest component of TEE, and the most variable accounting for 15% to 40% of TEE. DIT accounts for a small proportion of daily energy expenditure, the remainder being the energy cost of growth and obligatory losses.

REE varies between individuals, largely because of differences in body

size and composition. A large child has a higher REE than a small child. The difference in REE between two individuals is principally determined by the amount of fat-free mass (FFM). The variation in fat mass (FM) and FFM may have a significant impact on a child's energy requirements: a child with a high FFM is likely to have a higher energy requirement compared with a child with a high FM.[43] Moreover, FFM contains both internal organs and skeletal muscle. In a healthy child, organ mass is more metabolically active than skeletal muscle, therefore, organ mass contributes more to total REE than skeletal muscle. Boys tend to have higher REE than girls even when corrections for differences in body composition have been made. Several studies have demonstrated differences in REE in children from different ethnic backgrounds.[44] REE significantly decreases with age independent of other factors, partially because of the decrease in energy cost needed for tissue replacement with advanced age.

DIT is the ''obligate'' energy cost of processing food (e.g., chewing, peristalsis, digestion, absorption, postabsorptive processing). DIT represents the increase in energy expenditure that occurs after consumption of a mixed meal. DIT is a small component of TEE and is particularly difficult to measure. Therefore, by convention it is assumed to be about 10% of TEE.[45] Abnormalities in DIT are unlikely to be important contributors to variations in energy balance. DIT varies, depending on the macronutrient composition of the diet but also on body size and FM (nutritional state) of the child. When malnourished children experience rapid weight gain, they have a larger increase in energy expenditure after a meal compared with that when there is no weight gain or there are slower rates of weight gain.[46] This is likely the result of a higher energy requirement to synthesize new tissue during more rapid (catch-up) growth. During PN administration, DIT and the respiratory quotient (RQ) are affected by the mode of PN administration (continuously vs. cyclic).[47,48]

The energy cost of growth is inherently difficult to determine as growth is variable and influenced by age, pubertal status, illness, medications, and obviously nutritional status. Energy needs for growth contain two components: (1) the energy used to synthesize growing tissues, which is part of the total energy expenditure; and (2) the energy deposited in those tissues, basically as fat and protein. Studies in infants and young children recovering from malnutrition have estimated that the energy cost of growth is between 4 and 6 kcal/g of tissue deposited, with most of this attributable to the energy deposited (stored) and approximately 1 kcal/g being the energy cost of synthesis. However, there

are no studies to estimate the energy requirements of catch-up growth in older children and adolescents.[46,49] In clinical practice, during recovery from acute illness or in chronic disease, the children are usually supplied with 120%–150% of energy requirements to ensure adequate catch-up growth.

The measurement of physical activity is inherently difficult because of daily variation, but estimates can be made over several days with the use of accelerometers, heart rate monitors, or diaries.[50] An acutely unwell child will have very little physical activity, but as soon as recovery starts, their activity level will quickly return to normal. Obese children are difficult to assess because despite the sedentary lifestyle, the mechanical work done by their muscles to produce movement can be quite large. In general, obese subjects tend to underestimate intake and overestimate physical activity, whereas children with anorexia nervosa typically underreport the amount of exercise they perform and overestimate their intake.

Energy requirement can be measured directly using different techniques for TEE, REE, or physical activity. TEE can also be estimated by adding together the three component parts: BMR, DIT, and the energy expenditure of activity (activity thermogenesis).

TEE can be measured by using doubly labeled water (DLW) method. The use of the DLW ($^{2}H_2{}^{18}O$) technique to calculate total production of carbon dioxide (CO_2) over several days and, from this, total energy expenditure was originally developed for use in small mammals,[51] and its application was later validated in humans.[52] The subject ingests water with labeled isotopes of hydrogen and oxygen. The hydrogen-labeled isotope can only be cleared from the body through water loss, whereas the oxygen isotope is excreted through water loss and CO_2 production. By measuring the relative amounts of each isotope and the amount of CO_2 production, energy expenditure can be calculated. Although this method is considered the gold standard in TEE assessment, it is still subject to imprecision particularly in disease states.

REE can be measured by indirect calorimetry (IC) or determined using predictive equations, which are easily accessible without the need to use specialized equipment and appliances. The measurement of energy expenditure is the most accurate method to assess energy needs, especially in acutely ill patients. IC uses the measurement of carbon dioxide production (VCO_2) and oxygen (VO_2) based on the assumption that all

inspired oxygen is used to oxidize substrates and that all the expired CO_2 is produced from these reactions. Total average daily energy expenditure in kcal is usually calculated using the modified Weir equation with substitution of the measured VO_2 and VCO_2 values.[53] The urinary nitrogen component (uN_2) is often excluded when calculating energy expenditure because it only accounts for about 4% of the true energy expenditure and contributes to a small error of 1%–2% in the calculation of final energy expenditure in both inpatients and outpatients.[54] Thus the abbreviated equation is commonly used[55]:

Energy expenditure (kcal/d) = $[(VO_2 \times 3.941) + (VCO_2 \times 1.11)] \times 1440$

The technique requires adherence to strict conditions including environmental temperature, fasting, and rest before measurement to obtain measurements close to BMR.[56]

In addition to measurement, REE can be estimated from many equations. Several such equations have been developed to calculate REE, BMR, and TEE. Of these, the WHO equations (1985), Schofield (1985), and Harris Benedict (1919) equations are the most commonly used.[57–60] New equations for BMR estimation, Oxford equations, have been published recently.[61] The Oxford equations are based on the data available from 10,552 subjects (5794 males and 4702 females) from 166 separate investigations and report calculated BMR for a range of body weights and ages (0–3, 3–10, 10–18,18–30, 30–60, >60 years) for both males and females. For all available equations, the main predictor for each component of energy expenditure is body weight whereas height also accounts for some of the variability in energy needs (Table 68-1). These predictions of REE are derived at a population level and do not necessarily correlate with measured REE of an individual patient.

The performance of these equation compared to the actual measured REE has been estimated in several studies in healthy children populations. WHO and Schofield-wt/Ht have both underestimated[62] and overestimated[63] REE in healthy infants. Harris Benedict equation overestimates REE in infants and adults too.[64] In older children, 5–16 years, both WHO and Schofield-wt equations have been shown to be reasonably accurate.[65] The Schofield equations also provided the best estimates in another study in 7.8–16.6 years healthy children.[66] Overall the equations are not reliable in children with abnormal growth or body composition, but in children with failure to thrive, the Schofield-wt/Ht was

found to be the best predicting equation.[67]

The referred equations were never intended to predict REE in patients with complex disease processes. In sick children, the differences in actual energy needs versus calculated needs based on general equations arise from the special status of the patient, that is, energy losses from malabsorption, diarrhea, ostomies, underlying disease, inflammation, infection, impaired body composition (decreased lean body mass due to increased catabolism), and different energy routes of supplementation (oral, enteral feeding, continuous vs. intermittent feeding, and PN). Caution is also needed in applying estimates to individuals in whom height, weight, and physical activity deviate significantly from the pattern of the reference population. In addition, a hospitalized child lying in bed has reduced physical activity and growth is usually altered, further reducing the TEE. Measurement of REE in 398 chronically ill children found a wide variability for % measured REE/predicted REE (SD ± 27.2) and poor agreement between measured REE and predicted REE, as more than 50% of the children in this cohort would have been underfed if their daily energy requirements were prescribed based on predicted REE.[68] As most of the children in need of PN suffer from one or more of the above, the estimated energy needs based on current equations may be incorrect. However, a study in stable children on home PN found no difference in daily EE compared to age and gender matched controls.[69]

TABLE 68-1 Equations Used for Calculating REE/BMR (kcal/day) in Children 0–18 Years Old

Source	Gender and Age	Equations
Harris–Benedict[58]	Females	REE = 655.0955 + 9.5634*wt + 184.96*ht − 4.6756*age
	Males	REE = 66.4730 + 13.7516*wt +500.33 *ht − 6.7550*age
FAO/WHO/UNU[57]	Females 0–3	REE = 61*wt − 51
	Males 0–3	REE = 60.9*wt − 54
	Females 3–10	REE = 22.5*wt + 499
	Males 3–10	REE = 22.7*wt + 495
	Females 10–18	REE = 17.5*wt + 651
	Males 10–18	REE = 12.2*wt + 746
Schofield[59]	Females 0–3	BMR = 16.25*wt + 1023.23*ht − 413.5

	Males 0–3	BMR = 0.167*wt + 1517.43*ht − 617.6
	Females 3–10	BMR = 16.97*wt + 161.8*ht + 371.2
	Males 3–10	BMR = 19.60*wt + 130.3*ht + 414.9
	Females 10–18	BMR = 8.365*wt + 465*ht + 200
	Males 10–18	BMR = 16.25*wt + 137.2*ht + 515.5
Oxford[61]	Females 0–3	BMR = 30.4*wt + 703*ht − 287
	Males 0–3	BMR = 28.2*wt + 859*ht − 371
	Females 3–10	BMR = 15.9*wt + 210*ht + 349
	Males 3–10	BMR = 15.1*wt + 74.2*ht + 306
	Females 10–18	BMR = 9.4*wt + 249*ht + 462
	Males 10–18	BMR = 15.6*wt + 266*ht + 299

All equations are for kcal/day; wt, weight in kilograms; ht, height in meters; age in years.

Both approaches (measuring REE and calculating REE) are used in clinical practice. Whatever method is used, the need to make corrections for physical activity, DIT, and energy losses increases the likelihood of inappropriate estimates. Thus, adjustments are needed, according to the clinical situation and by observed responses to treatment (weight gain/growth). Providing nutrient supply by PN should also take into consideration the recommended intake of the different macronutrients (lipids, CHO, and protein) as well as possible intolerance and complications (hyperglycemia, hypertriglyceridemia, liver enzyme abnormalities, cholestasis). Average daily parenteral energy intakes per kg body weight considered adequate for a major proportion of patients are shown in Table 68-2.

Nutritional Support in Pediatric Intensive Care Unit

Children admitted to the pediatric intensive care unit (PICU) exhibit unpredictable metabolic response to critical illness depending on its nature and severity. Adequate nutritional support is critical for these children, as both overfeeding and underfeeding are associated with complications.[70] Pollack et al. reported that 16% to 20% of critically ill hospitalized children developed significant acute protein-energy malnutrition, some within 48 hours of admission to PICU.[71] Malnutrition is associated with decreased immunocompetence, increased risk of infection, poor wound healing, increased morbidity, and increased mortality, whereas overfeeding increases the risk of hyperglycemia, high RQ, and fatty liver.[71]

TABLE 68-2 Estimated PN Requirements for Energy,

Amino Acids, Fat and Glucose Supply					
	Preterm	0–12 Months	1–7 Years	7–12 Years	12–18 Years
Energy, kcal/kg/d	110–120	90–100	75–90	60–75	30–60
Amino acids[a,b], g/kg/d	1.5–4.0	1.5–3.0 < 2 months 1.0–2.0 > 2 months	1.0–2.5[a] < 3 years 1.0–2.0 > 3 years	1.0–2.0	1.0–2.0
Lipids[c,d], g/kg/d	1–4	3–4	2–3	2–3	2–3
Glucose[e], g/kg/d	8–18	16–18	10–14	10–12	<10

[a]A minimum of 1.5 g/kg/d (preterm and term newborns) to 1 g/kg/d (older children) amino acids is necessary in to prevent negative nitrogen balance.

[b]For critically ill older children the protein intake may be as high as 3 g/kg/d.

[c]The lower limit necessary to prevent EFAD is estimated at 0.25 g/kg/d in premature infants and 0.1 g/kg/d in term infants and older children, of linoleic acid. The intravenous lipid emulsion (ILE) dose necessary to prevent EFAD should be calculated according to ILE composition used, and is different for the different ILE formulations. For example, if the ILE is Intralipid (100% soybean oil, 53% linoleic acid), the minimal lipid intake in a premature infant is 0.5 g/kg/d and 0.2 g/kg/d in infants and older children. If the ILE is Clinoleic (20% soybean oil, 18.5% linoleic acid), the needed lipid intake necessary to avoid EFAD in a premature infant is 1.8 g/kg/d and 0.5 g/kg/d for full term and older children. For Lipofundin (50% LCT/50% MCT, 27% linoleic acid) the lipid intake necessary to avoid EFAD in a premature infant is 0.9 g/kg/d and 0.4 g/kg/d for full term and older children. For SMOFlipid (21.4% linoleic acid), the needed lipid intake necessary to avoid EFAD in a premature infant is 1.16 g/kg/d and 0.5 g/kg/d for full term and older children.

[d]Parenteral lipid delivery should be limited below 0.13–0.17 g/kg/hour in infants and 0.08–0.13 g/kg/hour in older children.

[e]The table shows the maximal glucose supply by age, under standard conditions. The glucose supply should, however, be adapted to child' clinical and nutritional status, type of PN delivery (continuous or cycling) and the presence of additional medications.

In some critically ill adults, it was shown that a hypermetabolic state occurs during critical illness, characterized by significantly greater energy expenditure than REE in the non-stressed state.[72] Extrapolating from the adult studies, it has been suggested that the energy requirements in critically ill children should also be estimated by adding a stress-related correction, "stress factor," to the REE. Stress or activity "correction factors" traditionally have been added to energy requirement estimates to adjust for the nature of illness, its severity, and the activity level of hospitalized subjects. The hypermetabolic state reported in adults is less prevalent in critically ill children. In addition, children admitted to PICU may have significant reduction in true energy expenditure, attributable to multiple factors including decreased activity, sedation, mechanical ventilatory support, and transient absence of growth during the acute illness. These patients may be at a risk of overfeeding when estimates of

energy requirements are based on age-appropriate equations developed for healthy children, especially if stress factors are incorporated.[73] Thus, the application of predetermined values of correction factor based on broad diagnostic groups of patients in the PICU is likely to be inaccurate and may increase the risk of overfeeding or underfeeding.[74,75] Studies in critically ill children have shown that REEs using presently available equations either overestimate the true energy expenditure or underestimate the energy requirements.[76–79] Tilden et al. and Coss-Bu et al. reported that measured REE was much higher than the predicted BMR estimated by either the Harris-Benedict or the Talbot equations.[80,81] In general, the estimates from prediction equations compare poorly to measured values; calculated values have shown an error range[82] of 7% to 55%.

In conclusion, it is recommended that in critically ill children, REE be measured rather than calculated due to individual variability and over or underestimation of energy requirements by the predicting equations.[83] Unfortunately, multiple factors have limited the application of this testing in everyday clinical practice, and IC is only sporadically used in the PICU. In a review of PICU practice across 111 centers in Europe, only 17% of centers reported the regular use of IC to measure energy expenditure.[84] Resource and technical restraints seem to preclude regular IC testing in most units. Selecting candidates for IC according to American Society for Parenteral and Enteral Nutrition (ASPEN) recommendations, a recent study in 150 pediatric ICU patients, showed that three of four patients were candidates for IC. Children less than 2 years of age, malnourished (underweight/overweight) on admission, or with >5 days ICU were at particular risk for nutritional imbalances, therefore, should be above all candidates for IC. Further studies are needed to examine the effectiveness of a model of selective use of IC and its impact on patient outcomes. Application of IC at the bedside to provide nutrition intake that is tailored to individual patients and adjusted dynamically to their illness course is a desirable goal in the future.[85]

Energy Requirements in Preterm and VLBW Infants

In preterm low-birth-weight (LBW) infants, energy requirements should cover both energy expenditure and energy needed for growth. Estimates of energy requirements are dependent on protein intake,[21] because energy is required for both protein metabolism (i.e., avoiding catabolism) and deposition (i.e., linear growth).[20] To achieve optimal growth, the premature infant on PN needs 90–100 kcal/kg/d. This estimate is based on

the minimum energy requirements that allow unlimited growth of lean body mass. Above this intake, the energy is presumably stored in adipose tissue.[21] Out of this, 40–60 (average 50) kcal/kg/d account for REE.[86,87] This is the minimum energy required to avoid protein catabolism when 1.5 g/kg/d protein intake is supplied, that is, the minimum protein intake required to induce positive nitrogen balance from the first day of life.[37] Energy expenditure appears to be greater with lower birth weights, associated morbidities, advancing postnatal age, and as the infant grows and gains weight.[86–88] To support further growth, an additional 20–25 kcal/kg/d should be provided in addition to the daily energy expenditure.[89] This results in a total of 90–100 kcal/kg/d of energy required for preterm infants on PN in the first days of life, considered as transitional period. It must be remembered that VLBW preterm infants on PN have lower energy requirements compared to infants on enteral nutrition (90–100 kcal/kg/d vs. 120–130 kcal/kg/d, respectively) due to less fecal energy losses, less energy required for activity and lower energy requirements for thermoregulation, as these infants are usually kept in neutral thermal environment at this stage. Later on, energy intake of 110–120 kcal/kg/d is required for optimal weight gain and growth in VLBW infants on PN. This is supplied by combined administration of lipid and glucose and can usually be achieved only after several days from birth, because of limited glucose and lipid tolerance, and the need to avoid hyperglycemia and possibly aggravate respiratory status.[21,90] Current recommendations endorse provision of 40 kcal/kg/d on the first day of life, 60–80 kcal/kg/d in the following days, and a target goal of 90–100 kcal/kg/d from PN by the end of the first week of life.[10] Balanced distribution of calories from CHO and lipids is also important as it favors protein accretion[91] and avoids protein oxidation that may occur if high glucose is given parenterally with low or without lipids.[92] Although the gold standard for caloric distribution is human milk (40%–45% calories from CHO and lipids), the energy distribution usually achieved in preterm infants' PN covers 45%–55% of the calories from CHO, 35%–40% from lipids, and 10%–15% from protein (amino acids). Keeping average protein/energy ratio of 3.3 g protein/100 kcal (range 2.7–3.9 for PN, higher ratios for lower gestational ages and birth-weights) is essential to maximize protein accretion, improve lipid and protein tolerance, and avoid acidosis.[20,21]

PROTEIN REQUIREMENTS

Proteins serve as the main structural element of the body. In addition, as biochemical catalysts and regulators of gene expression, proteins are essential elements for most metabolic functions. Proteins are complex structures that consist of chains of two to thousands amino acid subunits joined together by peptide bonds. There are 20 amino acids from which all proteins are synthesized. If these cannot be endogenously synthesized in sufficient amounts then dietary supply becomes essential. Classically, in healthy adults nine amino acids are regarded as dietary essential (or indispensable). During growth or during illness several other amino acids are regarded as conditionally essential (Table 68-3). If amino acids are not present in the appropriate proportions then protein synthesis will be reduced.[93,94] Furthermore, it has been shown that whole body protein breakdown is increased when less balanced mixtures of amino acids are fed.[95] In this case, the endogenous amino acids provide the missing amino acids for protein synthesis.[95,96]

TABLE 68-3 Essential, Conditionally Essential, and Nonessential Amino Acids

Essential Amino Acids	Conditionally Essential Amino Acids	Nonessential Amino Acids
Histidine	Arginine	Alanine
Isoleucine	Cysteine	Serinene
Leucine	Glycine	Aspartic acid
Lysine	Proline	Asparagine
Methionine	Tyrosine	Glutamic acid
Phenylalanine		Glutamine
Threonine		
Tryptophan		
Valine		

Clinical assessment of the adequacy of protein supply is done most easily in childhood through sequential height and weight measurements. Additional useful clinical tools are measurement of mid-upper arm parameters (mid-upper arm circumference and triceps skin-fold thickness) from which muscle area and circumference can be calculated. Body composition measurements using bone densitometry or bio electric impedance (BIA) are seldom done routinely and are usually part of a research protocol.

The classical approach to biochemical assessment of protein status has

been nitrogen balance studies in which excretion of nitrogen waste products in urine and feces are subtracted from dietary protein (nitrogen) intake.[97] This technique requires complete collections of urine and feces for three days and biochemical measurements which are not readily available in the clinical setting. A ‘‘partial nitrogen balance’’ can be obtained by measuring urea nitrogen in urine instead of total nitrogen. This wrongly assumes that urea nitrogen is a fixed proportion of total nitrogen.[98] For children, protein allowances were traditionally considered to be the amount of protein needed for sustaining positive nitrogen balance and therefore, growth. Recently, protein requirements have begun to encompass outcomes other than growth (i.e., immune function) and a more critical evaluation of protein requirements for children has been recommended.

Various blood proteins have been examined as possible markers of protein status. These include albumin, transferrin, prealbumin, and retinol-binding protein. Serum albumin represents the equilibrium between hepatic synthesis and albumin degradation and body losses. It is also influenced by intravascular and extravascular albumin compartments and water distribution (about one-third of the albumin pool is in the intravascular compartment, and two-thirds is in the extravascular compartment). Once albumin is released into plasma, its half-life is about three weeks and can be used for long-term monitoring. Serum albumin is a good predictor for outcome and thus reflects disease severity. However, albumin is a poor marker to assess nutritional status in inflammatory states. Prealbumin and retinol-binding protein, with half-lives of two days and 12 hours, respectively are regarded as being mostly reflective of acute protein intake. However, these markers also suffer from the influence of other nonnutritional factors such as hepatic and renal failure, hormone effects, infection, and inflammation.[99]

A number of different tracer techniques and protocols have been applied with the purpose of determining the requirements for specific indispensable amino acids. The advent of amino acid oxidation studies, and in particular, the indicator amino acid oxidation (IAAO), which is the current gold standard method, has been successfully applied to identify amino acid requirements in adults.[100] This method is based on the concept that when one indispensable amino acid is deficient in the diet, then all other amino acids, including the indicator amino acid, usually L-(1-^{13}C) phenylalanine, will be oxidized. With increasing intake of the limiting amino acid (or total protein), oxidation of the indicator amino acid decreases, reflecting increasing incorporation of the amino acid into

proteins. Once the requirement is met for the limiting amino acid, there will be no further change in the oxidation of the indicator amino acid. The inflection point at which the oxidation of the indicator amino acid stops decreasing and reaches a plateau is referred to as the “break point.” The break point identified with the use of two-phase linear regression analysis indicates the mean or estimated average requirement (EAR) of the limiting amino acid or total protein.[101] The initial application of IAAO to studies in humans involved intravenous indicator administration, blood sampling, and the use of up to nine days of amino acid–based experimental diet. To enable studies in children, the IAAO method has been adapted by administering the tracer orally, using urine instead of arterial blood sampling, and reducing the adaptation period to the experimental diet to four hour.[102] The minimally invasive IAAO have thus enabled the estimation of amino acid requirements in children[103,104] and neonates[105,106] over a shorter period of time and allowed the study of the test amino acid in 8–9-hour and over a range of 6–8 levels of the tested amino acid.

The amino acid utilization and therefore, requirements are different when provided via the enteral vs. the parenteral route. Although parenterally infused amino acids enter directly into the circulation, peptides and amino acids within the intestinal lumen undergo a variety of transformations, including utilization by enterocytes and conversion to other amino acids, within the enteric mucosa and liver.[107] Thus, the pattern of amino acids entering the circulation of the enterally fed individual reflects not only the pattern of the protein intake but also the extent of digestion and, subsequently, metabolism within the enteric mucosa and liver. In contrast, the pattern of individuals receiving only parenteral nutrients more closely reflects the amino acid pattern of the infused mixture.

The currently available pediatric parenteral amino acid mixtures were designed based on considerations of food protein composition (e.g., human milk proteins) and plasma pattern of amino acids observed in normally growing, breast-fed infants.[108,109] Their composition was designed to mimic the optimal amino acids profile based on fetal and neonatal cord blood amino acids concentrations (i.e., according to the factorial approach that uses estimations of weight gain and the composition of fetal tissues) (Primene) or based on plasma amino acids concentrations of healthy 30 days old breast-fed term infant (i.e., according to the approach that uses human breast milk as the gold standard) (TrophAmine). The rationale was

that this pattern reflects transformations of dietary protein and amino acids by enteric and hepatic metabolism as well as ongoing protein synthesis and breakdown of normally growing infants and, hence, should also be optimal for infants requiring parenteral nutrients.[110]

In addition to the eight classically indispensable amino acids and histidine, infants also require cysteine and tyrosine which most adults can synthesize from methionine and phenylalanine, respectively.[111] The requirement for cysteine is thought to result from low or absent hepatic activity of cystathionase, the rate-limiting enzyme for endogenous synthesis of cysteine from methionine. The hepatic activity of the rate-limiting enzyme for endogenous synthesis of tyrosine from phenylalanine, that is, phenylalanine hydroxylase, achieves adult levels early during gestation; thus, the reason for the apparent requirement for tyrosine in infants is not clear. The composition of different pediatric amino acids solutions compared to adult solutions is presented in Table 68-4.

Accretion of body protein in infants and children is dependent on the optimal intake of all of the indispensable and dispensable amino acids and adequate nonprotein energy. Energy supply of 30 to 40 kcal per 1 g amino acids is usually recommended. Stanley Dudrick originally conceived of PN as hyperalimentation, and provided nutrients in excess of normal requirements for patients with increased metabolic demands. The first infant to receive modern TPN, reported by Wilmore and Dudrick in 1968, was given 4 g/kg/d of protein hydrolysate.[9] Nitrogen retention of 0.200–0.300 g/kg/d and satisfactory weight gain were found in all patients when protein intake ranged from 2.0 to 5.0 g/kg/d.[112–115] Fox et al., however, reported optimal weight gain of 10–15 g/d in all babies with a protein intake of 1.5–2.7 g/kg/d.[116] As excessive supply of protein can have harmful effects it is important to achieve positive nitrogen balance with adequate amount and mixture of amino acids supply. The parenteral amino acids requirements for different age groups are presented in Table 68-2. No data endorse the common practice of gradually advancing either the amino acid or lipid dose.

Duffy et al. reported that amino acid and nitrogen utilization is enhanced in parenterally fed neonates when the energy intake is increased from suboptimal (60 kcal/kg/d) to optimal level (80–90 kcal/kg/d) by the addition of intravenous lipid to the amino acids and glucose.[95] However, other studies that investigated the optimal nonprotein fuel mix (glucose vs. lipid) for parenterally fed neonates showed no differences in nitrogen retention between the two energy sources.[117,118] Bresson et al., showed

however, that protein turnover, protein breakdown, and amino acid oxidation rates were significantly higher when glucose only was the energy substrate, compared to glucose-lipid supply, whereas protein-synthesis rates did not differ significantly.[91] These results suggest that the nature of energy substrates delivered to parenterally fed infants may affect protein metabolism.

TABLE 68-4 Amino Acid Composition of Several Parenteral Formulations for Infants and Children as Compared to Human Milk

	TrophAmine[a] (B.Braun)	Primene[a] (Baxter)	AminosynPF[a] (Abbott)	Travasol[b] (Baxter)	FreAmine[b] (B. Braun)	Aminoplasmal[b] (B. Braun)	Human Milk
Essential and Conditionally Essential Amino Acids							
Isoleucine	820	670	760	600	590	510	550
Leucine	1400	990	1200	730	770	890	1050
Lysine	820	1090	677	580	620	700	940
Methionine	340	240	180	400	450	380	160
Phenylalanine	480	420	427	560	480	510	510
Threonine	420	370	512	420	340	410	470
Tryptophan	200	200	180	180	130	180	190
Valine	<16	760	500	673	560	480	550
Cysteine	780	190	—	—	<14	—	240
Histidine	480	380	300	312	240	520	240
Tyrosine	240	45	240	40		30	590
Nonessential Amino Acids							
Alanine	540	800	698	2070	600	1370	390
Arginine	1200	840	1227	1120	810	920	490
Proline	680	300	722	812	950	890	960
Serine	380	400	530	495	500	240	480
Glycine	360	400	722	385	1190	790	250
Aspartate	320	600	700	527		130	990
Glutamate	500	990	738	820		460	1690
Taurine	25	60	—	70		—	30
Ornitine		249	—	—		320	—
Total amino acids (g/L)	100	100	100		85	100	
PH, average (range)	5.5 (5.0–6.0)	5.5	5.0 (5.0–6.5)		6.5 (6.0–7.0)		
Osmolarity (mOsm/L)	875	840		718	810	885	

All values as mg/100 mL. Data as provided by manufacturers.

[a]Recommended for infants less than 1 year.

[b]Recommended for children older than 1 year through adulthood.

The knowledge about safe levels of enteral amino acid intakes is sparse, and similarly there is limited knowledge of parenteral amino acids toxicity, although hyperammonemia, acidosis, and seizures have been well described.[108,119] Moss and colleagues have demonstrated that methionine

is hepatotoxic in a rabbit model in which animals received large amounts of parenteral methionine with or without enteral feedings.[120] L-Arginine, through L-ornithine production, is known to induce necrotizing pancreatitis in a rat model,[121] and L-lysine at large doses induces renal failure in dogs.[122]

Specific amino acid requirements are altered in many diseases, and recommendations for the general population may not apply in these conditions. Furthermore, unnecessary restrictions may cause aggravation of malnutrition.[123] In a retrospective study, Verbruggen et al. compared the parenteral amino acids intake in 116 critically ill children, with essential amino acids intakes recommended by the Institute of Medicine (IOM). The study found that the amino acid compositions of commercially available parenteral amino acid formulas provided to critically ill children are variable and unbalanced to support inflammatory/immune protein synthesis, resulting in excessive or deficient supply of amino acids compared with IOM recommendations, breast milk, and mixed muscle proteins.[124] Because there is no tailor-made recipe for amino acid supplementation in various disease states, periodic assessment of protein nutritional status for adequacy, including anthropometry and serum biomarkers is recommended.

Protein Requirements in VLBW Infants

Early high protein intake is the cornerstone of "early aggressive nutrition" (discussed above as part of the general approach to PN in preterm infants). Early high protein intake during the first week of life induces positive nitrogen balance, protein accretion, and also increases endogenous insulin secretion, thus improving glucose tolerance and preventing hyperglycemia.[20] Recent recommendations suggest providing 2–3 g/kg/d of amino acids on the first day of life, and progressively increasing up to 3.5–4.0 g/kg/d by the end of the first week.[10] This intake promotes weight gain and growth and improves neurodevelopmental outcome.[24,25] In extremely low-birth-weight (ELBW) infants (<1000 g) amino acids intakes up to 4.5 g/kg/d have been well-tolerated and have been associated with weight gain and lower rates of bronchopulmonary dysplasia (BPD).[36] Optimal nitrogen (protein) and nonnitrogen energy intake and their appropriate ratios are essential to achieve positive nitrogen balance. High early PN amino acids intake is well-tolerated with no significant acidosis or hyperammonemia, and only moderately elevated blood urea nitrogen levels that are transient.[35,36,125] There are some concerns regarding the

safety of early high amino acids intake in very premature infants, related to the concentrations of the different amino acids and the possible side effects, mainly neurodevelopmental long-term outcomes.[126] Extremely premature LBW infants who receive early and higher parenteral amino acids have higher plasma amino acid levels.[127] However, there is no consensus regarding the optimal profile of amino acids in these infants and currently there is no data about toxicity. Nevertheless, the proven benefits of providing early high amino acids intake outweigh these concerns, although, more long-term studies are needed to ensure safety.

The metabolic immaturity of the neonatal population, especially those born prematurely, requires the provision of optimal amounts of both essential and conditionally essential amino acids that preterm infants have limited ability to synthesize (cysteine, glutamine, glycine, taurine, and tyrosine). Glutamine is an amino acid abundant in human milk and may be considered as a conditionally essential amino acid in premature infants. It is not included in available amino acid solutions because of stability issues. Glutamine contributes to the maintenance of small intestinal interepithelial junction integrity, intestinal epithelial cells growth and proliferation, inflammatory responses, and mucosal healing.[128,129] However, in a multicenter study and meta-analysis, supplementation of glutamine to standard PN did not improve protein retention, weight gain or growth and did not reduce the risk of mortality, late-onset sepsis or NEC.[125] Cysteine supplementation was suggested as a means to improve glutathione (a major intracellular antioxidant) synthesis, and to help premature infants to face the oxidative stress that they encounter on their unanticipated early transition to extrauterine life.[34] Glutathione is synthesized from glutamate, cysteine, and glycine. In a meta-analysis, cysteine supplementation was associated with improved nitrogen balance, although it did not improve growth.[130] Addition of cysteine to PN solutions (done as its hydrochloride base, 30–40 mg/g amino acids, up to a maximum of 120 mg/kg) lowers mixture pH. This increases calcium and phosphorus solubility and allows higher concentrations of these minerals to be provided.[131]

LIPID REQUIREMENTS AND INTRAVENOUS LIPID EMULSIONS

The introduction of the first successful intravenous lipid emulsion (ILE) in 1961 was a major breakthrough in PN support. Besides being a high-

density nonglucose source of energy, lipids are increasingly recognized to influence various immune functions, according to physicochemical characteristics of its FA and emulsifier (egg phospholipid) concentration.[132] Fat emulsions provide both energy and essential FAs. In addition, partial replacement of glucose by fat emulsions as the source of nonprotein calories minimizes hyperglycemia and its detrimental effects. Compared to glucose, intravenous administration of fat was shown to have equal[133] or superior[134] effects on nitrogen economy in surgical patients receiving PN. Furthermore, providing glucose and fat to infants is advantageous to protein metabolism compared to providing glucose alone as discussed above.[91] In premature infants, FAs are also necessary for central nervous system development and retinal maturation.[135]

lLEs consist of three components: an aqueous phase (glycerol), a lipid phase (FA), and an emulsifier. The lipid phase supplies FAs and the majority of the calories. FAs are categorized based on several different characteristics. First is the number of carbons in the FA chain: 2–4 carbons, short-chain FAs; 6–12 carbons, medium-chain FAs; and ≥14 carbons, long-chain FAs. Second is the number of double bonds in the FA molecule. Saturated FAs have no double bonds, monounsaturated FAs (MUFA) have 1 double bond, and polyunsaturated FAs (PUFAs) have 2 or more double bonds. Unsaturated FAs are furthermore categorized according to the carbon position at which the first double bond occurs, counting from the methyl end of the molecule (the ω carbon). There are three principal families of unsaturated FAs in humans: ω-3, ω-6, and ω-9 in which the first double bond occurs at the third, sixth, or ninth carbon, respectively. Essential FAs, ω-6 linoleic acid (LA,18:2 n6) and ω-3 α-linolenic acid (ALA, 18:3n3) are polyunsaturated FAs (PUFAs) that cannot be synthesized in humans but are metabolized through two different pathways, that use and compete for the same enzymes with preference for ω-3 > ω-6 > ω-9. With some exceptions, high conversion of LA to arachidonic acid (AA) may lead to overproduction of proinflammatory cytokines; in contrast, the cytokines derived from ω-3 FAs are considered to be anti-inflammatory.[136]

Soybean oil–based ILEs

The first safe and effective ILE became available in Europe in 1961 with the introduction of a soybean oil–based ILE. Until today, soybean oil, rich in ω-6 FAs, continues to be one of the main lipid sources in the various ILE on the market. Soybean oil contains high concentrations of PUFAs,

around 60% of the total FAs, with 8:1 ratio of LA (ω-6) to ALA (ω-3) ratio. Many experimental reports have raised concerns about the safety of ω-6 rich ILEs. ω-6 FAs promote the generation of AA–derived eicosanoids, which could, potentially exaggerate the inflammatory response when administrated to critically ill patients and patients with compromised immune function, sepsis, and trauma. Proinflammatory eicosanoids increase synthesis of potent proinflammatory mediators such as, tumor necrosis factor α and interleukin-6. ω-6 PUFAs have been correlated with unwanted immunosuppressive actions such as impaired reticular endothelial system function and inhibition of lymphocytes, macrophages, and neutrophil functions, although the data are not always uniform. In addition, the high number of double bonds in ω-6 PUFAs predisposes them to lipid peroxidation. The generated lipid peroxides can lead to cell death and cause damage to DNA, lipids, and proteins. Lipid peroxidation may further impair immune cells function and has been involved in the pathogenesis of intestinal failure associated liver disease (IFALD). Phytosterols, an isomer of cholesterol, another toxic component of soybean oil, has also been suggested among the etiologies of IFALD.[137,138] Furthermore, more questions have been raised about providing infants and children with soybean oil–based ILEs as their sole lipid intake. Soybean oil contains 50% or more of its triglyceride FAs as LA. This may represent a "linoleic acid overload" that may compete for and inhibit synthesis of longer chain ω-3 FAs such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Especially important to the growing infant is adequate provision of DHA for normal retinal and brain development.[139]

Concerns about the association of ILE with infectious morbidity were raised after a report of an increased risk of bacteremia related to the use of ILE in neonates.[140] A recent meta-analysis investigated the influence of timing and type of LE on rate of weight gain and several secondary outcomes in VLBW infants.[141] Analysis of the type of LE included 10 studies (499 infants; 249 in the intervention groups, 250 in the control group) which compared different LEs (combinations of soybean oil with medium-chain triglycerides (MCT) and/or olive oil and/or fish oil) with pure soybean oil. The rate of weight gain, death before discharge, duration of respiratory support, hypertriglyceridemia, and hyperglycemia were not affected by the type of LE. Pooled analysis of the studies (three studies, n = 197) which reported the incidence of sepsis showed that the use of a lipid source which was not purely soybean reduced by 25% the number of sepsis episodes (RR: 0.75; 95% CI: 0.56, 1.00; P = .05). This borderline

significant difference was, however, not significant in the individual studies. Moreover, the overall data regarding the effects of soybean oil–based ILE on immune responses have been disparate, as many investigators in numerous clinical and experimental settings found no evidence of an effect on a wide range of features and functions of immune cells, cellular signal transduction, or membrane characteristics. Taken together, the different experimental approaches applied in animal and human studies most probably play an important role in the ongoing controversy.[142,143]

MCT/LCT–based ILEs

In 1984, a second generation of ILEs consisting of a physical mix of equal parts of soybean oil and MCT oil (from coconut oil) was introduced in Europe. This emulsion (Lipofundin®) contains 50% less ω-6 PUFA, still adequate for meeting the needs of adults and infants. The new LCT/MCT combination has a similar ratio of LA (ω-6) to ALA (ω-3) of approximately 8:1. Compared to LCT, MCT has the advantage of greater solubility, lower accumulation in adipose tissues and liver, faster clearance, and improved resistance to peroxidation. Compared to LCT, MCT are rapidly taken up by mitochondria, largely independent of carnitine, and are oxidized faster and to a greater proportion. When prescribing MCT/LCT ILE one must remember that the energy content per gram MCT fat is lower than of LCT. From a functional point of view MCTs do not promote the synthesis of proinflammatory mediators and, unlike LCTs, have been suggested to improve immune function. The clinical relevance of these findings, however, remains to be established.[144,145]

There are indications that using emulsions containing MCT is advantageous in increasing lipid utilization, but also in decreasing the severity of chronic cholestasis syndromes and other hepatotoxicities that have been linked to PN.[146,147] In clinical studies, ILEs containing MCT/LCT have been well tolerated and safe in low-birth weight[148,149] and older children[150] in ICU patients[151] and HPN patients.[152] The use of MCT/LCT–based ILE in pediatric surgical patients has been shown to improve immune response and spare protein better than traditional LCT-based ILE.[150] Side effects including metabolic acidosis and increased energy expenditure have been reported after administration of MCT to animals. High levels of MCT have also been reported to delay the utilization of LCT. MCTs are, however, always used in combination with

LCTs because MCTs do not contain EFAs.[153,154]

Olive oil–based ILEs

In the 1990s, a third generation of ILEs, containing olive oil and soybean oil in a ratio of 4:1, was developed in Europe. The new olive oil–based ILEs have a high content of the monounsaturated (MUFA) oleic acid and of biologically active vitamin E (α-tocopherol). The ratio between LA (ω-6) and ALA (ω-3) in this ILE is 9:1.

Several in vitro animal and human studies have shown that the impairment of immune function, oxidative stress, glucose metabolism, endothelial function, and inflammation that occurs with soybean oil–based ILEs may be attenuated with olive oil–based ILEs. However, it is unclear whether the use of olive oil–based lipid emulsions in PN results in improved clinical outcomes compared to PN containing conventional soybean oil–based formulations.[155,156]

In randomized controlled studies in children and infants, the olive oil–based emulsion showed comparable tolerance and safety to a soybean oil emulsion (premature infants, ill neonates, and HPN) or MCT/LCT emulsions (children after bone marrow transplantation), but resulted in more favorable FAs levels in serum, reduced markers of lipid peroxidation, and higher serum vitamin E levels.[157–160] Use of olive oil–based lipid emulsions have been also shown to be safe in various adult patients groups: HPN patients,[161] hemodialysis,[162] severe trauma,[163] and in burned patients.[164] However, at this point of time there is insufficient data on oxidative stress markers, immune function, and clinical outcomes advantages of olive oil–based ILE.

Fish oil–based ILEs

Fish oil (FO) containing emulsions represent the most recent development in ILEs. FO is currently found in three PN lipid emulsions: Omegaven® (pure FO emulsion); Lipoplus® (50% MCT, 40% soybean oil, 10% FO) and SMOFlipid® (30% soybean oil, 30% MCT, 25% olive oil, 15% FO). FO is rich in ω-3 PUFAs, particularly EPA and DHA. Compared to MCT and olive oil, ω-3 FAs, are highly bioactive compounds. FO–based ILEs are not only nutrients and source of energy but also have anti-inflammatory properties with potentially important pharmacological and clinical benefits.[165,166] Pure FO emulsion (Omegaven®) has been developed as an adjuvant pharmacological—anti-inflammatory agent in critically ill patients, regardless of PN indication. The isolated use of

parenteral fish oil LE as a pharmacological agent has been investigated as an approach for quickly modulating immune functions and improving clinical outcomes in different clinical settings (rheumatoid arthritis, cancer, postsurgery, and critically ill patients).[165–168] Addition of FO has been associated with several effects: (1) Suppression of ω-6 FAs tissue content and increased incorporation of ω-3 FAs in the phospholipids of plasma, platelets, red blood cells (RBCs), and leukocytes, as well into lung tissue, liver cells, and the intestinal mucosa; (2) lower PGI2 metabolites, TXA2, 6-keto PGF1α, and TXB2 and decreased tendency for thrombosis and platelet aggregation; (3) inhibition of the proinflammatory cytokines (TNF-α, IL-6, and IL-1β) production and modulation of the anti-inflammatory cytokine IL-10.[169,170]

A recent meta-analysis in surgical and ICU adults, which included 23 studies (*n* = 1502 patients, *n* = 762 admitted to the ICU) found no statistically significant difference in mortality rate between patients receiving ω-3 PUFA-enriched lipid emulsions and those receiving standard lipid emulsions (RR = 0.89; 0.59, 1.33). However, ω-3 PUFA-enriched emulsions were associated with a statistically and clinically significant reduction in the infection rate (RR = 0.61; 0.45, 0.84) and the lengths of stay, both in the ICU (−1.92; −3.27, −0.58) and in hospital overall (−3.29; −5.13, −1.45). Other beneficial effects included reduced markers of inflammation, improved lung gas exchange, liver function, antioxidant status, and FA composition of plasma phospholipids, and a trend toward less impairment of kidney function.[171] A meta-analysis published in 2013 (six RCTs, *n* = 390 patients, critically ill adults only) reported that FO-containing emulsions were associated with a trend toward a reduction in mortality (RR 0.71; 95% CI: 0.49,1.04; *P* = .08) and a reduction in the duration of mechanical ventilation (weighted mean difference in days [WMD] −1.41; 95% CI: −3.43, 0.61; *P* = .17). However, no effect on infections and intensive care unit length of stay was observed.[168]

Besides these immunomodulatory properties, ω-3 PUFAs also exerts metabolic effects, which are well documented in humans.[172–174] PUFAs decrease plasma triglyceride levels in healthy and hypertriglyceridemic subjects and, increase lipid oxidation through upregulation of mitochondrial uncoupling proteins (UCPs'3) gene expression in liver and skeletal muscle in animal models.[175] Lipid profiles of the 79 children who developed PN-related cholestasis while receiving standard PN with a soybean oil–based ILEs were examined before and after the switch to a FO–based ILE (Omegaven®). After a median 18.3 (9.4–41.4) weeks of

receiving the FO–based ILE, serum triglyceride, total cholesterol, LDL, and VLDL concentrations significantly decreased by 51.7%, 17.4%, 23.7%, and 47.9%, respectively. A trend toward increase in the favorable cholesterol HDL was observed.[176] SMOFLipid® emulsion was evaluated in a double-blind randomized study in 28 children on HPN allocated to receive either SMOFlipid 20% (n = 15) or a standard soybean oil–based (Intralipid 20%, n = 13). ILEs were administered four to five times per week (goal dose, 2.0 g/kg/d). SMOFlipid emulsion was safe and well tolerated, decreased plasma bilirubin, and increased ω-3 FA and α-tocopherol levels without changing lipid peroxidation.[177]

Use of FO–based ILEs seems to be advantageous in treating infants and children with IFALD. FO–based ILE as monotherapy has been recently used in patients with IF and cholestasis, with promising results.[178] To date, no prospective RCT have compared equal doses of soybean oil–based with FO–based ILEs in preexisting IFALD. Concerns with the use of FO monotherapy have included the development of essential fatty acids deficiency (EFAD) or bleeding. Neither complication has been reported to date, although EFAD can occur if doses <1 g/kg/d are used.[179] Recent evidence in a murine model suggests AA and DHA provision alone is sufficient to prevent biochemical and physiologic evidence of EFAD.[180] The use of mixed ILEs with some FO for the prevention of cholestasis and hyperbilirubinemia in VLBW infants has not been evaluated in RCT but is increasingly used in neonates, thus, calling urgently for more well-designed RCTs.[181,182]

A recent meta-analysis reviewed the evidence on the benefits and safety of ω-3 FO compared with standard LE in children. The authors identified five RCTs and three prospective cohort studies in premature and LBW infants (n = 6), children with heart disease (n = 1), and IF (n = 1). The strength of evidence was consistently low or very low across all ILE comparisons and outcomes. In young children, ω-3 emulsions resulted in improvement in some biochemical outcomes of liver disease but no difference in mortality. Few studies examined patient-important outcomes, such as hospital length of stay or intensive care days and important safety parameters including the effect on growth and development remain to be evaluated.[183] Table 68-5 shows the composition and characteristics of several ILEs for parenteral supply in infants and children.

Carnitine

Carnitine facilitates transport of long-chain FAs across the mitochondrial

membrane, and thus makes the FAs available for beta-oxidation.[184] Preterm infants on PN have low carnitine levels, although the clinical significance of this finding is not clear. Meta-analysis of studies on carnitine supplementation to infants on PN showed no effects on weight gain, ketogenesis, or lipid utilization.[185] Thus, there is no benefit in routine parenteral carnitine supplementation to neonates requiring PN. Carnitine supplementation should be considered on an individual basis in infants receiving exclusive PN for long periods, probably for more than four weeks.[10]

Essential Fatty Acids Deficiency

Essential FAs cannot be synthesized in human tissues and must be obtained from the diet. Essential FAs are needed for processes including maintaining the integrity of the skin, structure of cell membranes, and the synthesis of prostaglandins and leukotrienes. Essential FAs include LA (ω-6) and ALA (ω-3) acids. The body can derive other FAs, such as AA (20:4 ω-6) from LA and EPA (20:5 ω-3) and DHA (22:5 ω-3) from ALA, but FO is a more efficient source as it does not rely on the conversion of ALA to DHA and EPA or LA to AA. In fact, FO contains mainly EPA and DHA, whereas vegetable oils predominantly contain LA or ALA. Requirements of EFAs are 1%–2% of dietary calories for children and 4%–5% for infants. Previous recommendations suggest a balance of 10:1 of ω-6 to ω-3 FAs, but emerging data indicate that a ratio as low as 2:1 would be beneficial in ill patients.[186] Soybean and olive oil–based emulsions contain a ratio of approximately 7:1 and 9:1, respectively.

TABLE 68-5 Intravenous Lipid Emulsions for Parenteral Supply in Infants and Children[a]

	Intralipid 20%	Lipofundin 20%	Clinoleic 20%	Omegaven 10%	SMOFLipid 20%	Lipoplus 20%
Oil Source						
Soybean oil, g/L	200	100	40	—	60	80
MCT, g/L	—	100	—	—	60	100
Olive oil, g/L	—	—	160	—	50	—
Fish oil. g/L	—	—	—	100	30	20
Main Fatty Acids (% by weight)						
Linoleic acid (18:2ω-6)	44–62	27	18.5	4.4	21.4	25.7
Arachidonic acid (20:4ω-6)	0.2	0.2	0.3	2.4	0.5	0.2
Total ω-6 fatty acids	53.2	29.3	9.1	5.6	19.2	53.2
α-Linolenic acid (18:3ω-3)	4–11	4	2	1.8	2.5	3.4
Eicosapentaenoic acid (20:5ω-3)	—	—	—	19.2	3.0	3.7
Docosahexaenoic acid (22:6ω-3)	—	—	—	12.1	2.0	2.5
Total ω-3 fatty acids	8	4.5	1	45.8	7.7	8
Additives and Characteristics						
α-tocopherol, mg/L	38	85±20	30	150–296	200	190±30
Egg phospholipids, g/L	12	12	12	12	12	
Glycerol, g/L	22	25	22.5	25	25	
Phytosterols, mg/L	348±33	NA	327±8	0	47.6	NA
Energy, kcal/L	2000	1908	2000	1120	2000	1910
pH value	7–8	6.5–8.5	7.0–8.0	7.5–8.7	7.5–8.8	6.5–8.5
Osmolarity, mOsmol/L	350	380	270	273	380	410

[a]Data of commercial ILE according to the manufacturers.

Intralipid (Fresenius Kabi, Bad Homburg, Germany); Clinoleic (Baxter, Compton, United Kingdom), Omegaven 10% (Fresenius Kabi, Bad Homburg, Germany), SMOFLipid (Fresenius Kabi, Bad Homburg, Germany), Lipoplus (B. Braun Melsungen, Melsungen, Germany).

EFAD can develop when intake of EFAs is inadequate (<1%–2% of total calories). EFAD was prevented when at least 3.2% of total calories were given as intravenous fat or at least 15% as oral fat. To prevent EFAD a minimum LA intake of 0.25 g/kg per day should be given to preterm infants and 0.1 g/kg per day to term infants and older children.[187] Patients with EFAD may show adverse effects, including skin lesions, reproductive failure, growth retardation, reduced learning, impaired vision, and polydipsia, as well as susceptibility to infections.[188]

The decline in ω-3 and ω-6 FAs concentrations that is characteristic of EFAD is accompanied by a corresponding increase in the percentage of nonessential FAs (oleic acid, palmitic acid) derivates, produced as a result of the elongation and desaturation of oleic acid (to mead acid) and palmitic acid (to palmitoleic and vaccenic acid). This process occurs during de novo lipogenesis. As mead acid typically accumulates in the presence of EFAD, the ratio of this compound to AA (i.e., the triene:tetraene ratio, Holman's index) is used as a diagnostic marker for EFAD. Until recently, plasma values of triene:tetraene ratio above 0.2 were considered abnormal, whereas levels >0.4 were considered diagnostic for EFAD.[189] Using capillary-column-gas-liquid chromatography, Siguel et al. have developed

long ago a more sensitive criteria that suggests total essential FAs to total nonessential FAs <0.60 and mead acid to AA ratios >0.025 as more reflective of EFAD. It must also be noted that the triene:tetraene ratio does not reflect ω-3 FAs status.[190]

Biochemical abnormalities appear in 1–2 weeks in children, whereas physical signs and symptoms may not appear for 4–6 weeks. However, biochemical abnormalities occur within 72 hours of birth in neonates, and may manifest clinically in premature infants by the end of the first week of life.[191–193]

EFAD may be treated by a variety of modalities and the essential FAs may be administered orally, or intravenously. In the absence of ILEs, donor plasma has been administered to patients with EFAD. Topical application of safflower and corn oils has been used but is not currently recommended. These oils do not provide adequate LA, are associated with unpredictable results, may take 2–3 weeks to take effect, and requires permeable skin to be effective.[194,195] In the absence of enteral feedings, the most reliable therapy for EFAD consists of ILEs. The ILEs from soybean oil with or without safflower oils are rich in ω-6 FAs. Omegaven, consists of 100% FO and is rich in ω-3 FAs with very little ω-6 FAs. This lipid emulsion, however, dosed at approximately 1 g/kg/d, provides sufficient essential FAs to prevent EFAD.[179]

Lipids Dose

Lipid intake should usually provide 35%–40% of calories in fully parenterally fed patients. Age-based lipid intake requirements are presented in Table 68-2. The minimum lipid supply must cover the daily requirements of essential FAs. Excessive lipids supply, beyond the liver's ability to utilize CHO and fat can cause accumulation of by-products in the liver and is a risk factor for the development of IFALD. Although standard guidelines recommend administration of ILEs at a dose <2.5 g/kg/d, even a dose >1 g/kg/d has been associated with the development of IFALD.[196] A recent study showed that compared to a historical control group of infants receiving 3 g/kg/d, reduction of soybean-based ILE to 1 g/kg/d delivered twice weekly was associated with a significant reduction in total bilirubin levels. EFAD was however diagnosed in 8/13 infants in the intravenous fat emulsion reduction group, although clinical signs of EFAD were not detected.[197] Decreasing or withholding lipids for extended periods in neonates may be detrimental due to lack of fat reserves and the high risk of EFAD and growth retardation.[198] Furthermore, provision of 1 g/kg/d of a

soy-based ILE did not always prevent or delay the onset of cholestasis in neonates.[196]

Excess ILEs has also been associated with respiratory distress, coagulopathies, impaired reticuloendothelial system function, immunosuppression, and hypertriglyceridemia.[199,200] The upper limit of lipid administration is difficult to determine. In term infants, fat oxidation reaches a maximum at 4 g/kg per day, given that the maximum glucose intake does not exceed maximal oxidative glucose disposal of about 18 g/kg per day.[91] Fat oxidation depends on the total energy intake and expenditure, the total intake of CHO and triglycerides and the CHO/fat ratio administered. Maximal fat oxidation occurs when the ILE provides 40% of the nonprotein PN calories in newborns and 50% in infants.[91] In neonates and infants, assuming an intake of 2.5–3.0 g/kg/d amino acids and 18–20 g/kg per day glucose, the administration of 3 g/kg per day lipids would put the total calorific intake at ~110–120 kcal/kg per day. This would cover the needs in most cases. If there is need to achieve a total caloric intake of 140 kcal/kg per day and assuming an administration of 3.0 g/kg per day amino acids, there would be need to give 3–4 g/kg per day lipids with 24.5–22 g/kg per day glucose.[201]

The metabolic utilization of intravenous lipids varies with gestational and postnatal age, severity of disease, and other factors. Therefore, monitoring of indicators of lipid utilization such as plasma triglycerides (or ideally lipid oxidation) may help in defining adequate intakes. There is no need to withhold parenteral lipid administration for determining triglyceride serum levels. Upper triglyceride levels of 250 mg/dL during lipid infusion in newborns, and serum levels of triglycerides of 300–400 mg/dL in older children can be reached postprandially and are considered acceptable during infusion of lipid emulsions. Emulsions with a low phospholipid/triglyceride ratio, usually in 20% lipid emulsions, result in less hyperlipidemia than emulsions with a higher phospholipid/triglyceride ratio (10% emulsions).[202,203] Parenteral lipid intake should usually be limited to a maximum of 3–4 g/kg per day (0.13–0.17 g/kg per hour) in infants and 2–3 g/kg per day (0.08–0.13 g/kg per hour) in children.[10,204] In critically ill patients, with sepsis or acute respiratory failure, it might be prudent to limit lipid intake during the acute phase of respiratory failure. Closely monitoring plasma triglycerides and adjusting lipid infusion rate when necessary is recommended in septic patients. It is common practice to limit ILE intake to less than 3 g/kg/d in infants, especially premature, with extreme hyperbilirubinemia, approaching levels mandating exchange

transfusion.[205] This practice is based on a theoretical concern that FAs might displace bilirubin from albumin-binding sites thus increasing serum free unconjugated bilirubin levels that could increase the risk of kernicterus.[206]

Use of ILEs in VLBW Infants

For many years the use of ILEs in preterm infants, especially early within the first days of life, had been limited by concerns regarding impaired lipid tolerance that is increased with decreasing gestational age. Lipid infusion has been allegedly associated with an increased risk for sepsis,[207] increased albumin-bound bilirubin displacement and aggravation of hyperbilirubinemia,[206] impairment of oxygenation, and development of BPD.[207,208] However, recent data including meta-analyses suggest that the initiation of ILEs within the first two days of life, even on the first day of life, appears to be safe and well tolerated in VLBW infants.[209–211] Early initiation of ILE in this setting was not found to be associated with increased morbidity or mortality or with any significant short- or long-term sequels. Actually there is some evidence to suggest better long-term neurodevelopmental outcomes in these VLBW infants.[212]

A systematic review published in 2012 also found no differences related to the type of ILE used.[141] Using soybean oil–based ILEs and or mixed ILEs containing soybean oil in combination with other fat sources (olive oil, MCT, and/or FO) showed similar outcomes regarding weight gain, pulmonary morbidity, hypertriglyceridemia, hyperglycemia, or mortality before discharge. However, the use of non-purely soybean oil–based ILEs was significantly, although weakly, associated with reduced incidence of sepsis in preterm infants.[141] As sepsis in VLBW infants is associated with serious adverse long-term outcomes, this finding suggests that mixed ILEs that are not based only on soybean oil should be preferentially used in VLBW preterm infants.[181,182]

The recommended ILE dose in VLBW infants is 1 g/kg/d on the first day of life with gradual increase by 0.5–1.0 g/kg/d thereafter. If well-tolerated, without hypertriglyceridemia, the lipid dose can be increased up to maximum of 3.0–3.5 g/kg/d by the end of the first week of life.[10,213] A dose of 4 g/kg/d might be occasionally used in chronically ill infants with high energy needs.

Lipid Emulsions Stability

ILEs are complex, thermodynamically unstable pharmaceutical products manufactured as oil-in-water dispersions of individual oils or their mixtures that are emulsified with the aid of egg lecithin phospholipids. Each oil droplet is coated with a monolayer of phospholipids that aligns its hydrophobic tails toward the oil phase, whereas the negatively charged hydrophilic heads are oriented toward the water phase. Emulsion stability is maintained by mutual repulsion of the droplets' resultant negative surface charge (or zeta potential) from the hydrophilic part of the phospholipids. Emulsions are made isotonic (250–350 mOsm/L) by the addition of glycerol and thus can be infused directly by either peripheral or central veins. Parenteral emulsions have a mean droplet size of approximately 0.2–0.3 μm, which is similar to that of naturally occurring chylomicrons and a median pH of 7.5 (range, 6.0–9.0). The emulsions also contain other lipid soluble substances, such as vitamins E and K, lipid peroxidation products, and phytosterols.[214]

ILEs can be given either undiluted as a separate infusion or, more commonly, mixed together with other PN components (i.e., amino acids, glucose, electrolytes, vitamins, minerals) in a single container as an all-in-one (AIO) admixture. Industrially manufactured multichamber bags (MCB) containing the three macronutrient solutions (amino acids, glucose, and lipids) in separate chambers of a single closed plastic system are now rapidly gaining widespread acceptance around the world where standard PN regimens can be used.[215]

The use of in-line filters with all PN admixtures to prevent the passage of insoluble particulates and enlarged lipid droplets into the microvasculature, has been advocated. In-line 1.2-μm filters are preferred for AIO administration and adherence to maximum 24 hours hang-times is recommended. Light protection and oxygen exclusion by using multilayer bags are proven factors that can decrease lipid peroxides formation and improve stability, especially for long-term storage of AIO admixtures (e.g., home PN). It is recommended that all PN containers and infusion lines be protected from phototherapy light and daylight at all times, unless these are infused solely at night.

Structured ILEs

Structured-lipid emulsions are produced by mixing MCT and LCT, then allowing hydrolysis to form free FAs, followed by random transesterification of the FA into mixed triglyceride molecules. Studies in animals have shown an improvement in nitrogen balance with the use of these lipid emulsions. Few human clinical studies with these products have

been performed. The results of these studies have been less promising than the animal studies; however, an improvement in nitrogen balance and lipid metabolism has been reported with a physical mixture of LCT and MCT. Structured-lipid emulsion seems to induce less elevation in serum liver enzyme values compared with standard ILEs. In addition, structured-lipid emulsions seem to have no detrimental effect on the reticuloendothelial system. Further studies are necessary to recommend the use of structured lipid emulsions.[216]

CARBOHYDRATE REQUIREMENTS

Carbohydrates (CHO) are key ingredients of PN solutions, being the major and most immediate source of energy. CHO can improve the nitrogen balance by exerting a protein-sparing effect which is more effective when combining CHO with amino acids or CHO with amino acids and lipid emulsions.[217,218] In addition, glucose is an important carbon source for the de novo synthesis of FAs and nonessential amino acids. Under normal metabolic conditions, CHO metabolism is regulated predominantly by insulin and glucagon. Other important factors involved in the regulation of CHO metabolism, especially during stress, are the counter-regulatory hormones (epinephrine, cortisol) and cytokines (e.g., IL-6, TNF-α). In certain tissues, glucose uptake is insulin-independent (e.g., erythrocytes, renal medulla, bone marrow, leukocytes, and brain). Serum glucose level is influenced by various factors: glucose intake (enteral, parenteral); glucose oxidation via glycolysis or pentose phosphate cycle; gluconeogenesis mainly in the liver (but also occasionally in the kidneys); and glucose exchange with glycogen stores in the liver and muscles.

Generally, approximately 50%–70% of nonprotein energy should be supplied as CHO and blood glucose level should be maintained between 80–110 mg/dL. The upper and lower limits of CHO intake in infants and children are difficult to establish due to the paucity of available data. Acceptable intake of CHO may be however, inferred from the rate of endogenous production and utilization of glucose, response to glucose administration, glucose utilization by the brain, the relation between CHO intake and nitrogen sparing and the basal and total energy expenditure.

Estimation of the lower limit of CHO intake should take into consideration the energy needs of the brain and other glucose dependent organs, should avoid protein and nitrogen loss, minimize protein use for gluconeogenesis, and prevent ketosis. Glucose is the primary oxidative fuel for the brain. Compared to adults, infants are at danger during any

period of sustained fasting because of the high glucose demand of their nervous system. The minimum intake of dextrose, the CHO used in PN, required for normal brain function is about 2 mg/kg/min in adults and 5 mg/kg/min in neonate. Using published data on glucose metabolism in vivo, Kalhan and Kilic have published the estimates of glucose consumption by the brain in relation to age. These ranges probably reflect the amount of exogenous glucose required to prevent hypoglycemia under most circumstances.[219]

Hyperglycemia should also be avoided; serum glucose levels should be probably kept less than 130–150 mg/dL at any time (although there are no specific criteria for defining either hyper- or hypoglycemia among critically ill children), as higher levels have been associated with higher morbidity (infections rate and length of hospital stay) and mortality.[220,221] To avoid hyperglycemia, the exogenous glucose delivery should correspond to the normal endogenous glucose production and rate of substrate oxidation. The amount of glucose that can be infused safely depends on the clinical condition and maturity of the infant. The ability of neonates to metabolize glucose may be impaired by prematurity and low birth weight. The endogenous glucose production varies from about 2 mg/kg/min (3 g/kg/d) in adults to approximately 8 mg/kg/min (11.5 g/kg/d) glucose in premature infants.[219,222] During PN, the rate of parenteral glucose delivery must be kept constant without exceeding the maximum rate of glucose oxidation (RGO). Glucose administration to full term neonates and children up to 2 years of age should not exceed 12.5 mg/kg/min (18 g/kg/d).[223] The maximal rate of glucose infusion may exceed glucose oxidation rate during cyclical PN, but should not go beyond 20 mg/kg/min (1.2 g/kg/h) (Table 68-2).

Evaluation of substrate utilization by IC in normal infants has estimated oxidative glucose disposal rates to be 12.6 mg/kg/min. Studies using labeled glucose contradict these findings, revealing maximal rates of glucose oxidation similar to that of adults, even among preterm infants.[222,224] This difference could be explained by the fact that IC estimates energy utilization from the algebraic sum of glucose degradation and synthesis. By contrast, stable isotope glucose tracers facilitate closer inspection of glucose utilization beyond simple cellular uptake. By using this method, the fraction of parenterally infused glucose that actually is oxidized can be measured directly. In a study by Sheridan et al., 10 critically ill children (mean age: 5.2 years) with burns were evaluated by using a stable-isotope tracer technique. The authors found that maximal glucose oxidation occurred at glucose intakes of 5 mg/kg/min. An increase

in exogenous parenteral glucose infusion from 5 to 8 mg/kg/min reduced the fraction of total exogenous glucose being oxidized from 59% ± 5% to 48% ± 5% ($P < .05$).[225] The mechanisms behind this decreased capacity of glucose oxidation in critically ill patients are unclear. Hu et al. reported decoupling of the mitochondrial respiratory chain reaction after burn injury.[226] Demling et al. described relative hypoxia of liver parenchyma in critically ill patients, despite hemodynamic stability. This hypoxia could limit aerobic metabolism of glucose while other biochemical pathways for its degradation become favored resulting in tissue uptake of exogenous glucose, rather than increased oxidation.[227]

RQ expressed as the ratio between CO_2 production to O_2 consumption, reflects the proportion of substrate utilization in the body. The npRQ (nonprotein RQ) represents the ratio of glucose and fat utilization by excluding the participation of protein, and varies in value from 0.70–1.0, with values >1.0 indicating net fat biosynthesis from glucose (lipogenesis). It must be kept in mind that the RQ in healthy (enterally fed) infants and children with normal growth and development, is approximately 0.85. Studies of critically ill adults and children have shown that changes in the metabolic condition, or excessive energy intake in the form of glucose, modify the npRQ value.[220,228] Studies[220,221] have shown that 75% of the subjects who consumed high amounts of CHO had an npRQ value >1.0. Glucose provided in excess of oxidative capacity enters nonoxidative pathways including glycolysis, glycogenesis, and lipogenesis.[229] The risks associated with this process are many fold and include accumulation of the newly synthesized fat in the liver and aggravation of respiratory acidosis resulting from increased CO2 production, particularly in patients with compromised pulmonary function. In addition, infusion of excess glucose causes hyperglycemia, glucosuria, and dehydration and has been associated with higher risk of complications in adults and children alike.[230,231] Taking into consideration the complications associated with high glucose supply, Meguid et al. showed conclusively that substitution of 30% of glucose calories with fat avoids these complications.[232] Likewise, substitution of up to 50% of calories with fat appears equally safe and complication free. When the intake of glucose calories exceeds the REE, net fat oxidation is minimal regardless of fat intake.

Critically ill patients have major alterations in substrate utilization. Peripheral insulin resistance and accelerated hepatic gluconeogenesis are added to the preferential use of anerobic glycolysis in injured tissues. Although exogenous insulin can be used to achieve normoglycemia, forced

cellular uptake with insulin does not guarantee effective glucose utilization. In adults, maintenance of normoglycemia with insulin infusion has been shown to reduce morbidity of surgical and medical ICU patients, and reduces mortality of surgical ICU patients.[233] A randomized controlled study of 700 critically ill infants and children in PICU, showed that targeting blood glucose concentrations to an age-adjusted normal fasting concentration with insulin infusion improved morbidity and reduced mortality, despite a substantial risk of biochemical hypoglycemia. Intensive insulin therapy protected the cardiovascular system, prevented secondary infections, and attenuated the inflammatory response, reducing the need for extended stay in intensive care. Mortality was lowered by an absolute 3·1% (95% CI 0·2–6·0) due to prevention of lethal neurological and pulmonary damage, and possibly prevention of infection.[234] A recent study, however, showed that tight glycemic control after cardiac surgery in children can be achieved with a low hypoglycemia rate, but did not significantly change the infection rate, mortality, length of stay, or measures of organ failure, as compared with standard care.[235] Routine use of tight glycemic control in the PICU remains controversial, owing both to contradictory results of the trials evaluating this approach in adults as well as to concerns about potentially damaging effects of insulin-induced hypoglycemia on the developing brain of children. Glucose targets and insulin titration strategies to improve end points while avoiding harmful effects of hypoglycemia and glucose fluctuation are still to be established.

CHO in PN are provided in a hydrate form (dextrose), which provides 3.4 kcal/g of dextrose. Fructose use in PN solutions is not recommended as life-threatening complications may arise in the event of undiagnosed hereditary fructose intolerance. Glucose contributes substantially to osmolarity in the PN solution. The osmolarity of a glucose solution rises significantly with increasing concentration from 255 mOsm/L in a 5% glucose solution to 1020 mOsm/L in a 20% glucose solution. Glucose solutions higher than 12.5% must be administered via a central vein. Initiation of glucose infusion should occur in an incremental fashion to prevent hyperinsulinemia. The glucose infusion rate (GIR) in mg/kg/min can be calculated using the following equation: GIR = g/kg/d dextrose × 1,000/1,440 min/kg.

Parenteral Carbohydrate Supply in Premature Infants

Glucose is the major energy source in newborns because it is readily available for the brain.[20] In the fetus, glucose is transported across the

placenta from the mother. The amount of glucose transported across the placenta increases as pregnancy progresses, thus allowing glycogen and fat stores to be accumulated during the third trimester. In the healthy term newborn baby, hormonal and metabolic adaptation in the immediate perinatal period ensures that adequate fuel supply to the brain and other vital organs is delivered. However, in preterm infants glucose homeostasis is still immature on the first days of life, and hyperglycemia as well as hypoglycemia are common in these neonates. After initial hypoglycemia, due to limited glycogen and fat stores, VLBW preterm infants often become hyperglycemic because of insulin resistance combined with relative insulin deficiency.[20,236] Hyperglycemia is associated with increased morbidity and mortality in preterm infants. Although there is lack of agreement on the precise definitions of hypoglycemia and hyperglycemia, it is generally accepted that glucose blood levels should be maintained within the following range: above 40–45 mg/dL on the first day of life[237] and above 50 mg/dL thereafter; and below 200 mg/dL, preferably below 150–180 mg/dL.[238,239] There is great variation in terms of diagnosis and management of hyperglycemia in VLBW preterm infants.[239,240]

Approximately 4 mg/kg/min (7 g/kg/d) of glucose cross the placenta in the last trimester of pregnancy. Glucose production rate ranges from 5.5 mg/kg/min (8 g/kg/d) in full-term healthy newborns and up to 8 mg/kg/min (11.5 g/kg/d) in VLBW premature infants.[219,222,241] Accordingly, initial glucose infusion rates of 4–7 mg/kg/min are appropriate for most newborn infants, although extremely-low-birth-weight (ELBW) infants (less than 1000 g) may need higher rates of 8–10 mg/kg/min to match endogenous glucose production from the liver and preserve their limited CHO stores.[242] Unfortunately many of these ELBW infants will not tolerate these high glucose infusion rates in the first days of life without hyperglycemia. Thus lower glucose infusion rates of 4–5 mg/kg/min (6 g/kg/d) are initially given, usually by using solutions with 5% dextrose.[20] Initial glucose infusion rates as low as 3.5 mg/kg/min are used in VLBW, and more often in ELBW preterm infants, corresponding to 100/mL/kg of 5% dextrose-based PN solution.[243] Supply of balanced PN, including amino acids and lipids, with high amino acids intake of 2–3 g/kg/d from the first day of life was suggested as one of the contributing factors to the improved glucose tolerance in ELBW infants.[35] Providing lower glucose infusion rates as part of full early PN might be even more successful than administration of insulin, probably due to the relative

insulin resistance in VLBW infants during the first week of life.[20,240] Moreover, data on long-term outcomes including morbidity, mortality, and long-term neurodevelopmental outcomes comparing uncorrected hyperglycemia or insulin administration are lacking.[240] If low intake of intravenous glucose (4–5 mg/kg/min, i.e., 6 g/kg/d) is tolerated, glucose delivery may be gradually increased to 8, 10, and up to 12–16 g/kg/d over the first week of life.[20] This can usually be achieved with 10%–12.5% dextrose-based PN solutions. Infusion rate of 18 g/kg/d glucose is probably maximal, because higher rates will exceed glucose oxidative capacity.[243,244] If higher intakes of glucose are not tolerated, glucose intake should be reduced. Insulin administration should also be considered, according to the clinical and nutritional status of the infant, with an initial dose of 0.05 IU/kg/h. Insulin may help control plasma glucose concentration, achieve increased energy intake, and promote nitrogen retention and growth. However, the evidences on the use of insulin for decreasing hyperglycemia, especially in ELBW infants, are controversial, and currently do not support the routine use of insulin infusions to prevent hyperglycemia.[239,240] Early insulin administration to reduce hyperglycemia did not improve mortality, sepsis, growth, intracranial hemorrhage, NEC, or chronic lung disease.[245,246] Actually, there were some concerns regarding mortality at 28 days that might have been higher in the early-insulin group. Although early insulin was associated with decreased incidence of hyperglycemia, it was nevertheless associated with increased number of episodes of hypoglycemia.[244] Insulin administration has also raised concerns regarding the increased weight gain, probably FM, and not lean body mass; and regarding the fact that insulin infusion did not improve protein balance, yet caused unexpected lactic acidosis.[247] More data is required also on the safety and long-term consequences of insulin use, especially on growth and neurodevelopmental outcomes, compared to long-term effects of hyperglycemia.[245] Nevertheless, there are few ELBW infants on PN, who will remain hyperglycemic despite very low glucose infusion rates in the first days of life and in these infants exogenous insulin infusion seem to be justified for a short period to keep them normoglycemic. Close monitoring is mandatory to avoid hypoglycemia and provide the best possible balanced nutrition.

FLUIDS, ELECTROLYTES, AND MINERALS REQUIREMENTS FOR PN

The total body water (TBW) content is age-related and diminishes from about 90% in a preterm infant born after 24 weeks of gestation to 75% in a term newborn infant, less than 70% in a 1–year-old infant and about 50% in an adult.[10,248] TBW is divided into two compartments: intracellular fluid (ICF) and extracellular fluid (ECF). The total volume of ICF increases with body size during growth. ECF is subdivided into intravascular and extravascular components as well as a ‘‘third space’’ represented by the free fluid in preformed body compartments under physiological (such as urine in the bladder, cerebral spinal fluid, pleural, peritoneal, and synovial fluid etc.) and pathological conditions (such as ascites or pleural effusions). ECF decreases during growth.[249]

Fluid Requirements

Fluid requirements for infants and children are based on studies done half a century ago by Darrow.[250] These requirements are based on healthy infants and children and do not take into account the changes in losses that may occur during altered health states. Fluids provide the principal source of water; however, some water is derived from the oxidation of food and body tissues. Requirements for water are related to caloric consumption; therefore, infants must consume much larger amounts of water per unit of body weight than adults. In general, fluid requirements (mL/kg/d) are matched to calorie requirements (kcal/kg/d). Although there are several methods to estimate fluid requirements, the most commonly used is that of Holliday, based on weight.[251] This method of calculating fluid requirements is in very close agreement with several other methods based on formulas, body surface area, basal calories plus activity calories, and age:

100 mL/kg for an infant 3 to 10 kg
1000 mL + 50 mL/kg for every kg over 10 kg for a child 10 to 20 kg
1500 mL + 20 mL/kg for every kg over 20 kg for a child over 20 kg

Enteral and parenteral fluid requirements are essentially the same. Approximately 40 mL/100 kcal per 24 hours replaces insensible losses and 60 mL/100 kcal replaces urinary losses. In the absence of diarrhea, stool losses are minimal, however, if diarrhea is present, fluid loss can increase dramatically and fluids loss should be replaced milliliter for milliliter. Acute weight loss almost always reflects fluid loss; any acute weight deficit should be added to fluid requirements. Additional fluid losses (for instance from an ileostomy or biliary drain), should also be replaced on a daily basis. Fever increases insensible water loss via respiration and via the

skin. For each degree of temperature above 38°C, insensible water loss is increased by 10% to 12% per 24 hours. Monitoring of fluid status is vital, and should include assessment of clinical status, body weight, fluids intake and output, blood electrolytes, acid base status, hematocrit, urine-specific gravity, and urine electrolytes. As a remainder, urine osmotic load results from protein catabolism and electrolyte excretion, but is little affected by CHO and fat metabolism that produce metabolic water and CO_2.

Electrolytes Requirements

In an otherwise healthy child, essentially all electrolyte loss occurs via the urine. Therefore, recommendations for electrolyte requirements come from measurements of sodium, potassium, and chloride urinary losses in healthy children. As with fluids, unusual losses of electrolytes (diarrhea, stoma output, bums, exercise, diuretic therapy, renal abnormalities, etc.) need to be considered and replaced. Electrolytes are critically important in PN for the short term but also long term because normal muscle accretion cannot occur in the presence of insufficient electrolytes and minerals.[252]

Sodium (Na^+) is the principal cation of the ECF and modulates the maintenance of intravascular and interstitial volumes. Sodium intake can influence the ECF volume. Sodium excretion occurs primarily through urine, but also through sweat and feces. Chloride (Cl^-) is the major anion of the ECF. The intake and output of chloride usually parallels that of sodium, but external losses and excretion can occur independently, mainly in equilibrium with bicarbonate status. Potassium (K^+) is the major intracellular cation, and the potassium pool correlates well with the lean body mass. The potassium intake varies widely. Extracellular potassium concentration is not always related to intracellular concentration. Evidence concerning fluid and electrolyte needs in infants comes from relatively few well-performed studies. Recommended intakes,[10,253] based on recommended daily allowances (RDAs), endorsed by the ASPEN/ESPGHAN are shown in Table 68-6. In general, PN should not be used to correct acute fluid or electrolyte abnormalities. Maximum levels of 40 mEq/L of potassium through a peripheral line and 0.5 to 1 mEq/kg/h through a central line are generally accepted.

Calcium, Phosphorus, and Magnesium

In the context of pediatric PN solutions, calcium, phosphorus, and magnesium are termed “minerals.” Calcium, phosphorus, and magnesium provided in PN solutions are essential for various systemic functions.

Approximately 99% of total body calcium is found in bone, whereas 85% of phosphorus is in bone, and 50% to 60% of magnesium. Bone acts as a reservoir for calcium, phosphorus, and magnesium, therefore, serum levels often do not reflect total body content, which makes it difficult to identify deficiency states. The molar Ca:P ratio is 1.3 in the whole body and 1.67 in bone mineral apatite. Less than 1% of the total body calcium, phosphorus, and magnesium is found in the circulation. Because serum calcium is tightly controlled at the expense of bone, calcium deficiency is a chronic condition associated with bone loss. Calcium circulates in three forms: 45% is in the biologically active ionized form, 45% is bound to protein, mainly albumin, and 10% is complexed to phosphate, lactate, and citrate. Measurement of ionized calcium, because it is the active form, better reflects calcium homeostasis than does measuring total serum calcium. IOM updated recently dietary reference intakes (DRI) on calcium and vitamin D.[254] The recommendations for 0–12 months reflect AI value rather than the RDAs, as RDAs have not been established for infants. The amount of dietary calcium needed to satisfy the demand for skeletal growth and mineralization is greater than the theoretical accretion rate because of incomplete calcium absorption and losses from skin, urine, and gastrointestinal tract.

TABLE 68-6 Parenteral Electrolytes and Minerals Requirements

	Preterm	Term Newborn	Infants	1–2 Years	2–18 Years
Sodium, mEq/kg/d	2.0–5.0	2.0–3.0 (5.0)	2.0–3.0	1.0–3.0	1.0–3.0
Potassium, mEq/kg/d	1.0–3.0	1.5–3.0	1.0–3.0	1.0–3.0	1.0–3.0
Chloride, mEq/kg/d	2.0–3.0 (5.0)	2.0–3.0	2.0–3.0	1.0–3.0	1.0–3.0
Calcium[a], mg/kg/d (mEq/kg/d) 20 mg = 1 mEq = 0.5 mmol	60–100 (3.0–5.0)	60–80 (3.0–4.0)	20–40 (1.0–2.0)	20–40 (1.0–2.0)	10 (0.5) 10–20 mEq/d
Phosphorus[a], mg/kg/d (mmol/kg/d) 31 mg = 1 mmol	45–78 (1.5–2.5)	45–62 (1.5–2.0)	16–45 (0.5–1.5)	16–45 (0.5–1.5)	16–31 (0.5–1.0) 10–40 mmol/d
Magnesium, mg/kg/d (mEq/kg/d) 12.1 mg = 1 mEq	6–10.5 (0.5–0.8)	6–10.5 (0.5–0.8)	6–10.5 (0.5–0.8)	2.4–6 (0.2–	2.4–6 (0.2–

= 0.5 mmol		0.5)	0.5) 10–30 mEq/d

[a]Provide Ca:P ratio (mg/mg) in the range of 1.3–1.7 (1:1 mmol:mmol ratio) in the first year of life, thereafter equimolar Ca; P ratio is optimal.

Current recommendations include 0 to 6 months (M+F): 200 mg/d; 6 months to 1 year (M+F): 260 mg/d; 1 to 3 years (M+F): 700 mg/d; 4 to 8 years (M+F): 1000 mg/d; and 9 to 18 years (M+F): 1300 mg/d.

Phosphorus plays a vital role forming the high-energy bond within adenosine triphosphate (ATP), which fuels a wide range of physical and metabolic processes and acts as an essential regulator within the glycolytic pathway. Crucially involved in respiration as the integral part of 2,3-diphosphoglycerate for tissue oxygen supply, phosphorus also acts as a buffer for maintaining urinary and plasma pH. Fifty percent of phosphorus exists in circulation as free ions, 10% is protein bound, and 40% is complexed to calcium, magnesium, and sodium as salts. Phosphorus deficiency can be a serious acute problem when malnourished patients are refed and sufficient phosphorus is not provided. For PN, at least 10 to 15 mmol phosphate should be given for each 1000 kcal. Infants, neonates, and small babies require 1.5 mmol/kg per day. This requirement diminishes with increasing maturity, and after the third year of life it is around 1 mmol/kg per day. During phosphate therapy, care should be taken to monitor for hyperphosphatemia and hypocalcemia caused by a negative feedback effect on the parathyroid gland.[255] Magnesium is mainly an intracellular anion, therefore, serum magnesium levels may not accurately reflect total body magnesium status. Frequent monitoring of potassium, calcium, and magnesium status is essential in all clinical conditions.

Calcium and phosphate supplementation via PN is often limited by the lack of central venous access and calcium and phosphate solubility. The hypocalcemia associated with metabolic bone disease results from inadequate supply and from urinary calcium losses. Increased urinary calcium excretion has been noted to accompany high oral or parenteral protein, glucose, or sodium intake, acidosis, decreased phosphorus intake, or vitamin D deficiency, use of loop diuretics, corticosteroids, and insulin or cycling PN.[256–258] Hypophosphatemia resulting from phosphorus depletion and intracellular phosphorus shifting is a primary and potentially life-threatening manifestation of the refeeding syndrome. Many studies have evaluated the effects of different calcium and phosphate dosage and ratios on serum and urinary calcium and phosphorus concentrations, retention, and bone mineralization and it seems that sufficient bone

mineralization may occur in infants and children on PN. Optimal calcium to phosphate ratio depends on the route of administration, bone mineralization, and nitrogen retention. Optimal calcium phosphate ratio for PN is 1.3–1.7:1 Ca:P (weight:weight/mg:mg) and nearly 1:1 molar ratio.[10,259] Lower ratios result in elevated urinary and serum phosphorous that suggest inadequate utilization of phosphorous because of insufficient calcium intake. This Ca:P ratio appears to promote the highest retention of both minerals and to simulate in utero bone mineral accretion rates. This Ca:P ratio is typically provided throughout the first year of life or until the infant reaches approximately 10 to 12 kg in weight. Calcium and phosphate requirements decrease as children age. In older children, equimolar provision of calcium and phosphate is optimal. In adults, balance studies have reported that positive calcium and phosphate balances can be achieved with mean calcium intakes of 15 mEq per day and minimum phosphorus intake of 15 mmol per day.[260]

Calcium and phosphate precipitation is the primary limiting factor to delivering optimal amounts of these minerals via pediatric PN. Calcium phosphate precipitates can cause significant morbidity and even death. Calcium phosphate precipitates have been noted in the central line of a neonate and have been reported to cause microvascular pulmonary emboli and interstitial pneumonitis in patients receiving lipid-free HPN.[261–263] Attempts to circumvent this problem by providing separate infusions of calcium and phosphate failed and resulted in hyperphosphatemia and hypocalcemia during phosphate infusions and hypercalcemia and hypophosphatemia during calcium infusions.[264] The solubility and saturation point of a solution of calcium and phosphorus are determined by many factors including pH, temperature, time of storage, amino acid concentration, amino acid product, lipid emulsions, dextrose concentration, calcium, phosphate, and magnesium concentrations, type of phosphate salt (inorganic vs. organic: glucose-1-phosphate, glycerophosphate, and fructose-1,6-diphosphate), and the order of addition of calcium and phosphate.

At concentrations above the saturation point, the calcium and phosphorus form insoluble complexes identified as precipitates. The type of calcium salt used in compounding parenteral solution can increase the calcium ion concentration at which saturation occurs for a given phosphorus concentration. For example, calcium chloride is more soluble than calcium gluconate. Solution pH is another factor determining the solubility of calcium and phosphorus. Dibasic phosphate is the predominant ion when the pH increases above 6.5, with a low solubility of

30 mg/dL. Monobasic phosphate is the predominant ion at lower pH and has a solubility of 1800 mg/dL.[265] Amino acids vary in pH and have a buffering effect (titratable acidity) on parenteral solutions. Depending on the amino acid product and the concentration (ranging from 1%–4%), the pH can vary from 6.8 to 5.2. The addition of cysteine (pH 1) and fat emulsion (pH 9) also influences the solubility of calcium and phosphorus. Amino acid solutions designed for children under 1 year have added cysteine which lowers the pH enough to permit the addition of calcium and phosphorus in amounts that may meet daily requirements.[266] Increasing dextrose, amino acid, and magnesium concentration also lowers pH and increases calcium and phosphate solubility.[267] Temperature is another important factor for calcium and phosphate solubility. The higher the temperature, the more readily available the calcium ion becomes, increasing the probability of precipitation.[268,269] Calcium solubility is endothermic; solutions that are compounded and tested at room temperature differ in solubility from those tested at body temperature (room: 20–25°C; body: 37°C). In addition, the chances of precipitation are greater with increasing length of time the PN solution is stored and infused as well as with lower infusion rates.[270]

Many safeguards exist to prevent the infusion of calcium phosphate precipitates to patients receiving PN. The prescriber and the pharmacist responsible for compounding PN solutions should recognize the factors affecting calcium and phosphate solubility. Practitioners may predict solubility by using available software or by plotting the concentration of calcium and phosphorus in the compounded PN on a graph with a saturation curve specific for the amino acid concentration, temperature, and calcium and phosphorus concentration. These saturation curves can be approximated by a hyperbolic model and incorporated into computer-based PN ordering. These systems alert the pharmacy that the solution is over solubility limits, thereby allowing the prescriber to revise the formulation. A recent study that reviewed 38,019 hospital PN orders plotted the calcium and phosphate concentrations for each solution in relation to the published curves. In all solutions, calcium and phosphorus concentrations plotted below the precipitation limits predicted by the published curves despite a large range of concentrations of electrolytes and minerals, validating the solubility limits represented by published curves.[271] Generally, calcium and phosphate can be safely added to PN solution when the concentrations provided satisfy the following equation: Calcium (mEq) + Phosphate (mmol) < 30 (per 1000 mL of PN solution).

Recommendations for minimizing the risk of calcium phosphate precipitation in PN solutions have been published.[272,273] Recommended parenteral calcium, phosphorus, and magnesium intakes for infants and children are shown in Table 68-6.

Refeeding syndrome frequently accompanies the nutritional rehabilitation of malnourished individuals. The refeeding syndrome is a set of metabolic and functional complications that occurs as a result of intracellular shifts of elements. During rapid nutritional rehabilitation, profound hypophosphatemia, hypomagnesemia, and hypokalemia can precipitate acute respiratory and circulatory collapse. Neurologic manifestations of the refeeding syndrome include weakness, lethargy, paralysis, and confusion. Deaths have been documented. The pathophysiological results of phosphate deficiency are inadequate supplies of energy-rich phosphates and, in particular, inhibition of glyceraldehyde-3-phosphate dehydrogenase, which occupies a key position in glycolysis. The effect of this on the central nervous system, muscle, and erythrocyte energy metabolism is to reduce ATP and 2,3-diphosphoglycerate levels, leading energy deficit and left-hand displacement of the oxygen–hemoglobin dissociation curve with decreased peripheral oxygen uptake and transport. Phosphorus, magnesium, and potassium must be carefully monitored in malnourished patients. Aggressive supplementation and correction of low values before feeding initiation will prevent the adverse outcomes of refeeding syndrome.[274]

FLUIDS, ELECTROLYTES, AND MINERALS REQUIREMENTS IN VLBW INFANTS

Fluid volume per kg body weight is higher in newborn compared to older children. Handling fluids in VLBW is further complicated by renal immaturity and reduced ability to concentrate urine, higher energy expenditure, greater body surface area, skin immaturity, and thus increased insensible losses.[248] Immediately after birth a transition period that can extend from less than 12 hours to few days (up to 3–5 days) follows. During this transition period, water and electrolytes balance may be severely disturbed as a result of discontinuation of placental exchange, considerable increase in insensible water loss, and huge requirements for thermoregulation. This immediate postnatal phase is characterized by initial relative oliguria followed by a diuretic phase.[275] In these first hours of life, contraction of the ECF space should be allowed to take place, while

maintaining normal intravascular fluid volume and cardiovascular function, as well as maintaining as normal as possible serum electrolytes and urine output. Attention to avoiding fluid overload may be critical during this transition period in VLBW infants, because this was shown to decrease respiratory morbidity, patent ductus arteriosus (PDA), BPD, and chronic lung disease, later on.[276,277] This can be achieved many times by restricting fluid and sodium administration, even in face of very high urine output and urinary sodium excretion with net negative sodium balance. Fluid intake in the initial transition period should be started at 60–100 mL/kg/d and gradually increased over the next days.[10] VLBW and premature infants born at younger gestational age will require the higher fluid intakes within this range due to their increased losses. Weight loss during this transitional period is expected, yet it should be closely monitored, especially in VLBW infants, along with input and output balance and serum electrolytes. Subsequent adaptation involves the onset of autonomic renal regulation of fluids and electrolytes. Urine output will usually decrease and urine osmolarity will increase above serum osmolarity. By the end of this period which may extend up to 5–15 days in VLBW infants, birth weight will be regained. During this adaptation period, total fluid intake could already reach 150 mL/kg/d (range: 140–160 mL/kg/d), although some ELBW infants may need slightly more. The suggestion to avoid fluid overload, as discussed above, is still very applicable to this adaptation period. During the stable growing period that follows, fluid intake of 150 mL/kg/d (range: 140–160 mL/kg/d) for term and preterm infants should be appropriate, even for VLBW infants on PN.[10]

Sodium and potassium supplementation are usually not required during the early postnatal period in premature infants. This is because of their high initial ECF that needs to be decreased with sodium diuresis that occurs in the first 1–2 days. Most preterm infants born after 28 weeks gestation or less usually also get sodium from other sources, for example, transfusions, saline infusions for arterial line flushes, and drugs. At this stage, the ELBW infants are usually at risk of losing significantly more water than sodium with ensuing hyperosmolar hypernatremia.[10] ELBW are also at risk of developing nonoliguric hyperkalemia from immature distal tubular function.[10] Thus, close monitoring of serum electrolytes in the first days of life is mandatory even though Na and K are usually not given in PN during the first 1–3 days. Supplementation of Na and K is gradually started based on their decreasing serum levels. This usually coincides with the cessation of the initial weight loss, although it may be required earlier.

Maintenance intake of Na, K, and Cl are listed in Table 68-6. Sodium requirements may be higher in preterm infants due to renal immaturity with increased urinary Na excretion. Metabolic acidosis is quite common in preterm infants on PN. This is due to the relatively acidic amino acids admixtures (especially if cysteine is added) and to renal immaturity with decreased renal reabsorption of bicarbonate.[278] This can be corrected by adding acetate (1–2 mEq/kg/d) as sodium or potassium salt.[279]

Because most of the calcium and phosphorus transfer and retention occur during the third trimester of pregnancy, and because hormonal control systems (parathyroid hormone [PTH] and vitamin D) are also immature, preterm infants are at increased risk of hypocalcemia and hypophosphatemia. Inadequate intakes of calcium and phosphorus can lead to poor bone mineralization, osteopenia, and even frank rickets and fractures.[258,280] Calcium needs to be supplied to preterm infants from the first day because of the risk of early hypocalcemia. Phosphorous supplementation may sometimes be postponed for 1–2 days because initially phosphorus excretion maybe low due to low glomerular filtration rate resulting in normal or even slightly elevated serum phosphorus levels. However, phosphorus needs to be added soon after, to avoid hypophosphatemia thereafter.[281] Magnesium is rarely adjusted unless the infant has high levels secondary to magnesium treatment given to the mother before delivery, or persistent hypocalcemia secondary to hypomagnesemia. The currently recommended intakes of minerals for preterm infants on PN are shown in Table 68-6.[10,20] The parenteral calcium supply is 60%–80% of that deposited by the fetus during the last trimester of pregnancy (100–120 mg/kg/d), but quite similar to those provided by most currently available enteral preterm formulas.

VITAMIN REQUIREMENTS

In February 2009, at the annual ASPEN Research Workshop that focused on micronutrients in PN, a group of international experts reviewed the parenteral requirements of various nutrients.[282] The recommendations are similar to those found in the 2005 ESPEN/ESPGHAN guidelines[10] and the summary is presented here. Based on the recommended doses for the current parenteral multivitamin products, the daily dose of the fat-soluble vitamins is approximately the same as the oral RDA or AI for these vitamins, even though the bioavailability should be much greater when these vitamins are administered intravenously. The rationale for providing

a higher effective dose when given as part of PN therapy was that these patients have higher vitamin requirements due to malnutrition, baseline vitamin deficiencies, and metabolic changes secondary to acute and chronic illness. The current daily water-soluble vitamin parenteral doses are 2–5.5 times greater than the oral RDA or AI. The rationale for giving even higher relative doses for these vitamins was that in addition to the increased requirements, as discussed above, there is increased urinary excretion of water-soluble vitamins when administered intravenously. These relatively high doses of vitamins have been used in PN therapy for over 30 years, and no toxicity has been described. As the scientific data supporting the current dose recommendations for pediatric and neonatal parenteral vitamin administration are very limited, ASPEN Research Workshop concluded that there are insufficient data to make any recommended changes in the current pediatric and neonatal parenteral multivitamin dosages or products.

Vitamin A

The recommended amounts of vitamin A for the all pediatric age range is provided in Table 68-7. Vitamin A deserves special attention in premature infants.[283] Delivery of parenteral vitamin A to the preterm infant can be compromised by losses in the delivery system and photodegradation.[284,285] This can result in as little as 17%–66% of the expected amount being received. Greater plasma concentrations of vitamin A may be achieved when given mixed with the ILE.[286] In premature infants, vitamin A is necessary for normal lung growth and the integrity of respiratory tract epithelial cells. Preterm infants have low vitamin A status at birth, and this has been associated with increased risk of developing chronic lung disease (BPD). In a recent meta-analysis conducted to evaluate vitamin A supplementation on the incidence of death, neonatal chronic lung disease, and long-term neurodevelopmental disability in VLBW, it was found that vitamin A appears to be beneficial in reducing death or oxygen requirement at one month of age.[287]

Thiamine (Vitamin B1)

Thiamine is water soluble, stable at acidic pH, and unstable in alkaline solutions and with exposure to ultraviolet (UV) light. Urinary excretion is increased when serum levels are elevated and decreased when serum levels are low, with maximum excretion two hours after oral intake. The biological half-life of thiamine has been determined to be 9–18 days.[288]

Thiamine should never be omitted in PN regimens. Pediatric parenteral multivitamin solutions in the United States provide 1.2 mg thiamine per daily dose.[289,290] All of these products meet or exceed the RDA or AI levels. One recent study confirmed that current PN and EN regimens provide adequate thiamine for premature infants.[291]

Riboflavin (Vitamin B2) and Niacin (Vitamin B3)

The currently available pediatric parenteral multivitamin formulation provides 0.56 mg of riboflavin per day. In LBW infants, giving 0.56 mg/d, maintained riboflavin balance.[292] For 0–1 year, AI is 2–4 mg/d; age 1–8 years, 5–6 mg/d; age 9–13 years, 9–12 mg/d; and 14 mg and above, it is in the adult range which is 40 mg/d. Recommended intake for niacin is provided in Table 68-7. Although there are no good controlled studies to evaluate the exact need of riboflavin and niacin in PN, the available data suggest that the current formulation will meet requirements in stable patients.[293] For adults, the parenteral intake based on available formulations easily meets the requirements based on RDA, recommended daily intake (RDI) or DRI for riboflavin and niacin.[293]

TABLE 68-7 Daily Vitamins Recommendations for Parenteral Supply in Infants and Children

	Preterm Dose/kg/d	Term Newborn Dose/d	Infants Dose/kg/d	Children Dose/d
Fat-Soluble Vitamins				
Vitamin A[a], IU	700–1500	2300	500–1000	500
Vitamin D[b], IU	40–160	400	32	400
Vitamin E, IU	2.8–3.5	7	2.8–3.5	7
Vitamin K, µg	10	200	10	200
Water-Soluble Vitamins				
Vitamin B_1 (thiamine), mg	0.20–0.35	1.2	0.35–0.50	1.2
Vitamin B_2 (riboflavin), mg	0.15–0.20	1.5	0.15–0.20	1.4
Vitamin B_3 (niacin), mg	4.0–6.8	17	4.0–6.8	17
Vitamin B_5 (panthotenic acid), mg	1–2	5	1–2	5

Vitamin B_6 (pyridoxine), mg	0.15–0.20	1	0.15–0.20	1
Vitamin B_{12} (cyancobalamin), µg	0.3	1	0.3	1
Vitamin C, mg	15–25	80	15–25	80
Folate, µg	56	140	56	140
Biotin, µg	5–8	20	5-8	20

[a]1 µg RE (retinol equivalent) = 3.33 IU vitamin A.

[b]1 µg vitamin D = 40 IU vitamin D.

Source: Adapted from Koletzko B, et al.,[10] Laine L, et al.,[279] Levy R, et al.[292]

Pantothenic Acid (Vitamin B5)

RDA for pantothenic acid has not been established due to insufficient available scientific evidence. Pantothenic acid recommendations for PN are derived from the deliberations of expert panels.[10,294,295] Children, maintained on PN with commercial intravenous multivitamin products, showed no evidence of pantothenic acid deficiency or toxicity.[296,297] Pantothenic acid is stable in PN while stored in darkness at 2°C–8°C for 96 hours but only during 24 hours of simulated infusion initiated immediately after mixing.[298]

Pyridoxine (Vitamin B6)

For infants, the adequate oral intake (AI) is based on the amount of vitamin B6 in human milk. This dose was extrapolated, based on expected weight for older infants (7–12 months). For older children, EARs and RDAs are extrapolated based on adult data. The EAR of adults is 1.1 mg/d and DRI 1.3 mg/d.[299] As with other B vitamins, exposure of vitamin B6 to sunlight leads to destruction, although this has not been shown in all studies.[300,301] After intravenous infusion vitamin B6 reaches plasma steady-state concentration within 30 minutes. The serum half-life is only a few minutes; only 6.7% of the administered dose is detected unmetabolized in the urine.[302]

Cobalamin (Vitamin B12)

At birth, the liver of well-nourished babies contains only 25–30 mcg vitamin B12. If breast-fed from vegan mothers, these infants may develop B12 deficiency at 2–14 months.[303] The pediatric parenteral dose (1 mcg/d) is large enough to replenish the liver stores rapidly. There are no known

adverse consequences of providing this generous amount of B12.

Folate

The amount is expressed as mcg/kg/d (56 mcg/kg/d) for small infants (<3.0 kg), but thereafter the standard pediatric dose is expressed as mcg/d (140 mcg/d). The parenteral dose is 30% higher than the enteral dose until adulthood, whereas in adulthood enteral and parenteral recommendations are the same, 400 mcg/d.[295]

Biotin

RDA has not been set due to insufficient available scientific evidence. Daily AI of biotin levels has been established by IOM, Food and Nutrition Board based on extrapolation from the amount of biotin in human milk. Biotin deficiency has been reported in children on PN without biotin.[304] In children receiving HPN either total or partial, biotin deficiency was rare.[305] All plasma biotin levels normalized after supplementation with 60 mcg/d of parenteral biotin. The current recommended parenteral biotin supplementation is 20 mcg/d in infants and children and 8 mcg/kg/d in preterm infants. However, based on data on biotin-deficient children on biotin-free PN, some investigators have speculated that the biotin supplement currently recommended for pediatric patients (20 mcg/d) may not be adequate therapy for biotin deficiency and might not even be adequate to maintain normal biotin status during TPN.[306] Biotin is stable while stored in darkness at 2°C–8°C for 96 hours or during 24 hours of simulated infusion initiated immediately after mixing.[298]

Vitamin C

Ascorbic acid is a strong aqueous antioxidant, and it is a cofactor for several enzymes involved in formation of collagen, carnitine, iron, and sterol compounds metabolism, and neurotransmitters synthesis.[307] The 1998 ASPEN guidelines for full-term infants and children up to 11 years old, recommend a parenteral ascorbic acid dose of 80 mg/d for older children.[272] Friel et al., demonstrated that for most premature infants the recommended daily dosage of 25 mg/kg/d would be adequate.[308] Because antioxidant and anabolic requirements may vary, plasma concentrations of vitamin C should be monitored in long-term PN. In conventional bags stored at 4°C, the ascorbic content is reduced to 35% after 24 hours; at 21°C only 15% of vitamin C is left after 24 hours. Multilayered bags with

reduced gas permeability minimize the loss; at 4°C the loss is 5% at 48 hours and is 10% at 21°C for 48 hours.[309]

Vitamin D

Vitamin D is required throughout life for mineralization of the skeleton, prevention of rickets in childhood, and osteomalacia in the adult. In the last years, the finding of vitamin D receptor in other tissues, not involved with calcium and bone metabolism led to the discovery of new functions of vitamin D, mainly associated with immune system and cellular differentiation. Recently IOM published a new report on dietary requirements for calcium and vitamin D. Based on bone health, the RDA (covering requirements of 97.5% of the population) for vitamin D is 600 IU/d for ages 1–70 year, corresponding to a serum 25-hydroxyvitamin D level of at least 20 ng/mL (50 nmol/L). The recommendations for infants remain 400 IU/d.[254] For supply within PN, dosages of 400 IU/d have been recommended in children. In premature babies, the dosage of 160 IU/kg/d appears acceptable because this intravenous dose maintained plasma concentrations within the range recommended for normal term gestation infants consuming proprietary milk formulas.[310]

Vitamin E

Vitamin E (tocopherol) exists as several isomers, the most active being α-tocopherol. Vitamin E functions primarily as a biological antioxidant protecting the membrane phospholipids from peroxidation.[311] Toxicity in adults has not been described, but premature infants developed coagulopathy with progressive liver failure after one week of 25–100 mg/d of α-tocopherol.[312] A daily dose of 2.8 IU/kg α-tocopherol maintained plasma tocopherol blood levels within range of 23–58 μmol/L (1.0–2.5 mg/dL) in eight patients with birth weights between 450 and 1350 g. This dosage is therefore recommended in this population group.[313] The available data from term infants and children up to 30 kg indicate that blood levels between 12 and 35 μmol/L (0.5 and 1.5 mg/dL) can be achieved in all patients who received at least 7 mg/d of all-*rac*-α-tocopherol intravenously. This dose is therefore recommended for term infants and older children.[306] Vitamin E can be degraded by photooxidation in parenteral solutions.[314] The photooxidation of vitamin E may be prevented by the presence of ascorbic acid (competition for oxygen) and the type of bag used; multilayered bags substantially reduce oxidation reactions by preventing oxygen permeation from the air into the

solution. Skouroliakou et al. evaluated the stability of vitamin E in AIO mixtures for PN of neonates. In these AIO mixtures vitamin E (6.4 mg) remained stable (90%) within 24 hours, but started to decrease after 24 hours.[315]

Vitamin K

Phylloquinone of plant origin and menaquinone of bacterial origin are naturally occurring forms of vitamin K that are lipid soluble. Blood factors II, VII, IX, and X are dependent on the presence of vitamin K, which functions as posttranscription cofactor in activation of intracellular precursor proteins to plasma clotting proteins. The AAP recommends that 0.5–1.0 mg of vitamin K be given to all newborns and the Infant Formula Act requires that all formulas have a minimum of 27 μg/L.[316] For the past years, phylloquinone has been routinely supplied parenterally as 200 μg/vial of multivitamin infusion (MVI) pediatric. Because deficiency was not reported, based on plasma prothrombin time, or apparent toxicity from this dosage (the amount is much higher than that recommended enterally), the suggested daily intake should remain at 200 μg for infants and children. For preterm infants, doses of 130μg and 70μg/day have not been associated with abnormal prothrombin times or apparent complications. Therefore, the suggested daily dose for preterm infants is 100 μg phylloquinone/kg.

One potential problem with the introduction of vitamin K into the adult formulations is the possible interference with anticoagulants such as warfarin. Additionally, intravenous fat emulsions contain vitamin K in varying amounts (approximately 13–70 g/dL) which can make titrating anticoagulant therapy even more problematic.

Choline

Choline is an amine endogenously synthesized from the amino acid methionine. It is an essential component of all cell membranes and is necessary for DNA repair due to its role as a methyl donor. It also serves as the precursor for the neurotransmitter acetylcholine. Animal models have shown progression to steatohepatitis and even development hepatocellular carcinoma with long-term choline deficiency. Choline deficiency may cause or contribute to PN-associated hepatic steatosis. Currently, there is no commercially available parenteral choline product. The recommended daily choline doses for neonatal and pediatric PN supply are 0–6 months, 125 mg; 7–12 months, 150 mg; 1–3 years, 200 mg;

4–8 years, 250 mg; 9–13 years, 375 mg; >13 years, 550 mg/d (adult amounts).[317]

General Considerations

Vitamins are essential nutrients and must be provided by the PN solution, to avoid deficiency. The optimum time to begin vitamin supplementation in newborns and premature infants is not clear. It should be taken into consideration, that water-soluble vitamins, with the exception of vitamin B12, are inadequately stored. Insufficient thiamine intake can result in severe lactic acidosis within only a few days in children on PN. It is, therefore, recommended that vitamin substitution is started early with PN. Vitamin intake should be administered daily. All clinical studies have been carried out with commercially available vitamin preparations, therefore, existing intake recommendations are based on the composition of these available preparations. At this time there is no commercially produced vitamin supplement available on the market, which completely meets the current intake recommendations for premature infants as a supplement for long-term PN.

Vitamins can be degraded by oxygen, light, and heat. Degradation reactions can be accelerated by catalytically active TEs such as copper and iron. Fat-soluble vitamins can be adsorbed on specific synthetic materials, thus, the administered dose is uncontrolled and significantly reduced. Dissolving the vitamins in a lipid emulsion, reduce absorption on plastic. Furthermore, the formation of peroxides in lipid emulsions can be reduced by adding multivitamin preparations. Vitamin preparations should, if possible, be administered together with the lipid emulsion. The clinical impact of free radicals, which can develop in intravenous multivitamin emulsions when exposed to light, remains a concern.

Feed lines with light protection should be used when administering parenteral solutions with vitamins until the clinical significance of these results has been determined. Exposure to light of PN solutions increases oxidation products such as lipid peroxides and hydrogen peroxide and oxidative stress impairs glucose uptake and affects lipid metabolism. A prospective study in which preterm infants were allocated to light-exposed (n = 32) or light-protected (n = 27) PN solutions, blood glucose was higher and hypertriglyceridemia was twice as high in those who received the light-exposed solutions. The authors concluded that shielding PN solutions from light provides a potential benefit for preterm infants by avoiding hypertriglyceridemia and allowing increased substrate delivery.[318] Premature infants need higher amounts of some vitamins compared to term

infants, because of their increased requirements for growth, and possible greater losses.

Multivitamin formulations for parenteral use in infants and children are presented in Table 68-8.

Trace Elements

Routine addition of zinc, copper, selenium, chromium, and manganese to PN solutions is recommended to avoid deficiency. Although not a conventional addition, molybdenum supplementation may be appropriate in long-term TPN therapy.[319] In 1988, the subcommittee on Pediatric Parenteral Nutrient Requirements of The American Society for Clinical Nutrition published the recommendations on TEs requirements in the term infant and child for zinc, copper, selenium, chromium, manganese, and molybdenum. Intakes for preterm infants were also suggested.[295]

TE contamination must be considered when determining TEs supplementation. Blood concentrations of copper, chromium, and manganese may become elevated because of contamination of TPN solutions by these metals. Beginning in the late 1970s, several reports described contamination of PN formulations with zinc, copper, manganese, chromium, and selenium. Nonnutrient contaminants such as aluminum, arsenic, and strontium were also found. The greatest amounts of contamination were found in the amino acids, potassium chloride, calcium gluconate, and sodium chloride products.[320] Amounts varied between manufacturers and in different lots from the same manufacturer.

Pluhator-Murton et al. calculated the amount of TEs in a 2 L PN solution for adults made with the tested components, compared those numbers with the amounts expected to be provided in the same PN solution, and found an extra 1.1 mg zinc, 0.08 mg copper, 38 mcg manganese, 15 mcg chromium, and 21 mcg selenium. The amounts of contaminant manganese and chromium found in that study were approximately 63% of the current recommended daily requirement for manganese and 100% of the daily requirement for chromium.[320] Giving the change in the manufacturing processes over the years, ASPEN recommends that these contamination studies be repeated on the current PN component products.

Specific disease states will necessitate adjustments in the elements provided. Copper and manganese should be administered cautiously, if at all, in patients with impaired biliary excretion or cholestatic liver disease.[321–324] Because of renal excretion of these elements, dosage

reduction for selenium, molybdenum, and chromium should be evaluated in patients with renal dysfunction. Additional zinc supplementation is necessary in conditions of increased losses, such as persistent diarrhea or excessive ileostomy drainage.

The most recent ASPEN position paper (2012) has recommended several changes to the currently available parenteral TE products. These recommendations include greatly reduced daily doses of both copper and manganese based on the reports of manganese toxicity and excessive organ accumulation of both manganese and copper in patients who had received long-term PN.[282] TEs recommendations for parenteral supply in infants and children are shown in Table 68-9. TEs formulations for parenteral use in infants and children are presented in Table 68-10.

Zinc

Zinc is included in all the current parenteral TE products. There is strong data supporting the routine use of zinc supplementation.[325] Extra supplementation of zinc may be needed in patients with diarrhea, gastrointestinal fistulas, severe burns, and in preterm infants with NEC especially in the presence of bowel resection or ileostomy.[326,327]

Selenium

There is strong evidence that selenium should be routinely added to all PN formulas.[328] Selenium is especially important in VLBW preterm infants because it is a component of glutathione peroxidase.[329] Supplemental doses of selenium for VLBW preterm infants on PN should be provided early, probably from the first day and in higher doses than those currently used.[330] Selenium supplementation in VLBW preterm infants was associated with a significant reduction in sepsis episodes but not improved survival, neonatal chronic lung disease, or retinopathy of prematurity.[331]

TABLE 68-8 Multivitamins Formulations for Parenteral Use in Infants and Children

Content/Vial	M.V.I. Pediatric 5-mL Vial	Influvite PEDiatric 1-mL + 4-mL Vials	Vitalipid N Infant 10-mL Vial	Soluvit N Adult 10-mL Vial	Cernevit 5-mL Vial
Fat-Soluble Vitamins					
Vitamin A, IU	2300	2300	2300	—	3500

Vitamin D, Ergocalciferol (D_2), IU	400	400	400	—	220
Vitamin E, dl-α tocopherol acetate, IU	7	7	7	—	11.2
Vitamin K, phylloquinone (K_1), µg	200	200	200	—	—
Water-Soluble Vitamins					
Vitamin B_1(thiamine), mg	1.2	1.2	—	2.5	3.51
Vitamin B_2 (riboflavin), mg	1.4	1.4	—	3.6	4.14
Vitamin B_3 (niacin), niacinamide, mg	17	17	—	40	46
Vitamin B_5 (panthotenic acid), 15 mg dexpanthenol, mg	5	5	—	15	17.3
Vitamin B_6 (pyridoxine), mg	1	1	—	4	4.5
Vitamin B_{12} (cyancobalamin) µg	1	1	—	5	6
Vitamin C, mg	80	80	—	100	125
Folate, µg	140	140	—	400	414
Biotin, µg	20	20	—	60	69

M.V.I. Pediatric (Hospira); 5 mL is the dosage for infants and children from 3 kg to 11 years, for infants weighing 1–3 kg the daily dosage is 65% (3 mL) of the 5-mL vial, and for infants weighing less than 1 kg the daily dosage is 30% (1.5 mL) of the 5-mL vial.

Influvite PEDiatric (Baxter); is supplied in two single-dose vials, vial 2 (1 mL) contains folic acid, biotin, and vitamin B_{12}, and vial 1 (4 mL) contains all the other vitamins. 5 mL is the daily dose for infants and children from 3 kg up to age 11 years; for infants weighing 1–3 kg, the daily dosage is 65% (2.6 mL vial 1 and 0.65 mL vial 2); and for infants weighing less than 1 kg, the daily dosage is 30% (1.2 mL vial 1 and 0.3 mL vial 2).

Vitalipid N Infant (Fresenius Kabi); The dosage is 1 mL of Vitalipid N Infant per kg bodyweight per day up to a maximum of 10 mL.

Soluvit N Adult (Fresenius Kabi); supplement the water-soluble vitamins in adults and children 11 years of age and older. Soluvit N is not recommended for use in children under 11 years. Contains as preservative 500 µg methyl hydroxybenzoate.

Cernevit (Baxter); multivitamins supplement for children after 11 years.

However, not all the currently available parenteral TE products contain selenium. Selenium is also available as an individual parenteral product

that can be added to PN separately. ASPEN recommends that selenium be routinely added to PN formulas, either in a multiple TE product or as a separate component. The current recommended pediatric parenteral intake is 2 mcg/kg/d. The current parenteral dose of 20–60 mcg/d in adults seems to be too low, because patients receiving this level of daily intake frequently have low plasma concentrations of selenium.[328] It is recommended that the adult daily parenteral selenium requirement should be increased to 60–100 mcg/d.[282] Selenium is excreted in the kidneys, thus its intake should be reduced in patients with impaired renal function.

Manganese

Hypermanganesemia has been reported in 389 patients (17 studies) on PN (265 adults and 124 children). Most patients had no clinical symptoms, but few developed neurological symptoms (Parkinson-like syndrome, confusion, irritability, seizures). Serum and RBC manganese and basal ganglia magnetic resonance image (MRI) abnormalities regressed after discontinuation of manganese supply.[332] Additional patients were reported in 2008 by Hardy et al. Hypermanganesemia occurred particularly in infants with cholestasis, often within three weeks of the initiation of PN.[333] Manganese content in autopsy tissues were reported from eight patients who lived 2–21 years on PN, showed significant manganese elevation in liver and kidney samples, especially in patients who died of liver or renal failure.[323]

Currently, multiple TE products provide between 265 mcg to 500–800 mcg manganese per day (adults) and 2–10 mcg/kg/d (pediatric/neonatal TE products). Most patients who developed hypermanganesemia had received doses within these ranges. A dose-finding Japanese study showed that adult PN patients maintained a stable whole-blood manganese level on a supplement of 55 mcg/d, whereas an increase in RBC manganese and in MRI signal intensity was likely if 110 mcg or more was supplemented.[334]

TABLE 68-9 Daily Trace Elements Recommendations for Parenteral Supply in Infants and Children

	Preterm	Term Newborn	Infants	Children
Zinc, mg/kg/d	0.4	0.25	0.25 < 3 months 0.1 > 3	0.05 (max 5 mg/d)

			months	
Selenium, µg/kg/d	5–7	2	1–3	1–3 (max 30-60 µg/d)
Manganese[a], µg/kg/d	1	1	1	1 (max 50-100 µg/d)
Copper[b], µg/kg/d	20	20	20	20 (max 500 µg/d)
Chromium[c], µg/kg/d	0.05–0.2	0.2	0.2	0.2 (max 5–15 µg/d)
Molybdenum, µg/kg/d	0.25	0.25	0.25 (max 5 µg/d)	0.25 (max 5 µg/d)
Iron[d], mg/kg/d for PN >2 months	0.1–0.2	0.25–0.67	0.05–0.1	0.05–0.1 (max 1mg/d)
Iodine	1 µg/kg/d	0–1 µg/kg/d	0–1 µg/d	1 µg/d

[a]Regular monitoring rather than provision is recommended.

[b]Check serum copper and ceruloplasmin in children with cholestasis in order to decide whether copper should be reduced or removed from PN. There is insufficient data for PN supplementation in premature infants.

[c]ESPGHAN, ESPEN and ASPEN position papers states that chromium contamination of PN ingredients makes additional supplementation unnecessary and suggests regular monitoring rather than addition.

[d]Not usually added to PN solutions.

Source: Adapted from Koletzko B, et al.,[10] Laine L, et al.,[279] Levy R, et al.[292]

TABLE 68-10 Trace Elements Formulations for Parenteral Use in Infants and Children

Content/1 mL	Multitrace-4 Neonatal	Multitrace-4 Pediatric	Trace Elements Injection 4	Peditrace	Inzolen-Infantibu Sine NaK
Zinc, mg	1.5	1	0.5	0.25	0.097
Copper, mg	0.1	0.1	0.1	0.02	0.032
Selenium, µg	—	—	—	2	—
Chromium, µg	0.85	1	1	—	8
Molybdenum, µg	—	—	—	—	—
Manganese, mg	0.025	0.025	0.03	0.001	0.027
Iron, mg				—	0.091
Iodide, mg				0.001	—
Fluoride, mg				0.06	—

Cobalt, µg	—	14

Multitrace-4 Neonatal (American Regent), supplied as 2-mL vial. Does not contain selenium and iodine.
Multitrace-4 Pediatric (American Regent), supplied as 3-mL vial. Does not contain selenium and iodine.
Trace Elements Injection 4, USP—Pediatric (American Regent), supplied as 10-mL vial. Does not conta
Peditrace (Fresenius Kabi), supplied as 10-mL ampoule. Dosage 1 mL/kg up to 10 kg.
Inzolen-Infantibus sine NaK (Kohler), supplied as 10-mL ampoule. Does not contain selenium and iodin
Oligoelements Aguettant Pediatrique (Aguettant), supplied as 10-mL ampoule.

Current recommendations suggest that the dose of manganese in parenteral multiple TE products be decreased to 55 mcg/d for adults and 1 mcg/kg/d in pediatric and neonatal products with a maximum daily total dose of 55 mcg suggested by ASPEN[282] and 50 mcg suggested by ESPGHAN/ESPEN. Manganese dose should be further decreased or withheld in patients with significant cholestasis or hepatic dysfunction, elevated whole blood manganese levels, or in those with signs and symptoms of manganese toxicity.[282] In addition, ASPEN recommends the amount of manganese contamination in a standard adult composite PN formulation be limited to <40 mcg/d.[282] Contamination data for pediatric and neonatal PN formulas are not available and require further research.

Copper

Wilson's disease, an inborn error of copper metabolism leading to high concentrations of copper is the prototype of copper toxicity. Elevated hepatic copper, within the diagnostic range of Wilson's disease (copper >250 mcg/g dry weight), have been reported in 8/28 patients with cholestasis on PN, although the copper supply was not reported.[335] Autopsy studies reported significantly elevated copper in liver and kidney tissue, especially in patients who died of liver failure although receiving recommended 0.5–1.5 mg/d of copper within PN solution.[323] By analogy with cholestatic infants, it is likely that copper accumulates as a result of liver disease, but it is also possible that hepatic copper overload enhances the liver damage. However, two recent studies including neonates and premature infants on PN did not find elevated serum copper levels on standard 20 mcg/kg/d supplementation, although most of them were cholestatic. Moreover, most infants had low serum copper levels, therefore, regular monitoring of copper levels even in cholestatic children should guide the decision on copper supply or discontinuation.[336,337]

The adult parenteral multiple TE products currently available in the United States provide approximately 1 mg/d of copper. ASPEN recommends that the dose of copper in adult parenteral multiple TE

products be lowered to 0.3–0.5 mg/d and limit the amount of contamination in a composite PN formulation to less than 0.1 mg/d. Copper requirements are increased in severe burn patients.[338] No contamination data are available for pediatric and neonatal parenteral TE products; investigation of copper contamination in these products is desirable.

Chromium

Chromium toxicity varies depending on the chromium valency. Chromium IV, V, and VI are carcinogenic. Chromium in PN formulations, food, and oral supplements is trivalent and considered nontoxic.[339] There are several reports in PN patients, both adults and children, of serum and urine chromium concentrations 10–100 times normal values.[340,341] Autopsy tissue data found similar elevations in heart, muscle, liver, and kidney samples.[323]

A normal adult diet provides approximately 35 mcg/d of chromium but the absorbed amount of chromium from a standard adult oral diet is only 0.4–0.9 mcg/d. Therefore, the parenteral requirement is likely to be only one-tenth or less of the previously recommended amounts of 10–15 mcg/d. According to Pluhator-Murton et al. data, chromium contamination of a 2-L PN composite formula is approximately 15 mcg per day, mainly from the 70% dextrose solution, in addition to the 11 mcg/d intentionally added from a multiple TE product.[320] This amounts a parenteral chromium dose that is 30–60 times greater than the current estimated requirement.

The key question is whether these high chromium doses have clinical consequence or are potentially toxic. There are no reported cases of chromium toxicity in adult patients on long-term PN, suggesting that these high chromium concentrations are not toxic. Chromium toxicity may be more of a concern in pediatric patients. In 1992, Moukarzel et al. reported that glomerular filtration rates (GFR) in PN-dependent children were inversely correlated with their serum chromium concentration, cumulative parenteral chromium intake, and PN duration. One year later after discontinuation of parenteral chromium supplementation, serum chromium concentrations were lower but still elevated, and their reduced GFR was still below control patients who did not receive PN.[342] Renal tubular damage and high renal chromium deposits were also reported in rodents after receiving one week of PN.[343] Because PN components have significant chromium contamination, some investigators have stopped adding chromium to PN formulations, but the consequences of such

actions have not been evaluated.[344,345]

The current adult multiple TE products provide 10–15 mcg/d of chromium. Based on oral absorption in healthy individuals, the parenteral requirements may be as low as 0.14–0.87 mcg/d.[346] ASPEN recommends that in addition to the current TE products that deliver 10–15 mcg/d of chromium, a TE product without added chromium should be made available. Research on chromium requirements during PN and the effects of contamination on requirement for supplementation is urgently needed.[282] The pediatric and neonatal multiple TE products available provide 0.05–0.2 mcg/d of chromium. Studies of pediatric and neonatal PN patients on this level of supplementation report serum chromium concentrations 4–42 times that of normal.[347] Moukarzel recommended that the daily parenteral dose of chromium for neonates and pediatric patients should be radically reduced. In view of preliminary studies in newborns on PN that demonstrate chromium nephrotoxicity, and because PN formulations have significant chromium contamination, ASPEN recommends that the daily dose of chromium be reduced to the levels recommended by Moukarzel and that a pediatric/neonatal multiple TE product without added chromium should be available for use in this patient population.[342]

Molybdenum

Molybdenum functions as a cofactor for a limited number of enzymes in humans. The IOM's Food and Nutrition Board has developed AIs for infants (2–3 mcg/d) and RDAs for children and adults (17–50 mcg/d).[344] The published, estimated daily molybdenum PN requirements are listed in Table 68-9. Most professional groups indicate the need for molybdenum in long-term, home PN patients. While there are no reports of molybdenum deficiency in term infants, LBW infants might be at risk because they are born before adequate stores can be acquired, have rapid growth requiring increased intakes, and frequently receive PN. Friel and coworkers performed a molybdenum balance study in 16 LBW infants on PN. They speculate that an intravenous intake of 1 mcg/kg/d would be adequate for the LBW infant.[348] There are no significant data to warrant changes in molybdenum supplementation in PN formulas. Thus, no changes are recommended for adult, pediatric, and neonatal products. Molybdenum is present in most of the adult and some of the pediatric parenteral multiple TE products in Europe but none of the products in North America.

Iron

Some TEs products contain iron, but some do not. Instability of total nutrient admixtures containing iron has prevented its addition to TE mixtures. For patients not able to take oral iron supplements, addition of iron dextran to fat-free PN or separate infusions of IV iron have been recommended. Further research is required to examine the stability of iron additives to fat emulsion–containing admixtures.

Iron supplementation in pediatric and neonatal patients on PN is important in long-term therapy, and these infants should be monitored for iron status prior to and during parenteral therapy. Iron is probably not needed in preterm infants in the first few weeks, especially if they were transfused.[349] There are also some concerns regarding its possible contribution to oxidative stress.[350]

Iodide

Iodide is currently included in the parenteral multiple TE products available on the market in Europe, but not in the North America. The amount of iodide contamination in PN components is unclear but appears adequate to meet basal requirements as thyroid function remains normal in most long-term PN patients on iodide-free regimens.[351]

However, one study of 15 pediatric PN patients in Europe found an inverse correlation between duration of PN and urinary iodide concentrations. Although 9/15 (60%) of these patients had iodide deficiency based on urinary iodide concentrations, thyroid function tests remained normal. Use of povidone-iodine solutions for skin cleansing of catheter sites in the past may have maintained a normal iodide status due to iodide skin absorption.[352] As skin cleansing has widely moved to the use of 2% chlorhexidine, the risk of iodide deficiency and the need for parenteral supplementation need to be investigated. Routine supplementation of PN with iodide could be beneficial, but more research is needed for adult, pediatric, and neonatal products.

Fluoride

Fluoride is also present in the multiple TE products used in Europe but is not included in any products available in North America. The amount of fluoride contamination in PN is variable. Supplementation of PN with fluoride could be beneficial, but more research is needed for adult, pediatric, and neonatal patient populations.[353]

Trace Element Requirements

TEs should be supplemented in long-term PN. When PN is used partially, or is limited to one or two week, only Zn need be added to the infusate. If PN continues for >4 week, addition of selenium is advisable and in the absence of cholestasis, copper and manganese may also be added. Iodide and chromium may also be added at this time and iron supplementation needs to be determined. In the presence of suspected preexisting deficits or rapid growth, earlier use of TE supplements may be desirable. Requirements for some TEs have been narrowed down lately. There is no TE supplement for long-term PN, which meets the current recommendations.

TEs requirements and optimum time to begin supplementation in premature and full-term infants are not completely clear. Premature and ill full-term infants have an increased risk of developing TE deficiency. Premature infants are born with lower TEs stores as the accumulation of these stores occurs during the last trimester of the pregnancy. Rapid growth, unidentified requirement levels, and variable absorption are further risk factors contributing to the development of a TE deficiency in premature infants. Probably, the only TEs that need to be provided from the first day of life, when PN is initiated in VLBW premature infants, are zinc and selenium.[295,331] Supplementation with other TEs is probably not needed up to two weeks, although these are usually supplied with the TE preparations.

VASCULAR ACCESS

PN solutions are administered either via a central venous catheter (CVC) or over short term via peripheral venous access, depending on the condition of the patient (type of illness, current state of health, etc.), composition of the infused solution, amount of energy to be administered, and duration of PN.

Long-term vascular access is an essential component in the management of pediatric patients with a wide variety of chronic clinical conditions requiring IV blood sampling, drug therapy, or PN. PN is usually administered via a CVC because of the high osmolarity of nutrient admixtures and the inability to provide the prescribed amount of PN when using peripheral veins. The broad array of devices and techniques available highlights the importance of disease- and patient-specific selection to properly address the clinical needs of the child. In addition, the appropriate care of the device is crucial to decrease the risk of morbidity

and mortality due to device-related complications. Current CVC types, materials, and connectors and alternative access choices for delivery of long term PN are as follows:

1. Tunneled catheters (e.g., Hickman/Broviac silicone catheters with or without Groshong valve) offer acceptable rates of sepsis and thrombosis and ease of use. These catheters have a Dacron cuff attached to the outer surface and located near the proximal end. This cuff is meant to be placed in a subcutaneous tunnel and allows for the ingrowth of surrounding tissue during several weeks after placement to secure the catheter in place. This tissue ingrowth also putatively provides a physical barrier against bacterial migration along the catheter tract into the bloodstream. These catheters are available in single- and double-lumen variety and range in size from 2.7 to 12 Fr for pediatric patients. The visible length may be repaired if damaged.[354–356]
2. Fully implanted catheters: (e.g., Port-a-cath): These CVCs are accessed through the skin by a needle and are favored by some patients for functional and cosmetic reasons, for example, bathing and swimming, and could be considered for patients with a coexistent stoma. These catheters consist of a reservoir attached to a catheter. Most reservoirs have a compressed silicone self-sealing septum that is estimated to be able to maintain its integrity through approximately 2000 punctures. The port is accessed transcutaneously using a 90° angle 20–22 gauge noncoring (Huber) access needle. These catheters are better suited for intermittent rather than daily use.[357]
3. Peripherally inserted CVCs (PICCs): These are acceptable for short-term PN (can provide access for up to 12–18 months) and provide the advantage of avoiding general anesthesia and vein manipulation. These catheters are made of either Silastic or polyurethane, are single or double lumen, and come in sizes from 1.1 to 7 Fr, 40–60 cm in length. In a prospective study in adults, PICCs for the administration of HPN were found to be similar in terms of catheter-related complications (catheter-related blood stream infections [CRBIs] and thromboembolic complications) to other CVCs.[358,359] Recently, a retrospective study in infants with IF reported that the use of PICC was associated with low prevalence of CRBIs and venous thrombosis.[360]

4. Arteriovenous fistulae: This type of access can be useful when central venous access becomes impossible.

CVC Material

Standard silicone catheters are robust and soft and can last for 10–15 years. There is insufficient evidence to currently recommend alternative CVC materials (Teflon, antibiotic impregnated lines).

CVC Line Connectors

All connections should be Luer-locked to prevent accidental disconnection and air embolus. All external tunneled catheters should have a clamp and be locked under positive pressure.

CVC Placement and Positioning

To minimize complications or side effects, the smallest single-lumen catheter should be inserted into the largest possible vein. Surgical or radiologically guided percutaneous insertion using an aseptic technique should be used to insert a CVC. The location of the catheter tip should be radiologically documented. The recommended CVC tip position is just above the SVC–right atrium junction, parallel to the SVC to prevent complications such as arrhythmia, thrombosis, tricuspid valve damage, and cardiac tamponade.[361] Various helpful radiographic landmarks for safe positioning have been described. The carina is always located above the pericardial sac in adults and recommended as a landmark to guide the CVC placement. Although, the radiographic diagnosis may be more difficult in children (e.g., because of a large thymus gland), the carina seems a safe radiographic landmark for CVC positioning even in small children. The tip of the right sided CVC catheter should ideally lie just above the level of the carina.[362–364]

Exit Site Dressings

Dressings for the CVC exit site are recommended for about the first 10 days. Highly permeable transparent dressings or dry gauze are acceptable. The former should be changed weekly in the absence of a specific indication (blood, pus) and the latter daily. With a dry healed exit site, dressings are not necessary except for peripherally inserted CVC lines.[365]

Catheters' Flushing

In adults, routine flushing of nonused CVCs or port systems with heparin

is not recommended,[366] and there is no need for heparinization when the catheter is closed for a short period of time (<8 h). In children or when recommended by the manufacturer, implanted ports or opened-ended catheter lumens should be flushed and locked with heparin sodium flush solutions. The usual dosage based on patient weight is 1 U/mL for patients in the neonatal ICU, 10 U/mL for children less 15 kg; and 100 U/mL for children >15 kg.[367]

Infusion Pumps

PN should always be administered by an infusion pump in neonatal and pediatric patients. The advantage of such devices is first of all safety (precise control of the flow rate) and the ability to gradually start and stop the infusion, which may enhance PN tolerance.

Filters

The use of in-line filters during PN administration is effective for the mechanical removal of larger particles, precipitates, bacteria, fungi, larger lipid particles, and air. The greatest concern relates to AIO admixtures containing calcium phosphate precipitates, which can cause diffuse microvascular lung embolisms. The 1.2-µm filters should be used for the administration of lipid-containing solutions including AIO admixtures and changed every 24 hour. The 0.2 µm endotoxin-retaining filters should be used for the administration of non-lipid–containing solutions and can be changed every 96 hours. No evidence has been found to support the use of in-line filters for preventing infusate-related CRBSIs.[368,369]

Infusion Sets

The risk of colonization, and therefore CRBSI, is higher if the infusate administered supports proliferation of contaminating organisms. The administration of PN has been associated with an increased risk of catheter-related infection, as lipid emulsion is particularly suited to the growth of a wide range of microorganisms. A Cochrane systematic review concluded that administration sets that do not contain lipids, blood, or blood products may be left in place for intervals of up to 96 hours without increasing the incidence of infection. Administration sets that contain lipids should be changed every 24 hours, and is currently recommended.[370] A study in neonates has shown that microbial contamination of infusion sets is significantly more frequent with 72- than with 24-hour set changes in neonates receiving lipid solutions.[371]

The placement and long-term use of IV catheters exposes the patient to a wide variety of risks, which generally can be grouped into mechanical, infectious, and metabolic categories. The infectious and metabolic complications and how to avoid them are extensively discussed in the other sections of this chapter.

Mechanical complications can occur intraoperatively or postoperatively. Intraoperative complications include erroneous catheter tip positioning, arterial puncture (with possible subsequent soft tissue hematoma and hemomediastinum), lung puncture (with possible pneumothorax and hemothorax), thoracic duct injury, guide wire-induced arrythmia (stimulus in the area of the tricuspid valve), myocardial perforation, nerve palsy, air embolus, misplaced catheters, and, if the catheter is infused after erroneous placement in the pleural space, hydrothorax. Postoperative complications include thrombosis, fibrin sheath formation, catheter occlusion, catheter erosion through the vein or the heart, arteriovenous fistula formation, catheter embolus, and clot embolus. Retrieval of fragmented catheter emboli can now be safely and effectively accomplished percutaneously.

Long-term (>3 months) home parenteral nutrition (HPN) requires a long-term venous access device, such as a cuffed tunneled central catheter (Hickman, Broviac, Groshong, and Hickman-like catheters such as Lifecath, RedoTPN, etc.) or a totally implanted port. An alternative to central venous cannulation is a PICC using an ultrasound-guided cannulation of a peripheral vein in the upper arm. The advantages of these peripheral access sites are lower rates of acute complications such as pneumothorax, life-threatening bleedings, and vein manipulation even in small infants.[372]

INITIATION AND MONITORING OF PN IN HOSPITAL

An individualized plan for initiation, advancement, and nutritional support goals should be formulated before starting PN. A multidisciplinary team including pediatrician or surgeon, dietitian, pharmacist, and nurse (nutrition support team) should be involved in the process of patient' assessment and planning the nutrition support strategy. Exhaustion of all possible modalities of advanced enteral nutrition support (specialized formulas and diverse delivery protocols) should be attempted before the initiation of PN. PN is not indicated in patients with adequate small intestinal function in whom nutrition may be maintained by nasogastric

tube or gastrostomy feeding.

Baseline nutritional assessment including clinical examination and anthropometry, review of enteral nutrition supply and baseline laboratory parameters should be performed before planning PN therapy. The plan is based on estimates of fluids, energy, and protein requirements, should account for increased or decreased needs of each patient and consider other fluid requirements for intravenous drugs. Evaluation of energy requirements and goals should be defined, either using conventional equations or better by IC, especially for children in PICU.

The initiation protocols regarding timing and composition of PN in premature and VLBW infants are related to the general principles of early aggressive nutrition, as discussed in the previous sections. PN should be administered within the first day of life to provide adequate nutrition and minimize early weight loss and protein breakdown. Initial protein intake should be at least 2–3 g/kg/d, and preferably 3–3.5 g/kg/d. Glucose should be administered immediately after birth, preferably at rates of at least 6 mg/kg/min, but rates as low as 3.5 mg/kg/min are acceptable in the presence of hyperglycemia. Lipids should be provided from the first day at 1 g/kg/d to supply energy for protein metabolism and growth. Most advocate some fluid restriction on the first days of life as outlined in the section on fluid intake. Calcium and multivitamins should be provided from the first day, but other electrolytes are not included in the initial PN solutions. Close monitoring of weight loss and laboratory tests should dictate the rate of fluid intake and the timing for the addition of electrolytes. Protein and lipid intake should be gradually increased to meet the target intakes. Close monitoring of blood glucose levels should guide the increase in glucose infusion rates that can be achieved either by increasing volume (fluid intake), or by increasing the percent of dextrose in the PN solution (from 5% up to 10%, and possibly up to 12.5%). This approach may allow minimization of the cumulative nutrient deficits and reduction or even elimination of postnatal extrauterine growth restriction in VLBW preterm infants.[39,373]

The steps in devising a PN prescription are as follows:

1. Determine the amount of protein to be given and calculate the caloric/nitrogen ratio, infants 150–200 kcal/1 g nitrogen, in older children 100–150 kcal/1g nitrogen (or 30–40 kcal for each 1 g protein).
2. Determine the energy requirements and distribution between glucose and lipids. The distribution of calories derived from macronutrients depends on the clinical setting: 10% to 20% of

calories in the PN solution can be derived from protein, CHO and lipids may supply between 50%–70% and 30%–50% of nonprotein energy respectively.

3. Calculate the amount of dextrose to be used initially, during advancement, and for maintenance.
4. Determine the dose of lipid, no data endorse the common practice of gradually advancing the amino acid or lipid dose in pediatric PN.
5. Determine fluid volume and rate, based on the patient' fluid requirements.
6. Calculate the electrolytes and mineral requirements.
7. Determine the amounts of vitamins and TEs.

Suggested ranges for macro- and micronutrient supply in pediatric PN solutions are given in each section of the chapter.

To be able to correct dosages early and to minimize complications such as fluid and electrolyte imbalances, disturbances of acid-base balance, hyperglycemia, and hypertriglyceridemia, clinical and laboratory monitoring of the patients receiving PN is mandatory. Periodic nutritional assessment including weight, height, and mid-upper arm circumference (MUAC) should guide short-term and long-term nutritional support.[374–376] An in-hospital monitoring schedule is shown in Table 68-11.

Nutritional assessment of preterm infants in the stable growing phase, especially if they are on prolonged PN, should include the following:

- Growth parameters
  - Daily weight gain:
    - ≤35 weeks: 15 g/kg/d
    - >35 wks: 20–30 g/d
  - Once a week head circumference ~0.7–1.0 cm/week
  - Consider also once week length ~1 cm/week
- Daily follow-up of urine glucose (glycosuria can be present without hyperglycemia) Nutritional Lab (at least once a week for infants on PN):
  - Glucose
  - Complete Blood Count: Hemoglobin, Reticulocytes.
  - Electrolytes
  - Minerals: Calcium, Phosphorus, Alkaline Phosphatase (sensitive markers for mineralization and chemical osteopenia of prematurity)
  - Urea/BUN (as markers for protein status and amino acids intake)
  - Triglycerides (as marker of lipid tolerance ~up to 150–200 mg/dL,

may accept up to <250 mg/dL)
  - Urinary phosphorus
  - Consider also:
    - Creatinine (as marker of renal function)
    - Total protein, albumin (or preferably prealbumin, a better indicator of protein status)
    - Total and direct (conjugated) bilirubin and liver function tests, especially if PN-associated cholestasis is suspected
    - Zinc
- Further tests that may be occasionally considered, especially if there is evidence of chemical osteopenia of prematurity (because the maximum intake of calcium and phosphate in PN does not supply the minimal requirements, preterm infants on PN are at increased risk for osteopenia of prematurity):
  - X-Rays
  - Bone density/Ultrasound

TABLE 68-11 In Hospital Monitoring After PN Initiation

Timing	Monitoring
Baseline	Nutritional assessment including: 1. Evaluation of enteral nutrition support adequacy 2. Anthropometry 3. Biochemistry: electrolytes, blood urea, calcium, phosphorus, magnesium, albumin/prealbumin, liver enzymes, serum triglycerides, hemoglobin/Ht
Daily	Weight, input-output, urine glucose Glucose monitoring 2 times daily initially Inspection of catheter site
3–4 times/week initially	Biochemistry: electrolytes, blood urea, calcium, phosphorus, magnesium, albumin/prealbumin, liver enzymes, serum triglycerides Hemoglobin/hematocrit Urine glucose and electrolytes
Weekly thereafter	Idem and follow-up nutritional assessment

As stated earlier, multidisciplinary and qualified pediatric nutrition support teams are of paramount importance for provision of high quality nutrition care. Nutrition support teams should be involved in the initiation and monitoring of enteral and PN therapy in the hospital and at home. Several studies have documented that nutrition support teams improve

clinical and economic outcomes by reducing the incidence of PN–associated complications, encouraging the use of alternative feeding routes, and by reducing wastage of PN solutions and resources.

At the Royal Children's Hospital in Melbourne, the introduction of a formal Nutrition Support Service in 1994 was associated with a 61% reduction in PN costs over five years, despite a 23% increase in hospital admissions and a 24% increase in surgical procedures over the corresponding period.[377]

METABOLIC COMPLICATIONS

Liver, Biliary, Bone, and Renal Complications

Since its introduction, more than fifty years ago, PN has benefited and saved the lives of many patients with a variety of clinical conditions. PN, however, has been associated with a large spectrum of possible complications related to both the PN solution (metabolic complications) as well as the technique of intravenous delivery (catheter-related complications). Metabolic complications of PN can be divided into acute metabolic complications and chronic (long-term) metabolic complications. A comprehensive list of PN-related short-term metabolic complication is presented in Table 68-12. Acute complications, such as hypoglycemia or electrolytes disturbances may be life-threatening but can be easily managed. Chronic, long-term metabolic complications are however, more problematic, as the etiology of these long-term side effects is multifactorial and often not fully understood.

Intestinal Failure Associated Liver Disease

The reported prevalence of IFALD varies widely, but is estimated to be 40% to 60% in children and as high as 85% in neonates receiving PN for prolonged periods of time.[378,379] A recent survey from the United States, Pediatric Intestinal Failure Consortium reported that cholestasis was present in 75% of 272 infants included in the analysis.[380] In adults, serious hepatic dysfunction is much less common, occurring in 15% to 30% of patients.[381]

TABLE 68-12 Short-Term Metabolic Complications Associated with PN

	Cause	Prevention/Interventi

Fluid disturbances	Fluid overload	Refeeding syndrome	Reduce fluids supply at the start
	Osmotic diuresis	Cardiac or renal insufficiency or excessive infusion rate Hyperglycemia and glycosuria	Decrease fluid, increase nutrient concentration Reduce dextrose infusion ra or concentration and monitc give insulin
Acid-base disturbances	Metabolic acidosis	Renal or gastrointestinal losses; infusion of hydrogen	Increase acetate in solution Avoid excessive protein and/or calories; postinjury phase, increased metabolic rate
	Metabolic alkalosis	Base intake or gastric drainage	Provide additional fluid and NaCl, KCl, or NH_4 Cl
	Respiratory acidosis	Increased supply of calories as dextrose	Decrease fraction of calorie from dextrose and increase fraction of calories from lipic
Electrolytes and minerals abnormalities	Hyponatremia	Gastrointestinal or renal losses; inadequate supply	Increase sodium in solution decrease fluid volume; monitor intake and output,serum and urinary sodium and urine-specific gravity
	Hypokalemia	Gastrointestinal or renal losses; refeeding syndrome; high dextrose infusion; hypomagnesemia	Increase potassium up to 4 mEq K/kg/d; monitor serum levels, check and correct metabolic alkalosis, hypomagnesemia
	Hypomagnesemia	Inadequate magnesium in solution; refeeding syndrome; gastrointestinal or renal losses	Increase magnesium in solution; monitor serum leve
	Hypocalcemia	Refeeding syndrome; inadequate calcium	Increase calcium in solution monitor serum levels
	Hypophosphatemia	Inadequate phosphate; refeeding syndrome; high dextrose supply	Increase phosphate in solution; monitor serum leve
	Hyperphosphatemia	Renal insufficiency	Decrease phosphorus in nutrient solution but do not

			omit; eliminate other phosphate source
Glucose metabolism abnormalities	Hypoglycemia	Abrupt interruption of glucose/PN infusion	Graduate decrease of PN infusion rate during cycling
	Hyperglycemia	Excessive infusion rate or dextrose concentration	Reduce rate or concentratio of dextrose; rule out sepsis; concurrent steroid therapy; diabetes mellitus
Protein metabolism disturbances	Azotemia	Dehydration; calorie-nitrogen imbalance; renal dysfunction	Correct dehydration; increas nonprotein calorie-to-nitroge ratio or reduce amino acid concentration
	Hyperammonemia	Hepatic dysfunction; excessive amino acids infusion; insufficient arginine	Reduce protein concentration; increase nonprotein calorie-nitrogen ratio
Lipid metabolism	Hypertriglyceridemia	Lipid infusion dose too high; excess glucose supply; acute inflammatory state	Decrease total lipid supply o rate of infusion; decrease caloric/glucose supply
	EFAD	Fat free PN	Provide at least 2–4% of calories as fat (0.5–1.0 g/kg/d)
	Rash	EFAD or allergy to egg	The first time PN should be delivered in hospital under medical supervision
Vitamin disorders	Vitamins' deficiencies or excess	Inadequate intake	Provide vitamins from the fi day of PN; monitor serum levels
Trace elements disorders	Deficiency or excess	Inadequate or incomplete trace element supply; gastrointestinal losses (zinc)	Routine trace element supplementation, monitor a make changes according to medical condition

The development of IFALD significantly increases the risk of additional morbidity and mortality. Recent studies have shown that liver disease in patients on long-term PN is strongly correlated with survival; in a cohort of 78 children with short bowel syndrome (SBS), the survival rate among those with cholestasis (conjugated bilirubin >2 mg/dL), was close

to 20% compared with 80% in those without cholestasis.[382] Overall, approximately 15% of patients with IFALD progress to end-stage liver disease requiring either isolated or combined liver–small bowel transplantation, a treatment that is associated with a high morbidity and mortality.[383]

IFALD is a multifactorial entity with poorly understood pathogenesis and multiple risk factors. Contributing factors to PN-associated liver disease (PNALD) are patient related and PN related. These include prematurity and LBW, lack of enteral nutrition, immature bile secretion, inflammation, oxidative stress, infection, nutrient deficiencies or excesses, and the underlying disease. Prematurity and CRSBIs have been shown to be the most important factor involved in the development of IFALD. Successive episodes of sepsis have been shown to result in a 30% increase in the serum bilirubin level. Sepsis often results in inflammation and oxidative stress, but both of these processes can also occur without an underlying infectious etiology and are thought to have a role in the development of PNALD. The proinflammatory cytokines are potent inhibitors of hepatobiliary transporter genes expression, which may be responsible for impaired bile secretion resulting in hyperbilirubinemia and cholestasis. In addition, highly reactive oxygen species are capable of cell injury through lipid peroxidation, oxidation of proteins, and destruction of DNA.[384,385] Lack of EN lead to a decrease in the release of counterregulatory hormones secreted from the gut, thereby decreasing bile flow and delaying gut maturation. Deficiencies or excess of PN macro- and micronutrients and bacterial overgrowth are all contributing to the etiology of IFALD.

The clinical spectrum of IFALD includes hepatic steatosis (more prevalent in adults), cholestasis (more frequent in infants and children), fibrosis, and, in some, progression to hepatic cirrhosis with portal hypertension and end-stage liver disease.[386] A late occurrence of hepatocellular carcinoma has been also reported in children with IFALD. Clinically, hepatic dysfunction is manifested by jaundice, hepatomegaly, splenomegaly, ascites, edema, and signs of portal hypertension in children with cirrhosis (spider nevi, palmar erythema, and caput medusae). The laboratory diagnosis of IFALD is characterized by hyperbilirubinemia/conjugated hyperbilirubinemia (>2mg/dl), abnormal international normalized ratio (INR; hepatic synthetic function), alanine aminotransferase (ALT; hepatic injury), gamma-glutamyl transferase (GGT; cholestasis), alkaline phosphatase (biliary or bone disease), and reductions in platelet count and absolute neutrophil count. Sepsis can have

a confounding effect on these laboratory values, specifically causing increases in serum bilirubin, INR, and aminotransferase concentrations while lowering the platelet count, therefore, the presence of an infection at the time of laboratory studies should be taken into consideration.

The gold standard for diagnosis of IFALD is liver biopsy, but this is seldom done due to the invasive nature of this procedure and the critical condition of many of these children. Histology picture of IFALD shows usually cholestasis with periportal inflammation and bile duct proliferation and fibrosis progressing to cirrhosis in severe cases. However biochemical reversal of IFALD had been reported even in the presence of significant fibrosis.[387] Cholestasis should be, however, attributed to PN only after other causes have been excluded. Infection should always be considered in any patient receiving PN who develops hyperbilirubinemia. All infants with sustained conjugated hyperbilirubinemia should be evaluated for biliary obstruction (i.e., biliary atresia, choledochal cyst, cholelithiasis). In addition, metabolic, genetic liver disease and toxicity secondary to pharmacologic agents should be considered and investigated.

Prevention of IFALD should address all the possible amendable risk factors. This includes prompt treatment of CRBSIs and bacterial overgrowth, early institution of enteral nutrition/timely weaning from PN, PN cycling and balanced PN supply, considering the need for sufficient nutrient supply while avoiding excess of calories/fat/glucose.

There are no definite or proven treatments after the development of IFALD. The use of ursodeoxycholic acid (UDCA) has been examined in a systematic review which confirmed that administration of UDCA is generally followed by improvement in biochemical indices. However, the review also highlights the serious limitations of the currently available studies, including small sample sizes, absence of randomization and controls, short duration, and lack of control for confounding variables. There is no evidence that routine use of UCDA will improve clinical outcomes such as survival, rate of hospitalization, incidence of sepsis, or progression to cirrhosis. No definite data on prophylactic benefits of UDCA exist.[388] Erythromycin has been used to promote upper gastrointestinal motility and has been shown effective in promoting feedings in neonate. This agent has also been shown to lower significantly the incidence of IFALD.[389,390] Other medications that have been used empirically, including cholecystokinin, cholestyramine, corticosteroids, and phenobarbital, have little to no evidence to support their routine use.

Parenteral Nutrition Ingredients and IFALD

Unbalanced nutritional supply of energy and macronutrients, deficiencies as well as excesses, and toxicities have been associated with the developments of IFALD and all PN components have been implicated. Changes in PN composition and regimen (cycling) are key strategies for the reversal of established cholestasis.

Excess of energy delivered either as dextrose or fat calories may promote hepatic steatosis by stimulating insulin release, which inhibits FA oxidation and promotes lipogenesis.[391] In the presence of hypertriglyceridemia, fatty liver, or developing IFALD, reduction of energy supplied by PN while increasing the enteral nutrient intake, whenever possible, is advised. The development of IFALD has been also related to the toxicity or deficiency of several amino acids.[392] One study showed that premature infants with parenteral protein intake of 2.5 g/kg/d or 3.5 g/kg/d had similar incidence of cholestasis. Increasing parenteral protein intake, however, was associated with earlier onset and increased magnitude of cholestasis whereas increased dextrose intake increased the prevalence of cholestasis.[393] It seems that high cumulative amino acid doses may be associated with development of IFALD in children, although a definitive relationship has not been demonstrated. It is not clear what role the kind and dose of amino acids play in the development of IFALD in older children and adults, but this does not seem to be as significant as it is in the full term and more so in the premature infant. Deficiency of specific amino acids such as taurine and cysteine may play a role in the development of IFALD especially in premature infants.[394] Yet, studies found no significant differences between cholestasic and noncholestatic infants with respect to the relative percentages of protein, glucose, and lipid calories in the PN infusates.[395]

The role of ILE in the development of liver complications has been related to the fat dose, source of FA, and the phytosterol content. Both liver steatosis and cholestasis have been associated with excessive ILE supply. Multivariate analysis demonstrated that cholestasis and severe IFALD are strongly associated with ILEs supply above 1 g/kg/d.[396] Evidence of the value of lipid minimization comes only from retrospective or uncontrolled studies, however, all these studies reported improvement or disappearance of cholestasis when ILE were discontinued or lipid dose reduced to less than 1–1.5 g/kg/d.[397] A recent study showed that reduction of soybean-based ILE to 1 g/kg/d delivered twice weekly was associated with a significant reduction in total bilirubin levels compared to infants

receiving 3 g/kg/d (historical control).[398] However, in a retrospective study in neonates, provision of 1 g/kg/d of a soy-based ILE compared to 2–3 g/kg/d did not prevent (cholestasis incidence 51.7% in 1-g group; 43.8%, in 2- to 3-g group, not significant), or delayed the onset of cholestasis (32.6 ± 24.1 days, in 1-g group; 27.7 ± 10.6 days in 2- to 3-g group, not significant).[399] Soybean-based ILE, historically the most commonly used fat emulsion, contains mainly ω-6 PUFAs. Clinical and experimental evidence suggested a role for soybean oil–based ILE in the onset of IFALD.[400,401] Alternative ILEs based on combinations of soybean oil with MCT or olive oil–based ILE have been developed to reduce the ω-6 PUFA content. Animal studies have shown that ω-3 FAs reverse hepatic steatosis, reduce oxidative stress, have less proinflammatory effects and do not impair bile flow. Omegaven and SMOFlipid have both been used for the treatment of IFALD. Gura et al. have published the first report of two children with severe IFALD who achieved complete resolution of cholestasis after two months treatment with Omegaven.[178] To date, several case series from Boston and Canada have reported that use of Omegaven 1 g/kg/d or a combination of Omegaven (1 g/kg/d) with Intralipid (1 g/kg/d) improves cholestasis and hyperbilrubinemia in children with established IFALD.[402,403] The results of an ongoing trial on term and near term (>34 weeks) neonates comparing SMOFlipid with olive oil–based lipid emulsion are expected to provide new evidence of the advantage of the newer FO–based ILEs over other than soybean oil–based ILEs.[404]

Early reports of FO–based ILE used outcomes based on clinical presentation and biochemical markers of cholestasis (AST, ALT, total bilirubin, and direct bilirubin) and not based on liver biopsy results. Investigators from Boston have now reported 83 biopsy results taken in 66 children. Of the 83 biopsies, 74 demonstrated fibrosis; 8 of these also demonstrated cirrhosis. Forty-one of the 74 fibrotic biopsies (55%) were obtained in patients without biochemical evidence of cholestasis. Seventy percent of these patients were treated with FO–based ILE during their clinical course.[405]

Cyclic infusion of PN implies daily supply of PN components over less than 24-hour period, usually 8–12 hours. Allowing each day a period off PN may reduce the risk of liver complications, especially in patients requiring long-term PN. Metabolic studies have shown that 24-hour lipid oxidation was higher and 24-hour dextrose utilization lower during cyclic vs. continuous TPN. Compared with continuous infusion, cyclic infusion of PN has been shown to result in a reduction of serum liver enzyme and

conjugated bilirubin concentrations associated with reduced hyperinsulinemia as well as fat deposition in the liver in both adults and children.[406]

The role of carnitine and choline in the etiology, prevention, and treatment of PN-associated liver complications remains to be established. Previous studies showed that patients on long-term PN had carnitine deficiency, but the prophylactic effects of carnitine supplementation on hepatic steatosis have not been well documented.[407] Studies in humans have shown that choline supplementation can reverse hepatic abnormalities and even liver steatosis.[408] A decrease in antioxidants such as vitamin A, vitamin E, and selenium has also been reported in IFALD, although supplementation with antioxidants to normalize their levels does not guarantee a clinical improvement. With modern solutions, there is little evidence that other components of PN solutions such as aluminum, chromium, or other micronutrients are anymore responsible for PNALD.[409]

Progression to full enteral feeds remains the primary goal for all patients with IFALD. Surgical interventions may assist to improve gut absorptive capacity and shorten the duration of PN. Surgical procedures considered useful in a patient with SBS or associated IFALD include placement of a feeding tube (i.e., gastrostomy or jejunostomy tube) to allow for enteral feeds, tapering enteroplasty (in children with dilated loops and bacterial overgrowth) and several types of intestinal lengthening procedures: Bianchi and the serial transverse enteroplasty procedure.[410,411]

Patients with liver and IF who fail to achieve intestinal rehabilitation and for whom death is otherwise unavoidable may be offered a combined liver and intestinal transplant despite the formidable immediate and long-term risks. The University of Pittsburgh, a large transplant center, has recently reported its 5-year patient survival (for isolated intestinal transplants and combined liver/small intestinal transplants) at 77% with a corresponding graft survival rate[412,413] of 58%.

Biliary Tract Complications

The loss of enteric stimulation occurring in the absence on enteral intake impairs the gallbladder motor function, favoring the development of a sluggish gallbladder. This explains the frequent association of PN administration with gallbladder dysfunction, including abnormal gallbladder dilatation and the development of acalculous cholecystitis, biliary sludge, and gallstones. Ileal disease or resection, surgery, rapid

weight loss, drugs, and bacterial overgrowth have also been shown to have a role. Biliary sludge may disappear spontaneously, but frequently evolves into gallstones. Sludge may persist or recur in 50% of cases, and gallstones may form in up to 14% of affected subjects over the age of 3 years.[414]

Prevention of gallbladder stasis, avoiding drugs that decrease gallbladder contractility and slow small bowel transit, promoting enteral nutrition (protein and fat/LCTs), prevention and treatment of small bowel bacterial overgrowth, and use of UDCA are the currently available therapeutic options for prevention of biliary complications.

Bone Complications

Patients on long-term PN are at significantly increased risk of developing metabolic bone disease. Bone disease in patients on long-term PN can be asymptomatic or associated with bone pain or with fractures which may occur with no or minimal trauma. Examination of bone histology or bone mineral density (BMD) in patients on long-term PN reported 40% to 100% prevalence of bone disease.[415]

Studies of bone metabolism have found increased bone turnover at the start of PN which diminished in time in concert with decrease in bone mineralization.[416] Others showed an increase in the amount of osteoid, similar to the defect seen with osteomalacia.[417] Shike et al. showed in a long-term PN cohort of patients that osteoporosis was present rather than osteomalacia.[418]

The pathogenesis of the bone disease is not completely understood, but it is probably related to several factors. Some are PN-related, some are related to the primary disease (malabsorption of calcium and vitamin D, chronic inflammation, medication, in particular corticosteroids) or general lifestyle (sedentary lifestyle, smoking, and alcohol abuse). The possible PN-related factors are changes in the balance of calcium and phosphorus intake, acidosis, excess or low parenteral intake of vitamin D, deficiency of vitamin K, fluoride or copper, and aluminum toxicity.

A small number of the initial cases of PN-associated bone disease were likely related to aluminum contamination of amino acid solutions prepared from protein hydrolysates.[419] Aluminum is still present in calcium gluconate, phosphate salts, vitamins, and TE solutions; however, the amounts are far less than those encountered in casein hydrolysates, and they are not believed to make a significant contribution to bone disease.[420] Hypercalciuria is a frequent occurrence during PN and has been associated with excessive amino acids, dextrose, and sodium delivery. Low

phosphorus supply and cycling PN infusion also increase the renal loss of calcium.[421–423]

Evaluation of bone metabolic status should include a thorough history, physical examination, anthropometry, and laboratory assessment. Laboratory studies should include a complete blood count, serum chemistry panel to exclude kidney and liver disease, and a thyroid-stimulating hormone study (TSH) to evaluate for hyperthyroidism. Evaluation of parathyroid hormone (iPTH) and 25-hydroxyvitamin D is also mandatory. Baseline and periodical dual-energy absorptiometry (DEXA) should be considered in patients on HPN.

Providing adequate amounts of minerals and vitamins form the foundation for maintaining the integrity of bone. Optimizing the composition of PN infusion to avoid the development of negative calcium balance and minimize the risk of developing or exacerbating underlying bone disease is imperative.

Various attempts have been made to treat IF–associated bone disease, both by modifying PN composition and by administering substances such as calcitonin, iPTH, sex hormones, and bisphosphonates. Several reports and a placebo controlled study have shown that spinal and hip BMD can be improved in both adults and children, by administration of intravenous bisphosphonate, especially in the absence of corticosteroid treatment.[424–426]

Renal Complications

The etiology of renal dysfunction is most probably multifactorial and includes recurrent dehydration and electrolytes abnormalities, excessive protein load, solutions' contamination, and recurrent central line infections.[427,428] Impairment of glomerular and tubular function has been documented in both adults and children receiving PN.[429,430] Moreover, cycling delivery of PN has been shown to influence urinary volume and electrolyte excretion and has been associated with diurnal hypercalciuria in several studies published so far in adults.[431]

CATHETER-RELATED COMPLICATIONS

Catheter-Related Blood Stream Infections

The incidence of nosocomial CRBSIs in PICU patients, is reported to be as high as 9 per 1000 CVC-days.[432] In children on chronic PN, investigators have reported incidence rates of CRBSI of 2.1 per 1000 HPN days.[433] In

most children, CRBSIs typically arise from bacterial colonization of the internal surface of the intravascular catheter from migration of skin flora. In children with IF, however, bacterial translocation from the injured gut or small bowel bacterial overgrowth represents additional sources of bacteria that can result in catheter infection. Furthermore, daily catheter manipulation for PN or fluid administration increases the risk of the catheter serving as a port of entry for infection.[434] The most commonly reported causative pathogens are coagulase-negative staphylococci, *Staphylococcus aureus*, enterococci, and *Candida* spp. Gram negative bacilli account for 19% and 21% of CRBSIs reported to Center for Disease Control (CDC).[435,436]

Diagnosis of CRBSIs

In the presence of fever (38°C) in a child with a central line, whether or not another source of infection has been identified, central and peripheral cultures should be obtained, and the child should be treated with intravenous antibiotics covering both gram positive cocci and gram negative bacteria. Diagnosis of CRBSI is best achieved (a) by quantitative or semiquantitative culture of the catheter (when the CVC is removed or exchanged over a guide wire), or (b) by paired quantitative blood cultures or paired qualitative blood cultures from a peripheral vein and from the catheter, with continuously monitoring of the differential time to positivity (if the catheter is left in place).[437]

Treatment of Catheter-Related Infections

For salvage of the device in patients with uncomplicated CRBSI, two weeks antibiotic treatment with standard systemic therapy for catheter-related bacteremia due to *Staphylococcus aureus*, coagulase-negative staphylococci, and gram negative bacilli is recommended. Exit site infection (tenderness, erythema, or purulence at exit site or on dressing) should be treated for two weeks with empiric antibiotics for *Staphylococcus aureus.* The antibiotics should be adjusted based on culture and sensitivity. If no improvement is present after a second course of antibiotics, catheter removal is needed and replacement at other site.[437] Removal of the long-term venous access is required in case of (a) tunnel infection or port abscess, (b) clinical signs of septic shock, (c) paired blood cultures positive for fungi or highly virulent bacteria, and/or (d) complicated infection (e.g., evidence of endocarditis, septic thrombosis, or other metastatic infections). Reinsertion of long-term devices should be

postponed until after appropriate systemic antimicrobial therapy is begun, and after repeat cultures of blood samples yield negative results. If time permits, insertion of a new device in a stable patient ideally should be done after a systemic antibiotic course of therapy is completed, and repeat blood samples drawn 5–10 days later yield negative results.[437]

CRBSIs Prophylaxis

Reducing the risk of CRBSIs may have a significant impact on the rate of readmission, cost of care, and quality of life (QOL), and the progression of IFALD in patients on long-tem PN. Evidence indicates that the risk of catheter-related infection may be reduced by using particular devices (tunneled and implanted catheters, antimicrobial coated catheters, and single-lumen catheters dedicated only to PN). Additional recommendations are use of peripheral access whenever possible, ultrasound-guided vein puncture, and maximal barrier precautions during insertion as well as appropriate nurse stuff and patient training on proper catheter care (hand washing technique, use of 2% chlorhexidine as skin antiseptic, appropriate dressing of the exit site, disinfection of hubs, stopcocks, and needle-free connectors, and regular change of administration sets).[437] In-line filters, routine replacement of central lines, systemic or antibiotic lock prophylaxis, and the use of heparin have not been shown to be effective in reducing the risk of infection, and should not be adopted for this purpose according to ESPEN guidelines.[437]

The 2011 report from Centers for Disease Control and Prevention recommend, however, the use of antibiotic locks in patients with long-term catheters who have multiple line infections despite adherence to basic principles of infection prevention. There is no recommendation regarding what type of antibiotic or the duration of therapy. One of the main concerns with the routine use of antibiotic locks is the development of resistant strains of bacteria.[438] A systematic review that looked at the evidence on the effectiveness of antibiotic-based catheter lock solutions found 16 trials: nine in hemodialysis patients, six in oncology patients (mainly children), and one study on critically ill neonates. Three hemodialysis patients needed to be treated with antibiotics to prevent one CRBSI, given an average baseline risk of 3.0 events per 1000 catheter-days. In the oncology patients, the calculated number needed to treat (NNT) was of eight patients to prevent one CRBSI, given average baseline risk of 1.7 events per 1000 catheter-days. The conclusions of the systematic review favored the use of antibiotic-based lock solutions over

heparin lock for prevention of CRBSI in hemodialysis patients and were marginally effective in oncology patients.[439] A recent randomized controlled trial conducted in pediatric patients that compared taurolidine citrate to heparin locks in children receiving chemotherapy reported a significant decrease in CRBSIs incidence from 1.3 per 1000 catheter-days in the heparin group to 0.3 per 1000 catheter-days in the taurolidine/citrate group.[440]

An alternative to antibiotic locks are ethanol locks (ELs). ELs were first introduced in 2003 by Ball et al. and Danneberg et al. as treatment of frequent CRBSIs in PN-dependent and pediatric oncology patients.[441,442] Ethanol has two desired effects of a lock solution: it is antimicrobial and anti-fibrinolytic. It is readily available and has not shown antimicrobial resistance. Sherertz et al. characterized the antimicrobial efficacy of ethanol in an in vitro study: Ethanol concentrations of at least 30% for an incubation time of two hours, four hours, and 24 hours were shown to be similarly efficient in killing *Staphylococcus aureus*.[443] Mukherjee et al. reported that 10 minutes and 30 minutes of 70% ethanol exposure were enough to inhibit biofilm formation and *Candida albicans* growth.[444] Studies on the effect of ethanol on silicone or polyether urethane catheters integrity have shown only minimal changes in catheters quality most likely not clinically relevant.[445]

Several retrospective studies confirmed the effectiveness of ELs in CRBSIs treatment and prevention in mixed pediatric and home PN populations. ELs dwell time ranged from >2 hours per day to four hours three days per week. Lately, most studies use a 70% ethanol solution. The volume used for the catheter lock range from 0.2 mL to 3.0 mL. A recommended upper limit of absolute ethanol dosage of 1 mL/kg per day was ascertained. The CVC is flushed before and after administration of PN or other medications with 5 to 10 mL normal saline and the ethanol solution is withdrawn after the initial dose to avoid potential intoxication in pediatric patients. All studies reported significantly decrease in CRBSIs during EL prophylaxis from 8.0 mean CRBSIs rate per 1,000 catheter days before EL treatment to 1.3 after EL treatment ($P < .01$).[446–450] A recent meta-analysis of four prospective observational studies have revealed a mean rate difference (MRD) of CRBSIs of 7.67 (95% CI 5.87 to 9.47; $P = .00001$). ELs significantly reduced the risk of CRBSIs by 81% (RR 0.19, 95% CI 0.12, 0.32; $P = .00001$).[451] Adverse events were infrequent, but included serious complications, such as disseminated intravascular coagulation and catheter and deep vein thrombosis.[446,448,449] Two studies

also reported CVC-associated thrombus formation.[448,449] Infectious Diseases Society of America, however, states that although the role of ELs for CRBSIs prevention is becoming clearer, not enough evidence is available to promote ELs as a standard CRBSIs treatment or prevention option.[452]

Additional measures to prevent CRBSI in patients with recurrent infections that have been evaluated are antibiotic impregnated catheters or chlorhexidine dressing. The impregnated catheters are not widely used because of the associated increased cost and because of the CDC recommendation that these catheters should only be implemented for short-term use, particularly when there is need to enhance prevention of CRBSI after standard evidence-based strategies have been executed.

Retrospective analysis of the incidence of sepsis rates in a small group of patients on long-term PN, using cycled enteral antibiotics (CEA) reported significantly reduced rate of sepsis from 2.14 to 1.06 per 100 PN days (P = .014, median effect size 1.04, CI 95% 1.93 to 0.22), whereas the controls had no significant change (from 1.91 to 2.36 per 100 PN days (P = .402, median effect size 0.92 CI 95% 1.96 to 4.17).[453]

CVC-Related Mechanical Complications

CVC-related mechanical complications represent significant life-threatening events, for example, misplaced lines, lines that move, fall out, are pulled out, break, kink, or become occluded. CVC occlusion can be caused by anatomical or mechanical obstruction, drug precipitation or lipid residue, or thrombosis.[454]

Anatomical or mechanical obstructions can occur as a result of malpositioning of the catheter, catheter damage, or fracture due to traumatic insertion or kinking.

Calcium phosphate, diazepam, phenytoin, heparin, bicarbonate, mannitol, pentobarbital, and phenobarbital are all drugs that have been reported to be associated with precipitation. AIO PN solutions can produce a lipid residue that can result in CVC lumen occlusion.[455] When drug-precipitate occlusion is suspected, hydrochloric acid and sodium bicarbonate are used to alter the intraluminal pH to try to dissolve the drug precipitate.[456,457] Drugs with a pH > 7 are more soluble in an alkaline solution whereas drugs with a pH < 6 are more soluble in an acid solution. A 70% ethanol solution has been used to try and dissolve occlusions attributed to lipid residue.

Catheter occlusion due to thrombus formation is more frequent in

children than in adults and has been reported in 10% in children receiving HPN, 13% to 16% of oncology patients, and 31% of infants.[454] The differences in the reported incidence of CVC thrombosis are dependent on the population and setting studied and whether clinical or venographic evidence of thrombosis was reported. Clot may form as a fibrin sheath at the catheter tip, or a thrombus may form on the outside wall of the catheter, and/or on the wall of the vessel adjacent to the catheter. The various risk factors suggested for CVC-related thrombosis can be categorized into those related to the catheter, those related to the individual, and those related to the underlying disease and its treatment. The contribution of inherited and acquired thrombophilia to CV-related thrombosis is controversial.[458,459]

Most catheter-related thrombi develop in the upper extremities, as most catheters are located in the upper venous system. Patients with venous thrombosis may develop symptoms of pain, swelling of the arm and neck, edema, and discoloration of the arm. SVC syndrome may occur if thrombosis develops in the SVC or in both jugular veins. Symptoms of SVC syndrome include facial, extremity, and neck swelling, and dilated chest, neck, and facial veins. Most of the catheter-related thrombi are, however, asymptomatic and diagnosed solely by radiographic tests. The development of catheter-related thrombosis is usually a gradual process, permitting collaterals to form, which minimizes the acute symptoms. Although catheter-related thrombi are asymptomatic in most patients, serious complications may occur, including pulmonary embolism, increased risk of catheter infections, postthrombotic syndrome (limb pain and heaviness, swelling, cramps, edema, and varicosities), and loss of venous access.[454]

Diagnosing the cause for occlusion involves both clinical judgment based on recent medical history, review of medications and fluids received, and radiological investigations such as chest X-ray, venogram, and ultrasound. Ultrasonography is adequate for jugular thrombosis but inadequate for diagnosis of subclavian or SVC thrombosis. If the result is negative and clinical suspicion is high, venography is the method of choice.

Treatment of Catheter-Related Occlusion and Thrombosis

Many publications of nonrandomized, clinical studies, and case series suggest that thrombolytic drugs such as streptokinase, urokinase, and

tissue plasminogen activator are both safe and effective in treating CVC lumen occlusion caused by a thrombus, in adults and children alike. Treatment of thrombotic occlusions with tissue plasminogen has been shown to be more successful (90%) and has a lower complication rate compared with treatment with either streptokinase or urokinase. These agents catalyze the conversion of plasminogen to plasmin, which cleaves fibrin into fibrin degradation products to dissolve the thrombus. The major drawback of thrombolytic therapy is the increased number of major bleeding complications.[460,461] A recent Cochrane systematic review that assessed the efficacy and safety of different interventions used to restore patency of occluded CVC lumens in adults and children, identified only seven (two in children) randomized or quasi-randomized controlled trials (RCTs).[462,463] None of the included studies investigated chemical or surgical interventions for treating occluded CVCs. All seven studies investigated different comparisons or strengths of thrombolytic or anticoagulant therapies for treating CVC occlusion caused by a thrombus. There was some low evidence from two studies that investigated urokinase vs. placebo (RR 2.09, 95% CI 1.47 to 2.95) and tissue plasminogen activator 2 mg/2 mL vs. placebo (two studies, RR 4.19, 95% CI 2.44 to 7.20) that these two drug interventions may be effective in treating occlusion of CVC lumens caused by thrombosis.[464]

The recommended management of acute and symptomatic CVC-related thrombosis depends on the need to preserve the CVC, otherwise CVC removal is recommended. If the CVC is still required and is functioning, the CVC may be left in place, and anticoagulation therapy including low-molecular-weight heparin (LMWH) subcutaneously or unfractionated heparin (UFH) intravenously, started as the main initial therapy. The aims of initial anticoagulant therapy are to prevent thrombus extension and subsequent pulmonary embolization. After completion of the first three months of anticoagulant therapy, both vitamin K antagonists and LMWH can be used for long-term anticoagulation to prevent recurrent venous thrombosis. The American College of Chest Physicians recommends to treat catheter-related thrombosis for at least three months and if the catheter remains in place to continue anticoagulation treatment at a prophylactic dose. The recommended management of radiographically detected asymptomatic CVC related thrombosis is less clear.[465]

Prophylaxis

The prevention of catheter-occlusion is focused on methods to keep blood

out of the catheter. This can be obtained by flushing of the catheter and by use of antireflux valves. Many centers use routine flushing of the catheters with UFH or other solutions. Most studies in adults and children have shown no difference in efficacy between saline or heparin flushes. The 8th American College of Chest Physicians consensus conference on antithrombotic therapy recommends against routine primary prophylaxis for children of all age groups with central line catheters. Given potential safety concerns with the use of heparin (heparin-induced thrombocytopenia and loss of bone mineral), 0.9% sodium chloride may be the preferred flushing solution for short-term CVC maintenance.[466–468] In adults, three different meta-analyses of randomized controlled trials evaluating the effect of heparin on duration of catheter patency have concluded that intermittent flushing with heparin is no more beneficial than flushing with normal saline alone.[469–471]

Nevertheless, manufacturers of implanted ports or open-ended catheter lumens recommend heparin flushes for maintaining catheter patency and many clinicians feel that heparin flushes are appropriate for flushing devices that are infrequently accessed (less than eight hours). No randomized clinical trials have ever established the usefulness of heparinization or the ideal heparin concentration, though most authors suggest using a range of concentration between 10 and 100 units/mL in children.

In children on long-term PN, most on cycling regimen, consideration should be given to prophylactic flushing with heparin, or systemic anticoagulation (LMWH) that might decrease the rates of CVC-related thrombosis and thromboembolism, although there is no conclusive data on the benefit of these interventions.[472] A recent retrospective study in 18 children on long-term PN reported a 5-year cumulative thrombosis-free survival of 48% and 93% in the nonprophylaxis and prophylaxis (LMWH 16 children, vitamin K antagonists two children) groups, respectively (P = .047).[473] Further research is needed for more clearly defined guidelines and recommendations with regard to prophylaxis, treatment, and management of thrombosis and thromboembolism.

Needleless, antireflux devices can be attached to the hubs of most CVCs. These devices maintain positive pressure within the lumen of the catheter, which prevents backflow of blood. These devices have been studied mainly in adult and pediatric patients with non-tunneled CVCs and shown to decrease catheter-occlusion rates.[474]

CYCLING PN

Patients who are acutely ill or require short-term PN typically receive PN as a 24-hour continuous infusion. In stable patients, who are expected to have a longer course of PN or receive HPN, PN is often administered on a cyclic (discontinuous) schedule. Cycling implies that PN solution is administered over a shortened period, usually 8–16 hours, to allow patients a window of time off the infusion. Essentially, patients receive the same fluid volume, electrolytes, and nutrient load over a shorter period by increasing the infusion rate.

This technique, specifically nocturnal PN, was first proposed in adults in 1970 by Scribner et al. as a method to improve lifestyle.[475] The practice has subsequently been shown in both children (1.5 months to 20 years old) and adults to be a safe and effective means of nutritional support. Studies in adults have shown that cyclic and continuous PN infusions result in similar nitrogen balance and circulating counterregulatory hormone concentrations.[476] Furthermore, cyclic PN may be of benefit in the prevention and treatment of IFALD. Continuous PN infusion leads to continuously high serum insulin levels. This hyperinsulinemia causes hepatic lipid deposition and increased hepatic lipogenesis. Furthermore, EFAD may occur because insulin inhibits the release of free FAs from adipose tissue. These physiologic effects of continuous PN are reversed by cycling. The major proposed benefit is that of mimicry of normal physiology by altering circulating hormones and fuel source utilization. In adults, cyclic PN has been shown to induce variability in substrate utilization (as evidenced by measured RQ) that mimics more closely both the natural fasting state and the natural postprandial state as compared to traditional continuous PN. Following PN infusion, CHO oxidation decreases, fat oxidation increases, and the RQ decreases.[477–479] Diurnal variability in REE, circadian patterns of apolipoproteins, and a cyclic pattern of human growth hormone and insulin secretion have been demonstrated in children receiving cyclic PN.[480,481]

The use of cyclic PN as a means to normalize hyperbilirubinemia has been reported in both adults and children.[482,483] Cyclic PN was associated with a decreased incidence and delayed onset of hyperbilirubinemia in infants with SBS and after surgery for gastroschisis but not in a prospective RCT in VLBW infants.[406,484,485] A prospective study in infants who were switched from continuous to cyclic PN infusion demonstrated stabilization and possible decreases in serum bilirubin levels with initiation of cyclic PN infusion.[486] Cycling PN has been associated

with adverse events, too. These include hyperglycemia and respiratory distress due to the increase in the rate of dextrose and fluid infusion, hypoglycemia, and hypercalciuria. Therefore, cycling is more challenging in patients with underlying diabetes or renal, cardiac, or pulmonary dysfunction and in young children.

Cyclic PN in adults is usually performed in 12-hour intervals, but this is not generally possible in the neonatal population due to diminished glycemic reserves, which limits safe cycling to much shorter time intervals of up to six hours. Abrupt PN discontinuation in 15 children 2.4–11.4 years old led to no symptomatic hypoglycemia in a before–after study.[487] A randomized crossover study of 10 patients 1.5–36 months old showed significantly greater decreases in blood glucose levels with abrupt discontinuation compared with tapered discontinuation and higher rates of hypoglycemia (55% vs. 20%, *P* value not reported).[488] Blood glucose levels should be monitored following cessation of high glucose infusions. If not done by the infusion pump automatically, PN should be tapered by decreasing to half the rate for one hour and to one-fourth the original rate for another hour before complete discontinuation.[483] Although data are limited, cyclic PN infusion has not been associated with increased daily losses of calcium, phosphorus, magnesium, or vitamin D compared with continuous infusion.[489,490] Although calcium loss increases during cyclic PN infusion, there is no evidence that this contributes to metabolic bone disease.

Clinical data support the current widespread use of cyclic PN infusion in most populations. Generally, practicality is the primary drive for cyclic PN infusion and likely its greatest benefit. However, precautions must be taken to avoid hypoglycemia, particularly in young children and acutely ill patients who may not be suited to this modality.[491] The clinical significance of PN cycling-associated hypercalciuria should be further investigated.

HOME PARENTERAL NUTRITION

IF is defined as a reduction of functional gut mass below the minimum amount necessary for adequate absorption of nutrients, fluids, and electrolytes, to maintain health in adults and growth in children. The causes of IF include SBS, congenital or as a consequence of surgical resection; neuromuscular intestinal disorders, such as total aganglionosis or chronic intestinal pseudo-obstruction syndrome (CIPO); and severe protracted diarrhea syndromes due to congenital intestinal mucosal

disease, severe food allergies, enteroendocrine cells disorders, or immunologic diseases. The incidence of IF is estimated at 1 to 2 per million population.[492]

PN is the established therapy for chronic IF; it can be total or partial, permanent or temporary. In children and adults who depend on long-term PN, HPN is the only alternative to prolonged hospitalization and offers patients the best possible QOL. HPN offers children who would be otherwise institutionalized, the possibility of achieving their growth and developmental potential and best possible QOL at a reduced risk of nosocomial infections. As they get older, most can attend school and take part in normal social life. QOL scores from HPN-dependent children and siblings have been shown to be the same as those of healthy peers, suggesting effective use of coping strategies.[10] The increase in HPN use in the United States and in Europe has been demonstrated by several surveys performed over the past decade. A national point prevalence survey of HPN in United Kingdom in 2010 reported a fourfold increase in the number of children on HPN patients since 1993 (66 to 167). This reflects both increasing numbers and survival of patients with SBS, in addition to developments in home care.[493]

HPN Indications

SBS remains the main indication for HPN in children, accounting for 30% to 50% of the cases in most published surveys.[494] Pediatric studies have reported a mean HPN duration of about 2 to 10 years. Neonates with SBS had the best prognosis. Even after an extensive small bowel resection at birth, more than 80% of the children survive and acquire intestinal autonomy. The increase in the incidence of other indications, such as intractable diarrhea of infancy or CIPO, has led to an increase in the mean duration of HPN programs, some of which are “definitive,” the only possible alternative treatment being small bowel transplantation. Cancer, which is the main underlying diagnosis in adults on HPN in the United States and in Europe, is a marginal indication for HPN in children.[495,496]

Complications in HPN

Although PN is lifesaving to many pediatric patients and grants patients the ability to lead a life outside of the hospital, it is also a high-risk therapy with potential immediate and long-term complications:

IFALD

An increased risk of IFALD is associated with young gestational age, low birth body weight, young age at the start of PN, frequent episodes of bacterial infections or sepsis, long duration of PN, low enteral intake, very short intestinal length <10%, the presence of SBS with a stoma, and the presence of gastroschisis or intestinal atresia.[497–499] An appropriate PN regimen is also important as high parenteral energy intake, use of lipid emulsion at a dose of >1 g/kg, dextrose infusion >7 mg/kg/min, high amino acid load in premature infants, and deficiency of transsulfuration pathway products, most notably choline, have been reported to contribute to the development of IFALD.[500,501] Progression of IFALD to liver failure represents the greatest concern in terms of potential mortality for the long-term HPN patient. The risk of development of liver failure appears to be higher in children, specifically very young children, than in adults. In children, death due to IFALD accounted up to 36%, with a mortality rate greater in premature infants and in babies <1 year (38%) than in older children (30%).[502] Gupte et al. evaluated the survival rate of children referred for intestinal transplantation (ITx) and observed a significant difference depending on the severity of the cholestasis. The 6-year survival of children on HPN was 80% when the pretransplant total serum bilirubin concentration was <100 mmol/L, but when >100 mmol/L survival was only 40%.[503] The U.S., Pediatric Intestinal Failure Consortium also reported that the cumulative percentage of survival in children with cholestasis was lower, 79% vs. 95% at 1 year and 73% vs. 88% at 3 years, compared to infants without cholestasis.[380]

Catheter-Related Blood Stream Infections and Thrombosis

CRBSIs are common complications of HPN in children. The incidence of CRBSIs in most series is around 2/1000 HPN days (1/476 days, or 0.78 per patient-year). An increased risk for CRBSIs was found when HPN was begun during the first year of life, when first episode of CRBSI occurred shortly after starting HPN, in children with SBS and autoimmune enteropathy as the underlying disease.[433] Published series have shown that the death rate from CRBSIs was higher in children (up to 30% of total death in patients with SBS or CIPO) than in adults (8% of total death in mixed population). However, the CRBSI-related risk of death is reassuringly low. Pironi et al. observed 2.3% of deaths from 298 episodes of sepsis occurring in 527 patients, including 138 children who received a total of 1154 catheter-years of HPN.[495,496]

CVC-related deep vein thrombosis may cause HPN failure through the loss of venous access for CVC placement. The incidence of this is quite low (0.02–0.09 per catheter-year), with the highest rates reported in children.[495] In both adults and children, CVC-related deep vein thrombosis have accounted for a few deaths while on HPN. Mortality from CVC-related thrombosis is very rare (0%–3.9%), and represents a substantial risk only in those with a SVC syndrome (6.8%).[504] A study of 34 children reported a mortality rate of 11.8%, of which 26% had fatal pulmonary embolism.[505] CRBSIs may also be associated with CVC thrombosis as the clot becomes a nidus for bacteria. Loss of venous access is an important concern, although complete or impending loss of venous access has not been reported.[506]

Prognosis

Weaning of PN

Three studies in children have shown that the ability to successfully wean from HPN was greater in those with SBS (54%, 42%, and 73%), than for those with CIPO (38%, 25%, and 29%).[494,507–509] The U.S., Pediatric Intestinal Failure Consortium reported 47% cumulative percentage of intestinal autonomy at 3 years.[380] In children with SBS, the minimum required healthy residual small bowel was 40 cm in one study,[510] 15 cm in another,[511] or 10% of the expected normal length for a given gestational age in a third report.[512] In children with CIPO, neonatal onset, acute onset, urinary tract involvement, and requirement for surgery all predict a low probability of reaching intestinal autonomy.[513] In children with intractable diarrhea, Colomb et al. reported weaning in 48% of 25 children (40% with congenital mucosal disease, 60% protracted diarrhea of other causes), whereas Diamanti et al. observed weaning off HPN in 83% of 39 patients (18% with congenital mucosal disease and 82% with other types of protracted diarrhea).[494,508]

Survival and Prognostic Factors for Survival

Generally, the mean survival rate in children on long-term PN is better than in adults, with approximately 90% survival at 5 years.[494,508] However, the survival rates were only 49% at 1 year and 35% at 3 years in children referred for ITx.[503,514] Survival probabilities on HPN for SBS seem to differ on the basis of patient age and clinical condition at time of enrolment in the survey. The 5-year survival rate was 95% in children and

slightly lower, 88%, in neonates or infants <1 year old.[494,512] Starting PN at an early age, particularly in the neonatal period, was associated with greater mortality.[494,503,514] Primary non-digestive disease and NEC were the underlying conditions associated with the greatest mortality risk.[494,503] The mortality rate was highest in children with a very short small bowel remnant (duodenostomy or jejunostomy) and in the presence of a stoma.[512,513] Survival is slightly greater in patients with SBS, than in those with CIPO.[512,515] In patients with CIPO or aganglionosis, the risk of death was increased in cases with onset at birth, acute onset of the disease, myopathy as etiology, with intestinal malrotation, and in the presence of urinary tract involvement, and repeated surgery with or without resultant SBS.[515–517] Children with congenital mucosal disease or other types of protracted diarrhea had a lower survival than those with IF due to primary non-digestive disease.[494] The absence of a specialized nutrition care team was also a significant risk factor for mortality. The best outcome was described for patients with Crohn's disease.[494] Overall, CRBSIs, IFALD, and venous thrombosis remain the most common of potential serious risks.

Causes of Death in HPN

HPN-related causes (IFALD, infection, thromboembolic, and metabolic complications) accounted for the majority of deaths in three studies, whereas a fourth study reported that most deaths were related to underlying disease.[494,507,518,519] In those surveys where PN-related deaths prevailed, treatment had started within the first year of life. In the survey reporting underlying disease as the primary cause of death, the mean age at the beginning of HPN was 3 years. This suggests an increased risk of life-threatening HPN complications during the first years. PN-related death was the main cause of death in children with SBS or CIPO. In patients with protracted diarrhea, the cause of death was related to the underlying disease or to PN. The underlying disease was the main cause of death in patients with IF due to primary non-digestive disease (cancer).[494,507,518,519]

Discharge on HPN

PN dependency no longer mandates inpatient management. Clinical stability and severity of underlying disease must be taken into account when planning for HPN, including fluid losses, enteral tolerance, comorbidity, and the ability to tolerate cyclic PN. Children on HPN are provided binary admixtures (lipids being administered separately) or "all-in-one" admixtures. PN is usually infused over night for 12 hours and the

CVC heparin-locked during the day. This “cyclical” PN is important not only for protecting against IFALD but also for the psychosocial development of the child.

With the development of hospital nutritional support teams (NST) and commercial home care companies, HPN has become a practical option. However, the necessary organizational infrastructure remains complex. Parents need to be highly motivated wishing to assume responsibility for the medical care of their child. They must cope not only with the emotional demands of HPN, but also master the technical aspects of catheter and infusion care. Assessment of whether a family is able to take on the responsibility of HPN should be undertaken by a NST staff member in conjunction with the hospital ward staff and community team. A teaching program should be implemented to ensure that parents are confident and competent in all aspects of their child’s care. This includes hand washing and aseptic technique, knowledge of CVCs, catheter care and dressing changes, care of the infusion pump, running of infusion solutions through and connection to the CVC, setting up the feed and making the necessary corrections, urine testing, temperature taking, blood glucose monitoring, problem solving, and what to do in an emergency.[520]

Patients on HPN must be seen and assessed on a regular basis by the multidisciplinary team (optimally a nurse, dietitian, pharmacist, and physician). The timing will depend on the status of the patient and may range from weekly to every 4–6 months. Supervision by a multidisciplinary NST can optimize outcomes and facilitate HPN. Suggested schedule for monitoring during long-term PN is shown in Table 68-13.

Long-Term PN Monitoring

Patients receiving long-term PN must be monitored to optimize nutrition outcomes and to prevent complications that may develop over time, either related to their disease state or the PN. For patients who remain on long-term PN, it is important to monitor growth and pertinent laboratory assessments to ensure that nutrition and outcome goals are being met.[10,521] A comprehensive review of the evaluation and monitoring that should be considered for pediatric patients receiving specialized nutrition support is available.[522]

TABLE 68-13 Suggested Schedule and Monitoring During Long-Term PN

Interval	Clinical Assessment	Tests
1–3 months	Physical examination	Complete blood count, acid-base balance, and serum electrolytes
	Weight, height, head circumference in children <2 years	Nutritional biochemistry: glucose, urea, creatinine, albumin, prealbumin, lipid profile
	Enteral assessment	Calcium-phosphorous metabolism: calcium, phosphorus, magnesium, alkaline phosphatase
	Extra renal losses (e.g., stoma losses)	
	Line status and adequacy of care	Liver enzymes and bilirubin: alanine amino transferase, γ-glutamyl transpeptidase, total and direct bilirubin
	Functional status	Urinanalysis and Na, K, PO_4, Ca, creatinine
6–12 months	Idem	PT, PTT, ferritin
		Vitamin A, D, E, B_{12}, folate
		Zinc, copper and iron status
		Parathormone and thyroid hormones
		Hepatobiliary and renal ultrasound
12–24 months	Idem	Aluminium, chromium, manganese, selenium
		Chest X-ray (catheter location)
		Echocardiogram (if recurrent CRBSIs)
		Bone densitometry (DEXA scan)

Quality of Life

Three studies evaluated the QOL of children on HPN and their families. The first study found that the QOL scores were high in patients of all ages and were not significantly different from scores in a reference population of healthy children and adolescents. Lower QOL scores were recorded in the spheres related to hospital, health, doctors, medications, and obligations. The QOL was not affected in siblings but was significantly impaired in parents, especially in mothers, who showed a lower level of satisfaction for items related to work, inner life, and freedom. Presence of an ileostomy was the only factor that influenced QOL, especially of adolescents.[523] Overall the studies found that despite the problems they may face, it is possible for pediatric patients on long-term PN to develop and maintain a positive outlook, and cope well with illness-related

demands. The ability to cope so well may be related to the lack of experience with any other way of life. Children were also enabled to cope by support from parents, siblings, friends, and household pets. Participation in normal childhood activities and possession of a good understanding of their illness was also helpful.[524,525]

Cost

HPN is one of the more expensive ambulatory therapies, but the only possible alternative for many patients would be long-term hospitalization. The few cost-benefit studies have demonstrated that HPN is more cost-effective (by 50%–75%) than inpatient hospital PN. In the absence of published economic appraisals of pediatric HPN, one must consider costs to be similar to those of adult HPN. The annual cost of HPN, including nutrients, pump, and disposable equipment, has been estimated in 1990, to be approximately UK£ 60,000 pounds (about US$ 100,000) per patient in the United Kingdom, and ranges from US$ 80,000 to US$ 140,000 in the United States.[526–528] The HPN costs rise significantly in the presence of complications. A pediatric study, however, showed that HPN was associated with a fourfold decrease in the incidence of septic episodes as compared to hospital, leading to major savings.[529]

PERIPHERAL PN

Peripheral parenteral nutrition (PPN) is now regarded as a safe and effective alternative to central vein administration of nutrients for many patients. It avoids the risks of central venous catheterization, simplifies nursing care, reduces cost, and may shorten the time to initiation of nutritional support. PPN is useful for supplementation of inadequate oral or tube feeding intake, or transitional support until central access can be obtained. In general, it is difficult to provide adequate support via PPN for a patient with severe metabolic stress or nutritional depletion requiring electrolyte replacement and high calorie or protein intake. Other major limitations to the use of PPN are the availability of suitable peripheral veins, the development of peripheral vein thrombophlebitis, and the frequent need to replace the vein access, which limits the effective delivery of fluids and nutrients.

PN administered via peripheral vein catheters should last for no more than 10–14 days as the rate of complications increases after this period. Thrombophlebitis is one of the most significant complications limiting PPN support. There are many factors involved in its pathogenesis. There is

no consensus about optimal feed composition, choice of cannula, infusion technique, or pharmacological adjuncts. The incidence of thrombophlebitis is related to the osmotic content of infused solutions as well as to osmolality rate (product of osmolality and infusion rate). In most nutrient solutions, the major osmotic components are glucose, amino acids, and electrolytes. Early studies of PPN using crystalline amino acid and glucose-based solutions found phlebitis rates of almost 100% when the osmolarity was increased above 600 mOsmol/L.[530,531] Correlation between hyperosmolality and phlebitis is not, however, apparent when lipid-based solutions are used. Lipid emulsion appears to provide protection to the vein and allows tolerance of higher osmolarity infusates. Kane et al. randomized 36 patients to receive lipid-based PN solutions of osmolarity 1700 or 1200 mOsmol/L. They found no difference in the incidence of thrombophlebitis.[532] In adults, Everitt et al. have used successfully lipid-based formulations with an osmolality greater than 1100 mOsmol/kg, with phlebitis rates[533] ranging between 7% and 26%. Vein tolerance to hyperosmolar formulations may be further improved by admixture of the lipid emulsion with the dextrose-amino acid solution, AIO admixture.[534]

Addition of low-dose heparin and hydrocortisone to PPN formulations may increase tolerance to hyperosmolar formulations.[535] The incidence of vein thrombosis appears to be reduced by addition of low-dose heparin (1 unit/mL) to peripherally administered fluids. Premature infants receiving one unit of heparin per milliliter of PPN had about one-third the incidence of thrombophlebitis (5.7% vs. 17.2%) and nearly double duration of catheter patency compared to those without heparin.[536] Significant reduction of peripheral vein thrombophlebitis has been reported in randomized placebo controlled trials with use of transdermal glyceryl trinitrate in both PPN and non-PPN patients.[537] In healthy subjects, application of a topical nonsteroidal anti-inflammatory agent to the cannulation site on one arm reduced the incidence of phlebitis by nearly half in comparison with the site on the other arm, which served as a placebo.[538]

The risk of phlebitis seems lower when the cannula is placed on the back of the hand compared to venous access sites in the wrist or upper arm.[539] A Cochrane systematic review and a recent randomized, non-blinded equivalence trial showed that, in adults, there is no evidence of benefit in changing catheters every 72 to 96 hours.[540,541] Taking into consideration the small number of peripheral vascular access sites, pain,

and distress associated with sites changes, and the limitations in delivering optimal nutrition (osmolarity constraints and frequent interruptions) make routine change of vascular access in children, in the absence of clinical indications, an unwarranted practice favoring the use of CVCs.

Composition of PPN

Macronutrient concentrations are typically limited in PPN formulations. The final concentration of amino acids is generally between 2.5% (25 g/L) and 5% (50 g/L) for PPN. The generally recognized upper limit for dextrose concentrations in PPN formulations is 10% for adults (approximately 500 mOsm/L) and 12.5% for pediatric patients (625 mOsm/L). At these concentrations, dextrose provides 340 kcal/L in adults and 425 kcal/L in pediatric patients. The micronutrient components of PPN are similar to those of centrally administered PN formulations, with a few exceptions. Electrolytes such as sodium chloride, potassium, magnesium, phosphate, and calcium are essential for muscular and central nervous system functions, as well as for metabolism and maintenance of fluid status. All these and vitamins should be administered daily. TEs can be routinely added to all PN formulation, although the short duration of most PPN therapy makes this unnecessary unless the patient has preexisting deficiencies.

Monitoring of PPN therapy is essentially in the same way as for PN administered via a central venous access device. Metabolic complications parallel those associated with central PN. The typical duration of PPN therapy and limitations on CHO provision in PPN—with a subsequent increase in calories from lipid emulsion—may alter the incidence of hyperglycemia and hypertriglyceridemia. The frequency of monitoring should be based on the overall condition of the patient, on the presence of factors that may contribute to complications, and response to alterations in the nutrient prescription. As PPN patients tend to be under less metabolic stress than patients who require full support, the frequency of laboratory monitoring may be reduced.

Standard vs. Individually/Tailored Parenteral Solutions

PN can be provided as a standard, usually commercial, formulation that is designed to meet the nutritional needs of most patients of the same age group with a similar condition. Alternatively, an individually tailored PN compound, adapted to the individual patient’s nutritional needs, can be prescribed and prepared, usually on a daily basis and in the hospital

pharmacy. Both types of PN preparations have advantages and disadvantages associated with their use.

The main advantage of individually prescribed PN solutions is that they are tailored to suit a specific patient, and to achieve the best possible nutrition and precise biochemical control needed. The prescription can be changed on a daily basis, reflecting the medical condition and most recent laboratory tests.[31] In contrast to individualized prescriptions, standardization carries the risk of turning into "cookbook medicine" that lacks continuous clinical judgment of the patients' changing nutritional requirements.[542] This risk may be increased when the patient is a tiny fragile VLBW premature infant with enormous nutritional requirements and tendency for developing major nutrient deficits and sometimes life-threatening, metabolic disturbances (e.g., hypo- or hyperglycemia, hypo- or hypernatremia, hypo- or hyperkalemia). Studies in VLBW infants as well as in pediatric patients suggested that compared to infants on standard PN, infants who got individually tailored PN got more optimal nutrition, achieved better growth without clinical or laboratory complications, had a shorter period of exclusive PN, and required less electrolyte corrections.[542–544] Furthermore, Smolkin et al. suggested that individualized PN preparations were more optimal for the current more aggressive nutritional approach for PN in VLBW infants.[544] Yet, these authors admitted that the currently available standardized PN admixtures with adequate nutritional composition should be considered as appropriate alternatives. This is in contrast to "historical" standardized PN solutions that did not meet all the nutritional requirements of neonates, and could have resulted in inadequate nutrition and poor growth if used beyond short periods.[545]

PN is an intravenous medication, with more than 50 ingredients and additives, and as such is very liable to medication errors, especially in pediatric patients, where all the calculations are weight based.[546] The complexity of providing the nutritional requirements to children and neonates turns the task of ordering PN into a difficult process for the inexperienced clinician. Development of an optimal PN order form, including age and weight-specific nutrient requirements with guidelines for advancing substrates may aid the clinician, improve PN prescriptions, and decrease prescribing errors.[547,548] The stability and compatibility of PN solutions is also a safety topic of great concern. The most significant pharmaceutical issues involve the stability of ILE and the compatibility of calcium and phosphate salts to avoid precipitates. The existence of a

hospital NST, well-established communication channels between the prescribing clinicians, and the pharmacy team dedicated to PN preparation, and the use of compounding devices decrease these risks, but do not abolish them.[548–551]

Recent technology has advanced computerized PN ordering systems based on the guidelines contained in the program.[552] Such computerized nutritional software provides low cost and easy to use effective data of correctly calculated nutrient dosages. When data is both updated and reliable it results in better growth and better biochemical control, as evidenced in blood samples from the newborn studied.[553] In addition, electronic ordering systems can still allow individualization of PN prescription for each patient, thus improving biochemical control and decreasing wastage. Computer-assisted PN prescribing programs are also a valuable educational tool for the junior clinician, not experienced in clinical nutrition. These tools also facilitate the communication between the prescribing clinical team and the pharmacy department. Puangeo et al. found that automating the process of writing and delivering PN orders saved time and resulted in improved nutrient content of the PN solutions. This helped achieving caloric and protein goals earlier, and improving mineral status in premature infants.[554] However, in practice not all the alleged advantages of individualized computer-assisted prescriptions could be proved in premature infants.[555] A possible disadvantage of a computer-based prescription program is that it might encourage trivial adjustments in PN prescriptions, based on laboratory results that in clinical practice are irrelevant.[543] Based on these observations, it was suggested that higher proportion of PN could be standardized, if modified to reflect the practice pattern.

Krohn et al. evaluated the use of standard PN solutions in a PICU, and found that standard PN orders could be used in the majority of the patients. These solutions were usually nutritionally adequate and the intake of most macronutrients and electrolytes was similar to individually prescribed PN. Actually, calcium and phosphate intakes were better with standard PN compared to the individualized PN, and electrolyte imbalances occurred less frequently with the standard PN.[556] Yeung et al. evaluated retrospectively the difference in nutrient intakes and biochemical responses in premature infants who received standardized versus individualized PN between days 2–7 of life. They could not demonstrate any clinical advantage or improved biochemical control with individualized PN regimes.[557] In that study, the increase in protein was

accompanied by proportional increases in the intakes of glucose, electrolytes, and acetate. They also found that in infants who were on standardized PN, the cumulative deficit in protein intake by the end of the first week was 35% less compared to those who were on the individualized regimes. Infants on standardized PN had also higher intakes of calcium and phosphate, resulting in less commutative deficits and better bone mineralization. Lenclen et al. also showed that in premature infants standardized parenteral formulations provided higher early intakes of amino acids and glucose, and better calcium phosphate ratio during the first week of life while maintaining the same biochemical parameters.[558] In addition, it was suggested that the standardization strategy should be considered as part of the approach for improving quality control and good professional practice for the preparation of PN solutions. Batch-produced standardized PN bags can be readily available as ward stocks in the neonatal intensive care units, thus allowing initiation of early PN immediately after the delivery of a premature infant.

Commercially prepared standard PN bags decrease the risk of ordering errors, as well as the risk of compounding errors in the hospital pharmacy that has to deal with many different PN prescriptions on a daily basis. Large-scale commercial production of standard PN bags can be further eased by using automated compounding technology that can assure better pharmaceutical control of the physicochemical stability and compatibility of PN admixtures. This can decrease the risk of potentially adverse outcomes from infusion of incompatible nutrient admixtures (e.g., precipitated calcium phospate).[549–551] Large-scale commercial production of standard PN bags can also offer better aseptic manufacturing conditions than the average pharmacy hospital, thus decreasing the risk of PN-associated infections.[546] Commercially batch-produced standardized PN bags may also reduce the large costs of individualized PN production.[559] The need to add the parenteral multivitamins to the standard PN bag shortly before infusion is a limitation, that requires proper handling to assure aseptic conditions and avoid errors.

Based on European guidelines on pediatric PN, we suggested that standard PN solutions can be used safely in most pediatric and newborn patients, including VLBW premature infants for short periods (up to 2–3 weeks).[10,560] A range of standard regimens to suit different clinical conditions should always be available. Adequate monitoring of the metabolic and nutritional status of an infant on standardized PN should be assured, and the most suitable available standard PN formulation for the infant’s condition should be ordered at least once daily. Individually

tailored PN solution should be preferred when the nutritional needs cannot be met by the available range of standard PN formulations (i.e., in very sick and metabolically unstable patients [such as those with abnormal fluid and electrolyte losses]; and in infants and children requiring PN for prolonged periods [such as those with SBS]). Computers-assisted prescribing software for PN should become readily available, as they can save time, decrease prescription and compounding errors, and improve the quality of nutritional care. Such computerized programs should guide the physician to the most adequate standardized solution and optimize the use of individualized solutions.

Recently, Rigo et al. evaluated the efficacy, safety, flexibility, and ease of use of industrially manufactured ready-to-use (RTU) multichamber PN bag system containing three sterilized macronutrient solution chambers (for amino acids, glucose, and optional lipid bag activation system) specially designed for administration to preterm infants.[561] This technologically advanced multichamber PN bag system was easy to use, guaranteed sterility and longer shelf life, and provided well-balanced and safe nutritional support. Nutritional intakes and weight gain were within the recent PN recommendations for preterm infants. The combination of computerized prescription and the use of multichamber PN bags may enhance our ability to rely on standardized PN with minimal usage of individualized prescriptions.

SAFETY OF PN

PN is one of the most complex prescription drug preparations used for routine patient care. A single patient PN admixture contains macronutrients and micronutrients, solubilizers, preservatives, and other excipients. Giving this complexity, PN is classified among the “high-alert medications,” a term that describes drugs that can place patients at risk for significant harm when used in error.[562]

The options for delivery of pediatric PN solutions are RTU compounded bags prepared either by the hospital pharmacy or an outside service provider, and standard formulations prepared by the hospital pharmacy or outside service, to be modified on the ward according to the patient’s needs. All of these formulations are subject to human error. Isolated cases have reported of tragic but preventable outcomes from PN prescription errors. Otherwise, there is scant data in the literature describing PN-related medication errors.[563] Flynn and colleagues reported a significant error rate in manually and partially automated compounded

PN of 37% and 22%, respectively.[564] Other risks, such as stability issues and contamination and prescribing errors, have also been reported with compounded PN.[564] A recent retrospective analysis of adverse drug events in pediatric patients found that replacement preparations and total PN represented the largest category of adverse drug events (37%).[565]

PN fits easily into the model of regular medical decisions: decision to use drug therapy; selecting the best regimen for a patient; preparing, dispensing, and administering the drug; and then monitoring the patient and reassessment of the initial decision. Many relevant practice recommendations, guidelines, and standards for PN prescription and use are available for clinicians.[566–568] These include the following:

1. Use of a standardized PN ordering process that can reduce prescribing errors, provide prescriber education, and improve the efficiency of patients' nutrition care and the clinicians' follow-up. Although computerized order entry may reduce errors (e.g., associated with legibility), it does not eliminate errors, including those leading to an adverse drug event. Handwritten, and in some cases, nonstandardized forms are still being used in many centers, despite expert guidelines that have addressed mandatory components of the PN order form.
2. Verifying and reviewing the submitted PN order by a knowledgeable pharmacist. The review process includes two aspects: a *clinical* review verifies that dosing of each PN component is appropriate for the individual patient and a *pharmaceutical* review that the ordered components are compatible and that the preparation is expected to be stable.
3. Strictly adherence to the recommendations of professional organizations on PN compounding. Use of electronic interface offers the benefit of automatization and build in dose-warning limits that should remain active for all PN orders.
4. Standardization of the dispersing process involves labeling, delivery, and storage of the PN admixture prior to administration. The label should match all the elements of the PN order using the same terms and in the same order, with all ordered ingredients included. A beyond-use date and storage directions for the admixture should also be included.
5. Connection of PN solution to the patient should be made only after patient identification and corroboration of the label components and dosage with the written order from the patient'

file.

6. Each step along the PN use process needs to be readily documented in the patient's chart or in the pharmacy system as appropriate, whether using electronic medical record or paper documentation.

PN DELIVERY BAGS

All-In-One

In the past, lipids, amino acids, and glucose were administered in separate bottles and infused through separate intravenous lines (multi-bottle system). Presently, health-care institutions increasingly use AIO systems, with which nutrients are mixed in a single bag and infused simultaneously through a single intravenous line. The hypothetical advantage is that AIO systems reduce the manipulations. This system results in reduced manipulation and savings of material, time, and personnel workload. AIO admixtures require only one intravenous line, and the risk of infection is lowered by the closed system. The complete and simultaneous administration of all substrates reduces the risk for metabolic complications. AIO admixtures offer a convenient system for patients, clinicians, and nursing staff. The associated savings may overcome the higher manufacturing costs of the multi-compartment bags.

AIO PN bags can be compounded by hospital pharmacies (hospital-compounded AIO) and their composition can be standardized or tailor-made. These allow for the provision of patient-specific RTU admixtures, individually adapted according to energy, volume, and substrate needs. AIO bags can also be manufactured industrially, in a standardized way, as two or three-compartment bags. Industrially manufactured standard AIO can be provided as two- or three-chamber bags: in the case of a two-chamber bag, a lipid emulsion is admixed with a transfer set shortly before its administration to the patient, thus creating an AIO admixture, or through a separate line. Three-chamber bags contain all the macronutrients and electrolytes in three separate compartments. The substrates are mixed together immediately prior to intravenous delivery by breaking the separation seals between the bag chambers. Vitamins and TEs are injected into the bag prior to administration or infused by a separate intravenous line. When not mixed together, these standard MCBs have usually a shelf life of more than 12 months.

Multichamber Bags

Industrially prepared MCBs, with established standard compositions, often are an attractive method of providing PN. They are predominantly used in the short-term PN treatment of hospitalized adults. Individual compounding is, however, still required.[552] Colomb et al. described the use of the first pediatric triple-chamber PN bag (Ped3CB) in a prospective, open-label, multicenter, phase III clinical trial. The study enrolled 46 children, who were treated for five consecutive days with formulations designed for term infants and toddlers up to 2 years of age (Ped3CB-B) or for children and adolescents 2–18 years of age (Ped3CB-C). The volume of Ped3CB administered was based on the patient's individual needs. Majority of pharmacists and nurses found the Ped3CB system easy to use, and favored Ped3CB formulations over bags compounded on the ward, RTU compounded bags, and premixes prepared by the pharmacy and tailored to patient needs. Ped3CB delivered 91% of the PN energy intake among the Ped3CB-B group and 68% in the Ped3CB-C group. PN administered using the Ped3CB system was well tolerated in both groups. Four adverse events, all of which occurred in the Ped3CB-B group, were related to study treatment, two cases of hypertriglyceridemia and two cases of mild hyperglycemia. No serious adverse events considered to be related to study treatment were reported.[569]

The safety of an industrially manufactured three-chamber PN system specially designed for preterm infants was evaluated in a recent prospective, multicenter, open-label phase III clinical trial. The researchers used Ped3CB-A three-chamber PN that provide 3.1 g amino acids, 13.3 g glucose, 2.5 g lipid, and 91 kcal/100 mL with the ternary admixture and 3.9 g AA, 16.7 g glucose, and 83 kcal/100 mL with the binary admixture. Additional supplementation of sodium, potassium, calcium, and organic phosphate may be added to the basic admixtures, increasing concentrations by 78%, 67%, 118%, and 78%, respectively. Vitamins and TEs are not included in the Ped3CB-A bag and were added to the activated bag or provided separately, according to the general practice of the NICU. One hundred and thirteen preterm infants were enrolled in the study from five centers. The triple-chamber PN system was easy to use, has the flexibility required to meet the varied and changing nutritional needs of preterm infants, provided levels of nutrients within the ranges of recent recommendations, and resulted in appropriate weight gain rates. The majority of prescription days were successfully fulfilled using Ped3CB-A as is, with the addition of vitamins and TEs as usually performed in adult patients. Few additional supplementations into the bag, mainly sodium,

were necessary, representing only 23.1% of the perfusion days. Few abnormal biological parameters were reported as adverse events, including hyperglycemia, hypertriglyceridemia, and electrolytes deviation, but no serious adverse events related to PN supply. In conclusion, the triple-chamber bag for premature infants was easy to use and provided well-balanced and safe nutritional support in premature infants.[561]

CONCLUSIONS

Pediatric IF or immaturity is a complex and unique problem. Nutritional support in these children should be met by balancing both parenteral and enteral routes, while maintaining adequate growth and development. Efforts must be made to deliver a balanced PN solution, avoid catheter-related infections, and the development of IFALD. Additional research is needed to refine the enteral formulas and parenteral macro- and micronutrients for this pediatric population.

REFERENCES

1. Vinnars E, Wilmore D. Jonathan Rhoads symposium papers. History of parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2003;27:225–231.
2. Cuthbertson DP. Observation on the disturbance of metabolism produced by injury to the limbs. *Q J Med*. 1932;25:233–246.
3. Cuthbertson DP. Second Annual Jonathan E. Rhoads Lecture. The metabolic response to injury and its nutritional implications: retrospect and prospect. *JPEN J Parenter Enteral Nutr*. 1979;3:108–129.
4. Matas RM. The continued intravenous “drip.” *Ann Surg*. 1924;79: 643–661.
5. Rose WC. The significance of amino acids in nutrition. *Harvey Lect*. 1934;30:45–65.
6. Elman R. Parenteral replacement of protein with the amino-acids of hydrolyzed casein. *Ann Surg*. 1940;112:594–602.
7. Wretlind A. Free amino acids in a dialyzed casein digest. *Acta Physiol Scand*. 1947;13:45–54.
8. Schuberth O, Wretlind A. Intravenous infusion of fat emulsions, phosphatides, and emulsifying agents. *Acta Chir Scand Suppl*. 1961;278:1–21.
9. Dudrick SJ, Wilmore DW, Vars HM, et al. Long-term total parenteral nutrition with growth, development, and positive nitrogen balance. *Surgery*. 1968;64:134–142.
10. Koletzko B, Goulet O, Hunt J, et al. Guidelines on Paediatric Parenteral Nutrition of the European Society of Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) and the European Society for Clinical Nutrition and Metabolism (ESPEN), Supported by the European Society of Paediatric Research (ESPR). *J Pediatr Gastroenterol Nutr*. 2005;41:Suppl 2, S1–S87.
11. Cahill GF Jr. Starvation in man. *N Engl J Med*. 1970;282:668–675.
12. Owen OE, Morgan AP, Kemp HG, et al. Brain metabolism during fasting. *J Clin Invest*. 1967;46:1589–1595.

13. Long CL, Shiller WR, Geiger JW, et al. Gluconeogenesis response during glucose infusions in patients following skeletal trauma or during sepsis. *JPEN J Parenteral Enteral Nutr*. 1978;2:619–626.
14. Wilmore DW. Hormonal responses and their effect on metabolism. *Surg Clin North Am*. 1976;56:999–1018.
15. Heird WC, Driscoll JM, Schollinger JN, et al. Intravenous alimentation in pediatric patients. *J Pediatr*. 1972;80:351–372.
16. Gamble GL. *Chemical Anatomy, Physiology and Pathology of Extracellular Fluid: a Lecture Syllabus 1947*. Cambridge, MA: Harvard University Press; 1954.
17. O'Connell RC, Morgan AP, Aoki TT, et al. Nitrogen conservation in starvation: graded responses to intravenous glucose. *J Clin Endocrin Metab*. 1974;39:555–563.
18. Elwyn DH, Gump FE, Munro HN, et al. Changes in nitrogen balance of depleted patients with increasing infusions of glucose. *Am J Clin Nutr*. 1979;32:1597–1611.
19. American Academy of Pediatrics Committee on Nutrition. Nutritional needs of low-birth-weight infants. *Pediatrics*. 1985;75:976–986.
20. DeCurtis M, Rigo J. The nutrition of preterm infants. *Early Hum Dev*. 2012;88(Suppl 1):S5–S7.
21. Ziegler EE. Meeting the nutritional needs of the low-birth-weight infant. *Ann Nutr Metab*. 2011;58 (Suppl 1):8–18.
22. Colvin M, McGuire W, Fowlie PW. Neurodevelopmental outcomes after preterm birth. *BMJ*. 2004;329(7479):1390–1393.
23. Ehrenkranz RA, Das A, Wrage LA, et al. Early nutrition mediates the influence of severity of illness on extremely LBW infants. *Pediatr Res*. 2011;69:522–529.
24. Embleton NE, Pang N, Cooke RJ. Postnatal malnutrition and growth retardation: an inevitable consequence of current recommendations in preterm infants? *Pediatrics*. 2001;107:270–273.
25. Stephens BE, Walden RV, Gargus RA, et al. First-week protein and energy intakes are associated with 18-month developmental outcomes in extremely low birth weight infants. *Pediatrics*. 2009;123:1337–1343.
26. Lucas A, Fewtrell MS, Davies PS, et al. Breastfeeding and catch-up growth in infants born small for gestational age. *Acta Paediatr*. 1997;86:564–569.
27. Bhatia J. Post-discharge nutrition of preterm infants. *J Perinatol*. 2005;25(Suppl 2):S15–S16.
28. Ehrenkranz RA. Early, aggressive nutritional management for very low birth weight infants: what is the evidence? *Semin Perinatol*. 2007;31:48–55.
29. Wilson DC, Cairns P, Halliday HL, et al. Randomised controlled trial of an aggressive nutritional regimen in sick very low birthweight infants. *Arch Dis Child Fetal Neonatal Ed*. 1997;77:F4–F11.
30. Ziegler EE, Thureen PJ, Carlson SJ. Aggressive nutrition of the very low birthweight infant. *Clin Perinatol*. 2002;29:225–244.
31. Beecroft C, Martin H, Puntis JW. How often do parenteral nutrition prescriptions for the newborn need to be individualized? *Clin Nutr*. 1999;18:83–85.
32. Te Braake FW, van den AkkerCH, Wattimena DJ, et al. Amino acid administration to premature infants directly after birth. *J Pediatr*. 2005;147:457–461.
33. Van Goudoever JB, Colen T, Wattimena JL, et al. Immediate commencement of amino acid supplementation in preterm infants: effect on serum amino acid concentrations and protein kinetics on the first day of life. *J Pediatr*. 1995;127:458–465.

34. Te Braake FW, Schierbeek H, de GK, et al. Glutathione synthesis rates after amino acid administration directly after birth in preterm infants. *Am J Clin Nutr*. 2008;88:333–339.
35. Thureen PJ, Melara D, Fennessey PV, et al. Effect of low versus high intravenous amino acid intake on very low birth weight infants in the early neonatal period. *Pediatr Res*. 2003;3:24–32.
36. Porcelli Jr PJ, Sisk PM. Increased parenteral amino acid administration to extremely low-birth-weight infants during early postnatal life. *J Pediatr Gastroenterol Nutr*. 2002;34:174–179.
37. Embleton ND. Optimal protein and energy intakes in preterm infants. *Early Hum Dev*. 2007;83:831–837.
38. Te Braake FW, van den Akker CH, Riedijk MA, et al. Parenteral amino acid and energy administration to premature infants in early life. *Semin Fetal Neonatal Med*. 2007;12:11–18.
39. Senterre T, Rigo J. Reduction in postnatal cumulative nutritional deficit and improvement of growth in extremely preterm infants. *Acta Paediatr*. 2012;101:e64–e70.
40. Senterre T, Rigo J. Optimizing early nutritional support based on recent recommendations in VLBW infants and postnatal growth restriction. *J Pediatr Gastroenterol Nutr*. 2011;53:536–542.
41. Martin CR, Brown YF, Ehrenkranz RA, et al. Nutritional practices and growth velocity in the first month of life in extremely premature infants. *Pediatrics*. 2009;124:649–657.
42. Greenberg GR, Marliss EB, Anderson H, et al. Protein-sparing therapy in postoperative patients. Effects of added hypocaloric glucose or lipid. *N Engl J Med*. 1976;294:1411–1416.
43. Wells JC, Cole TJ, Davies PS. Total energy expenditure and body composition in early infancy. *Arch Dis Child*. 1996;75:423–426.
44. Tershakovec AM, Kuppler KM, Zemel B, et al. Age, sex, ethnicity, body composition, and resting energy expenditure of obese African American and white children and adolescents. *Am J Clin Nutr*. 2002;75:867–871.
45. Reed GW, Hill JO. Measuring the thermic effect of food. *Am J Clin Nutr*. 1996;63:164–169.
46. Spady DW, Payne PR, Picou D, Waterlow JC. Energy balance during recovery from malnutrition. *Am J Clin Nutr*. 1976;29:1073–1088.
47. Just B, Messing B, Darmaun D, et al. Comparison of substrate utilization by indirect calorimetry during cyclic and continuous total parenteral nutrition. *Am J Clin Nutr*. 1990;51:107–111.
48. Lerebours E, Rimbert A, Hecketsweiler B, et al. Comparison of the effects of continuous and cyclic nocturnal parenteral nutrition on energy expenditure and protein metabolism. *JPEN J Parenter Enteral Nutr*. 1988;12:360–364.
49. Ashworth A. Energy balance and growth: experience in treating children with malnutrition. *Kidney Int*. 1978;14:301–305.
50. Kohl HW, Fulton JE, Caspersen CJ. Assessment of physical activity among children and adolescents a review and synthesis. *Prevent Med*. 2000;31:S54–S76.
51. Lifson N, Gordon GB, McClintock R. Measurement of total carbon dioxide production by means of D2O. *J Appl Physiol*. 1955;7:704–710.
52. Schoeller DA, van Santen E. Measurement of energy expenditure in humans by doubly labeled water method. *J Appl Physiol*. 1982;53:955–959.
53. Weir JB. New methods for calculating metabolic rate with special reference to protein metabolism. *J Physiol*. 1949;109:1–9.

54. Ultman JS, Bursztein S. Analysis of error in the determination of respiratory gas exchange at varying FIO2. *J Appl Physiol*. 1981;50:210–216.
55. Cunningham JJ. Body composition as a determinant of energy expenditure: a synthetic review and a proposed general prediction equation. *Am J Clin Nutr*. 1991;54:963–969.
56. Haugen HA, Chan LN, Li F. Indirect calorimetry: a practical guide for clinicians. *Nutr Clin Pract*. 2007;22:377–388.
57. World Health Organization. Energy and protein requirements: report of a joint FAO/WHO/UNU expert consultation. (WHO Technical Report Series No. 724). Geneva, Switzerland: World Health Organization; 1985.
58. Harris JA, Benedict FG. *A Biometric Study of Basal Metabolism in Man*. Washington, DC: Carnegie Institute of Washington; 1919.
59. Schofield WN. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr*. 1985;39(suppl 1):5–41.
60. Schofield WN, Schofield C, James WPT. Basal metabolic rate—review and prediction, together with an annotated bibliography of source material. *Hum Nutr Clin Nutr*. 1985;39C(suppl 1):1–96.
61. Henry CJK. Basal metabolic rate studies in humans: measurement and development of new equations. *Public Health Nutr*. 2005;8:1133–1152.
62. Duro D, Rising R, Cole C, et al. New equations for calculating the components of energy expenditure in infants. *J Pediatr*. 2002;140:534–539.
63. Thomson MA, Bucolo S, Quirk P, et al. Measured versus predicted resting energy expenditure in infants: a need for reappraisal. *J Pediatr*. 1995;126:21–27.
64. Daly JM, Heymsfield SB, Head CA, et al. Human energy requirements: overestimation by widely used prediction equation. *Am J Clin Nutr*. 1985;42:1170–1174.
65. Firouzbakhsh S, Mathis RK, Dorchester WL, et al. Measured resting energy expenditure in children. *J Pediatr Gastroenterol Nutr*. 1993;16:136–142.
66. Rodriguez G, Moreno LA, Sarria A, et al. Resting energy expenditure in children and adolescents: agreement between calorimetry and prediction equations. *Clin Nutr*. 2002;21:255–260.
67. Sentongo TA, Tershakovec AM, Mascarenhas MR, et al. Resting energy expenditure and prediction equations in young children with failure to thrive. *J Pediatr*. 2000;136:345–350.
68. Shakur YA, Richards H, Encharz ABP. Is it necessary to measure resting energy expenditure in clinical practice in children? *J Pediatr*. 2008;152:437–439.
69. Beghin L, Michaud L, Hankard R, et al. Total energy expenditure and physical activity in children treated with home parenteral nutrition. *Pediatr Res*. 2003;53:684–690.
70. Merritt RJ, Sunkind RM. Nutritional survey of hospitalized pediatric patients. *Am J Clin Nutr*. 1979;32:1320–1325.
71. Pollack MM, Wiley JS, Holbrook PR. Early nutritional depletion in critically ill children. *Crit Care Med*. 1981;9:580–583.
72. Kinney JM. Metabolic responses of the critically ill patient. *Crit Care Clin*. 1995;11:569–586.
73. Briassoulis G, Venkataraman S, Thompson AE. Energy expenditure in critically ill children. *Crit Care Med*. 2000;28:1166–1172.
74. Mehta NM, Bechard LJ, Leavitt K, et al. Cumulative energy imbalance in the pediatric intensive care unit: role of targeted indirect calorimetry. *JPEN J Parenter Enteral Nutr*. 2009;33:336–344.
75. llman HE, Mehta NM. Nutrition therapy in the critically ill child. *Curr Opin Crit*

Care. 2012;18:192–198.
76. Winthrop AL, Wesson DE, Penchars PB, et al. Injury severity, whole-body protein turnover and energy expenditure in pediatric trauma. *J Pediatr Surg*. 1987;22:534–537.
77. Chwals WJ, Lally KP, Woolley MM, et al. Measured energy expenditure in critically ill infants and young children. *J Surg Res*. 1988;44:467–472.
78. Gebara BM, Gelmini M, Sarnik A. Oxygen consumption, energy expenditure, and substrate utilization after cardiac surgery in children. *Crit Care Med*. 1992;20:1550–1554.
79. Selby AM, McCauley JC, Schell DN, et al. Indirect calorimetry in mechanically ventilated children: a new technique that overcomes the problem of endotracheal tube leak. *Crit Care Med*. 1995;23:365–370.
80. Tilden SJ, Watkins S, Tong TK, et al. Measured energy expenditure in pediatric intensive care patients. *Am J Dis Child*. 1989;143:490–492.
81. Coss-Bu JA, Jefferson LS, Walding D, et al. Resting energy expenditure in children in a pediatric intensive care unit: comparison of Harris-Benedict and Talbot predictions with indirect calorimetry values. *Am J Clin Nutr*. 1998;67:74–80.
82. Reid CL. Poor agreement between continuous measurements of energy expenditure and routinely used prediction equations in intensive care patients. *Clin Nutr*. 2007;26:649–657.
83. Mehta NM, Compher C, A.S.P.E.N. Board of Directors. A.S.P.E.N. clinical guidelines: nutrition support of the critically ill child. *JPEN J Parenter Enteral Nutr*. 2009; 33:260–276.
84. van der Kuip M, Oosterveld MJ, van Bokhorst de van der Schueren MA, et al. Nutritional support in 111 pediatric intensive care units: a European survey. *Intensive Care Med*. 2004;30:1807–1813.
85. Kyle UG, Arriaza A, Esposito M, et al. Is indirect calorimetry a necessity or a luxury in the pediatric intensive care unit? *JPEN J Parenter Enteral Nutr*. 2012;36:177–182.
86. Bauer J, Maier K, Hellstern G, et al. Longitudinal evaluation of energy expenditure in preterm infants with birth weight less than 1000 g. *Br J Nutr*. 2003;89:533–537.
87. Weintraub V, Mimouni FB, Dollberg S. Effect of birth weight and postnatal age upon resting energy expenditure in preterm infants. *Am J Perinatol*. 2009;26:173–177.
88. Torine IJ, Denne SC, Wright-Coltart S, et al. Effect of late-onset sepsis on energy expenditure in extremely premature infants. *Pediatr Res*. 2007;61:600–603.
89. Denne SC. Protein and energy requirements in preterm infants. *Semin Neonatol*. 2001;6:377–382.
90. Piedboeuf B, Chessex P, Hazan J, et al. Total parenteral nutrition in the newborn infant: energy substrates and respiratory gas exchange. *J Pediatr*. 1991;118:97–102.
91. Bresson JL, Bader B, Rocchiccioli F, et al. Protein-metabolism kinetics and energy-substrate utilization in infants fed parenteral solutions with different glucose-fat ratios. *Am J Clin Nutr*. 1991;54:370–376.
92. Salas-Salvado J, Molina J, Figueras J, et al. Effect of the quality of infused energy on substrate utilization in the newborn receiving total parenteral nutrition. *Pediatr Res*. 1993;33:112–117.
93. Brunton JA, Ball RO, Pencharz PB. Determination of amino acid requirements by indicator amino acid oxidation: applications in health and disease. *Curr Opin Clin*

Nutr Metab Care. 1998;1:449–453.
94. Zello GA, Wykes LJ, Ball RO, et al. Recent advances in method of assessing amino acid requirements for adult humans. *J Nutr*. 1995 125:2907–2915.
95. Duffy B, Gunn T, Collinge J, et al. The effect of varying protein quality and energy intake on the nitrogen metabolism of parenterally fed very low birthweight (<l600 g) infants. *Pediatr Res*. 1981;15: 1040–1044.
96. Jones EM, Baumann CA, Reynolds MS. Nitrogen balances of women maintained on various levels of lysine. *J Nutr*. 1956;60:549–562.
97. Nakagawa I, Takahashi T, Suzuki T, et al. Amino acid requirements in children: nitrogen balance at the minimal level of essential amino acids. *J Nutr*. 1964;83:115–118.
98. Ziegler EE, O'Donnell AM, Stearns G, et al. Nitrogen balance studies with normal children. *Am J Clin Nutr*. 1977;30:939–934.
99. Golden MH. Transport proteins as indices of protein status. *Am J Clin Nutr*. 1982;35(5 Supp l):1159–1165.
100. Pencharz PB, Ball RO. Different approaches to define individual amino acid requirements. *Annu Rev Nutr*. 2003;23:101–116.
101. Elango R, Ball RO, Pencharz PB. Indicator amino acid oxidation: concept and application. *J Nutr*. 2008;138:243–246.
102. Bross R, Ball RO, Pencharz PB. Development of a minimally invasive protocol for determination of phenylalanine and lysine kinetics in humans during the fed state. *J Nutr*. 1998;128:1913–1919.
103. Humayun MA, Turner JM, Elango R, et al. Minimum methionine requirement and cysteine sparing of methionine in healthy school-age children. *Am J Clin Nutr*. 2006;84:1080–1085.
104. Elango R, Humayun MA, Ball RO, Pencharz PB. Lysine requirement of healthy school-age children determined by the indicator amino acid oxidation method. *Am J Clin Nutr*. 2007;86:360–366.
105. Courtney-Martin G, Chapman KP, Moore AM, et al. Total sulfur amino acid requirement and metabolism in parenterally fed postsurgical human neonates. *Am J Clin Nutr*. 2008;88:115–124.
106. Chapman KP, Courtney-Mart G, Moore AM, et al. Threonine requirement of parenterally fed postsurgical human neonates. *Am J Clin Nutr*. 2009;89:134–141.
107. Stoll B, Henry J, Reeds PJ, et al. Catabolism dominates the first-pass intestinal metabolism of dietary essential amino acids in milk-replacer-fed piglets. *J Nutr*. 1998;128:606–614.
108. Heird WC, Dell RB, Helms RA, et al. Amino acid mixture designed to maintain normal plasma amino acid patterns in infants and children requiring parenteral nutrition. *Pediatrics*. 1987;80:401–408.
109. Wu PYK, Edwards NB, Storm MC. The plasma amino acid pattern of normal term breast-fed infants. *J Pediatr*. 1986;109:347–349.
110. Brunton JA, Ball RO, Pencharz PB. Current total parenteral nutrition solutions for the neonate are inadequate. *Curr Opin Clin Nutr Metab Care*. 2000;3:299–304.
111. Snyderman SE, Holt LE Jr. Amino acid requirements of infants. *Am J Dis Child*. 1965;110:108–109.
112. Das JB, Filler RM. Amino acid utilization during total parenteral nutrition in the surgical neonate. *J Pediatr Surg*. 1973;8:793–799.
113. Peden VH, Karpel JT. Total parenteral nutrition in premature infants. *J Pediatr*. 1972;81:137–144.
114. Driscoll JM, Heird WC, Shullinger JN, et al. Total intravenous alimentation in low-birth-weight infants: a preliminary report. *J Pediatr*. 1972;81:145–153.

115. Asch MJ, Sperling M, Fiser R, et al. Metabolic and hormonal studies comparing three parenteral nutrition regimens in infants. *Ann Surg*. 1975;182:62–65.
116. Fox HA, Krasna IH. Total intravenous nutrition by peripheral vein in neonatal surgical patients. *Pediatrics*. 1973;52:14–20.
117. Van Aerde JEE, Sauer PJJ, Pencharz PB, et al. The effect of replacing glucose with lipid on the energy metabolism of newborn infants. *Clin Sci*. 1989;76:581–588.
118. Paluzzi M, Meguid MM. A prospective randomized study of the optimal source of nonprotein calories in total parenteral nutrition. *Surgery*. 1987;102:711–717.
119. O'Neill JA, Meng HC, Caldwell MD, Stahlman MT. Metabolic evaluation of a synthetic amino acid mixture for parenteral nutrition in infants and children. *J Pediatr Surg*. 1976;11:979–985.
120. Moss RL, Haynes AL, Pastuszyn A, Glew RH. Methionine infusion reproduces liver injury of parenteral nutrition cholestasis. *Pediatr Res*. 1999;45:664–668.
121. Rakonczay Z Jr, Hegyi P, Dosa S, et al. A new severe acute necrotizing pancreatitis model induced by L-ornithine in rats. *Crit Care Med*. 2008;36:2117–2127.
122. Asanuma K, Adachi K, Sugimoto T, Chiba S. Effects of lysine-induced acute renal failure in dogs. *J Toxicol Sci*. 2006;31:87–98.
123. Soeters PB, van de Poll G, Van Gemert WG, Dejong CH. Amino acid adequacy in pathophysiological states. *J Nutr*. 2004;134:1575S–1582S.
124. Verbruggen S, Sy J, Arrivillaga A, et al. Parenteral amino acid intakes in critically ill children: a matter of convenience. *JPEN J Parenter Enteral Nutr*. 2010;34:329–340.
125. Poindexter BB, Ehrenkranz RA, Stoll BJ, et al. Parenteral glutamine supplementation does not reduce the risk of mortality or late-onset sepsis in extremely low birth weight infants. *Pediatrics*. 2004;113: 1209–1215.
126. Clark RH, Chace DH, Spitzer AR. Effects of two different doses of amino acid supplementation on growth and blood amino acid levels in premature neonates admitted to the neonatal intensive care unit: a randomized, controlled trial. *Pediatrics*. 2007;120:1286–1296.
127. Blanco CL, Gong AK, Green BK, et al. Early changes in plasma amino acid concentrations during aggressive nutritional therapy in extremely low birth weight infants. *J Pediatr*. 2011;158:543–548.
128. Potsic B, Holliday N, Lewis P, et al. Glutamine supplementation and deprivation: effect on artificially reared rat small intestinal morphology. *Pediatr Res*. 2002;52:430–436.
129. DeMarco V, Dyess K, Strauss D, et al. Inhibition of glutamine synthetase decreases proliferation of cultured rat intestinal epithelial cells. *J Nutr*. 1999;129:57–62.
130. Soghier LM, Brion LP. Cysteine, cystine or N-acetylcysteine supplementation in parenterally fed neonates. *Cochrane Database Syst Rev*. 2006;(4):CD004869.
131. Dunham B, Marcuard S, Khazanie PG, et al. The solubility of calcium and phosphorus in neonatal total parenteral nutrition solutions. *JPEN J Parenter Enteral Nutr*. 1991;15:608–611.
132. Wretlind A. Development of fat emulsions. *JPEN J Parenter Enteral Nutr*. 1981;5:230–235.
133. Nordenstrom J, Askanazi J, Elwyn DH, et al. Nitrogen balance during total parenteral nutrition. Glucose vs. fat. *Ann Surg*. 1983;197:27–33.
134. MacFie J, Smith RC, Hill GL. Glucose or fat as a nonprotein energy source? *Gastroenterology*. 1981;80:103–107.

135. Driscoll DF, Bistrian BR, Demmelmair H, et al. Pharmaceutical and clinical aspects of parenteral lipid emulsions in neonatology. *Clin Nutr*. 2008;27:497–503.
136. Waitzberg DL, Torrinhas RS, Jacintho TM. New parenteral lipid emulsions for clinical use. *JPEN J Parenter Enteral Nutr*. 2006;30: 351–367.
137. Yaqoob P. Lipids and the immune response: from molecular mechanisms to clinical applications. *Curr Opin Clin Nutr Metab Care*. 2003;6:133–150.
138. Siqueira J, Smiley D, Newton C, et al. Substitution of standard soybean oil with olive oil-based lipid emulsion in parenteral nutrition: comparison of vascular, metabolic, and inflammatory effects. *J Clin Endocrinol Metab*. 2011;96:3207–3216.
139. Koletzko B. Lipid supply and metabolism in infancy. *Curr Opin Clin Nutr Metab Care*. 1998;1:171–177.
140. Freeman J, Goldmann DA, Smith NE, et al. Association of intravenous lipid emulsion and coagulase-negative staphylococcal bacteremia in neonatal intensive care units. *N Engl J Med*. 1990;323:301–308.
141. Vlaardingerbroek H, Veldhorst MA, Spronk S, et al. Parenteral lipid administration to very-low-birth-weight infants—early introduction of lipids and use of new lipid emulsions: a systematic review and meta-analysis. *Am J Clin Nutr*. 2012;96:255–268.
142. Wanten GJ, Naber AH, Netea M. Modulation of inflammatory cytokine production by parenteral lipid emulsions. *Clin Nutr*. 2005;24: 164–165.
143. Calder PC. Hot topics in parenteral nutrition. Rationale for using new lipid emulsions in parenteral nutrition and a review of the trials performed in adults. *Proc Nutr Soc*. 2009;68:252–260.
144. Bach AC, Frey A, Lutz O. Clinical and experimental effects of medium-chain-triglyceride-based fat emulsions—a review. *Clin Nutr*. 1989;8:223–235.
145. Johnson RC, Cotter R. Metabolism of medium-chain triglyceride lipid emulsions. *Nutr Int*. 1986;2:150–158.
146. Baldermann H, Wicklmayr M, Rett K, et al. Changes of hepatic morphology during parenteral nutrition with lipid emulsions containing LCT or MCT/LCT quantified by ultrasound. *J Parenter Enteral Nutr*. 1991;15:601–603.
147. Jaurrieta E, Biondo S, Rafecas A, et al. A comparative study of hepatic cholestasis after infusion of long chain triglycerides and a mixture of medium and long chain triglycerides. *Nutr Hosp*. 1991;152–155.
148. Rubin M, Moser A, Naor N, et al. Effect of three intravenously administered fat emulsions containing different concentrations of fatty acids on the plasma fatty acid composition of premature infants. *J Pediatr*. 1994;125:596–602.
149. Lehner F, Demmelmair H, Röschinger W, et al. Metabolic effects of intravenous LCT or MCT/LCT lipid emulsions in preterm infants. *J Lipid Res*. 2006;47:404–411.
150. Lai HS, Chen WJ. Effects of medium-chain/long-chain triglyceride in pediatric surgical patients. *Nutrition*. 2000;16:401–406.
151. Ball MJ. Parenteral nutrition in the critically ill: use of a medium chain triglyceride emulsion. *Intensive Care Med*. 1993;19:89–95.
152. Goulet O, Postaire M, De Potter S, et al. Medium-chain triglycerides and long-term parenteral nutrition in children. *Nutrition*. 1992;8:333–337.
153. Ulrich H, Pastores SM, Katz DP, et al. Parenteral use of medium-chain triglycerides: a reappraisal. *Nutrition*. 1996;12:231–238.
154. Bach AC, Storck D, Meraihi Z. Medium-chain triglyceride-based fat emulsions: an alternative energy supply in stress and sepsis. *JPEN J Parenter Enteral Nutr*. 1988;12:82S–88S.

155. Yaqoob P. Monounsaturated fatty acids in parenteral nutrition; evaluation of risks and benefits. *Br J Nutr*. 2005;94:867–868.
156. Umpierrez GE, Spiegelman R, Zhao V, et al. A double-blind, randomized clinical trial comparing soybean oil-based versus olive oil-based lipid emulsions in adult medical-surgical intensive care unit patients requiring parenteral nutrition. *Crit Care Med*. 2012;40:1792–1798.
157. Goebel Y, Koletzko B, Böhles HJ, et al. Parenteral fat emulsions based on olive and soybean oils: a randomized clinical trial in preterm infants. *J Pediatr Gastroenterol Nutr*. 2003;37:161–167.
158. Webb AN, Hardy P, Peterkin M, et al. Tolerability and safety of olive oil-based lipid emulsion in critically ill neonates: a blinded randomized trial. *Nutrition*. 2008;24:1057–1064.
159. Goulet O, de Potter S, Antébi H, et al. Long-term efficacy and safety of a new olive oil-based intravenous fat emulsion in pediatric patients: a double-blind randomized study. *Am J Clin Nutr*. 1999;70:338–345.
160. Hartman C, Ben-Artzi E, Berkowitz D, et al. Olive oil-based intravenous lipid emulsion in pediatric patients undergoing bone marrow transplantation: a short-term prospective controlled trial. *Clin Nutr*. 2009;28:631–635.
161. Reimund JM, Rahmi G, Escalin G, et al. Efficacy and safety of an olive oil-based intravenous fat emulsion in adult patients on home parenteral nutrition. *Aliment Pharmacol Ther*. 2005;21:445–454.
162. Cano NJ, Saingra Y, Dupuy AM, et al. Intradialytic parenteral nutrition: comparison of olive oil versus soybean oil-based lipid emulsions. *Br J Nutr*. 2006;95:152–159.
163. Huschak G, Zur Nieden K, Hoell T, et al. Olive oil based nutrition in multiple trauma patients: a pilot study. *Intensive Care Med*. 2005;31:1202–1208.
164. García-de-Lorenzo A, Denia R, Atlan P, et al. Parenteral nutrition providing a restricted amount of linoleic acid in severely burned patients: a randomised double-blind study of an olive oil-based lipid emulsion vs. medium/long-chain triacylglycerols. *Br J Nutr*. 2005;94:221–230.
165. Bahadori B, Uitz E, Thonhofer R, et al. Omega-3 fatty acids infusions as adjuvant therapy in rheumatoid arthritis. *J Parenter Enteral Nutr*. 2010;34:151–155.
166. Khor BS, Liaw SJ, Shih HC, Wang LS. Randomized, double blind, placebo controlled trial of fish-oil-based lipid emulsion infusion for treatment of critically ill patients with severe sepsis. *Asian J Surg*. 2011;34:1–10.
167. de Miranda Torrinhas RS, Santana R, Garcia T, et al. Parenteral fish oil as a pharmacological agent to modulate post-operative immune response: a randomized, double-blind, and controlled clinical trial in patients with gastrointestinal cancer. *Clin Nutr*. 2013;32:503–510.
168. Manzanares W, Dhaliwal R, Jurewitsch B, et al. Parenteral fish oil lipid emulsions in the critically ill: a systematic review and meta-analysis. *JPEN J Parenter Enteral Nutr*. 2014;38:20–28.
169. Serhan CN, Chiang N, Van Dyke TE. Resolving inflammation: dual anti-inflammatory and pro-resolution lipid mediators. *Nat Rev Immunol*. 2008;8:349–361.
170. Serhan CN, Yacoubian S, Yang R. Anti-inflammatory and proresolving lipid mediators. *Annu Rev Pathol*. 2008;3:279–312.
171. Pradelli L, Mayer K, Muscaritoli M, et al. n-3 Fatty acid-enriched parenteral nutrition regimens in elective surgical and ICU patients: a meta-analysis. *Crit Care*. 2012;16:R184.
172. Qi K, Seo T, Al-Haideri M, et al. Omega-3 triglycerides modify blood clearance

and tissue targeting pathways of lipid emulsions. *Biochemistry*. 2002;41:3119–3127.
173. Griffin MD, Sanders TA, Davies IG, et al. Effects of altering the ratio of dietary n-6 to n-3 fatty acids on insulin sensitivity, lipoprotein size, and postprandial lipemia in men and postmenopausal women aged 45–70 y: the OPTILIP Study. *Am J Clin Nutr*. 2006;84:1290–1298.
174. Skulas-Ray AC, Kris-Etherton PM, Harris WS, et al. Dose-response effects of omega-3 fatty acids on triglycerides, inflammation, and endothelial function in healthy persons with moderate hypertriglyceridemia. *Am J Clin Nutr*. 2011;93:243–252.
175. Waitzberg DL, Torrinhas RS. Fish oil lipid emulsions and immune response: what clinicians need to know. *Nutr Clin Pract*. 2009;24:487–499.
176. Le HD, de Meijer VE, Robinson EM, et al. Parenteral fish-oil-based lipid emulsion improves fatty acid profiles and lipids in parenteral nutrition-dependent children. *Am J Clin Nutr*. 2011;94:749–758.
177. Goulet O, Antébi H, Wolf C, et al. A new intravenous fat emulsion containing soybean oil, medium-chain triglycerides, olive oil, and fish oil: a single-center, double-blind randomized study on efficacy and safety in pediatric patients receiving home parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2010;34:485–495.
178. Gura KM, Duggan CP, Collier SB, et al. Reversal of parenteral nutrition-associated liver disease in two infants with short bowel syndrome using parenteral fish oil: implications for future management. *Pediatrics*. 2006;118:e197–e201.
179. de Meijer VEL HD, Meisel JA, Gura KM, Puder M. Parenteral fish oil as monotherapy prevents essential fatty acid deficiency in parenteral nutrition dependent patient. *J Pediatr Gastroenterol Nutr*. 2010; 50:212–218.
180. Le HD, Meisel JA, de Meijer VE, et al. The essentiality of arachidonic acid and docosahexaenoic acid. *Prostaglandins Leukot Essent Fatty Acids*. 2009;81:165–170.
181. Koletzko B. Intravenous lipid emulsions for infants: when and which? *Am J Clin Nutr*. 2012;96:225–226.
182. Koletzko B, Goulet O. Fish oil containing intravenous lipid emulsions in parenteral nutrition-associated cholestatic liver disease. *Curr Opin Clin Nutr Metab Care*. 2010;13:321–326.
183. Seida JC, Mager DR, Hartling L, et al. Parenteral w-3 fatty acid lipid emulsions for children with intestinal failure and other conditions: a systematic review. *JPEN J Parenter Enteral Nutr*. 2013;37:44–55.
184. Borum PR. Carnitine and lipid metabolism. *Bol Asoc Med PR*. 1991;83:134–135.
185. Cairns PA, Stalker DJ. Carnitine supplementation of parenterally fed neonates. *Cochrane Database Syst Rev*. 2000;(4):CD000950.
186. Simopoulos AP. Importance of the ratio of omega-6/omega-3 essential fatty acids: evolutionary aspects. *World Rev Nutr Diet*. 2003;92:1–22.
187. Barr LH, GD Dunn, MF Brennan. Essential fatty acid deficiency during total parenteral nutrition. *Ann Surg*. 1981;193:304–311.
188. Frieman Z, Damon A, Stahlman MT, et al. Rapid onset of essential fatty acid deficiency in the newborn. *Pediatrics*. 1976;58:640–649.
189. Holman R. The ratio of trienoic:tetranoic acids in tissue lipids as a measure of essential fatty acid requirements. *J Nutr*. 1970;60:405–410.
190. Siguel EN, Chee KM, Gong JX, et al. Criteria for essential fatty acid deficiency in plasma as assessed by capillary column gas-liquid chromatography. *Clin Chem*. 1987;33:1869–1873.

191. Farrell PM, Gutcher GR, Palta M, et al. Essential fatty acid deficiency in premature infants. *Am J Clin.Nutr*. 1988;48:220–229.
192. Foote KD, MacKinnon MJ, Innis SM. Effect of early introduction of formula vs fat-free parenteral nutrition on essential fatty acid status of preterm infants. *Am J Clin Nutr*. 1991;54:93–97.
193. Gutcher GR, Farrell PM. Intravenous infusion of lipid for the prevention of essential fatty acid deficiency in premature infants. *Am J Clin Nutr*. 1991;54:1024–1028.
194. Bohles H, Bieber MA, Heird WC. Reversal of experimental essential fatty acid deficiency by cutaneous administration of safflower oil. *Am J Clin Nutr*. 1976;29:398–401.
195. Skolnick P, Eaglestein WH, Ziboh VA. Human essential fatty acid deficiency: treatment of topical applications of linoleic acid. *Arch Dermatol*. 1977;113:939–941.
196. Nehra D, Fallon EM, Carlson SJ, et al. Provision of a soy-based intravenous lipid emulsion at 1 g/kg/d does not prevent cholestasis in neonates. *JPEN J Parenter Enteral Nutr*. 2013;37:498–505.
197. Cober MP, Killu G, Brattain A, et al. Intravenous fat emulsions reduction for patients with parenteral nutrition-associated liver disease. *J Pediatr*. 2012;160:421–427.
198. Crawford MA, Costeloe K, Ghebremeskel K, et al. The inadequacy of the essential fatty acid content of present preterm feeds. *Eur J Pediatr*. 1998;157:S23–S27.
199. Allen TM, Murray L. Effect of long-term intralipid administration in mice. *Can J Physiol Pharmacol*. 1986;64:1006–1010.
200. Sobrado J, Moldawer LL, Pomposelli JJ, et al. Lipid emulsions and reticuloendothelial system function in healthy and burned guinea pigs. *Am J Clin Nutr*. 1985;42:855–863.
201. Pierro A, Carnielli V, Filler RM, et al. Metabolism of intravenous fat emulsion in the surgical newborn. *J Pediatr Surg*. 1989;24:95–101.
202. Roulet M, Wiesel PH, Pilet M, et al. Effects of intravenously infused egg phospholipids on lipid and lipoprotein metabolism in postoperative trauma. *JPEN J Parenter Enteral Nutr*. 1993;17(2):107–112.
203. Haumont D, Deckelbaum RJ, Richelle M, et al. Plasma lipid and plasma lipoprotein concentrations in low birth weight infants given parenteral nutrition with twenty or ten percent lipid emulsion. *J Pediatr*. 1989;115:787–793.
204. Krohn K, Koletzko BV. Parenteral lipid emulsions in pediatrics. *Curr Opin Clin Nutr Metab Care*. 2006;9:319–323.
205. Spear ML, Stahl GE, Paul MH, et al. The effect of 15-hour fat infusions of varying dosage on bilirubin binding to albumin. *JPEN J Parenter Enteral Nutr*. 1985;9:144–147.
206. Rubin M, Naor N, Sirota L, et al. Are bilirubin and plasma lipid profiles of premature infants dependent on the lipid emulsion infused? *J Pediatr Gastroenterol Nutr*. 1995;21:25–30.
207. Park W, Paust H, Schroder H. Lipid infusion in premature infants suffering from sepsis. *JPEN J Parenter Enteral Nutr*. 1984;8:290–292.
208. Hammerman C, Aramburo MJ. Decreased lipid intake reduces morbidity in sick premature neonates. *J Pediatr*. 1988;113:1083–1088.
209. Hammerman C, Aramburo MJ, Hill V. Intravenous lipids in newborn lungs: thromboxane-mediated effects. *Crit Care Med*. 1989;17:430–436.
210. Drenckpohl D, McConnell C, Gaffney S, et al. Randomized trial of very low birth

weight infants receiving higher rates of infusion of intravenous fat emulsions during the first week of life. *Pediatrics*. 2008;122:743–751.
211. Simmer K, Rao SC. Early introduction of lipids to parenterally-fed preterm infants. *Cochrane Database Syst Rev*. 2005(2):CD005256.
212. di Trolli SE, Kermorvant-Duchemin E, Huon C, et al. Early lipid supply and neurological development at one year in very low birth weight (VLBW) preterm infants. *Early Hum Dev*. 2012;88 Suppl 1:S25–S29.
213. Fusch C, Bauer K, Bohles HJ, et al. Neonatology/pediatrics—Guidelines on parenteral nutrition. Chapter 13. *Ger Med Sci*. 2009;7:Doc15.
214. Driscoll DF. Lipid injectable emulsions: pharmacopoeial and safety issues. *Pharm Res*. 2006;23:1959–1969.
215. Mühlebach SF. Practical aspects of multichamber bags for total parenteral nutrition. *Curr Opin Clin Nutr Metab Care*. 2005;8:291–295.
216. Chambrier C, Lauverjat M, Bouletreau P. Structured triglyceride emulsions in parenteral nutrition. *Nutr Clin Pract*. 2006;21:342–350.
217. Humberstone DA, Koea J, Shaw JH. Relative importance of amino acid infusion as a means of sparing protein in surgical patients. *JPEN J Parenter Enteral Nutr*. 1989;13:223–227.
218. Tappy L, Schwarz JM, Schneiter P, et al. Effects of isoenergetic glucose-based or lipid-based parenteral nutrition on glucose metabolism, de novo lipogenesis, and respiratory gas exchanges in critically ill patients. *Crit Care Med*. 1998;26:860–867.
219. Kalhan SC, Kilic I. Carbohydrate as nutrient in the infant and child: range of acceptable intake. *Eur J Clin Nutr*. 1999;53:S94–S100.
220. Wintergerst KA, Buckingham B, Gandrud L, et al. Association of hypoglycemia, hyperglycemia, and glucose variability with morbidity and death in the pediatric intensive care unit. *Pediatrics*. 2006;118:173–179.
221. Hirshberg E, Larsen G, Van Duker H. Alterations in glucose homeostasis in the pediatric intensive care unit: hyperglycemia and glucose variability are associated with increased mortality and morbidity. *Pediatr Crit Care Med*. 2008;9:361–366.
222. Lafeber HN, Sulkers EJ, Chapman TE, Sauer PJ. Glucose production and oxidation in preterm infants during total parenteral nutrition. *Pediatr Res*. 1990;28(2):153–157.
223. Jones MO, Pierro A, Hammond P, et al. Glucose utilization in the surgical newborn infant receiving total parenteral nutrition. *J Pediatr Surg*. 1993;28:1121–1125.
224. Sauer PJ, Van Aerde JE, Pencharz PB, et al. Glucose oxidation rates in newborn infants measured with indirect calorimetry and [U-13C] glucose. *Clin Sci*. 1986;70:587–593.
225. Sheridan RL, Yu YM, Prelack K, et al. Maximal parenteral glucose oxidation in hypermetabolic young children: a stable isotope study. *JPEN J Parenter Enteral Nutr*. 1998;22:212–216.
226. Hu H, Greif RL, Goodwin CW. The effects of thermal injury on mitochondrial oxygen consumption and the glycerol phosphate shuttle. *Metabolism*. 1994;43:913–916.
227. Demling R, Saldinger P, LaLonde C, et al. Multiple-organ dysfunction in the surgical patient: pathophysiology, prevention and treatment. *Curr Prob Surg*. 1003;30:347–411.
228. Frayn KN. Calculation of substrate oxidation rates in vivo from gaseous exchange. *J Appl Physiol*. 1983;55:628–634.
229. Askanazi J, Rosenbaum SH, Hyman AI, et al. Respiratory changes induced by the

large glucose loads of total parenteral nutrition. *JAMA*. 1980;243:1444–1447.
230. Giovannini I, Boldrini G, Castagneto M, et al. Respiratory quotient and patterns of substrate utilization in human sepsis and trauma. *JPEN J Parenter Enteral Nutr*. 1983;7:226–230.
231. Nose O, Tipton JR, Ament ME, et al. Effect of the energy source on changes in energy expenditure, respiratory quotient, and nitrogen balance during total parenteral nutrition in children. *Pediatr Res*. 1987;21:538–541.
232. Meguid M, Akahoshi MP, Jeffers S, et al. Amelioration of metabolic complications of conventional TPN. *Arch Surg*. 1984;119:1294–1298.
233. van den Berghe G, A Wilmer, G Hermans, et al. Intensive insulin therapy in the medical ICU. *N Engl J Med*. 2006;354:449–461.
234. Vlasselaers D, Milants I, Desmet L, et al. Intensive insulin therapy for patients in paediatric intensive care: a prospective, randomised controlled study. *Lancet*. 2009;373:547–556.
235. Agus MS, Steil GM, Wypij D, et al; SPECS Study Investigators. Tight glycemic control versus standard care after pediatric cardiac surgery. *N Engl J Med*. 2012;367:1208–1219.
236. Ogilvy-Stuart AL, Beardsall K. Management of hyperglycaemia in the preterm infant. *Arch Dis Child Fetal Neonatal Ed*. 2010;95:F126–F131.
237. Cornblath M, Hawdon JM, Williams AF, et al. Controversies regarding definition of neonatal hypoglycemia: suggested operational thresholds. *Pediatrics*. 2000;105:1141–1145.
238. Beardsall K, Vanhaesebrouck S, Ogilvy-Stuart AL, et al. Prevalence and determinants of hyperglycemia in very low birth weight infants: cohort analyses of the NIRTURE study. *J Pediatr*. 2010;157:715–719.
239. Beardsall K, Vanhaesebrouck S, Ogilvy-Stuart AL, et al. Early insulin therapy in very-low-birth-weight infants. *N Engl J Med*. 2008;359:1873–1884.
240. Sinclair JC, Bottino M, Cowett RM. Interventions for prevention of neonatal hyperglycemia in very low birth weight infants. *Cochrane Database Syst Rev*. 2011;(10):CD007615.
241. Denne SC, Karn CA, Wang J, et al. Effect of intravenous glucose and lipid on proteolysis and glucose production in normal newborns. *Am J Physiol*. 1995;269:E361–E367.
242. Hertz DE, Karn CA, Liu YM, et al. Intravenous glucose suppresses glucose production but not proteolysis in extremely premature newborns. *J Clin Invest*. 1993;92:1752–1758.
243. Sunehag AL, Haymond MW, Schanler RJ, et al. Gluconeogenesis in very low birth weight infants receiving total parenteral nutrition. *Diabetes*. 1999;48:791–800.
244. Chessex P, Belanger S, Piedboeuf B, et al. Influence of energy substrates on respiratory gas exchange during conventional mechanical ventilation of preterm infants. *J Pediatr*. 1995;126:619–624.
245. Bottino M, Cowett RM, Sinclair JR. Interventions for treatment of neonatal hyperglycemia in very low birth weight infants. *Cochrane Database Syst Rev*. 2011;(10):CD007453.
246. Collins JW Jr. Hoppe M, Brown K, et al. A controlled trial of insulin infusion and parenteral nutrition in extremely low birth weight infants with glucose intolerance. *J Pediatr*. 1991;118:921–927.
247. Poindexter BB, Karn CA, Denne SC. Exogenous insulin reduces proteolysis and protein synthesis in extremely low birth weight infants. *J Pediatr*. 1998;132:948–953.

248. Fusch C, Hungerland E, Scharrer B, Moeller H. Water turnover of healthy children measured by deuterated water elimination. *Eur J Pediatr*. 1993;152:110–114.
249. Friis-Hansen B. Body water compartments in children: changes during growth and related changes in body composition. *Pediatrics*. 1961;28:169–181.
250. Darrow DC. Physiologic basis for estimating requirements for parenteral fluids. *Pediatr Clin North Am*. 1959;6:29–41.
251. Holliday MA, Segar WE. The maintenance need for water in parenteral fluid therapy. *Pediatrics*. 1957;19:823–832.
252. Bower TR, Pringle KC, Soper RT. Sodium deficit causing decreased weight gain and metabolic acidosis in infants with ileostomy. *J Pediatr Surg*. 1988;23:567–572.
253. Mirtallo J, Canada T, Johnson D, et al. Safe practices for parenteral nutrition. Task force for the revision of safe practices for parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2004;28:S39–S70.
254. Ross AC, Manson JE, Abrams SA, et al. The 2011 report on dietary reference intakes for calcium and vitamin D from the Institute of Medicine: what clinicians need to know. *J Clin Endocrinol Metab*. 2011;96:53–58.
255. Hicks W, Hardy G. Phosphate supplementation for hypophosphataemia and parenteral nutrition. *Curr Opin Clin Nutr Metab Care*. 2001;4(3):227–233.
256. Schenk WG, Moylan JA. Hypercalciuresis in trauma patients on high-nitrogen enteral and parenteral diets. *J Trauma*. 1979;19:227–233.
257. Adelman RD, Abern SB, Merten D, Halsted CH. Hypercalciuria with nephrolithiasis: a complication of total parenteral nutrition. *Pediatrics*. 1977;59:473–475.
258. Koo WW. Parenteral nutrition-related bone disease. *JPEN J Parenter Enteral Nutr*. 1992;6:386–394.
259. Pelegano JF, Rowe JC, Carey DE, et al. Effect of calcium/phosphorus ratio on mineral retention in parenterally fed premature infants. *J Pediatr Gastroenterol Nutr*. 1991;12:351–355.
260. Sloan GM, White DE, Murray MS, Brennan F. Calcium and phosphorus metabolism during total parenteral nutrition. *Ann Surg*. 1983;197:1–6.
261. Flurkey H. A case presentation: central venous line: what went wrong? *Neonatal Netw*. 1994;13:51–55.
262. Knowles JB, Cusson G, Smith M, Sitrin MD. Pulmonary deposition of calcium phosphate crystals as a complication of home total parenteral nutrition. *J Parenter Enteral Nutr*. 1989;13:209–213.
263. Hill SE, Heldman LS, Goo EDH, et al. Fatal microvascular pulmonary emboli from precipitation of a total nutrient admixture solution. *J Parenter Enteral Nutr*. 1996;20:81–87.
264. Kimura S, Nose O, Seino Y, et al. Effects of alternate and simultaneous administrations of calcium and phosphorus on calcium metabolism in children receiving total parenteral nutrition. *J Parenter Enteral Nutr*. 1986;10:513–516.
265. Wong JC, McDougal AR, Tofan M, et al. Doubling calcium and phosphate concentrations in neonatal parenteral nutrition solutions using monobasic potassium phosphate. *J Am Coll Nutr*. 2006;25:70–77.
266. MacKay MW, Fitzgerald KA, Jackson D. The solubility of calcium and phosphate in two specialty amino acid solutions. *J Parenter Enteral Nutr*. 1996;20:63–66.
267. Boulet M, Marier JR, Rose D. Effect of magnesium on formation of calcium phosphate precipitates. *Arch Biochem Biophysics*. 1962;96:629–636.
268. Robinson LA, Wright BT. Central venous catheter occlusion caused by body-heat-

mediated calcium phosphate precipitation. *Am J Hosp Pharm*. 1982;39:120–121.
269. Chwals WJ. Effect of ambient temperature on physical compatibility of neonatal total parenteral nutrition admixtures. *Pediatr Crit Care Med*. 2009;10:273–274.
270. Driscoll DF, Bhargava HN, Li L, et al. Physicochemical stability of total nutrient admixtures. *Am J Health Syst Pharm*. 1995;52:623–634.
271. MacKay M, Jackson D, Eggert L, et al. Practice-based validation of calcium and phosphorus solubility limits for pediatric parenteral nutrition solutions. *Nutr Clin Pract*. 2011;26:708–713.
272. ASPEN Board of Directors, National Advisory Group on Standards and Practice Guidelines for Parenteral Nutrition. Safe practices for parenteral nutrition formulations. *J Parenter Enteral Nutr*. 1998;22:49–66.
273. Lumpkin MM. Safety alert: hazards of precipitation associated with parenteral nutrition. *Am J Hosp Pharm*. 1994;51:1427.
274. Lichtman MA, Miller DR, Cohen J, Waterhouse C. Reduced red cell glycolysis, 2,3-diphosphoglycerate and adenosine triphosphate concentration and increased haemoglobin-oxygen affinity caused by hypophosphataemia. *Ann Intern Med*. 1971;74:562–568.
275. Modi N. Development of renal function. *Br Med Bull*. 1988;44:935–956.
276. Kugelman A, Durand M. A comprehensive approach to the prevention of bronchopulmonary dysplasia. *Pediatr Pulmonol*. 2011;46: 1153–1165.
277. Reller MD, Lorenz JM, Kotagal UR, et al. Hemodynamically significant PDA: an echocardiographic and clinical assessment of incidence, natural history, and outcome in very low birth weight infants maintained in negative fluid balance. *Pediatr Cardiol*. 1985;6:17–23.
278. Mitton SG. Amino acids and lipid in total parenteral nutrition for the newborn. *J Pediatr Gastroenterol Nutr*. 1994;18:25–31.
279. Laine L, Shulman RJ, Pitre D, et al. Cysteine usage increases the need for acetate in neonates who receive total parenteral nutrition. *Am J Clin Nutr*. 1991;54:565–567.
280. MacMahon P, Blair ME, Treweeke P, et al. Association of mineral composition of neonatal intravenous feeding solutions and metabolic bone disease of prematurity. *Arch Dis Child*. 1989;64:489–493.
281. Prestridge LL, Schanler RJ, Shulman RJ, et al. Effect of parenteral calcium and phosphorus therapy on mineral retention and bone mineral content in very low birth weight infants. *J Pediatr*. 1993;122:761–768.
282. Vanek VW, Borum P, Buchman A, et al. Novel Nutrient Task Force, Parenteral Multi-Vitamin and Multi–Trace Element Working Group; American Society for Parenteral and Enteral Nutrition (A.S.P.E.N.) Board of Directors. A.S.P.E.N. position paper: recommendations for changes in commercially available parenteral multivitamin and multi-trace element products. *Nutr Clin Pract*. 2012;27:440–491.
283. Greene HL, Smith R, Pollack P, et al. Intravenous vitamins for very-low-birth-weight infants. *J Am Coll Nutr*. 1991;10:281–288.
284. Shenai JP, Stahlman MT, Chytil F. Vitamin A delivery from parenteral alimentation solution. *J Pediatr*. 1981;99:661–663.
285. Thomas DG, James SL, Fudge A, et al. Delivery of vitamin A from parenteral nutrition solutions in neonates. *J Paediatr Child Health*. 1991;27:180–183.
286. Haas C, Genzel-Boroviczeny O, Koletzko B. Losses of vitamin A and E in parenteral nutrition suitable for premature infants. *Eur J Clin Nutr*. 2002;56:906–912.
287. Darlow BA, Graham PJ. Vitamin A supplementation to prevent mortality and

short- and long-term morbidity in very low birthweight infants. *Cochrane Database Syst Rev*. 2011;(10):CD000501.
288. Institute of Medicine, Food and Nutrition Board. Thiamine. In: Dietary Reference Intakes for Thiamine, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline. Washington, DC: National Academy of Sciences; 1998.
289. Infuvite Adult and Infuvite Pediatric Multiple Vitamins for Infusion [product description]. Sandoz, Canada: Boucherville; 2007.
290. M.V.I. Pediatric Multi-Vitamins for Infusion [package insert]. Lake Forest, IL: Mayne Pharma. http://www.hospira.com/Files/MVI_pediatric_PI.pdf. Accessed August 20, 2013.
291. Friel JK, Bessie JC, Belkhode SH, et al. Thiamine, riboflavin, pyridoxine, and vitamin C status in premature infants receiving parenteral and enteral nutrition. *J Pediatr Gastroenterol Nutr*. 2001;33:64–69.
292. Levy R, Herzberg GR, Andrews WL, et al. Thiamine, riboflavin, folate, and vitamin B12 status of low birth weight infants receiving parenteral and enteral nutrition. *JPEN J Parenter Enteral Nutr*. 1992;16:241–247.
293. Vanamee P, Shils ME, Burke AW, et al. Multivitamin preparations for parenteral use: a statement by the nutrition advisory group. *JPEN J Parenter Enteral Nutr*. 1979;3:258–261.
294. Mirtallo J, Canada T, Johnson D, et al. Safe practices for parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2004;28:S39–S70.
295. Greene HL, Hambidge KM, Schanler R, et al. Guidelines for the use of vitamins, trace elements, calcium, magnesium, and phosphorus in infants and children receiving total parenteral nutrition: report of the Subcommittee on Pediatric Parenteral Nutrient Requirements from the Committee on Clinical Practice Issues of the American Society for Clinical Nutrition [published errata appear in *Am J Clin Nutr*. 1989;49:1332 and 1989;50:560, *Am J Clin Nutr*. 1988;48:1324–1342.
296. Moore MC, Greene HL, Phillips B, et al. Evaluation of a pediatric multiple vitamin preparation for total parenteral nutrition in infants and children, I: blood levels of water-soluble vitamins. *Pediatrics*. 1986;77: 530–538.
297. Norton JA, Nichoalds GE, Maher MM, et al. Water-soluble vitamins in cancer patients on parenteral nutrition: a prospective study. *JPEN J Parenter Enteral Nutr*. 1987;11:243–249.
298. Dahl GB, Jeppsson RI, Tengborn HJ. Vitamin stability in a TPN mixture stored in an EVA plastic bag. *J Clin Hosp Pharm*. 1986;11:271–279.
299. Institute of Medicine, Food and Nutrition Board, Institute of Medicine. Vitamin B6. In: *Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline*. Washington, DC: National Academies Press; 1998
300. Chen MF, Boyce HW Jr, Triplett L. Stability of the B vitamins in mixed parenteral nutrition solution. *JPEN J Parenter Enteral Nutr*. 1983;7:462–464.
301. Van der Holst A, Martens HJM, de Goede PNEC. Analysis of water-soluble vitamins in total parenteral nutrition solution by high pressure liquid chromatography. *Pharm Weekly*. 1989;11:169–174.
302. Zempleni J, Kubler W. The utilization of intravenously infused pyridoxine in humans. *Clin Chim Acta*. 1994;229:27–36.
303. Specker BL, Black A, Allen L, Morrow F. Vitamin B12: low milk concentrations are related to low serum concentrations in vegetarian women and to methylmalonic aciduria in their infants. *Am J Clin Nutr*. 1990;52:1073–1076.
304. Marinier E, Gorski AM, de Courcy GP, et al. Blood levels of water-soluble

vitamins in pediatric patients on total parenteral nutrition using a multiple vitamin preparation. *JPEN J Parenter Enteral Nutr*. 1989;13:176–184.
305. Vargas JH, Ament ME, Berquist WE. Long-term home parenteral nutrition in pediatrics: ten years of experience in 102 patients. *J Pediatr Gastroenterol Nutr*. 1987;6:24–32.
306. Mock DM, Baswell DL, Baker H, et al. Biotin deficiency complicating parenteral alimentation: diagnosis, metabolic repercussions, and treatment. *J Pediatr*. 1985;106:762–769.
307. Standing Committee on the Scientific Evaluation of Dietary Reference Intakes Food and Nutrition Board. Vitamin C, vitamin E, selenium, and carotene and other carotenoids: methods. In: Institute of Medicine, ed. *DRI—Dietary Reference Intakes for Vitamin C, Vitamin E, Selenium, and Carotenoids*. Washington, DC: National Academy Press; 2000: pp. 58–72.
308. Friel JK, Bessie JC, Belkhode SL, et al. Thiamine, riboflavin, pyridoxine, and vitamin C status in premature infants receiving parenteral and enteral nutrition. *J Pediatr Gastroenterol Nutr*. 2001;33:64–69.
309. Dupertuis YM, Ramseyer S, Fathi M, et al. Assessment of ascorbic acid stability in different multilayered parenteral nutrition bags: critical influence of the bag wall material. *JPEN J Parenter Enteral Nutr*. 2005;29:125–130.
310. Baeckert PA, Greene HL, Oelberg DG, et al. Blood levels of vitamins A, D, E and riboflavin in very low birth weight infants receiving total parenteral nutrition with vitamins added to the lipid emulsion. *J Pediatr*. 1988;113:1057–1065.
311. Tappel AL. Vitamin E as the biologic lipid antioxidant. *Vitam Horm*. 1962;20:493–510.
312. Martone WJ, Williams WW, Mortensen ML, et al. Illness with fatalities in premature infants association with an intravenous vitamin E preparation E-ferol. *Pediatrics*. 1986;78:591–600.
313. Greene HL, Moore MC, Phillips B, et al. Evaluation of a pediatric multiple vitamin preparation for total parenteral nutrition. II Blood levels of vitamins A, D and E. *Pediatrics*. 1986;77:539–540.
314. Allwood MC, Martin HJ. The photodegradation of vitamins A and E in parenteral nutrition mixtures during infusion. *Clin Nutr*. 2000;19: 339–342.
315. Skouroliakou M, Matthaiou C, Chiou A, et al. Physicochemical stability of parenteral nutrition supplied as all-in-one for neonates. *JPEN J Parenter Enteral Nutr*. 2008;32:201–209.
316. American Academy of Pediatrics, Committee on Nutrition. Vitamin and mineral supplement needs in normal children in the United States. *Pediatrics*. l980;66:1015–1021.
317. Buchman AL. The addition of choline to parenteral nutrition. *Gastroenterology*. 2009;137(supp l):S119–128.
318. Khashu M, Harrison A, Lalari V, et al. Impact of shielding parenteral nutrition from light on routine monitoring of blood glucose and triglyceride levels in preterm neonates. *Arch Dis Child Fetal Neonatal Ed*. 2009;94:F111–115.
319. Leung FY. Trace elements in parenteral micronutrition. *Clin Biochem*. 1995;28:561–566.
320. Pluhator-Murton MM, Fedorak RN, Audette RJ, et al. Trace element contamination of total parenteral nutrition, 1: contribution of component solutions. *JPEN J Parenter Enteral Nutr*. 1999;23:222–227.
321. Friel JK, MacDonald AC, Mercer CN, et al. Molybdenum requirements in low-birth-weight infants receiving parenteral and enteral nutrition. *JPEN J Parenter Enteral Nutr*. 1999;23:155–159.

322. Dickerson RN. Manganese intoxication and parenteral nutrition. *Nutrition*. 2001;15:689–693.
323. Howard L, Ashley C, Lyon D, Shenkin A. Autopsy tissue trace elements in 8 long-term parenteral nutrition patients who received the current U.S. Food and Drug Administration formulation. *JPEN J Parenter Enteral Nutr*. 2007;31:388–396.
324. Blaszyk H, Wild PJ, Oliveira A, et al. Hepatic copper in patients receiving long-term total parenteral nutrition. *J Clin Gastroenterol*. 2005;39:318–320.
325. Jeejeebhoy K. Zinc: an essential trace element for parenteral nutrition. *Gastroenterology*. 2009;137(supp l):S7–S12.
326. Harper JI, Thompson D, Kovar IZ, et al. Zinc deficiency in a preterm neonate with necrotizing enterocolitis. *J R Soc Med*. 1984;77 (Suppl 4):40–41.
327. Herson VC, Phillipps AF, Zimmerman A. Acute zinc deficiency in a premature infant after bowel resection and intravenous alimentation. *Am J Dis Child*. 1981;135:968–969.
328. Shenkin A. Selenium in intravenous nutrition. *Gastroenterology*. 2009;137(supp l):S61–S69.
329. Lockitch G, Jacobson B, Quigley G, et al. Selenium deficiency in low birth weight neonates: an unrecognized problem. *J Pediatr*. 1989;114:865–870.
330. Makhoul IR, Sammour RN, Diamond E, et al. Selenium concentrations in maternal and umbilical cord blood at 24–42 weeks of gestation: basis for optimization of selenium supplementation to premature infants. *Clin Nutr*. 2004;23:373–381.
331. Darlow BA, Austin NC. Selenium supplementation to prevent short-term morbidity in preterm neonates. *Cochrane Database Syst Rev*. 2003;(4):CD003312.
332. Dickerson RN. Manganese intoxication and parenteral nutrition. *Nutrition*. 2001;15:689–693.
333. Hardy IJ, Gillanders L, Hardy G. Is manganese an essential supplement for parenteral nutrition? *Curr Opin Clin Nutr Metab Care*. 2008;11:289–296.
334. Takagi Y, Okada A, Sando K, et al. Evaluation of indexes of in vivo manganese status and the optimal intravenous dose for adult patients undergoing home parenteral nutrition. *Am J Clin Nutr*. 2002;75:112–118.
335. Blaszyk H, Wild PJ, Oliveira A, et al. Hepatic copper in patients receiving long-term total parenteral nutrition. *J Clin Gastroenterol*. 2005;39:318–320.
336. Corkins MR, Martin VA, Szeszycki EE. Copper levels in cholestatic infants on parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2013;37:92–96.
337. Frem J, Sarson Y, Sternberg T, et al. Copper supplementation in parenteral nutrition of cholestatic infants. *J Pediatr Gastroenterol Nutr*. 2010;50:650–654.
338. Berger MM, Cavadini C, Bart A, et al. Cutaneous copper and zinc losses in burns. *Burns*. 1992;18:373–380.
339. National Academy of Sciences, Institute of Medicine, Food and Nutrition Board. *Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium, and Zinc*. 2001.
340. Leung FY. Elevated serum chromium in patients on total parenteral nutrition and the ionic species of contaminant chromium. *Biol Trac Elem Res*. 1995;50:221–228.
341. Bougle D, Bureau F, Deschrevel G, et al. Chromium and parenteral nutrition in children. *J Pediatr Gastroenterol Nutr*. 1993;17:72–74.
342. Moukarzel A, Song MK, Buchman AL, et al. Excessive chromium intake in children receiving total parenteral nutrition. *Lancet*. 1992;339:385–388.

343. Buchman AL, Neely M, Grossie VB, et al. Organ heavy-metal accumulation during parenteral nutrition is associated with pathologic abnormalities in rats. *Nutrition*. 2001;17:600–606.
344. Jeejeebhoy KN, Chu RC, Marliss EB, et al. Chromium deficiency, glucose intolerance, and neuropathy reversed by chromium supplementation in a patient receiving long-term total parenteral nutrition. *Am J Clin Nutr*. 1977;30:531–538.
345. Brown RO, Forloines-Lynn S, Cross RE, et al. Chromium deficiency after long-term total parenteral nutrition. *Dig Dis Sci*. 1986;31:661–664.
346. Moukarzel A. Chromium in parenteral nutrition: too little or too much? *Gastroenterology*. 2009;137(5)(supp l):S18–S28.
347. Mouser JF, Hak EB, Helms RA, et al. Chromium and zinc concentrations in pediatric patients receiving long-term parenteral nutrition. *Am J Health Syst Pharm*. 1999;56:1950–1956.
348. Friel JK, MacDonald AC, Mercer CN, et al. Molybdenum requirements in low-birth-weight infants receiving parenteral and enteral nutrition. *JPEN J Parenter Enteral Nutr*. 1999;23;155–159.
349. Dani C, Martelli E, Bertini G, et al. Effect of blood transfusions on oxidative stress in preterm infants. *Arch Dis Child Fetal Neonatal Ed*. 2004;89:F408–F411.
350. Perrone S, Negro S, Tataranno ML, et al. Oxidative stress and antioxidant strategies in newborns. *J Matern Fetal Neonatal Med*. 2010;23 Suppl 3:63–65.
351. Zimmermann MB. Iodine: it's important in patients that require parenteral nutrition. *Gastroenterology*. 2009;137(supp l):S36–S46.
352. Below H, Behrens-Baumann W, Bernhardt C, et al. Systemic iodine absorption after preoperative antisepsis using povidone-iodine in cataract surgery—an open controlled study. *Dermatology*. 2006;212 (suppl 1):41–46.
353. Nielsen FH. Micronutrients in parenteral nutrition: boron, silicon, and fluoride. *Gastroenterology*. 2009;137(supp l):S55–S60.
354. Broviac JW, Cole JJ, Scribner BH. A silicone rubber atrial catheter for prolonged parenteral alimentation. *Surg Gynecol Obstet*. 1973;136: 602–606.
355. Hickman RO, Buckner CD, Clift RA, et al. A modified right atrial catheter for access to the venous system in marrow transplant recipients. *Surg Gynecol Obstet*. 1979;148:871–875.
356. Thomas JH, MacArthur RI, Pierce GE, et al. Hickman-Broviac catheters: Indications and results. *Am J Surg*. 1980;140:791–796.
357. Näslund E, Wadström C, Backman L. Long term home parenteral nutrition using a subcutaneous venous access. *Clin Nutr*. 1992;11:93–96.
358. Braswell LE. Peripherally inserted central catheter placement in infants and children. *Tech Vasc Interv Radiol*. 2011;14:204–211.
359. Botella-Carretero JI, Carrero C, Guerra E, et al. Role of peripherally inserted central catheters in home parenteral nutrition: a 5-year prospective study. *JPEN J Parenter Enteral Nutr*. 2013;37:544-549.
360. Piper HG, de Silva NT, Amaral JG, et al. The use of peripherally inserted central catheters for long-term parenteral nutrition in infants with intestinal failure. *J Pediatr Gastroenterol Nutr*. 2013;56:578–581.
361. Task Force for the Revision of Safe Practices for Parenteral Nutrition and the A.S.P.E.N. Board of Directors. Safe practices for parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2004;28:S39–S70. Errata 2006; 30:177.
362. Fletcher SJ, Bodenham AR. Safe placement of central venous catheters: where should the tip of the catheter lie? *Br J Anaesth*. 2000;85:188–191.
363. The carina as a landmark for central venous catheter placement in small children. *Eur J Pediatr*. 2006;165:264–266.

364. Na HS, Kim JT, Kim HS, et al. Practical anatomic landmarks for determining the insertion depth of central venous catheter in paediatric patients. *Br J Anaesth*. 2009;102:820–823.
365. Maki DG, Stolz SS, Wheeler S, et al. A prospective, randomized trial of gauze and two polyurethane dressings for site care of pulmonary artery catheters: implications for catheter management. *Crit Care Med*. 1994;22:1729–1737.
366. Mitchell MD, Anderson BJ, Williams K, et al. Benefit of heparin in peripheral venous and arterial catheters: systematic review and meta-analysis of randomised controlled trials. *J Adv Nurs*. 2009;65:2007–2021.
367. Hanson RL, Grant AM, Majors KR. Heparin-lock maintenance with ten units of sodium heparin in one milliliter of normal saline solution. *Surg Gynecol Obstet*. 1976;142:373–376.
368. Ball PA. Intravenous in-line filters: filtering the evidence. *Curr Opin Clin Nutr Metab Care*. 2003;6:319–325.
369. Bethune K, Allwood M, Grainger C, et al. Use of filters during the preparation and administration of parenteral nutrition: position paper and guidelines prepared by a British pharmaceutical nutrition group working party. *Nutrition*. 2001;17:403–408.
370. Gillies D, O'Riordan L, Wallen M, et al. Optimal timing for intravenous administration set replacement. *Cochrane Database Syst Rev*. 2005; (4):CD003588.
371. Matlow AG, Kitai I, Kirpalani H, et al. A randomized trial of 72-versus 24-hour intravenous tubing set changes in newborns receiving lipid therapy. *Infect Control Hosp Epidemiol*. 1999;20:487–493.
372. Duerksen DR, Papineau N, Siemens J, et al. Peripherally inserted central catheters for parenteral nutrition: a comparison with centrally inserted catheters. *JPEN J Parenter Enteral Nutr*. 1999;23:85–89.
373. Lapillonne A, Fellous L, Mokthari M, Kermorvant-Duchemin E. Parenteral nutrition objectives for very low birth weight infants: results of a national survey. *J Pediatr Gastroenterol Nutr*. 2009;48:618–626.
374. Shulman RJ, Phillips S. Parenteral nutrition in infants and children. *J Pediatr Gastroenterol Nutr*. 2003;36:587–607.
375. Meadows N. Monitoring and complications of parenteral nutrition. *Nutrition*. 1998;14:806–808.
376. Puntis JW. Nutritional support at home and in the community. *Arch Dis Child*. 2001;84:295–298.
377. Newton R, Timmis L, Bowling TE. Changes in parenteral nutrition supply when the nutrition support team controls prescribing. *Nutrition*. 2001;17:347–348.
378. Btaiche IF, Khalidi N. Parenteral nutrition-associated liver complications in children. *Pharmacotherapy*. 2002;22:188–211.
379. Buchman A. Total parenteral nutrition-associated liver disease. *JPEN J Parenter Enteral Nutr*. 2002;26(Suppl 5):S43–48.
380. Squires RH, Duggan C, Teitelbaum DH, et al. Pediatric Intestinal Failure Consortium. Natural history of pediatric intestinal failure: initial report from the pediatric intestinal failure consortium. *J Pediatr*. 2012;161:723–728.
381. Chan S, McCowen KC, Bistrian BR, et al. Incidence, prognosis, and etiology of end-stage liver disease in patients receiving home total parenteral nutrition. *Surgery*. 1999;126:28–34.
382. Kelly DA. Liver complications of pediatric parenteral nutrition–epidemiology. *Nutrition*. 1998;14:153–157.
383. Quiros-Tejeira RE, Ament ME, Reyen L, et al. Long-term parenteral nutritional

support and intestinal adaptation in children with short bowel syndrome: a 25-year experience. *J Pediatr*. 2004;145:157–163.
384. Willis TC, Carter BA, Rogers SP, et al. High rates of mortality and morbidity occur in infants with parenteral nutrition-associated cholestasis. *JPEN J Parenter Enteral Nutr*. 2010;34:32–37.
385. Beath SV, Davies P, Papadopoulou A, et al. Parenteral nutrition-related cholestasis in postsurgical neonates: multivariate analysis of risk factors. *J Pediatr Surg*. 1996;31:604–606.
386. Trauner M, Fickert P, Stauber RE. Inflammation-induced cholestasis. *J Gastroenterol Hepatol*. 1999;14:946–959.
387. Zambrano E, El-Hennawy M, Ehrenkranz RA, et al. Total parenteral nutrition induced liver pathology: an autopsy series of 24 newborn cases. *Pediatr Dev Pathol*. 2004;7:425–432.
388. San Luis VA, Btaiche IF. Ursodiol in patients with parenteral nutrition-associated cholestasis. *Ann Pharmacother*. 2007;41:1867–1872.
389. Ng PC, So KW, Fung KS, et al. Randomised controlled study of oral erythromycin for treatment of gastrointestinal dysmotility in preterm infants. *Arch Dis Child Fetal Neonatal Ed*. 2001;84:F177–182.
390. Ng PC, Lee CH, Wong SP, et al. High-dose oral erythromycin decreased the incidence of parenteral nutrition–associated cholestasis in preterm infants. *Gastroenterology*. 2007;132:1726–1739.
391. Tulikoura I, Huikuri K. Morphological fatty changes and function of the liver, serum free fatty acids and triglycerides during parenteral nutrition. *Scand J Gastroenterol*. 1982;17:177–185.
392. Drongowski RA, Coran AG. An analysis of factors contributing to the development of total parenteral nutrition-induced cholestasis. *JPEN J Parenter Enteral Nutr*. 1989;13:586–589.
393. Vileisis RA, Inwood RJ, Hunt CE. Prospective controlled study of parenteral nutrition-associated cholestatic jaundice: effect of protein intake. *J Pediatr*. 1980;96:893–897.
394. Cooper A, Betts JM, Pereira GR, et al. Taurine deficiency in the severe hepatic dysfunction complicating total parenteral nutrition. *J Pediatr Surg*. 1984;19:462–466.
395. Blau J, Sridhar S, Mathieson S, et al. Effects of protein/nonprotein caloric intake on parenteral nutrition associated cholestasis in premature infants weighing 600–1000 grams. *JPEN J Parenter Enteral Nutr*. 2007;31:487–490.
396. Cavicchi M, Beau P, Crenn P, et al. Prevalence of liver disease and contributing factors in patients receiving home parenteral nutrition for permanent intestinal failure. *Ann Intern Med*. 2000;132:525–532.
397. Colomb V, Jobert-Giraud A, Lacaille F, et al. Role of lipid emulsions in cholestasis associated with long term parenteral nutrition in children. *J Parenteral Enteral Nutr*. 2000;24:345–350.
398. Cober MP, Killu G, Brattain A, et al. Intravenous fat emulsions reduction for patients with parenteral nutrition-associated liver disease. *J Pediatr*. 2012;160:421–427.
399. Nehra D, Fallon EM, Carlson SJ, et al. Provision of a soy-based intravenous lipid emulsion at 1 g/kg/d does not prevent cholestasis in neonates. *JPEN J Parenter Enteral Nutr*. 2013;37:498–505.
400. Cowles RA, Ventura KA, Martinez M, et al. Reversal of intestinal failure-associated liver disease in infants and children on parenteral nutrition: experience with 93 patients at a referral center for intestinal rehabilitation. *J Pediatr Surg*.

2010;45:84–87.
401. Diamond IR, de Silva NT, Tomlinson GA, et al. The role of parenteral lipids in the development of advanced intestinal failure-associated liver disease in infants: a multiple-variable analysis. *JPEN J Parenter Enteral Nutr*. 2011;35:596–602.
402. Diamond IR, Sterescu A, Pencharz PB, et al. Changing the paradigm: omegaven for the treatment of liver failure in pediatric short bowel syndrome. *J Pediatr Gastroenterol Nutr*. 2009;48:209–215.
403. Cheung HM, Lam HS, Tam YH, et al. Rescue treatment of infants with intestinal failure and parenteral nutrition-associated cholestasis (PNAC) using a parenteral fish-oil-based lipid. *Clin Nutr*. 2009;28:209–212.
404. Deshpande G, Simmer K, Madhur R, et al. Efficacy and safety of a novel fish oil based lipid emulsion (SMOF) compared with olive oil based lipid emulsion (Clinoleic) in term and near-term (>34 weeks) neonates: a randomized controlled trial. http://www.anzctr.org.au/trial_view.aspx?ID=83596. Accessed November 27, 2012.
405. Fitzgibbons SC, Jones BA, Hull MA, et al. Relationship between biopsy-proven parenteral nutrition-associated liver fibrosis and biochemical cholestasis in children with short bowel syndrome. *J Pediatr Surg*. 2010;45:95–99.
406. Jensen AR, Goldin AB, Koopmeiners JS, et al. The association of cyclic nutrition and decreased incidence of cholestatic liver disease in patients with gastroschisis. *J Ped Surg*. 2009;44:183–189.
407. Bowyer BA, Miles JM, Haymond MW, et al. L-Carnitine therapy in home parenteral nutrition patients with abnormal liver tests and low plasma carnitine concentrations. *Gastroenterology*. 1988;94:434–438.
408. Buchman AL, Dubin MD, Moukarzel AA, et al. Choline deficiency: a cause of hepatic steatosis during parenteral nutrition that can be reversed with intravenous choline supplementation. *Hepatology*. 1995;22:1399–1403.
409. Bishay M, Pichler J, Horn V, et al. Intestinal failure-associated liver disease in surgical infants requiring long-term parenteral nutrition. *J Pediatr Surg*. 2012;47:359–362.
410. Bianchi A. Longitudinal intestinal lengthening and tailoring: results in 20 children. *J R Soc Med*. 1997;90:429–432.
411. Kim HB, Fauza D, Garza J, et al. Serial transverse enteroplasty (STEP): a novel bowel lengthening procedure. *J Pediatr Surg*. 2003;38:425–429.
412. Mazariegos GV, Squires RH, Sindhi RK. Current perspectives on pediatric intestinal transplantation. *Curr Gastroenterol Rep*. 2009;11:226–233.
413. Avitzur Y, Grant D. Intestine transplantation in children: update 2010. *Pediatr Clin North Am*. 2010;57:415–431.
414. Cano N, Cicero F, Ranieri F, et al. Ultrasonographic study of gallbladder motility during total parenteral nutrition. *Gastroenterology*. 1986;91:313–317.
415. Buchman AL, Moukarzel A. Metabolic bone disease associated with total parenteral nutrition. *Clin Nutr*. 2000;19:217–231.
416. Shike M, Harrison JE, Sturtridge WC, et al. Metabolic bone disease in patients receiving long term total parenteral nutrition (TPN). *Ann Intern Med*. 1980;92:343–350.
417. Klein GL, Ament ME, Bluestone R, et al. Bone disease associated with TPN. *Lancet*. 1980;2:1041–1044.
418. Shike M, Shils ME, Heller A, et al. Bone disease in prolonged parenteral nutrition: osteopenia without mineralization defect. *Am J Clin Nutr*. 1986;44:89–98.
419. Diamanti A, Bizzarri C, Basso MS, et al. How does long-term parenteral nutrition impact the bone mineral status of children with intestinal failure? *J Bone Miner*

Metab. 2010;28:351–358.
420. Klein GL. Aluminum in parenteral products: medical perspective on large and small volume parenterals. *J Parenter Sci Technol*. 1989;43:120–124.
421. Bengoa JM, Sitrin MD, Wood RJ, et al. Amino acid induced hypercalciuria in patients on TPN. *Am J Clin Nutr*. 1983;83:264–269.
422. Wood RJ, Sitrin MD, Cusson GJ, et al. Reduction of TPN induced urinary calcium loss by increasing the phosphorous in the TPN prescription. *JPEN J Parenter Enteral Nutr*. 1996;10:188–190.
423. Wood RJ, Bengoa JM, Sitrin MD, et al. Calciuretic effect of cyclic versus continuous TPN. *Am J Clin Nutr*. 1985;41:614–619.
424. Raman M, Aghdassi E, Baun M, et al. Metabolic bone disease in patients receiving home parenteral nutrition: a Canadian study and review. *JPEN J Parenter Enteral Nutr*. 2006;30:492–496.
425. Haderslev KV, Tjellesen L, Sorensen HA, et al. Effect of cyclical intravenous clodronate therapy on bone mineral density and markers of bone turnover in patients receiving home parenteral nutrition. *Am J Clin Nutr*. 2002;76:482–488.
426. Pastore S, Londero M, Barbieri F, et al. Treatment with pamidronate for osteoporosis complicating long-term intestinal failure. *J Pediatr Gastroenterol Nutr*. 2012;55:615–618.
427. Tack I, Wilcox CS, Tran-Van T, et al. Amino acid loading disrupts whole kidney proximal glomerular-tubular balance in humans. *Clin Nephrol*. 1996;45:42–50.
428. Lauverjat M, Hadj Aissa A, Vanhems P, et al. Chronic dehydration may impair renal function in patients with chronic intestinal failure on long-term parenteral nutrition. *Clin Nutr*. 2006;25:75–81.
429. Moukarzel AA, Ament ME, Buchman A, et al. Renal function of children receiving long-term parenteral nutrition. *J Pediatr*. 1991;119:864–868.
430. Buchman AL, Moukarzel A, Ament ME, et al. Serious renal impairment is associated with long-term parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 1993;17:438–444.
431. Boncompain-Gérard M, Robert D, Fouque D, et al. Renal function and urinary excretion of electrolytes in patients receiving cyclic parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2000;24:234–239.
432. Elward AM, Fraser VJ. Risk factors for nosocomial primary bloodstream infection in pediatric intensive care unit patients: a 2-year prospective cohort study. *Infect Control Hosp Epidemiol*. 2006;27:553–560.
433. Colomb V, Fabeiro M, Dabbas M, et al. Central venous catheter-related infections in children on long-term home parenteral nutrition: incidence and risk factors. *Clin Nutr*. 2000;19:355–359.
434. Marra AR, Opilla M, Edmond MB, et al. Epidemiology of bloodstream infections in patients receiving long-term total parenteral nutrition. *J Clin Gastroenterol*. 2007;41:19–28.
435. Sengupta A, Lehmann C, Diener-West M, et al. Catheter duration and risk of CLA-BSI in neonates with PICCs. *Pediatrics*. 2010;125:648–653.
436. Miller SJ, Dickerson RN, Graziani AA, et al. Antibiotic therapy of catheter infections in patients receiving home parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 1990;14:143–147.
437. Pittiruti M, Hamilton H, Biffi R, et al. ESPEN guidelines on parenteral nutrition: central venous catheters (access, care, diagnosis and therapy of complications). *Clin Nutr*. 2009;28:365–377.
438. O'Grady NP, Alexander M, Burns LA, et al. Healthcare Infection Control Practices Advisory Committee. Guidelines for the prevention of intravascular

catheter-related infections. *Am J Infect Control*. 2011;39(4 Suppl 1): S1–34.
439. Snaterse M, Rüger W, Scholte O, et al. Antibiotic-based catheter lock solutions for prevention of catheter-related bloodstream infection: a systematic review of randomised controlled trials. *J Hosp Infect*. 2010;75:1–11.
440. Dümichen MJ, Seeger K, Lode HN, et al. Randomized controlled trial of taurolidine citrate versus heparin as catheter lock solution in paediatric patients with haematological malignancies. *J Hosp Infect*. 2012;80:304–309.
441. Dannenberg C, Bierbach U, Rothe A, et al. Ethanol-lock technique in the treatment of bloodstream infections in pediatric oncology patients with broviac catheter. *J Pediatr Hematol Oncol*. 2003;25:616–621.
442. Ball PA, Brokenshire E, Parry B, et al. Ethanol locking as a possible treatment for microbial contamination of long-term central venous catheters. *Nutrition*. 2003;19:570.
443. Sherertz RJ, Boger MS, Collins CA, et al. Comparative in vitro efficacies of various catheter lock solutions. *Antimicrob Agents Chemother*. 2006;50:1865–1868.
444. Mukherjee PK, Mohamed S, Chandra J, et al. Alcohol dehydrogenase restricts the ability of the pathogen *Candida albicans* to form a biofilm on catheter surfaces through an ethanol-based mechanism. *Infect Immun*. 2006;74:3804–3816.
445. Guenu S, Heng AE, Charbonné F, et al. Mass spectrometry and scanning electron microscopy study of silicone tunneled dialysis catheter integrity after an exposure of 15 days to 60% ethanol solution. *Rapid Commun Mass Spectrom*. 2007;21:229–236.
446. Cober MP, Kovacevich DS, Teitelbaum DH. Ethanol-lock therapy for the prevention of central venous access device infections in pediatric patients with intestinal failure. *JPEN J Parenter Enteral Nutr*. 2011;35:67–73.
447. Jones BA, Hull MA, Richardson DS, et al. Efficacy of ethanol locks in reducing central venous catheter infections in pediatric patients with intestinal failure. *J Pediatr Surg*. 2010;45:1287–1293.
448. Mouw E, Chessman K, Lesher A, et al. Use of an ethanol lock to prevent catheter-related infections in children with short bowel syndrome. *J Pediatr Surg*. 2008;43:1025–1029.
449. Wales PW, Kosar C, Carricato M, et al. Ethanol lock therapy to reduce the incidence of catheter-related bloodstream infections in home parenteral nutrition patients with intestinal failure: preliminary experience. *J Pediatr Surg*. 2011;46:951–956.
450. Opilla MT, Kirby DF, Edmond MB. Use of ethanol lock therapy to reduce the incidence of catheter-related bloodstream infections in home parenteral nutrition patients. *JPEN J Parenter Enteral Nutr*. 2007;31:302–305.
451. Oliveira C, Nasr A, Brindle M, et al. Ethanol locks to prevent catheter-related bloodstream infections in parenteral nutrition: a meta-analysis. *Pediatrics*. 2012;129:318–329.
452. Mermel LA, Allon M, Bouza E, et al. Clinical practice guidelines for the diagnosis and management of intravascular catheter-related infection: 2009 update by the Infectious Diseases Society of America. *Clin Infect Dis*. 2009;49:1–45.
453. Dobson R, McGuckin C, Walker G, et al. Cycled enteral antibiotics reduce sepsis rates in paediatric patients on long-term parenteral nutrition for intestinal failure. *Aliment Pharmacol Ther*. 2011;34:1005–1011.
454. Jacobs BR. Central venous catheter occlusion and thrombosis. *Crit Care Clin*. 2003;19:489–514.
455. Hardy G, Ball P. Avoiding catheter complications with lipid containing parenteral

nutrition. *Br J Int Care*. 2006;16:64–67.
456. Shulman RJ, Reed T, Pitre D, et al. Use of hydrochloric acid to clear obstructed central venous catheters. *JPEN J ParenterEnteral Nutr*. 1988;12:509–510.
457. Duffy LF, Kerzner B, Gebus V, et al. Treatment of central venous catheter occlusions with hydrochloric acid. *J Pediatr*. 1989;114:1002–1004.
458. Schmidt-Sommerfeld E, Snyder G, Rotti TM, et al. Catheter-related complications in 35 children and adolescents with gastrointestinal disease on home PN. *JPEN J Parenter Enteral Nutr*. 1990;14:148–151.
459. Borrow M, Crowley JG. Evaluation of central venous catheter thrombogenicity. *Acta Anaesth Scand Suppl*. 1985;81:59–64.
460. Jacobs BR, Haygood M, Hingl J. Recombinant tissue plasminogen activator in the treatment of central venous catheter occlusion in children. *J Pediatr*. 2001;139:593–596.
461. Kerner JA Jr, Garcia-Careaga MG, Fisher AA, et al. Treatment of catheter occlusion in pediatric patients. *JPEN J Parenter Enteral Nutr*. 2006;30 (1 Supp l):S73–81.
462. Deitcher SR, Fraschini G, Himmelfarb J, et al. Dose-ranging trial with a recombinant urokinase for occluded central venous catheters in oncology patients. *J Vascular Interventional Radiol*. 2004;15:575–579.
463. Fink JM, Capozzi D, Shermock KM, et al. Alteplase for central catheter clearance: doses 1 mg/mL versus 2 mg/2 mL. *Ann Pharmacotherapy*. 2004;38:351–352.
464. van Miert C, Hill R, Jones L. Interventions for restoring patency of occluded central venous catheter lumens. *Cochrane Database Syst Rev*. 2012;4:CD007119.
465. Monagle P, Chan A, Massicotte P, et al. Antithrombotic therapy in children. *Chest*. 2004;126:645S–687S.
466. Smith S, Dawson S, Hennessey R, et al. Maintenance of the patency of indwelling central venous catheters: is heparin necessary? *Am J Pediatr Hemat Oncol*. 1991;13:141.
467. Monagle P, Chalmers E, Chan A, et al. Antithrombotic therapy in neonates and children: American College of Chest Physicians Evidence-Based Clinical Practice Guidelines (8th Edition). *Chest*. 2008;133:887S–986S.
468. Revel-Vilk S, Ergaz Z. Diagnosis and management of central-line-associated thrombosis in newborns and infants. *Semin Fetal Neonatal Med*. 2011;16:340–344.
469. Randolph AG, Cook DJ, Gonzales CA, et al. Benefit of heparin in peripheral venous and arterial catheters: systematic review and meta-analysis of randomised controlled trials. *Br Med J*. 1998;316:969–975.
470. Goode CJ, Titler M, Rakel B, et al. A meta-analysis of effects of heparin flush and saline flush: quality and cost implications. *Nurs Res*. 1991;40:324–330.
471. Peterson FY, Kirchoff KT. Analysis of the research about heparinized versus non-heparinized intravascular lines. *Heart Lung*. 1991;20:631–640.
472. Bertolino G, Pitassi A, Tinelli C, et al. Intermittent flushing with heparin versus saline for maintenance of peripheral intravenous catheters in a medical department: a pragmatic cluster-randomized controlled study. *World Views Evid Based Nurs*. 2012;9:221–226.
473. Vegting IL, Tabbers MM, Benninga MA, et al. Prophylactic anticoagulation decreases catheter-related thrombosis and occlusion in children with home parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2012;36:456–462.
474. Jacobs BR, Schilling S, Doellman D, et al. Central venous catheter occlusion: a prospective, controlled trial examining the impact of a positive-pressure valve device. *JPEN J Parenter Enteral Nutr*. 2004;28:113–118.

475. Scribner BH, Cole JJ, Christopher TG, et al. Long-term total parenteral nutrition. The concept of an artificial gut. *JAMA*. 1970;212:457–463.
476. Byrne WJ, Halpin TC, Asch MJ, et al. Home total parenteral nutrition: an alternative approach to the management of children with severe chronic small bowel disease. *J Pediatr Surg*. 1977;12:359–366.
477. Just B, Messing B, Darmaun D, et al. Comparison of substrate utilization by indirect calorimetry during cyclic and continuous total parenteral nutrition. *Am J Clin Nutr*. 1990;51:107–111.
478. Pullicino E, Goldberg GR, Elia M. Energy expenditure and substrate metabolism measured by 24 h whole-body calorimetry in patients receiving cyclic and continuous total parenteral nutrition. *Clin Sci (Lond)*. 1991;80:571–582.
479. Lerebours E, Rimbert A, Hecketsweiler B, et al. Comparison of the effects of continuous and cyclic nocturnal parenteral nutrition on energy expenditure and protein metabolism. *JPEN J Parenter Enteral Nutr*. 1988;12:360–364.
480. Matuchansky C, Fabre J, Guillard O, et al. Effects of cyclic (nocturnal) total parenteral nutrition and continuous enteral nutrition on circadian rhythms of blood lipids, lipoproteins and apolipoproteins in humans. *Am J Clin Nutr*. 1985;41:727–734.
481. Yokoyama S, Hirakawa H, Soeda J, et al. Twenty-four-hour profile of growth hormone in cyclic nocturnal total parenteral nutrition. *Pediatrics*. 1997;100:973–976.
482. Hwang TL, Lue MC, Chen LL. Early use of cyclic TPN prevents further deterioration of liver functions for the TPN patients with impaired liver function. *Hepatogastroenterology*. 2000;47:1347–1350.
483. Collier S, Crough J, Hendricks K, et al. Use of cyclic parenteral nutrition in infants less than 6 months of age. *Nutr Clin Pract*. 1994;9:65–68.
484. Meehan JJ, Georgeson KE. Prevention of liver failure in parenteral nutrition-dependent children with short bowel syndrome. *J Pediatr Surg*. 1997;132:473–475.
485. Salvador A, Janeczko M, Porat R, et al. Randomized controlled trial of early parenteral nutrition cycling to prevent cholestasis in very low birth weight infants. *J Pediatr*. 2012;161:229–233.
486. Hwang TL, Lue MC, Chen LL. Early use of cyclic TPN prevents further deterioration of liver functions for the TPN patients with impaired liver function. *Hepatogastroenterology*. 2000;47:1347–1350.
487. Werlin SL, Wyatt D, Camitta B. Effect of abrupt discontinuation of high glucose infusion rates during parenteral nutrition. *J Pediatr*. 1994;124:441–444.
488. Bendorf K, Friesen CA, Roberts CC. Glucose response to discontinuation of parenteral nutrition in patients less than 3 years of age. *JPEN J Parenter Enteral Nutr*. 1996;20:120–122.
489. Boncompain-Gérard M, Robert D, Fouque D, et al. Renal function and urinary excretion of electrolytes in patients receiving cyclic parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2000;24:234–239.
490. Wood RJ, Bengoa JM, Sitrin MD, et al. Calciuretic effect of cyclic versus continuous total parenteral nutrition. *Am J Clin Nutr*. 1985;41:614–619.
491. Longhurst C, Naumovski L, Garcia-Careaga M, et al. A practical guideline for calculating parenteral nutrition cycles. *Nutr Clin Pract*. 2003;18:517–520.
492. Goulet O, Ruemmele F, Lacaille F. Irreversibile intestinal failure. *J Pediatr Gastroenterol Nutr*. 2004;38:250–269.
493. Beath SV, Gowen H, Puntis JW. Trends in paediatric home parenteral nutrition and implications for service development. *Clin Nutr*. 2011;30:499–502.

494. Colomb V, Dabbas-Tyan M, Taupin P, et al. Long-term outcome of children receiving home parenteral nutrition: a 20-year single-center experience in 302 patients. *J Pediatr Gastroenterol Nutr*. 2007;44: 347–353.
495. Pironi L, Joly F, Forbes A, et al. Long-term follow-up of patients on home parenteral nutrition in Europe: implications for intestinal transplantation. *Gut*. 2011;60:17–25.
496. Pironi L, Goulet O, Buchman A, et al. Home Artificial Nutrition and Chronic Intestinal Failure Working Group of ESPEN. Outcome on home parenteral nutrition for benign intestinal failure: a review of the literature and benchmarking with the European prospective survey of ESPEN. *Clin Nutr*. 2012;31:831–845.
497. Hsieh MH, Pai W, Tseng HI, et al. Parenteral nutrition-associated cholestasis in premature babies: risk factors and predictors. *Pediatr Neonatol*. 2009;50:202–207.
498. Christensen RD, Henry E, Wiedmeier SE, et al. Identifying patients, on the first day of life, at high-risk of developing parenteral nutrition-associated liver disease. *J Perinatol*. 2007;27:284–290.
499. Robinson DT, Ehrenkranz RA. Parenteral nutrition-associated cholestasis in small for gestational age infants. *J Pediatr*. 2008;152:59–62.
500. Lloyd DA, Zabron AA, Gabe SM. Chronic biochemical cholestasis in patients receiving home parenteral nutrition: prevalence and predisposing factors. *Aliment Pharmacol Ther*. 2008;27:552–560.
501. Goulet O, Joly F, Corriol O, et al. Some new insights in intestinal failure-associated liver disease. *Curr Opin Organ Transplant*. 2009;14:256–261.
502. Willis TC, Carter BA, Rogers SP, et al. High rates of mortality and morbidity occur in infants with parenteral nutrition-associated cholestasis. *J Parenter Enteral Nutr*. 2010;34:32–37.
503. Gupte GL, Beath SV, Protheroe S, et al. Improved outcome of referrals for intestinal transplantation in the UK. *Arch Dis Child*. 2007;92:147–152.
504. Buchman AL, Misra S, Moukarzel A, et al. Catheter thrombosis and superior/inferior vena cava syndrome are rare complications of long term parenteral nutrition. *Clin Nutr*. 1994;13:356–360.
505. Dollery CM, Sullivan ID, Bauraind O, et al. Thrombosis and embolism in long-term central venous access for parenteral nutrition. *Lancet*. 1994;344:1043–1045.
506. Pironi L, Forbes A, Joly F, et al. Survival of patients identified as candidates for intestinal transplantation: a 3-year prospective follow-up. *Gastroenterology*. 2008;135:61–71.
507. Guarino A, De Marco G, Italian National Network for Pediatric Intestinal Failure. Natural history of intestinal failure, investigated through a national network-based approach. *J Pediatr Gastroenterol Nutr*. 2003;37:136–141.
508. Diamanti A, Basso MS, Castro M, et al. Irreversible intestinal failure: prevalence and prognostic factors. *J Pediatr Gastroenterol Nutr*. 2008;47:450–457.
509. Goday PG. Short bowel syndrome: how short is too short? *Clin Perinatol*. 2009;36:101–110.
510. Goulet O, Baglin-Gobet S, Talbotec C, et al. Outcome and long-term growth after extensive small bowel resection in the neonatal period: a survey of 87 children. *Eur J Pediatr Surg*. 2005;15:95–101.
511. Quirós-Tejeira RE, Ament ME, Reyen L, et al. Long-term parenteral nutritional support and intestinal adaptation in children with short bowel syndrome: a 25-year experience. *J Pediatr*. 2004;145:157–163.
512. Spencer AU, Kovacevich D, McKinney-Barnett M, et al. Pediatric short-bowel syndrome: the cost of comprehensive care. *Am J Clin Nutr*. 2008;88:1552–1559.
513. Faure C, Goulet O, Ategbo S, et al. Chronic intestinal pseudoobstruction

syndrome. Clinical analysis, outcome and prognosis in 105 children. *Dig Dis Sci*. 1999;44:953–959.
514. Bueno J, Ohwada S, Kocoshis S, et al. Factors impacting the survival of children with intestinal failure referred for intestinal transplantation. *J Pediatr Surg*. 1999;34:27–32.
515. Mousa H, Hyman P, Cocjin J, et al. Long-term outcome of congenital intestinal pseudo-obstruction. *Dig Dis Sci*. 2002;47:2298–2305.
516. Pini Prato A, Rossi V, Avanzini S, et al. Hirschsprung's disease: what about mortality? *Pediatr Surg Int*. 2011;27:473–478.
517. Kimura O, Ono S, Furukawa T, et al. Management strategies for infants with total intestinal aganglionosis. *J Pediatr Surg*. 2009;44:1564–1567.
518. Ksiazyk J, Lyszkowska M, Kierkus J, et al. Home parenteral nutrition in children: the Polish experience. *J Pediatr Gastroenterol Nutr*. 1999;28:152–156.
519. Candusso M, FaragunaD, Di LeoG, et al. Home parenteral nutrition in children: outcome and quality of life. *Clin Nutr*. 2001;20(Suppl 2):7–10.
520. A.S.P.E.N. Board of Directors and the Clinical Guidelines Task Force. Guidelines for the use of parenteral and enteral nutrition in adult and pediatric patients. *JPEN J Parenter Enteral Nutr*. 2002;26(1 supp l): 1SA–138SA.
521. Szeszycki E, Cruse W, Strup M. Evaluation and monitoring of pediatric patients receiving specialized nutrition support. In: Corkins MR, ed. *The A.S.P.E.N. Pediatric Nutrition Support Core Curriculum*. Silver Spring, MD: American Society for Parenteral and Enteral Nutrition; 2010: pp. 460–476.
522. Norman JL, Crill CM. Optimizing the transition to home parenteral nutrition in pediatric patients. *Nutr Clin Pract*. 2011;26:273–285.
523. Engstrom I, Bjornestam B, Finkel Y. Psychological distress associated with home parenteral nutrition in Swedish children, adolescents, and their parents: preliminary results. *J Pediatr Gastroenterol Nutr*. 2003;37:246–250.
524. Gottrand F, Staszewski P, Colomb V, et al. Satisfaction in different life domains in children receiving home parenteral nutrition and their families. *J Pediatr*. 2005;146:793–797.
525. Emedo MJ, Godfrey EI, Hill SM. A qualitative study of the quality of life of children receiving intravenous nutrition at home. *J Pediatr Gastroenterol Nutr*. 2010;50:431–440.
526. Puntis JW. The economics of home parenteral nutrition. *Nutrition*. 1998;14:809–812.
527. Reddy P, MaloneM. Cost and outcome analysis of home parenteral and enteral nutrition. *JPEN J Parent Enter Nutr*. 1998;22:302–310.
528. Howard L. Home parenteral nutrition: survival, cost, and quality of life. *Gastroenterology*. 2006;130(2 Suppl 1):S52–S59.
529. Melville CAS, Bisset WM, Long S, et al. Counting the cost: hospital versus home central venous catheter survival. *J Hosp Infect*. 1997;35:197–205.
530. Maki DG, Ringer M. Risk factors for infusion-related phlebitis with small peripheral venous catheters: a randomized controlled trial. *Ann Int Med*. 1991;114:845–853.
531. Tager IB, Ginsberg MB, Ellis SE, et al. An epidemiologic study of the risks associated with peripheral intravenous catheters. *Am J Epidemiol*. 1983;118:839–851.
532. Kane KF, Cologiovanni L, McKiernan J, et al. High osmolality feedings do not increase the incidence of thrombophlebitis during peripheral IV nutrition. *JPEN J Parenter Enteral Nutr*. 1996;20:194–197.
533. Everitt NJ, McMahon MJ. Peripheral intravenous nutrition. *Nutrition*.

1994;10:49–57.
534. Pineault M, Chessex P, Piedboeuf B, Bisaillon S. Beneficial effect of coinfusing a lipid emulsion on venous patency. *J Parenter Enteral Nutr*. 1989;13:637–640.
535. Tighe MJ, Wong C, Martin IG, et al. Do heparin, hydro-cortisone, and glyceryl trinitrate influence thrombophlebitis during full intravenous nutrition via a peripheral vein? *J Parenter Enteral Nutr*. 1995;19:507–509.
536. Alpan G, Eyal F, Springer C, et al. Heparinization of alimentation solutions administered through peripheral veins in premature infants: a controlled study. *Pediatrics*. 1984;74:375–378.
537. Khawaja H T, Williams JD, Weaver PC. Transdermal glyceryl trinitrate to allow peripheral total parenteral nutrition: a double-blind placebo controlled feasibility study. *J R Soc Med*. 1991;84:69–71.
538. Payne-James JJ, Bray MJ, Kapadia S, et al. Topical nonsteroidal anti-inflammatory gel for the prevention of peripheral vein thrombophlebitis. *Anaesthesia*. 1992;47:324–326.
539. Shimandle RB, Johnson D, Baker M, et al. Safety of peripheral intravenous catheters in children. *Infect Control Hosp Epidemiol*. 1999;20:736–740.
540. Webster J, Osborne S, Rickard C, et al. Clinically-indicated replacement versus routine replacement of peripheral venous catheters. *Cochrane Database Syst Rev*. 2010;CD007798.
541. Rickard CM, Webster J, Wallis MC, et al. Routine versus clinically indicated replacement of peripheral intravenous catheters: a randomised controlled equivalence trial. *Lancet*. 2012;380:1066–1074.
542. Lehmann CU, Miller MR. Standardization and the practice of medicine. *J Perinatol*. 2004;24:135–136.
543. Dice JE, Burckart GJ, Woo JT, et al. Standardized versus pharmacist-monitored individualized parenteral nutrition in low-birth-weight infants. *Am J Hosp Pharm*. 1981;38:1487–1489.
544. Smolkin T, Diab G, Shohat I, et al. Standardized versus individualized parenteral nutrition in very low birth weight infants: a comparative study. *Neonatology*. 2010;98:170–178.
545. Mutchie KD, Smith KA, MacKay MW, et al. Pharmacist monitoring of parenteral nutrition: clinical and cost effectiveness. *Am J Hosp Pharm*. 1979;36:785–787.
546. Bethune K. The use of standard parenteral nutrition solutions in pediatrics: a UK perspective. *Nutrition*. 2001;17:357–359.
547. Porcelli P. A survey of neonatal parenteral nutrition design practices in North Carolina. *J Perinatol*. 2004;24:137–142.
548. Driscoll DF. Clinical delivery of nutritional therapy: automated compounders and patient-specific feeding. *Nutrition*. 2006;12:461–462.
549. Driscoll DF. Physicochemical assessment of total nutrient admixture stability and safety: quantifying the risk. *Nutrition*. 1997;13:166–167.
550. Driscoll DF. Compounding TPN admixtures: then and now. *JPEN J Parenter Enteral Nutr*. 2003;27:433–438.
551. Driscoll DF. Stability and compatibility assessment techniques for total parenteral nutrition admixtures: setting the bar according to pharmacopeial standards. *Curr Opin Clin Nutr Metab Care*. 2005;8:297–303.
552. Ochoa-Sangrador C, Brezmes-Valdivieso MF, Gil-Valino C. Pediatric parenteral nutrition mixtures design program: validity and stability study. *Comput Biomed Res*. 1995;28:271–281.
553. Migliori C, Verdura C, Belloni C. Experience with the use of a computerized data processing program on solutions for parenteral nutrition of neonates. *Minerva*

Pediatr. 1997;49:229–234.
554. Puangco MA, Nguyen HL, Sheridan MJ. Computerized PN ordering optimizes timely nutrition therapy in a neonatal intensive care unit. *J Am Diet Assoc*. 1997;97:258–261.
555. Cade A, Thorp H, Puntis JW. Does the computer improve the nutritional support of the newborn? *Clin Nutr*. 1997;16:19–23.
556. Krohn K, Babl J, Reiter K, et al. Parenteral nutrition with standard solutions in paediatric intensive care patients. *Clin Nutr*. 2005;24:274–280.
557. Yeung MY, Smyth JP, Maheshwari R, et al. Evaluation of standardized versus individualized total parenteral nutrition regime for neonates less than 33 weeks gestation. *J Pediatr Child Health*. 2003;39:613–617.
558. Lenclen R, Crauste-Manciet S, Narcy P, et al. Assessment of implementation of a standardized parenteral formulation for early nutritional support of very preterm infants. *Eur J Pediatr*. 2006;165:512–518.
559. Richardson DK, Zupancic JA, Escobar GJ, et al. A critical review of cost reduction in neonatal intensive care. II. Strategies for reduction. *J Perinatol*. 2001;21:121–127.
560. Riskin A, Shiff Y, Shamir R. Parenteral nutrition in neonatology-to standardize or individualize? *Isr Med Assoc J*. 2006;8:641–645.
561. Rigo J, Marlowe ML, Bonnot D, et al. Benefits of a new pediatric triple-chamber bag for parenteral nutrition in preterm infants. *J Pediatr Gastroenterol Nutr*. 2012;54:210–217.
562. Institute for Safe Medication Practices. List of high-alert medications, 2008. http://ismp.org/Tools/highalertmedications.pdf. Accessed September 12, 2012.
563. Boitano M, Bojak S, McCloskey S, et al. Improving the safety and effectiveness of parenteral nutrition: results of a quality improvement collaboration. *Nutr Clin Pract*. 2010;25:663–671.
564. Flynn EA, Pearson RE, Barker KN. Observational study of accuracy in compounding i.v. admixtures at five hospitals. *Am J Health Syst Pharm*. 1997;54:904–912.
565. Long AL, Horvath MM, Cozart H, et al. Tailoring adverse drug event surveillance to the pediatric inpatient. *Qual Saf Health Care*. 2010;19:e40.
566. Mirtallo J, Canada T, Johnson D, et al. Safe practices for parenteral nutrition. *JPEN J Parenter Enteral Nutr*. 2004;28:S39–S70.
567. Seres D, Sacks GS, Pedersen CA, et al. Parenteral nutrition safe practices: results of the 2003 American Society for Parenteral and Enteral Nutrition survey. *JPEN J Parenter Enteral Nutr*. 2006;30:259–265.
568. Kochevar M, Guenter P, Holcombe B, et al. A.S.P.E.N. statement on parenteral nutrition standardization. *JPEN J Parenter Enteral Nutr*. 2007;31:441–448.
569. Colomb V, Marlowe ML, Bonnot D, et al. Practical use of a new three-chamber bag for parenteral nutrition in pediatric patients. *e-SPEN Journal* 2012;7:e93–e99.

Nutrition in the Athlete

69

Kathryn E. Ackerman, MD, MPH and Madhusmita Misra, MD, MPH

INTRODUCTION

Approximately 35 million children in the United States ages 5 to 18 years are involved in organized sports, including 66% of boys and 52% of girls in this age range (http://www.statisticbrain.com/youth-sports-statistics/). In Europe, depending on the country, sports participation rates among children reach as high as 98% for at least occasional involvement in sports activities.[1] In general, physically active children and adolescents tend to have healthier nutritional habits than their nonathletic peers.[2] However, many are unaware of the effects of dietary choices on performance, injury prevention or recovery, much less the effects on growth, maturation, and overall health. Some attempt excessive caloric supplement and protein ingestion in an effort to bulk up for sports. Others take their dietary habits to the other extreme, restricting caloric content and suffering nutritional deficiencies with a goal of achieving a thin, lean physique. Although many nutritional issues in the young athlete are similar to those of the adult athlete, there are several physiologic and metabolic characteristics that distinguish child and adolescent athletes from adults.[3] Therefore, specific nutritional issues need to be addressed.

This chapter will review the nutritional needs for normal growth and development of children and adolescents and the special considerations for athletic youth. It will discuss macro- and micronutrient requirements, vitamin and mineral recommendations, special dietary restrictions (such as vegetarianism), in addition to hydration, supplementation, and appropriate timing of meals to optimize health and performance. It will also review some of the negative sequelae of energy imbalance and the importance of fueling for sport.

GROWTH

Infancy and early childhood are typically times of rapid growth. Later childhood is associated with a slowing, but consistent gain in stature and weight, with adolescence being a time again of rapid growth. A general rule is that a child grows 25 cm in the first year of life, half that in the second year, and then 5–6 cm until puberty.[4] Girls average a peak height velocity of 9 cm/year around age 12, with a total height gain of 25 cm during the pubertal growth period.[4,5] Boys have a later but greater peak height velocity of 10.3 cm/year around age 14, with a total height gain of 28 cm during the pubertal growth period.[4,5] Approximately 90% of peak bone mass is attained by the second decade in boys and girls, with 26% of adult bone mineral content acquired during the 2 years around peak height velocity.[6–8]

School-aged children and adolescents require adequate caloric intake to ensure appropriate growth and maturation, and these caloric needs are generally higher in active children versus their sedentary counterparts. Although it is difficult to provide specific caloric recommendations because of large individual variability, on average, during growth spurts, adolescents should consume an additional 500 kcal/d above their usual intake.[9] Growth and maturation are influenced by numerous factors that act independently or synergistically to impact any individual's inherent potential for these parameters. Worldwide, the most common cause of growth retardation is poverty-related malnutrition. In the United States, in upper-middle and upper-class teens, the most common cause for poor growth and delayed pubertal development is self-induced energy restriction.[4]

Participation in sports activities that do not require weight control does not delay puberty or alter growth. In fact, children involved in sports not associated with weight control are normal if not advanced in their rates of growth and maturation. It is quite possible that because strength and power advantages associated with earlier development are beneficial for such sports, the earlier maturing athletes are self-selected for participation.[10,11]

However, there are activities associated with an increased risk for prolonged inadequate energy intake (EI) relative to expenditure. These include wrestling, gymnastics, distance running, and dance.[12] A sustained energy-deficit state may result in short stature, delayed puberty, menstrual irregularity or amenorrhea, poor bone health, and increased rates of injury.[13] Roemmich et al. found that statural growth and advancement of skeletal maturation of wrestlers was similar to controls both during and

after wrestling season. However, increase in fat-free mass and other soft tissues slowed during wrestling season, followed by catch-up growth postseason when caloric intake increased.[14,15]

A muscular and short frame provides competitive advantage in gymnastics. Performance often declines after the onset of puberty secondary to increases in height and fat mass, and development of secondary sexual characteristics. Thus, gymnasts may try to modify activity levels and caloric consumption to maintain a lean physique, maximize their strength to weight ratio, and potentially to delay puberty and growth.[12] Studies of gymnasts have found reduced insulin-like growth factor-I (IGF-I) concentrations, relatively shorter lower limbs in both males and females, and delayed age of menarche in female gymnasts.[16–20] Delay in growth and sexual maturation is most notable in gymnasts, dancers, and long-distance runners 10. However, there are few longitudinal studies of such athletes, and it is also important to consider the role of genetic predisposition, with short stature often being familial, and menarchal age correlating positively between mothers and daughters.[10,21]

Many of the hormones most strongly linked to pubertal development [such as growth hormone (GH), IGF-I, and sex steroids] are also linked to energy metabolism during exercise.[22] The antiquated theory that elevations in pubertal hormones (GH and testosterone) during exercise cause increased growth and development during adolescence is yet to be confirmed through controlled, longitudinal studies. Transient increases in hormones have been documented in exercising adolescents, but their long-term effects have not been determined.[22–25]

MACRONUTRIENTS

Carbohydrates

The daily recommended intake for carbohydrates in children is based on the amount of carbohydrate needed to provide sufficient glucose for brain function and does not take into account the amount needed for muscle glycogen repletion.[1,3] In adults, carbohydrates are the dominant fuel source for maintaining prolonged or high-intensity exercise. As exercise intensity decreases, a greater proportion of energy is derived from oxidation of free fatty acids, particularly from muscle triglycerides. In addition, with more aerobic training, the energy derived from fat increases, while that from carbohydrates decreases,[26] as seen in better trained athletes. Of note, children are more dependent on fat oxidative pathways

than adults. This may be due to higher intramuscular triglyceride levels, higher free fatty acid turnover during exercise, or an underdeveloped glycolytic system in children compared with adults.[27] Fat may play as important a role as carbohydrates for child athletes, whereas adolescent athletes behave more like adults.[28] In a study of 13- to 15-year-old teens, there was no significant difference in muscle glycolytic enzymes compared to adults.[29]

It is well-established that puberty is associated with a period of insulin resistance. Although the exact cause in unclear, increases in GH and sex steroids are thought to oppose the effects of insulin in skeletal muscle, adipose tissue, and liver, leading to a decrease in insulin sensitivity of about 30%.[3,22]

Parts of the human body that almost exclusively require glucose for metabolic functions (such as the brain, red blood cells, and a few other tissues) have a daily need in adolescents and adults of about 180 g of carbohydrates/day. Gluconeogenesis from noncarbohydrates (e.g., fat, protein, lactate) generates about 130 g/day. Most child athletes get at least 200–500 g/day of carbohydrate.[1,3] However, female athletes, particularly endurance athletes, are less likely than their male counterparts to consume recommended amounts of carbohydrate, often due to restriction of total EI from a desire to achieve or maintain a lower body weight and/or body fat level.[30]

The Institute of Medicine's Dietary Reference Intakes (DRIs) are the recommended dietary allowances for food types. The DRI of carbohydrates for children and adults to maintain brain function is 130 g/day, with a suggestion that 45%–65% of total calories be derived from carbohydrates (http://fnic.nal.usda.gov/dietary-guidance/dietary-reference-intakes/dri-tables). It is important to remember that during heavy exercise, adolescents may utilize up to 1–1.5 g/kg/h of total carbohydrates.[31]

Fat

Fat is a necessary component of any diet, providing energy and essential elements of cell membranes, in addition to facilitating the absorption of fat-soluble vitamins such as vitamins A, D, E, and K.[32] Children and early and mid-pubertal adolescents oxidize relatively more fat than carbohydrate at a given relative exercise intensity compared to late pubertal adolescents and adults.[27,33] This is based on studies using the respiratory exchange ratio during and after bouts of exercise, in addition to blood byproducts of fat oxidation (such as plasma-free glycerol and free fatty acids) and of

carbohydrate metabolism (such as lactate). As mentioned above, the phenomenon appears to be inversely correlated with maturation, with an adult-like metabolic pattern observed by the end of puberty.[27,29,33]

Yet even with this increased reliance on fat as a fuel source, there is no evidence that young endurance athletes benefit from higher fat content in their diets.[33,34] In fact, ingestion of high-fat food prior to exercise may decrease GH secretion during exercise from an increase in free fatty acids. If GH secretion during exercise is important for muscle adaptation and hypertrophy, high fat ingestion could have negative consequences.[33]

The American Heart Association recommends that dietary fat constitute 25%–35% of the daily calories of individuals ages 4–18 years.[35] There are no Institute of Medicine DRIs for fat other than recommending amounts of linoleic (10, 12, and 16 g/d for ages 4–8 years, 9–13 years, and 14–18 years, respectively) and α-linolenic acid (0.9, 1.2, and 1.6 g/d for ages 4–8 years, 9–13 years, and 14–18 years, respectively) (http://fnic.nal.usda.gov/dietary-guidance/dietary-reference-intakes/dri-tables).

Protein

Children and adolescents must maintain a positive nitrogen balance for growth and development. Although it is important to ingest enough protein, particularly essential amino acids, it is just as important to ingest enough calories, to spare protein from being used as a substrate for energy. Amino acids contribute 5%–15% of the fuel burned during exercise. There have been few studies to determine the optimal protein intake in child/adolescent athletes. In a recent study by Aerenhouts et al.,[36] 60 adolescent sprint athletes were followed over 2 years. Body height, weight, and skeletal muscle mass increased in the participants throughout the study, and it was concluded that about 1.5 g/kg/d of protein (1.46 g/kg/d for girls and 1.35 g/kg/d for boys) was sufficient to maintain a positive nitrogen balance, even during periods of peak growth.[36]

MICRONUTRIENTS

Micronutrients are required in small amounts for normal bodily functions including hemoglobin synthesis, bone health, immune defense, and protection from oxidative damage. They include vitamins and minerals such as vitamins A, C, D, E, K, the B vitamins, calcium, iron, zinc, magnesium, β-carotene, and selenium. A few of those of most concern to

young athletes will be discussed below.

Antioxidants

Antioxidant vitamins C and E, β-carotene, and selenium protect cell membranes from oxidative damage.[26] Because of increased oxygen consumption with exercise, it has been hypothesized that long-term, intense exercise may increase oxidative stress on the body, resulting in lipid peroxidation of membranes.[26] However, studies in well-trained adult and adolescent athletes have demonstrated enhanced antioxidant systems.[37,38] Whether or not adolescent athletes truly benefit from increased antioxidant consumption remains to be determined.

B Vitamins

The B vitamins, namely thiamine (vitamin B_1), riboflavin (vitamin B_2), niacin (vitamin B_3), pantothenic acid (vitamin B_5), pyridoxine (vitamin B_6), and biotin (vitamin B_7) are important for energy production during exercise, while folate (vitamin B_9) and vitamin B_{12} are important for red blood cell production, protein synthesis, and tissue repair and maintenance, including of the central nervous system.[26] Research in adults suggests that exercise may increase requirements for riboflavin and vitamin B_6, but data for folate and vitamin B_{12} are limited,[39] and there are no confirmatory studies in otherwise healthy child/adolescent athletes. Any increased needs in young athletes can likely be met with appropriate EI. However, those on restrictive diets or with low EI may potentially suffer from severe B_{12} and folate deficiency, resulting in anemia, with subsequent endurance performance impairment.[26,40] Young amenorrheic athletes with the female athlete triad have been shown to benefit from folate supplementation, with improved endothelial vascular function.[41]

Vitamin D

Vitamin D is critical for absorption of calcium, regulation of serum calcium, phosphorus, and parathyroid hormone levels, and promotion of bone health. Vitamin D is also important for nervous system and skeletal muscle function.[42] As little as 10–15 minutes of direct sunlight can generate 10,000 to 20,000 IU of vitamin D. However, many factors can significantly affect vitamin D synthesis, including latitude, amount of skin exposed, and skin pigmentation.[43] Children with darker skin pigmentation

require 5–10 times more sunlight exposure to achieve the same serum concentrations of 25-hydroxy (25-OH) vitamin D as those with lighter skin.[43,44] Vitamin D intake reduces the risk of stress fractures, infections, inflammation, and impaired muscle function[45]; however, adolescent athletes often consume less than the recommended amount of vitamin D.[46–48] In 2008, the American Academy of Pediatrics increased their recommendations for daily intake of vitamin D from 200 to 400 IU for adolescents,[49] and the Institute of Medicine has recommended a daily intake of 600 IU of vitamin D in this age group. Most bone experts consider 25-OH vitamin D concentrations <20 ng/mL (50 nmol/L) to represent vitamin D "deficiency," with levels of 21–29 ng/mL (52–72 nmol/L) considered "insufficient," and levels >30 ng/mL (>70 nmol/L) "sufficient."[45,48] Some believe that current recommendations for daily vitamin D intake are still not quite adequate and suggest that up to 1000 IU daily intake may be necessary to maintain 25-OH vitamin D levels in an optimal range.[50] Vitamin D levels should be checked in adolescent athletes, particularly those living in northern latitudes, training primarily indoors (such as gymnasts and figure skaters), and those who do not consume foods fortified with vitamin D.[26,51]

Calcium

The Institute of Medicine's recommendations for daily calcium intake in children ages 9–18 is 1300 mg daily (http://fnic.nal.usda.gov/dietary-guidance/dietary-reference-intakes/dri-tables). The maximal absorption of elemental calcium in a single dose is approximately 500 mg; therefore, calcium intake needs to be divided into multiple doses over the course of the day. Prospective, placebo-controlled calcium supplementation trials (1000 mg calcium/d) in otherwise healthy adolescents with inadequate baseline calcium intake have reported increases in total body bone mineral density (BMD).[52,53] Although dairy products are obvious sources of calcium, many foods including juices, cereals, and cereal bars contain added calcium with the percent recommended daily allowance (%RDA) listed on foods being based on 1000 mg daily. Of note, in amenorrheic athletes, calcium intake per recommendations by itself is not sufficient to optimize BMD.[52]

Iron

During rapid growth and maturation, iron stores are influenced by changes in muscle mass and nutrition. Exercise affects body composition and

increases iron loss in urine, stool, sweat, and by hemolysis. In addition, young female athletes may have increased iron losses with the onset of menses.[3] Overt iron deficiency anemia is rare, but depleted iron stores (low serum ferritin) without decreased hemoglobin is prevalent in 25%–50% of male and female adolescent athletes, particularly endurance athletes.[3,54,55] Animal studies have demonstrated impaired endurance with iron deficiency even without associated anemia. Human studies have shown that iron supplementation maintains the ventilatory threshold and improves aerobic capacity and energy efficiency in iron deficient, nonanemic young athletes.[3,56]

Food sources of iron include enriched grain products, meat, fish, poultry, vegetables, legumes, nuts, and soy. However, adolescents need to consume up to 3000 kcal in order to achieve the RDA for iron of 18 mg/d[3] (http://fnic.nal.usda.gov/dietary-guidance/dietary-reference-intakes/dri-tables). Iron occurs in food in two separate forms: heme and nonheme. Heme iron comprises about 40% of the iron in meat, fish, and poultry. It is more efficiently absorbed than nonheme iron, which comprises the remaining iron in meat, fish, and poultry, as well as all iron in other foods.[57] Nonheme iron absorption is enhanced by tissue protein factor (in meat, fish, and poultry), vitamin C, alcohol, and potentially retinol and carotenoids, but is inhibited by phytic acid (in whole grains, legumes, lentils, and nuts), polyphenols (in tea, coffee, red wines, and certain vegetables), soy protein, eggs, and calcium and phosphate salts.[58,59] Thus, while vegetarian diets often have enhancers that include increased vitamin C and carotenoids, this frequently does not make up for the increase in inhibiting substances and the absence of tissue protein factor.[58]

Adolescent athletes may have higher iron needs than the RDA and should be encouraged to consume iron-rich foods. Nutritional restrictions, declining performance, and/or fatigue should alert medical providers to check ferritin levels of athletes.

FLUID AND ELECTROLYTES

Hydration (thermoregulation)

Prior reports suggested that exercising children do not adapt to extremes of climatic temperature as effectively as adults, and need more time to acclimatize. Reasons include children's greater surface area-to-body mass ratio, which could lead to greater heat gain on a hot day and more heat loss on a cold day. Children are also believed to produce more heat per kg of

body weight than adults during physical activity, and to have a lower sweating capacity, thus reducing the ability to dissipate body heat effectively by evaporation. Therefore, higher air temperature and humidity would potentially lead to lower exercise capacity of a child.[60] However, recent studies in 9- to 12-year-old children and similarly fit and heat-acclimatized adults exposed to similar relative intensity exercise workloads and environmental conditions (while minimizing dehydration) have demonstrated similar rectal and skin temperatures, cardiovascular responses, and exercise-tolerance time during exercise in the heat in the two groups. Thirst-driven drinking behavior also appeared to be similar, as *ad libitum* fluid intake was comparable.[61–65]

In children, maturation, body size, diet, medications, illnesses, and chronic health conditions all play a role in the amount of water needed to maintain optimal cardiovascular, thermoregulatory, and other physiologic functions.[66] With exercise, individuals sweat in order to mitigate rises in core body temperature, thus losing water, sodium, potassium, iron, calcium, and magnesium, in addition to other trace elements. Significant dehydration can lead to premature fatigue, impaired sports performance, cognitive changes, electrolyte abnormalities, and increased risk of heat illness.[66] For the average child engaged in routine physical activity, water and a healthy diet should suffice in replenishing losses in sweat. When a child drinks in response to thirst alone during exercise, he/she experiences a fluid deficit of 0%–35% of incurred fluid losses. It is also possible that the inexperienced young athlete will not respond to normal thirst cues because of distraction, sympathetic stimulation, and/or lack of motivation to drink.[65] Therefore, coaches and athletic trainers should encourage appropriate hydration before, during, and after exercise. Weight checks before and after exercise are also helpful to assess fluid status, minimizing risk of both dehydration and overhydration.

The American Academy of Pediatrics recommends that fluids be available and consumed at regular intervals before, during, and after physical activities to offset sweat loss and to maintain adequate hydration while avoiding overdrinking. The Academy suggests about 3–8 oz. every 20 minutes for 9- to 12-year-olds and up to 34–50 oz. per hour for adolescents. In addition, long-duration (≥1 hour) or repeated same-day sessions of physical activity might warrant electrolyte-supplemented beverages.[61]

Sports Drinks

"Sports drinks" are typically beverages containing carbohydrates, electrolytes, minerals, and flavoring, and are intended to replenish water and electrolytes lost through sweat during exercise. "Energy drinks" are very different types of beverages, as they also contain non-nutritive stimulants, such as caffeine, guarana, taurine, ginseng, l-carnitine, creatine, and/or glucuronolactone, with claims of ergogenic or performance-enhancing benefits.[66]

As mentioned above, there may be utility in young athletes consuming sports drinks that contain carbohydrates and electrolytes during prolonged, high intensity, and/or frequent exercise sessions. Sports drinks contain 2–19 g of carbohydrates (glucose and fructose) per 8 oz., whereas energy drinks contain 0–67 g per 8 oz. In addition, the caloric content of sports drinks range from 10 to 70 calories, with that of energy drinks ranging from 10 to 270 calories.[66] Excessive intake of carbohydrate-containing beverages beyond what is needed to replenish blood glucose and muscle glycogen stores should be discouraged, as such excessive intake can increase the risk of overweight and obesity in children and adolescents, as well as dental erosion.[66]

Caffeine has been shown to improve physical performance in adults by increasing aerobic endurance and strength, improving reaction time and delaying fatigue. However, effects are quite variable and have not been studied in children or adolescents. Caffeine can increase heart rate, blood pressure, diuresis, temperature, and anxiety, and may have a negative impact on the developing neurologic and cardiovascular systems in children, in addition to causing physical dependence and addiction.[66,67] Therefore, there is no role for caffeine or other stimulants in the diet of the child or adolescent athlete.

Other legitimate ingredients in energy drinks marketed to enhance performance, such as amino acids/protein and vitamins/minerals can usually be obtained adequately from diet. Thus, although there may be utility in some sports drinks due to carbohydrate, caloric, and electrolyte needs, energy drinks are not warranted in this population.

Performance-enhancing Substances

A "performance-enhancing substance" (PES) or "performance- enhancing drug" is defined by the American Academy of Pediatrics as "*…any substance when taken in non-pharmacological doses specifically for the purposes of improving sport performance. A substance should be considered performance enhancing if it benefits sports performance by*

increasing strength, power, speed or endurance (ergogenic) or by altering body weight or body composition."[68] Sports activities require varying levels of strength and power. As with adults, children and adolescents may be tempted to use what they believe to be a PES to help them achieve the Olympic motto, "*faster (citius), higher (altius), stronger (fortius).*" This motto was introduced by the father of the Modern Olympics, Pierre de Coubertin, and often overshadows another Olympic concept introduced at the 1908 Games, "*The most important thing is not to win, but to take part!*" Our society places a huge emphasis on sports, with children getting scouted at an earlier and earlier age, and regional, state, national, and international competitions becoming a bigger and bigger industry. Parents and coaches are often dissatisfied with the rate of growth, ability to gain lean mass, and/or the predicted adult height of a child. Children may want to please the adults in their lives and also feel the thrill of victory. Thus, some will seek an athletic advantage at any cost.

In a study of preadolescent athletes, 3% admitted to experimenting with doping agents in the prior 6 months,[69] and reports suggest that 4%–12% of adolescent high school athletes use anabolic steroids.[70] Although the perceived benefit of such substances is beyond the scope of this chapter, suffice it to say that athletes use PES often with little information regarding associated risks. Such substances are often obtained illegally, do not meet Food and Drug Administration standards, and have potential serious adverse effects. Examples include a theoretical risk of cancer with human GH and IGF-I; decreased blood flow and increased blood clots from erythropoietin; and psychological disturbances, premature growth plate closure, and liver dysfunction from steroids.[70]

SPECIAL DIETS AND SUPPLEMENTS

As with adults, young athletes may follow specific diets for health, ethical, or religious reasons. Examples include gluten-free diets for those with celiac disease, modified carbohydrate diets for those with diabetes, vegetarian diets for those who oppose consumption of animals and/or animal products, and so on. Regardless of the reason for the food type omission, it can be very instructive for a young athlete following a restrictive diet to meet with a dietician/nutritionist. No research to date has shown that if well-planned and followed properly, any of the above types of dietary restrictions inhibit performance. However, there are special considerations and challenges to keep in mind. Those with celiac disease may have issues with malabsorption of vitamins and minerals.[71] Those

with diabetes have to strike a delicate balance with carbohydrate ingestion and insulin administration to avoid hyper- and hypoglycemia, the latter being a risk up to 15 hours postexercise.[72] Vegetarians are at risk for nonanemic iron deficiency and typically have lower mean muscle creatine stores.[58] Additionally, it is important to consider that some athletes choose specific diets (e.g., vegetarianism) for weight control and this practice may, therefore, be used to conceal disordered eating.[58]

Although most adolescent athletes can obtain the requisite macro- and micronutrients through food sources, some on restrictive diets may benefit from supplementation. Examples include iron and/or calcium supplementation in those who have difficulty obtaining appropriate amounts of these micronutrients through food. Decisions about supplements should be made on a case by case basis after careful dietary analysis.

TIMING OF MEALS

Although timing, size, and type of meal can vary greatly depending on the preferences and gastrointestinal characteristics of each athlete, there are some general principles to follow. There is more tolerability to a variety of meals and snacks prior to a low-intensity workout compared to a high-intensity training session or competition. Eating prior to exercise, rather than working out in a fasting state, has been shown to correlate with performance improvement.[73,74] The meal or snack consumed prior to intense exercise should fuel the athlete for the upcoming activity, while leaving him/her neither hungry nor with undigested food in the stomach. Most athletes do not like to compete on a full stomach; therefore, the pre-exercise meal should be small in order to allow for gastric emptying, while a larger meal may be consumed many hours prior to competition. The pre-exercise meal or snack should include enough fluid to maintain hydration, food relatively low in fat and fiber to facilitate gastric emptying and minimize gastrointestinal distress, food high in carbohydrates to maintain blood glucose and maximize glycogen repletion in muscles, and food with moderate protein to provide a positive net protein balance. Most importantly, the meal should be familiar to the athlete, so that he/she has experienced the consistent effects of such fueling in past similar exercise bouts.[42] This is important both physiologically and psychologically.

During exercise, fluid and glycogen losses can be replenished with water and easily digested carbohydrate sources that may include fruits,

sports gels, and bars.[75] The amount varies depending on duration and intensity of exercise. Postworkout recovery snacks that include carbohydrate and protein shortly after exercise have been shown to positively affect subsequent exercise performance and could specifically be helpful for athletes involved in multiple training or competition sessions on the same or consecutive days.[76]

ENERGY BALANCE

Energy balance is achieved when EI equals energy expenditure (EE). To achieve this, athletes need to appropriately replenish the calories expended during athletic activity in addition to maintaining their basal needs.

Obesity

Although the prevalence of childhood obesity and its consequences are at an all-time high in the United States, adolescent athletes suffer from body weight concerns at both extremes of the nutritional spectrum. These extremes include the morbidly obese, adolescent, offensive lineman with type 2 diabetes, and the adolescent dancers and runners suffering from anorexia nervosa. Emphasis on size for sports performance has driven young athletes to extremes of weight-related behavior.

In a study of 653 male American football players 8.7–14.6 years of age, 45% were overweight or obese according to Centers for Disease Control and Prevention body mass index (BMI) criteria, with the prevalence being highest in early maturing boys and more commonly in offensive and defensive linemen.[77] In the 1960s, National Collegiate Athletic Association (NCAA) Division I collegiate linemen averaged about 90.7 kg (200 lbs).[78] Now they average 136.1 kg (300 lbs).[79] Noel et al.[80] found that Division I offensive and defensive linemen average >25% body fat, with much of that fat being concentrated in the abdominal region, putting these young athletes at higher risk for the metabolic syndrome, heart disease, and stroke.79, When anthropometric measurements and blood biomarkers were obtained in 70 NCAA Division I, II, and III football lineman, 49% were found to have the metabolic syndrome.[81] Larger athletes often generate more force, thus contributing to increased musculoskeletal injuries and concussions.[79] Increased BMI also leads to increased oxygen demand and a decreased ability to dissipate heat, thus making larger players more susceptible to heat-related injuries.[82] It is paramount that larger athletes are educated about health consequences of

obesity and are given the tools to maintain healthier weight both during their competitive years and after retirement.

Energy Restriction

Energy restriction refers to decreased caloric intake often for the purpose of weight loss or relative to EE. In athletes, energy restriction may be a means to achieve a perceived ideal sports physique (as in gymnastics, figure skating, and dance) or to make a weight class (as in wrestling or lightweight rowing). Mild energy restriction may be beneficial in those who are overweight or those who need to lose only a small percentage of weight to make a weight class. However, all too often, energy restriction for sports may lead to detrimental health consequences.

Wrestling is often cited as the classic example of the dangers of rapid energy restriction combined with dehydration. For a perceived competitive advantage, wrestlers may plan to compete in a weight category lower than their preseason weight and utilize rapid weight-losing techniques to achieve this goal. Collegiate wrestlers have died from restricting food and fluid intake and exercising vigorously in hot environments prior to weigh-ins.[83] In 2005, the *British Journal of Sports Medicine* reported the case of a 5-year-old boy who had already learned dangerous habits for weight-making. On the day of weigh-ins for the final competition, the boy was overheard saying to his teammates, ‘‘Why are all of you eating before weigh-ins? I bet that I was the only one in the sauna last night. I haven’t had anything to eat today, or yesterday.” The boy, who weighed 22.7 kg, had been pressured by his father to wrestle in a lower weight class, which would require a weight loss of 2.27 kg or 10% of his usual body weight.[84,85] Thus, it’s never too early to discuss nutrition, disordered eating, and health consequences of bad habits for sport.

Female Athlete Triad

The female athlete triad (Triad) is the interrelatedness of low-energy availability, menstrual dysfunction, and low bone density.[86] Energy availability is the amount of dietary energy available after accounting for resting EE and that needed for exercise, standardized for lean body mass. Energy availability of greater than 30 kcal/kg lean body mass is believed to be necessary for normal reproductive function. In athletes with restrictive eating habits and/or those who overexercise, caloric intake may not be sufficient to meet the energy needs for the individual’s normal bodily functions, causing suppression of the reproductive axis.

Gonadotropin-releasing hormone pulsatility is diminished, leading to decreased levels of the gonadal steroids estradiol and testosterone, both of which are important antiresorptive hormones with possible bone anabolic effects. In addition, other bone anabolic hormones, such as IGF-1 and leptin, are decreased in conditions of energy deficit.[46] Thus, athletes with decreased energy availability and subsequent menstrual dysfunction are at higher risk for decreased BMD, stress fractures, and fragility fractures.[86]

Sometimes adolescent female athletes inadvertently slip into an underfueled state, because they are unaware of how much energy they are expending through physical activity. In other cases, they are trying to lose too much weight, lose weight too quickly, or maintain a low weight in an attempt to look a certain way or perform better. In a recent review of 65 studies on Triad prevalence, it was noted that only a small percentage of athletes exhibited all three components of the Triad simultaneously (0%–15.9%), but 2.7%–27.0% had two Triad conditions, and 16.0–60.0% had one condition.[87] It is important to note that having one component of the Triad is a risk factor for developing the other two components.

The greatest bone mass accrual occurs during puberty.[88] In general, healthy weight-bearing athletes should have higher BMD than their nonathletic counterparts.[89] However, low bone density has been noted in 22%–50% of female athletes, compared to 12% of the general population.[90] In young female athletes with the Triad, a compromise in bone strength and impaired bone accrual can result in short-term injuries (e.g., stress fractures) and long-term consequences (e.g., osteoporosis and fragility fractures).[86]

Disordered eating and more serious eating disorders, such as anorexia nervosa, can also lead to non-bone-related problems. These include decreased speed, strength, and exercise recovery. More serious consequences include depression, anxiety, electrolyte abnormalities, cardiac arrhythmias, and even death.[91] Because disordered eating behaviors often start in childhood or adolescence, it is important to monitor growth and dietary behavior, coupled with good nutrition and body image education, during these critical years.

DIETARY COUNSELING

When helping a young athlete determine energy requirements, a variety of questions should be asked: Does the athlete need to gain or lose weight to achieve a healthy BMI? What type of activity and how much is the athlete

performing? Is the athlete at risk for over- or undernutrition because of preconceived notions about physique and his/her particular sport? Does the athlete fear body changes during maturation and adolescence? Is there an optimal caloric and fluid intake before, during, and after training sessions and competition? Does the athlete use PES and/or follow specific diets (high protein, vegetarian, vegan, gluten free)?

Laboratory methods and equations are available to help calculate energy needs. In the laboratory, indirect calorimetry may be employed during both rest and exercise to measure EE. Outside a laboratory setting, the Harris–Benedict[92] equation is often used. The Harris–Benedict equations for males and females is detailed below:

MALE: basal energy expenditure (BEE) in kcal = 66.47 + 13.75 (weight in kg) + 5.00 (height in cm) − 6.76 (age in years)

FEMALE: BEE in kcal = 655.10 + 9.56 (weight in kg) + 1.85 (height in cm) − 4.68 (age in years)[92]

Total Caloric Requirements equal the BEE multiplied by the sum of stress and activity factors. Stress plus activity factors range from 1.2 to over 2. Activity levels include:

Little to no exercise: BEE × 1.2

Light exercise (1–3 days per week): BEE × 1.375

Moderate exercise (3–5 days per week): BEE × 1.55

Heavy exercise (6–7 days per week): BEE × 1.725

Very heavy exercise (twice per day, extra heavy workouts): BEE × 1.9

(http://www.myfitnesspal.com/topics/show/256851-harris-benedict-equation-for-maintenance)

Modifications to these equations as well as other formulas have also been utilized. Individual nutritional assessments should be performed with frequent adjustments, as these equations and methods only serve as guides.

CONCLUSION

When providing nutritional guidance to young athletes, it is essential to keep in mind the age, growth rate, level of physical maturity, specific sports involvement, and special dietary restrictions or preferences of the individual child or adolescent. This knowledge helps guide the athlete, parents, and coaches involved in the athlete's sports participation. To achieve change in fueling behavior, rather than overemphasizing effects on growth and maturation, focusing on the relationship between good dietary habits and athletic performance, injury prevention and recovery can be a powerful and effective approach in the child/adolescent athlete.

REFERENCES

1. Petrie HJ, Stover EA, Horswill CA. Nutritional concerns for the child and adolescent competitor. *Nutrition*. 2004;20(7–8):620–631.
2. Cavadini C, et al. Food habits and sport activity during adolescence: differences between athletic and non-athletic teenagers in Switzerland. *Eur J Clin Nutr*. 2000;54(suppl 1):S16–S20.
3. Nemet D, Eliakim A. Pediatric sports nutrition: an update. *Curr Opin Clin Nutr Metab Care*. 2009;12(3):304–309.
4. Rogol AD, Clark PA, Roemmich JN. Growth and pubertal development in children and adolescents: effects of diet and physical activity. *Am J Clin Nutr*. 2000;72(2 suppl):521S–528S.
5. Marshall WA, Tanner JM. Variations in the pattern of pubertal changes in boys. *Arch Dis Child*. 1970;45(239):13–23.
6. Recker RR, et al. Bone gain in young adult women. *JAMA*. 1992;268(17):2403–2408.
7. Henry YM, Fatayerji D, Eastell R. Attainment of peak bone mass at the lumbar spine, femoral neck and radius in men and women: relative contributions of bone size and volumetric bone mineral density. *Osteoporos Int*. 2004;15(4):263–273.
8. Bailey DA, et al. Calcium accretion in girls and boys during puberty: a longitudinal analysis. *J Bone Miner Res*. 2000;15(11): 2245–2250.
9. Hoch AZ, Goossen K, Kretschmer T. Nutritional requirements of the child and teenage athlete. *Phys Med Rehabil Clin N Am*. 2008;19(2): 373–398, x.
10. Malina RM. Physical growth and biological maturation of young athletes. *Exerc Sport Sci Rev*. 1994;22:389–433.
11. Malina RM. Physical activity and training: effects on stature and the adolescent growth spurt. *Med Sci Sports Exerc*. 1994;26(6):759–766.
12. Roemmich JN, Richmond RJ, Rogol AD. Consequences of sport training during puberty. *J Endocrinol Invest*. 2001;24(9):708–715.
13. Jeukendrup A, Cronin L. Nutrition and elite young athletes. *Med Sport Sci*. 2011;56:47–58.
14. Roemmich JN, Sinning WE. Weight loss and wrestling training: effects on growth-related hormones. *J Appl Physiol*. 1997;82(6):1760–1764.
15. Roemmich JN, Sinning WE. Sport-seasonal changes in body composition, growth, power and strength of adolescent wrestlers. *Int J Sports Med*. 1996;17(2):92–99.
16. Bass S, et al. Short stature and delayed puberty in gymnasts: influence of selection bias on leg length and the duration of training on trunk length. *J Pediatr*. 2000;136(2):149–155.
17. Buckler JM, Brodie DA. Growth and maturity characteristics of schoolboy gymnasts. *Ann Hum Biol*. 1977;4(5):455–463.
18. Nichols DL, et al. The effects of gymnastics training on bone mineral density. *Med Sci Sports Exerc*. 1994;26(10):1220–1225.
19. Jahreis G, et al. Influence of intensive exercise on insulin-like growth factor I, thyroid and steroid hormones in female gymnasts. *Growth Regul*. 1991;1(3):95–99.
20. Theintz GE, et al. Evidence for a reduction of growth potential in adolescent female gymnasts. *J Pediatr*. 1993;122(2):306–313.
21. Baxter-Jones AD, et al. Menarche in intensively trained gymnasts, swimmers and tennis players. *Ann Hum Biol*. 1994;21(5):407–415.
22. Riddell MC. The endocrine response and substrate utilization during exercise in children and adolescents. *J Appl Physiol*. 2008;105(2): 725–733.
23. Ekblom B. Effect of physical training in adolescent boys. *J Appl Physiol*.

1969;27(3):350–355.
24. Galassetti P, et al. Effect of a high-fat meal on the growth hormone response to exercise in children. *J Pediatr Endocrinol Metab*. 2006;19(6): 777–786.
25. Fahey TD, et al. Pubertal stage differences in hormonal and hematological responses to maximal exercise in males. *J Appl Physiol*. 1979;46(4): 823–827.
26. Rodriguez NR, et al., Position of the American Dietetic Association, Dietitians of Canada, and the American College of Sports Medicine: nutrition and athletic performance. *J Am Diet Assoc*., 2009;109(3):509–527.
27. Phillips SM. Carbohydrate supplementation and prolonged intermittent high-intensity exercise in adolescents: research findings, ethical issues and suggestions for the future. *Sports Med*. 2012;42(10):817–828.
28. Almquist J, et al., Summary statement: appropriate medical care for the secondary school-aged athlete. *J Athl Train*. 2008;43(4):416–427.
29. Haralambie G. Enzyme activities in skeletal muscle of 13-15 years old adolescents. *Bull Eur Physiopathol Respir*. 1982;18(1):65–74.
30. Burke LM, et al. Guidelines for daily carbohydrate intake: do athletes achieve them? *Sports Med*. 2001;31(4):267–299.
31. Riddell MC, et al. Substrate utilization in boys during exercise with [13C]-glucose ingestion. *Eur J Appl Physiol*. 2000;83(4–5):441–448.
32. Blaner WS. The fat-soluble vitamins 100 years later: where are we now? *J Lipid Res*. 2013;54(7):1716–1718.
33. Meyer F, et al. Nutrition for the young athlete. *J Sports Sci*. 2007; 25(suppl 1):S73–S82.
34. Burke LM, et al. Nutrition for distance events. *J Sports Sci*. 2007; 25(suppl 1):S29–S38.
35. Gidding SS, et al., Dietary recommendations for children and adolescents: a guide for practitioners: consensus statement from the American Heart Association. *Circulation*. 2005;112(13):2061–2075.
36. Aerenhouts D, et al. Influence of growth rate on nitrogen balance in adolescent sprint athletes. *Int J Sport Nutr Exerc Metab*. 2013;23(4):409–417.
37. Carlsohn A, et al. Exercise increases the plasma antioxidant capacity of adolescent athletes. *Ann Nutr Metab*. 2008;53(2):96–103.
38. Watson TA, MacDonald-Wicks LK, Garg ML. Oxidative stress and antioxidants in athletes undertaking regular exercise training. *Int J Sport Nutr Exerc Metab*. 2005;15(2):131–146.
39. Woolf K, Manore MM. B-vitamins and exercise: does exercise alter requirements? *Int J Sport Nutr Exerc Metab*. 2006;16(5):453–484.
40. Lukaski HC. Vitamin and mineral status: effects on physical performance. *Nutrition*. 2004;20(7–8):632–644.
41. Lanser EM, Zach KN, Hoch AZ. The female athlete triad and endothelial dysfunction. *PMR*. 2011;3(5):458–465.
42. American Dietetic Association, et al. American College of Sports Medicine position stand. Nutrition and athletic performance. *Med Sci Sports Exerc*. 2009;41(3):709–731.
43. Casey CF, Slawson DC, Neal LR. Vitamin D supplementation in infants, children, and adolescents. *Am Fam Physician*. 2010;81(6):745–748.
44. Clemens TL, et al. Increased skin pigment reduces the capacity of skin to synthesise vitamin D3. *Lancet*. 1982;1(8263):74–76.
45. Holick MF. Vitamin D deficiency. *N Engl J Med*. 2007;357(3):266–281.
46. Christo K, et al. Bone metabolism in adolescent athletes with amenorrhea, athletes with eumenorrhea, and control subjects. *Pediatrics*. 2008;121(6):1127–1136.

47. Larson-Meyer DE, Willis KS. Vitamin D and athletes. *Curr Sports Med Rep*. 2010;9(4):220–226.
48. Stoffman N, Gordon CM. Vitamin D and adolescents: what do we know? *Curr Opin Pediatr*. 2009;21(4):465–471.
49. Wagner CL, et al. Prevention of rickets and vitamin D deficiency in infants, children, and adolescents. *Pediatrics*. 2008;122(5):1142–1152.
50. Holick MF. Vitamin D: extraskeletal health. *Endocrinol Metab Clin North Am*. 2010;39(2):381–400, table of contents.
51. Willis KS, Peterson NJ, Larson-Meyer DE. Should we be concerned about the vitamin D status of athletes? *Int J Sport Nutr Exerc Metab*. 2008;18(2):204–224.
52. Rozen GS, et al. Calcium supplementation provides an extended window of opportunity for bone mass accretion after menarche. *Am J Clin Nutr*. 2003;78(5):993–998.
53. Prentice A, et al. Calcium supplementation increases stature and bone mineral mass of 16- to 18-year-old boys. *J Clin Endocrinol Metab*. 2005;90(6):3153–3161.
54. Dubnov G, Constantini NW. Prevalence of iron depletion and anemia in top-level basketball players. *Int J Sport Nutr Exerc Metab*. 2004;14(1):30–37.
55. Beals KA. Eating behaviors, nutritional status, and menstrual function in elite female adolescent volleyball players. *J Am Diet Assoc*. 2002;102(9):1293–1296.
56. Hinton PS, Sinclair LM. Iron supplementation maintains ventilatory threshold and improves energetic efficiency in iron-deficient nonanemic athletes. *Eur J Clin Nutr*. 2007;61(1):30–39.
57. Hunt JR. Bioavailability of iron, zinc, and other trace minerals from vegetarian diets. *Am J Clin Nutr*. 2003;78(3 suppl):633S–639S.
58. Barr SI, Rideout CA. Nutritional considerations for vegetarian athletes. *Nutrition*. 2004;20(7–8):696–703.
59. Hallberg L, Hulthen L. Prediction of dietary iron absorption: an algorithm for calculating absorption and bioavailability of dietary iron. *Am J Clin Nutr*. 2000;71(5):1147–1160.
60. American Academy of Pediatrics. Climatic heat stress and the exercising child and adolescent. American Academy of Pediatrics. Committee on Sports Medicine and Fitness. *Pediatrics*. 2000;106(1 Pt 1):158–159.
61. Council on Sports Medicine and Fitness and Council on School Health. Policy statement—Climatic heat stress and exercising children and adolescents. *Pediatrics*. 2011;128(3):e741–e747.
62. Inbar O, et al. Comparison of thermoregulatory responses to exercise in dry heat among prepubertal boys, young adults and older males. *Exp Physiol*. 2004;89(6):691–700.
63. Rivera-Brown AM, et al. Exercise tolerance in a hot and humid climate in heat-acclimatized girls and women. *Int J Sports Med*. 2006;27(12):943–950.
64. Rowland T, Garrison A, Pober D. Determinants of endurance exercise capacity in the heat in prepubertal boys. *Int J Sports Med*. 2007;28(1):26–32.
65. Rowland T. Fluid replacement requirements for child athletes. *Sports Med*. 2011;41(4):279–288.
66. Committee on Sports Medicine and Fitness. Sports drinks and energy drinks for children and adolescents: are they appropriate? *Pediatrics*. 2011;127(6):1182–1189.
67. Nawrot P, et al. Effects of caffeine on human health. *Food Addit Contam*. 2003;20(1):1–30.
68. American Academy of Pediatrics Committee on Sports Medicine and Fitness. Use

of performance-enhancing substances. *Pediatrics*. 2005;115(4):1103–1106.
69. Laure P, Binsinger C. Doping prevalence among preadolescent athletes: a 4-year follow-up. *Br J Sports Med*. 2007;41(10):660–663; discussion 663.
70. Rogol AD. Drugs of abuse and the adolescent athlete. *Ital J Pediatr*. 2010;36:19.
71. Mancini LA, Trojian T, Mancini AC. Celiac disease and the athlete. *Curr Sports Med Rep*. 2011;10(2):105–108.
72. MacDonald MJ. Postexercise late-onset hypoglycemia in insulin-dependent diabetic patients. *Diabetes Care*. 1987;10(5):584–588.
73. Jentjens RL, et al. Effects of pre-exercise ingestion of differing amounts of carbohydrate on subsequent metabolism and cycling performance. *Eur J Appl Physiol*. 2003;88(4–5):444–452.
74. Moseley L, Lancaster GI, Jeukendrup AE. Effects of timing of pre-exercise ingestion of carbohydrate on subsequent metabolism and cycling performance. *Eur J Appl Physiol*. 2003;88(4–5):453–458.
75. Mujika I, Burke LM. Nutrition in team sports. *Ann Nutr Metab*. 2010;57(suppl 2):26–35.
76. Beelen M, et al. Nutritional strategies to promote postexercise recovery. *Int J Sport Nutr Exerc Metab*. 2010;20(6):515–532.
77. Malina RM, et al. Overweight and obesity among youth participants in American football. *J Pediatr*. 2007;151(4):378–382.
78. Berg K, Latin RW, Baechle T. Physical and performance characteristics of NCAA Division I football players. *Res Q Exerc Sport*. 1990;61(4):395–401.
79. Jacobson BH. Anthropometric cross-sectional comparisons of college football players and potential health implications. *J Strength Cond Res*. 2012;26(12):3358–3364.
80. Noel MB, et al. Body composition in Division I football players. *J Strength Cond Res*. 2003;17(2):228–237.
81. Buell JL, et al. Presence of metabolic syndrome in football linemen. *J Athl Train*. 2008;43(6):608–616.
82. Yamamoto JB, et al. Epidemiology of college athlete sizes, 1950s to current. *Res Sports Med*. 2008;16(2):111–127.
83. Centers for Disease Control and Prevention. Hyperthermia and dehydration-related deaths associated with intentional rapid weight loss in three collegiate wrestlers—North Carolina, Wisconsin, and Michigan, November-December 1997. *MMWR Morb Mortal Wkly Rep*. 1998;47(6):105–108.
84. Cunningham E. Where can I find resources to address healthful weight practices for wrestlers? *J Am Diet Assoc*. 2006;106(5):761.
85. Sansone RA, Sawyer R. Weight loss pressure on a 5 year old wrestler. *Br J Sports Med*. 2005;39(1):e2.
86. Nattiv A, et al. American College of Sports Medicine position stand. The female athlete triad. *Med Sci Sports Exerc*. 2007;39(10):1867–1882.
87. Gibbs JC, Williams NI, De Souza MJ. Prevalence of individual and combined components of the female athlete triad. *Med Sci Sports Exerc*. 2013;45(5):985–996.
88. Gilsanz V, et al. Age at onset of puberty predicts bone mass in young adulthood. *J Pediatr*. 2011;158(1):100–105, 105 e1–e2.
89. McKay H, et al. Physical activity positively predicts bone architecture and bone strength in adolescent males and females. *Acta Paediatr*. 2011;100(1):97–101.
90. Khan KM, et al. New criteria for female athlete triad syndrome? As osteoporosis is rare, should osteopenia be among the criteria for defining the female athlete triad syndrome? *Br J Sports Med*. 2002;36(1):10–3.

91. Yager J, Andersen AE. Clinical practice. Anorexia nervosa. *N Engl J Med*. 2005;353(14):1481–1488.
92. Harris JA, Benedict FG. A biometric study of human basal metabolism. *Proc Natl Acad Sci USA*. 1918;4(12):370–373.

Special Diets

70

Sharon Evans, MSc, Tracey Johnson, BSc, SRD, Katharina Dokoupil, MSc, and Anita MacDonald, BSc, PhD

INTRODUCTION

Nutritional intervention and therapeutic diets play a central role in the treatment of many pediatric disorders. The aims and principles of the dietary management include the following:

1. Obtaining relief of symptoms by avoidance of the "offending" or source of the substrate food/additive
2. Preventing inadvertent exposure to the "offending" foods
3. Preventing unnecessary avoidance of foods
4. Supporting normal growth and development for age and gender in children
5. Ensuring that the diet is safe and does not lead to metabolic instability
6. Providing an adequate, healthy, nutritionally dense and balanced diet with appropriate alternatives for the excluded food allergens to minimize the impact on quality of life

Dietary treatment may be used as a monotherapy or in combination with drug or exercise therapy. Additional emergency treatment (EpiPen [epinephrine] Auto-Injector for anaphylaxis in food allergy or administration of emergency glucose solutions either orally or intravenously in some inherited metabolic disorders [IMDs]) may be necessary. The diet may require exclusion of one or more substrate "offending" foods or a controlled and regular food intake to maintain normoglycemia and prevent secondary complications. Some patients may require additional nutritional support with dietary supplements or tube feeds. Diet therapy is often arduous and unrelenting and may create

additional anxieties and difficulties for caregivers. Feeding problems may be more common in children on special diets than in healthy children.

This chapter describes commonly used special diets for selected pediatric disorders. Special diets for clinical conditions such as food allergy, gastrointestinal (GI) disease, faltering growth, hyperlipidemia, selected inborn errors of metabolism, weight control, and mineral deficiency are found in this chapter.

PRINCIPLES OF MANAGEMENT OF FOOD ALLERGY AND INTOLERANCE

The only treatment of any food intolerance is the avoidance of the causative allergen or offending food or ingredient. Individuals will tolerate different thresholds to offending foods; the most sensitive food allergic individuals will experience reactions from exposure to trace quantities only.

In general, the following stringent dietary advice is necessary:

- Avoidance of food/ingredient sources containing allergenic ingredients, even in small amounts.
- Avoidance of cross-contamination from allergen food sources.
- Avoidance of unrefined oil derived from allergenic foods.
- Advice on meals or foods of uncertain composition (meals eaten in restaurants or “Takeaways”).
- Meal planning and appropriate food choices.
- Monitoring nutritional adequacy of diet therapy.
- Regular observation of symptom control, dietary adherence/indiscretions and associated consequences, growth.

Targeted information may also be required about lactation diets, infant feeding and choice of special formula, and solid food introduction for lactating mothers and infants.

Individuals require verbal and detailed written advice about reading and interpreting food labels and understanding how to avoid cross-contamination during food preparation,[1] with regular updates and retraining. It is important to effectively liaise and teach the caregivers at nurseries, school, and school meal services to eliminate or reduce exposure to food allergens. All those involved with caring or teaching a child need to be aware of the food allergy, treatment, and any emergency treatment procedures in case of food reactions. A training program for school nurses with educational materials is important so they can help cascade

information to teachers and other caregivers at school.[2] Children also require education about which foods should be avoided. Role-play with plastic foods, about foods permitted and not permitted, is helpful. Also teaching a child that they should not accept food from others is important.

Many food allergy reactions are caused by allergens eaten from unlabeled sources. Detailed information is required about purchasing unpackaged food from retail outlets, from restaurants, or travelling abroad. There can be no venue considered a completely safe place to eat for those with food allergies.[3] Accidental exposures to food allergens can lead to reactions at home, school, work, and restaurants.

Some patients or caregivers may choose to avoid too many foods and therefore over restrict their diets. This may be caused by anxiety regarding unfamiliar foods and fear of inadvertent allergy reactions.[4] Patients with milk allergy may unnecessarily avoid nuts and soy or with peanut allergy may avoid all tree-nut sources too.

There is evidence that persistent food allergy and food hypersensitivity may be an important cause of anxiety[5] and negatively impact on quality of life.[6] Disrupted family activities include family social events, sleepovers, and visits to friend's houses without parental supervision. Children report anxiety about going on holiday, attending parties, and using public transport. Activities such as eating out are seen as frightening.[7] Parents often become hypervigilant but worry about the effect this overprotectiveness may have on their child. More than 30% of parents of children with food allergies make more than one visit per month to school to discuss issues surrounding their child's allergy.[8]

Allergen Food Labeling and European Legislation

The EU (EC 2003[9]; Food Labelling Regulations EC 2006[10]) state that the common food allergens or ingredients made from them should be identified on prepackaged food including alcoholic drinks. These foods include:

- Cereals containing gluten, that is, wheat, rye, barley, oats, spelt or their hybridized strains and products.
- Eggs and egg products.
- Fish and fish products.
- Crustaceans.
- Mollusks.
- Peanuts and peanut products.
- Soybeans and products.

- Lupin.
- Milk and milk products (lactose included).
- Tree-nuts and nut products.
- Celery and products.
- Mustard and products.
- Sesame seeds and products.
- Sulfur dioxide and sulfites in concentrations of >10 mg/kg or 10 mg/L.

Similar labeling laws have been adopted by the United States.[11]

These commonly allergenic foods account for over 90% of all moderate to severe allergic reactions to foods, although more than 160 foods are associated with sporadic allergic reactions.[12] Allergic reactions to fresh fruits, the so called oral allergy syndrome, are also rather common,[13] but these foods are not included on the EU list because the symptoms are typically mild and confined to the oropharyngeal region and the allergens are unstable to heating and digestion.

The EU[14] (Commission Directive 2007/68/EC) further published a very highly processed list of food ingredients that are unlikely to be associated with adverse reactions in susceptible individuals. These ingredients are exempt from EU labeling requirements and include the following:

- Wheat-based glucose syrups including dextrose
- Wheat-based maltodextrins
- Glucose syrups based on barley
- Cereals used for making distillates or ethyl alcohol of agricultural origin for spirit drinks and other alcoholic beverages
- Fish gelatin used as a carrier for vitamin or carotenoid preparations
- Fish gelatin or isinglass used as fining agent in beer and wine
- Fully refined soybean oil and fat, vegetable oils derived from phytosterols, esters from soybean sources, plant stanol ester produced vegetable oil, sterols from soybean sources
- Whey used for making distillates or ethyl alcohol of agricultural origin for spirit drinks and other alcoholic beverages
- Lactitol
- Nuts used for making distillates or ethyl alcohol of agricultural origin for spirit drinks and other alcoholic beverages

A further EC Directive[15] (No 1169/2011) will introduce a new requirement for allergen information to be provided for foods sold

nonpacked or prepacked for direct sale. At the end of 2014, allergy information on food sold unpackaged in catering outlets, deli-counters, bakeries, and sandwich bars should be provided.

The EU also request that mandatory information will be clearly visible on food labels, and identification of mandatory allergens should be in a type set that clearly distinguishes it from the rest of the ingredients. In the United Kingdom, the mandatory allergens will be identified in **bold type** on the label.[16]

Also in the United Kingdom, many manufacturers have provided information about the possible and unintentional presence of substances or products causing allergies or intolerances that may be in a food product. The EU suggests that in many cases such voluntary food information is provided to the detriment of the clarity of the mandatory food information. In the United Kingdom, such statements will be replaced with an Allergy Advice box containing the following statement: 'for allergens, see ingredients in bold.' There will also be consistent presentation of information about cross-contamination.

EGG-FREE DIET (Tables 70-1 and 70-2)

- Egg allergy usually presents in infancy with a prevalence of approximately 2% in children and 0.1% in adults. It is induced by egg protein (usually ovalbumin and/or ovomucoid).[17]
- Ingestion of raw or undercooked egg may trigger more severe clinical reactions than well-cooked egg. Children with mild egg allergy may be able to tolerate extensively heated egg products.[17]
- Ingredients and allergy warning labels on prepackaged foods should be checked. Non-prepackaged foods may be a hazard, particularly bread and pastries that may have been glazed or cross-contaminated with egg.[17]
- Breast-feeding: egg protein from the maternal diet is detectable in breast milk.[18] Therefore, eczema in breast-fed babies with egg allergy may improve if their mothers avoid eating eggs.

TABLE 70-1 Egg or Egg Derivatives

Egg proteins: Albumin, globulin, ovalbumin, ovomucin, ovomucoid, ovovitellin, livetin,
Egg white, egg yolk
Dried egg, egg powder, egg solids, pasteurized egg
Lysozyme

Egg lecithin (E322)

TABLE 70-2 Egg-Free Diet

Foods Allowed	Foods to Avoid
Milk, milk products and milk derivatives Cows, goat, sheep and other animal milks (including whole, skimmed, dried, evaporated and condensed milk). Yogurt, cream, sour cream, crème fraiche, fromage frais, cream cheese, whipped toppings Rice drink, soy drink, almond drink, hazelnut drink, potato drink, coconut milk, (cream made from these milks). Soy cheese, soy yogurt, soy desserts	*Milk, milk products and milk derivatives, Greek yogurt*
Cheese All cheese	*Cheese* Blue cheese, cream cottage cheese
Fats and oils Butter, margarines Vegetable oils Lard, dripping, suet	*Fats and oils* Butter, salad dressing with egg, mayonnaise, tartar sauce, hollandaise sauce
Meat and fish Plain beef, lamb, veal, pork, wild game, poultry, liver, kidney, offal, fish, shellfish (fresh or frozen)	*Meat and fish* Commercially breaded meats, fish, or poultry. Meatballs, meat loaf, croquettes, some sausages, rissoles, pâté, prepared meat dishes containing egg
Egg-free substitutes only	Eggs Eggs from all birds Scotch eggs, quiche, scrambled or omelette, meringue, soufflés, egg curry
Non-meat alternatives Peas, beans, lentils (dried, canned, or frozen e.g. dhal or hummus); all nuts and seeds, tofu, peanut butter.	*Non-meat alternatives* Quorn products, vegetarian burgers or sausages and other similar vegetarian products
Cereal, flour, pasta All grains; wheat, oats, corn, cornmeal, barley, maize, sago, rye, tapioca, bulgar, quinola, millet, arrowroot. Custard power (original), semolina Carob	*Cereal, flour, pasta* Egg noodles or pasta. Fried rice containing eggs Pancakes, crepes, doughnuts, waffles, muffins, bread crumbs and pretzels. Baking mixes, fritter batter, or batter-fried food. Yorkshire pudding

Breakfast cereals Most are suitable, e.g. Weetabix, Cornflakes, Rice Krispies	*Breakfast cereals*
Bread and yeast products Plain bread, white, wholewheat, rye bread (without egg products or brushing with egg for glazing).	Bread and yeast products French toast, waffles, muffins, egg glaze on bread, bread rolls, croissants, egg roll wrappers
Cakes, biscuits, crackers Egg-free biscuits. Oatcakes, Rye crackers	*Cakes, biscuits, crackers* Many cakes and biscuits contain egg, cream filled pies, pastries, scones, choux pastry, pastry glazed with egg, fruit flans. *Royal icing, Icing or frostings*
Desserts Sorbet, jelly, yogurt	*Desserts* Many desserts contain egg in some form. Ice cream (especially fresh and deluxe types), custard containing egg, dessert powders, Mousse, some sorbets, Bread and butter pudding, custard tart, egg custard, cream caramels, frittata, tiramisu
Fruit All fresh, frozen, tinned or dried fruit All fruit juices	*Fruit*
Vegetables All fresh, frozen, tinned or dried vegetables (without added milk or milk products) Most dried and tinned pulses, e.g. red kidney beans, chick peas, lentils Tomato paste and puree All vegetable juices Olives	*Vegetables* Coleslaw, potato salad, vegetables with mayonnaise and salad cream. Any vegetables prepared with a sauce or breaded that contains eggs in any form (such as hollandaise sauce, vegetable soufflé, or batter style vegetables)
Savoury snacks Plain popcorn, plain pretzels, plain potato and tortilla chips, fruit crisps	*Savoury snacks*
Seasonings Herbs, spices, salt, pepper, vinegar, mustard	*Seasonings*
Soups, gravies and sauces Bouillon, broth, meat, or vegetable stock soups; bisques and chowders made with water or milk. Stock cubes/concentrates, gravy granules, ketchup, sauces made without egg, salsa, relish, pickles.	*Soups, gravies, and sauces* Mayonnaise, Aioli, Remoulade, Béarnaise sauce Hollandaise sauce, tartar sauce, salad cream Any stock cleared with egg (consommé, broth, bouillon), canned soups Cream sauces made with egg.

Sugar, sweet spreads Sugar, honey, jams, jellies, maple and corn syrup, fruit based pouring syrup, molasses.	Sugar, sweet spreads Lemon curd
Confectionery Boiled or hard sweets/candy, most mints, plain fruit lollies, chewing gum, gum drops	Confectionery Chewy sweets, marshmallow, jelly beans brushed with egg white, chocolate bars (e.g. Mars Bar, Snickers, Wagon Wheel), nougat
Drinks Milk shake syrup or powders Carbonated, squash, fruit juice Cocoa powder, tea, most coffee Flavored water	*Drinks* Malted beverages, eggnog
Miscellaneous – used in baking Baking powder, yeast, gelatine Food essence and food coloring	*Miscellaneous—used in baking* Marzipan Alcohol clarified with egg
Food labels should always be checked because ingredients might change.	

Possible Sources of Egg

- A shiny glaze or yellow baked goods may have been brushed with an egg wash. Simplesse is a protein-based fat substitute derived from egg whites and milk (or whey protein) that allows energy reductions of up to 80%. It may be used in ice cream, yogurt, cheese spread, salad dressings, margarine, mayonnaise, coffee creamer, soups, and sauces.
- Egg shells or whites may have been used to clarify soup stocks, consommés, wine, and coffee drinks.
- Breaded foods may be dipped in egg before dipping in breadcrumbs.

Egg-Free Ingredients and Substitutions

Cake, muffin, and other recipes can be adapted by using egg replacers or replacing a teaspoon of baking powder for each egg. Alternatively, replace one egg with one teaspoon of Xantham gum or gelatin. Egg replacers have little nutritional value and are based on potato, tapioca starch, and raising agents. They can be purchased from health food shops or large supermarkets. Egg-free mayonnaise, cake and muffin mixes, omelets, and quiche mixes are available.

Nonfood Items Containing Egg

These can be identified by reading the ingredient label. U.K. law requires

cosmetics, toiletries, perfumes, finger paints, and medications to include a list of ingredients on their packaging.

Vaccines

Yellow fever vaccine and influenza vaccine should be avoided by those with egg allergy.

SOY-FREE DIET (Tables 70-3 and 70-4)

- The soy bean belongs to the legume family, which includes peas, beans, carob, liquorices, and peanut. Reactions to more than one member of the legume family are rare. It is usually unnecessary to avoid all foods from the plant family as cross-reactivity is uncommon.
- Soy is widely used in foods and so is difficult to avoid. As many as 60% of manufactured foods contain soy. Soy can be ingested as whole beans, soy flour, soy sauce, or soy oil. Soy can also be used in foods as a texturizer (texturized vegetable protein), emulsifier (soy lecithin), or protein filler. Soy flour is widely used in foods including breads, cakes, processed foods (ready meals, burgers and sausages), and baby foods.
- Soy lecithins are commonly used in the food industry as emulsifiers. Lecithin is a lipid, which stabilizes foods that contain water and fats, which do not normally mix. Evidence suggests that proteins present in soy lecithin and soy oil have low allergenicity[19] and most individuals with soy allergy should tolerate it.
- In the United Kingdom, the Food Standard Agency advises that refined soy oil (the main component of vegetable oil) should be safe for people with soy allergy, because the proteins that cause allergic reactions are removed during the refining process. However, cold pressed soy oil, usually sold from delicatessen counters or health food shops, may contain soy protein and should be avoided.
- Most soy sauces contain very small amounts of soy, with most of the protein in the sauce being derived from fermented wheat.[20] Soy sauce is well known as a Japanese traditional seasoning, namely shoyu.

TABLE 70-3 Soy Derivatives
Soy protein isolate
Soy protein concentrate
Soy shortening
Soy protein
Soy albumin
Soy beans
Soy flavoring
Soy flour
Soy gum
Soy lecithin (E322)
Soy milk
Soy nuts
Soy oil
Soy starch

Terms That May Indicate the Presence of Soy

Vegetable broth, vegetable oil, vegetable protein, vegetable paste, textured vegetable protein (TVP), hydrolyzed vegetable protein (HVP), hydrolyzed plant proteins (HPP), vegetable starch, vegetable gum.

Foods That Contain Soy

Miso, soy infant formula, soy margarine, soy or Shoyu sauce, Tamari, soy flour, grits, soy nuts, soy drink, soy sprouts, soy yogurts, soy cheese, soy desserts, tempeh, tofu/tofutti, TVP, Edamame, Natto, bean curd, Kinako, Nimane, yuba, and Okara.

Foods That May Contain Soy

Baby foods, bread, breakfast cereals, cakes and biscuits, confectionary with a biscuit base, cheese substitutes, canned and tinned soup, Chinese foods, chocolates (especially cream centers), commercial fruit products, crackers, crisps, dessert mixes, flavored crisps, frozen dessert, ice cream, liquid meal replacers, margarine, meat products (cold meats, beef burgers, meat pasta/pies, minced beef, sausages, hotdogs, coffee whiteners or cream replacers, pancake and waffle mixes, pasta, pizza bases, ready meals, sauces (including Worcester sauce, sweet and sour, Teriyaki sauce, stock cubes, gravy powders, and some cook in sauces), seasoned salt, snack bars, soups (canned or packet), sandwich spread, mayonnaise, salad creams, vegetable products, and vegetarian meals.

Alternatives to Soy

- Avocado, tahini (sesame paste), and nut butters can make useful alternatives to margarine for spreading on sandwiches and toast.
- Olive oil, sunflower or safflower oil, canola oil can be used as alternatives to undefined vegetable oil.

PEANUT-FREE DIET

- Peanut allergy is an IgE-mediated allergic disease, associated with fatal anaphylaxis. It typically presents with symptoms after one of the first few exposures to the peanut source. Only about 20% of individuals acquire tolerance[3] but about 8% may then experience a resensitization. The prevalence rate of peanut allergy is estimated to be 1% of the population.[3,21] There are no parameters that can reliably predict with certainty those at risk of anaphylaxis.[22]
- Peanut (*Arachis hypogaea*) is a member of the legume family but not nut family. Legumes are edible seeds enclosed in pods; others include soy beans, lima beans, carob, and sweet clover.
- Eight peanut allergens have been identified that are termed as Ara h 1 to Ara h 8.[23] Individuals may react to minute amounts of peanut allergen (100 μg–50 mg). The protein fraction of peanuts makes up 25%–30% of the kernel. The allergenicity of peanut proteins is very resistant to thermal, chemical, and proteolytic denaturation.
- Long-term management consists of complete avoidance of all sources of peanut. It requires constant vigilance.
- Unrefined (crude) peanut oil should be avoided by peanut allergic people as the process by which the oil is made means that low levels of protein can contaminate the oil. Highly refined peanut oils are unlikely to cause a reaction for the majority of people with peanut allergy[24] but it may be difficult to identify the type of oil used in a product.
- Roasting and heating of peanuts (not boiling) may increase allergenicity.[25]
- Some individuals who are peanut allergic may also react to lupin.[26] Lupin flour may be added to some baked goods. Under European law, lupin must be labeled when it is added to prepacked food.
- In the United Kingdom, in the past, the Department of Health

advised against the consumption of peanuts during pregnancy, breast-feeding, and weaning. Since 2009, the advice is that peanuts can be consumed during pregnancy or breast-feeding. However, peanuts should be avoided in the following situations: if a mother suffers from peanut allergy, if there is a history of allergy in the immediate family, or if the child already has an allergy. Medical advice should be sought before giving peanut for the first time.[27]

TABLE 70-4 Soy-Free Diet

Foods Allowed	Foods to Avoid
Milk, milk products and milk derivatives Cows, goat, sheep and other animal milks (including whole, skimmed, dried, evaporated and condensed milk. Yogurt, cream, sour cream, crème fraiche, fromage frais, cream cheese made from animal milk Rice drink, almond drink, hazelnut drink, oat drink, quinola drink and hemp drink, coconut milk, (cream made from these milks).	*Milk, milk products and milk derivatives* Soy drink, soy infant milk, non-dairy creamers, Soy yogurt, soy desserts, soy cheese, some processed cheeses.
Fats and oils Butter Pure sunflower or olive oil margarines Home-made salad dressings	*Fats and oils* Vegetable oil, margarine, salad dressing, mayonnaise, vegetable oils, vegetable sprays, shortening unless the type of oil is specified as non-soy oil.
Meat and fish Plain beef, lamb, veal, pork, wild game, poultry, liver, kidney, offal, fish, shellfish (fresh or frozen)	*Meat and fish* Commercially breaded meats, fish, or poultry, sausages, hamburgers, rissoles, pate, prepared meat dishes, hot dogs, meat loaf containing soy, Deli meats
Eggs Plain eggs	*Eggs*
Non-meat alternatives Peas, beans, lentils (dried, canned, or frozen e.g. dhal or hummus); all nuts and seeds.	Non-meat alternatives Textured vegetable protein, vegetarian ‘meat’ or ‘chicken’ burgers or sausages and other similar vegetarian products, soy nuts, peanut butter and peanut butter substitutes, sandwich spread, commercial Seitan.

Cereal, flour, pasta All grains; wheat, oats, corn, cornmeal, barley, maize, sago, rye, tapioca, bulgar, quinola, millet, arrowroot, pasta, semolina.	*Cereal, flour, pasta* Soy containing noodles, pasta, commercial pasta and rice in sauces, dry pizza mixes Pancake mixes
Breakfast cereals Commercial cereals not containing soy	Breakfast cereals Granolas and natural cereals containing soy.
Bread and yeast products Plain bread, white, whole-wheat, rye bread (without soy flour or soy milk).	*Bread and yeast products* Breads, bread rolls, stuffing, doughnuts, waffles containing soy flour.
Cakes, biscuits, crackers Soy-free biscuits, oatcakes and wheat and rice crackers	*Cakes, biscuits, crackers* Cakes, biscuits, cookies, muffins containing soy.
Desserts Home-made ice cream. Pastries, puddings, pies without soy. Frostings, sauces, without soy.	*Desserts* Many desserts contain soy in some form. Ice cream, dessert powders, ice cream cones, pies, puddings, pastries, frostings, creamy desserts containing soy.
Fruit All fresh, frozen, tinned or dried fruit All fruit juices	*Fruit* Fruits in commercial baked goods, fruit bars
Vegetables All fresh, frozen, tinned or dried vegetables (without added milk or milk products) Most dried and tinned pulses, e.g. red kidney beans, chick peas, lentils Tomato paste and puree All vegetable juices Olives	*Vegetables* Soy sprouts, commercially prepared vegetables that are breaded or contain soy sauce, canned and frozen Oriental style vegetables, canned and frozen vegetables in any sauce.
Savoury snacks Plain popcorn popped with corn, sunflower, rice cakes	*Savoury snacks* Pretzels, crisps, corn chips
Seasonings Herbs, pure spices and seasonings	*Seasonings* Seasoned salt
Soups, gravies and sauces Home-made soup made with fresh ingredients and vegetable/meat stock.	*Soups, gravies, and sauces* Commercial soups, canned and dry mixes, noodle soups, soups containing tofu or nondairy creamer, pesto, cook in sauces, curry sauces, marinades. Worcester sauce, Oyster sauce, Teriyaki sauce, stock cubes, gravy powders

Sugar, sweet spreads Sugar, honey, jams, jellies, maple and corn syrup, molasses.	*Sugar, sweet spreads*
Confectionery Home-made sweets without soy.	*Confectionery* Sweets containing soy oil/undefined oil.
Drinks Carbonated, squash, fruit juice Cocoa powder, tea, most coffee *Flavored water*	*Drinks* Soy drink
Food labels should always be checked because ingredients might change.	

Possible Sources of Peanuts (Table 70-5)

- Foods that could contain peanuts or peanut products include African, Chinese, Indonesian, and Thai dishes, peanut brittle, peanut slab, dried fruit mixes, trail mix, baked goods (pastries, cookies), sweets, chili and spaghetti sauce (which may be thickened with peanut butter); chocolate spread, chocolate bars, egg rolls (may be sealed with peanut butter), hydrolyzed plant vegetable protein, vegetable burgers and sausages, nutmeats, ice cream, desserts, marzipan (which can be a mixture of nuts), and nougat. Foods most likely to contain hidden peanut products include cakes, cupcakes, fruit cakes, delicatessen cakes containing vegetable oil, cereal/granola bars, some breakfast cereal, granola, satay sauce, peanut sauce, confectionery, curries, salad dressings, breakfast cereals, sweet mincemeat, salad dressings, flavorings (natural), blended oils, and curries. Restaurant and takeaway meals may be a particular risk. Peanut shoots may be used in stir-fry dishes.
- Contamination can occur through the use of shared production lines or the same utensils. Manufacturers will commonly voluntarily identify on food labels if a product has been produced in the same factory as other foods containing peanuts to indicate possible cross-contamination. However, some of this advice may be disregarded by parents or patients. Research has indicated that many parents or patients are unlikely to purchase food products if the label indicated “not suitable for nut allergy sufferers” or “may contain nuts” but if the label stated “this product does not contain

nuts but is made in a factory that uses nuts," "cannot guarantee is nut free," and "may contain traces of nut" this would only be avoided by 50%. Large numbers of parents did not read labels for the presence of nuts in nonfood products. There is an assumption that risk is dependent on the wording of the label warning.[28] Teenagers in particular are shown to take risks with label interpretation and continue to eat foods labeled as "may contain" an allergen as they perceive the food is very unlikely to contain an allergen.[29]

- The peanut allergen appears to persist on the tabletop surface for at least 110 days if no cleaning occurs.[30] Therefore, regular cleaning of surfaces before and after eating should be reinforced as a safety measure for all individuals with peanut allergy.
- Foods sold in catering outlets or at in store bakeries and delicatessen counters, are generally unlabeled and so are problematic. Also there is inadequate knowledge about peanut allergy in restaurant staff.[31] A study undertaking chemical analysis of the peanut content of meals sold on food premises found that one in five provided a meal that could have triggered a fatal reaction. Most "front of house" staff did not check the allergy status of the meal with those doing the cooking.[32]
- Even cases of peanut allergy reactions via kissing are reported.[33] A study assessing the risk of peanut antigen in the mouth, found one hour after ingestion of peanut butter that 13% of participants had detectable levels.[34]
- Cosmetics, shampoos, medications, creams, and other topical products may contain arachis oil.

TABLE 70-5 Peanut Sources

Peanut flour, peanut meal
Peanut butter
Peanuts
Beer nuts
Mixed nuts
Ground nuts
Monkey nuts
Earth nuts
Goober nuts/peas
Mandalona nuts
Cold, pressed, expressed or expelled peanut oil
Arachis oil

TREE-NUT-FREE DIET (Table 70-6)

- Tree-nut allergy is less clearly characterized than peanut allergy.[3] The prevalence of allergy to tree-nuts is about half that of peanut allergy but it is associated with fatal anaphylaxis.[35] Only about 10% of children achieve tolerance to their tree-nut allergy.
- Accidental exposure to tree-nut allergens are common[36,37] and are predominantly associated with eating food outside the home, particularly restaurants, school cookies, and confectionary.
- In the United States and United Kingdom, the most common tree-nut allergies are walnut, cashew, almond, pecan, Brazil nut, hazelnut, macadamia nut, pistachio, and pine nut.[37,38] It has been reported that 45% of tree-nut allergic patients react to more than one nut.[37]
- The most common tree-nut allergies reported are walnut, cashew, almond, pecan, Brazil nut, and macadamia nut.[37]
- 25%–50% of peanut sensitive individuals are also allergic to tree-nuts.[37,39]
- Tree-nuts are available in their natural state (with or without shells) as ingredients in dishes or processed into butters, flours, oils, pastes, milks, or coffees.
- The tree-nuts identified by European/US legislation on prepackaged product food ingredient labels are almond, hazelnut, walnut, cashew, pecan nut, Brazil nut, macadamia, or Queensland nut.
- Although not mandatory, many manufacturers also use voluntary labeling (may contain tree-nuts) to indicate possible cross-contamination with tree-nuts.
- The following foods may contain tree-nuts: vegetarian sausages and burgers, Caponata, salads (commonly as garnishes), marinades, barbeque sauce, pesto, baked goods, cakes, biscuits, granola bars, pastries, Baklava, Macaroons, Amaretto biscuits, confectionery, praline (confectionery products made with nuts), fudge, trail mix, cereals, ice cream, desserts, marzipan, mortadella, natural flavoring, nougat, Turron, Nutella (or other chocolate nut spread), sweet mincemeat, and flavored coffee. Salad dressing, flavored natural extracts (e.g., almond), and nut distillates/alcoholic extracts are potential sources of tree-nuts.
- Specialty oils such as walnut oil contain significant levels of protein and should be avoided.

- Artificial nuts (e.g., Nu-Nuts) are ground and reshaped tree-nuts and should be avoided.
- Many African, Chinese, Indonesian, Mexican, Thai, and Vietnamese dishes contain tree-nuts.
- Coconut, nutmeg, water chestnuts, butternuts are not in the tree-nut family. Coconut is classified as a fruit. Although allergic reactions to coconut have been documented, most people allergic to tree-nuts can safely eat coconut.
- Lotions and sunscreens may contain shea butter or other nut butter. Nut oils such as almond may be used for massage.

TABLE 70-6 Tree-Nuts

Acorns
Almonds/almond paste
Artificial nuts
Beechnuts
Brazil nuts
Cashews
Hazelnuts (filberts)
Hickory nuts
Macadamia nuts (Queensland nut)
Pecans (mashuga nuts)
Pine nuts (pignoli)
Pistachios
Shea nut, shea butter
Walnuts
Nut oils, e.g., walnut oil
Natural nut extract (e.g., almond, walnut)
Nut meal
Nut meat
Nut paste
Nut liquors (Frangelico, Amaretto, and Nococello)
Nut butter (e.g., almond butter, hazelnut butter, brazil nut butter, macadamia nut butter, pistachio nut butter, shea nut butter, karike butter) Gianduja: a sweet chocolate hazel nut paste

SHELLFISH-FREE DIET

- Shellfish allergy affects about 1% of the population with significant consumption of shellfish. About 60% of people with shellfish allergy develop it in adulthood and it is usually lifelong.
- Tropomyosin, a heat stable muscle protein, is the major allergen in shellfish.[40]
- Edible shellfish are usually divided into two categories: mollusks

and crustaceans. Mollusks, such as clams and mussels, have two shells, but also include the abalone, which has a shell covering and a soft under part; crustaceans such as the lobster have segmented bodies that are covered with armor-like sections of thick and thin shells.

- Elimination of the following ingredients is necessary:
  – Mollusks: abalone, calamari, clams, cockle, cuttlefish, escargot/snails, mussels, octopus, oysters, periwinkle, sea urchin, scallops, sea cucumber, squid, and whelk.
  – Crustaceans: barnacle, crab, crawfish, crayfish, ecrevisse, krill, lobster, shrimp, prawn, and crevette.
- Caution should be exercised at salad bars where cross-contamination may occur, as well as restaurants with deep fryers shared with shellfish. It may be found in fish stock, seafood flavoring (crab extract), oyster sauce, fish sauce, sea food dips, sushi, sashimi, surimi, and prawn crackers. In the United States, many pasta sauces contain crustaceans.
- A shellfish allergy is distinct from allergy to fish. Those who are allergic to shellfish do not necessarily react to fish and vice versa.
- Individuals with shellfish allergy should avoid seafood restaurants where cross-contamination may occur.
- When shellfish is cooked, its proteins may become airborne and for some sensitive individuals inhalation of the cooking vapors (especially grilling and on the barbecue) may cause a reaction. Some people have an allergy reaction just with the smell of shellfish.
- The medication glucosamine is derived from the outer coating of shellfish such as crustaceans. Sometimes, chondroitin sulfate is added, usually derived from shark cartilage.

FISH ALLERGY

- Fish is a relatively common and important cause of food allergy, presenting with a high rate of anaphylaxis. The prevalence of fish allergy is dependent on amount of fish eaten in the local diet. In Spain, it is the third most frequent allergy, after milk and eggs in children under 2 years of age.[41] Fish allergy affects about 0.4% of populations with significant consumption of fish.
- Most cases are IgE-mediated due to ingestion, contact, or

inhalation of cooking vapors.[41]

- The major fish allergen across fish species is parvalbumin, which is resistant to thermal and enzymatic degradation. There are reports of sensitization to multiple species (50%–92% of individuals) or monosensitization to a single fish type.[42]
- Cross-reactivity between crustaceans and fish is not reported, so those with fish allergy do not need to avoid shellfish. However, up to 50% of individuals may be sensitive to both shellfish and fish.[43]
- Fish is often found in Worcester sauce and Caesar salad, and is sometimes found in imitation crab products in the form of surimi, a processed food mainly from Alaskan Pollack. Fish is also prevalent in Asian cuisine, which uses fish-based stock for many dishes.

SULFUR DIOXIDE AND SULFITES

- Sulfur dioxide and sulfites are preservatives widely used in food industry as antibrowning agents, antioxidants, and preservatives.
- Sulfites can cause allergy-like symptoms in people with underlying asthma and allergic rhinitis and is as high as 3%–10%.[44] The severity of the reactions varies and steroid-dependent asthmatics, those with marked airway hyperresponsiveness, and children with chronic asthma appear at greater risk.[44] Skin reactions through both topical exposure and ingestion of sulfite additives can also occur.
- EU food labeling rules require prepacked food sold to indicate on the label if it contains sulfur dioxide or sulfites at levels above 10 mg/kg or 10 mg/L (or if one of its ingredients contains it).

Possible Sources of Sulfur Dioxide and Sulfites (Table 70-7)

Drinks: Bottled soft drinks and fruit juice, cordials, cider, beer, wine (including sparkling wine), tea.

TABLE 70-7 Food Additives Containing Sulfites

E220: sulfur dioxide
E221: sodium sulfite
E222: sodium hydrogen sulfite

E224: potassium metabisulfite
E223: sodium metabisulfite
E225: potassium sulfite
E226: calcium sulfite
E227: calcium hydrogen sulfite
E228: potassium hydrogen sulfite
E150b: caustic sulfite caramel
E150d: sulfite ammonia caramel

Other liquids: commercial preparations of lemon and lime juice, vinegar, grape juice, vegetable juices.

Fruit: dried apricots, fruit bars.

Commercial foods: canned vegetables, dried or precut potatoes, gravies, bottled sauces, dressings, guacamole, fruit toppings, maraschino cherries, pickled onions, sauerkraut, pickles, jams, maple syrup, sugar syrups, jellies, biscuits, bread, pie, pizza dough, soup mixes, potato crisps, tomato paste, and tomato puree.

Meats: delicatessen meats, mincemeat, sausages.

Other foods: gelatin, coconut.

- Sulfites may be present in hair colors or bleaches, skin lighteners, fake tanning lotions, shampoos, and shower washes, antiageing creams, facial cleansers, body washes, and moisturizers. They may be found in topical creams and ointments, eye drops, and parenteral medications such as adrenaline.
- There may be individual, varying tolerance to sulfite-containing foods. Some people with mild sensitivity may need to avoid foods with the highest sulfite content only.

LACTOSE-FREE DIET (Tables 70-8–70-12)

NB: This diet is not intended for those with cow's milk protein intolerance.

- The lactose-free diet is intended for individuals with lactose intolerance (with low levels of the enzyme lactase) or the IMD, galactosemia (requiring a lifelong lactose-free diet).
- Patients with lactose intolerance experience GI symptoms such as bloating, flatulence, abdominal pain, and diarrhea which may occur when undigested lactose reaches the colon and is fermented by gas-producing bacteria.[45]
- Galactosemia is a disorder of galactose metabolism causing abnormal glycosylation of glycoproteins and glycolipids, causing

liver failure and cataracts if untreated.

- Intolerance to lactose is individual and people will vary in their ability to absorb lactose-containing foods depending on the degree of lactase deficiency and the form of food substance in which the lactose is ingested.[46] Individuals with galactosemia require a lifelong low-lactose diet.
- Cow's milk contains 4.5%–5.5% lactose. All milk, milk products, and manufactured foods containing milk need to be avoided.
- Constituents of milk are also likely to be found in foods such as biscuits, some sweets, and tinned and processed meats. Many spreads (including reduced fat) and mayonnaise include milk components to improve their flavor. The lactose content of whey derivatives such as whey powder is high, comprising 70% of total solids. In contrast, the lactose content of casein is low, with the U.K. Casein and Caseinates Regulations[47] stating the maximum anhydrous lactose content of casein should not exceed 1% by weight.[48] Butter oil and ghee contain only minimal lactose, but the recommendation is to avoid these.
- Lactose powder is added to a diverse range of food products including bakery goods, confectionery, dry mixes, dried vegetables, and crisps. It is often added to prepared foods to prevent caking or as a coating. It may be used in bakery goods for enhancing browning and, as it has one sixth the sweetness of sucrose, it is added instead of sucrose to reduce the sweetness. It is commonly used as a filler and flowing agent in seasoning mixes for foods such as instant pot noodles. It may act as a carrier for flavors and seasonings and may be added to ham, bacon, and salami. It may also be added to some artificial tabletop or tablet sweeteners.

TABLE 70-8 Lactose Content of Foods[48]

Food	Lactose Content (g/100 mL or g/100 g)
Milk, skimmed	4.4
Milk, whole	4.5
Cream, single	2.2
Yogurt, low fat, fruit	4.4
Butter	0.6
Chocolate, milk	10.1
Ice cream, dairy	5.2

Lactofree milk (Arla)[a]	0.02

[a] Data analyzed by UK Galactosemia Support Group (unpublished data)

TABLE 70-9 Milk, Milk Products, and Milk Derivatives

Milk and milk products
Cow's milk, goat's milk, sheep's milk
Cheese, cream, butter
Ice cream, yogurt, fromage frais, crème fraiche
Chocolate
Milk derivatives
Skimmed milk powder, milk solids, milk protein, non-fat milk solids, separate milk solids
Whey, hydrolysed whey protein, margarine or shortening containing whey, whey syrup sweetener, casein, hydrolysed casein, sodium caseinate, calcium caseinate
Lactose
Buttermilk, butterfat, butter oil, milk fat, animal fat (may be butter), ghee, artificial cream
Cheese powder
Lactose as a filler may be used in:
Flavorings
Table top or tablet artificial sweeteners
Medicines

Milk Substitutes

Breast milk and cow's milk-based infant formulas are contraindicated because they contain lactose. It is important that infants and young children are provided with a suitable nutritionally adequate formula. Several factors influence the choice of milk substitute including its nutritional composition, the child's clinical and nutritional status, safety and palatability of the product, ease of preparation, cost, and availability.

Low-lactose Cheeses Allowed for Galactosemia

A small number of hard mature cheeses are very low in lactose and are considered suitable for galactosemia by the U.K. medical advisory panel for galactosemia. Most cheese types have high levels of casein (which contains no more than 1% lactose) and low levels of whey (which contains 70% lactose). In the United Kingdom, the following low-lactose cheeses are permitted in galactosemia: Emmental, Gruyere, Jarlsberg, Parmigiano Reggiano, and Grana Padano Italian Parmesans with an EU PDO (Protected Designation of Origin) seal on the label. All of these cheeses have been extensively tested for their lactose and galactose content[49] and have been shown to contain minimal lactose and galactose on repeated

analysis. *All other dairy cheeses are not permitted.*

Vitamin and Mineral Supplementation

- It is important to ensure that patients meet the reference nutrient intake (RNI) for all bone-related nutrients. Calcium intake is commonly low in galactosemia[50–52] and lactose-free diets.
- Wherever possible, calcium requirements should be achieved from food sources. Soy infant formula normally provides adequate calcium in the first one to two years if sufficient amounts are taken, but it may be necessary to change to a calcium-enriched soy drink (preferably supplemented with vitamin D), encourage low-lactose cheese, soy yogurts, sardines, mackerel, sesame seeds, lentils, and milk-free fortified bread to ensure adequate calcium intake.
- Dietary assessment should be performed annually to ensure sufficiency. If calcium intake is low, it is necessary to give a calcium supplement and vitamin D to aid calcium absorption.

Lactose in Medications

- Lactose is an excipient with properties that make it ideal as a tablet filler or binder and it is found in many medications including laxatives.
- Some toothpastes and shampoos may contain milk derivatives. No recommendation is given to avoid these. Although a little of these may be swallowed by young children, the amount is unlikely to be significant.

TABLE 70-10 Lactose-Free Diet Guidelines

Foods Allowed	Foods to Avoid
Soy milk and soy products Infant soy formula Liquid soy milk – calcium enriched (not before 1 year of age), rice milk, almond milk, hazelnut milk, potato milk, coconut milk, (cream and soy made from these milks). Soy cheese, soy yogurt, soy desserts	*Milk, milk products and milk derivatives* Cows, goat, sheep and other animal milks (including whole, skimmed, dried, evaporated and condensed milk, breast milk) Yogurt, cream, sour cream, crème fraiche, fromage frais, cream cheese, whipped toppings
Cheese Dairy free cheese	*Cheese* Some hard cheeses
Fats and oils	*Fats and oils*

Milk free margarine Vegetable oils Lard, dripping, suet Salad dressing without cheese or milk	Butter, many margarines and low fat spreads contain milk
Meat and fish Plain beef, lamb, veal, pork, wild game, poultry, liver, kidney, offal, fish, shellfish (fresh or frozen)	*Meat and fish* Many processed meat or fish products such as sausages, salami, frankfurters, burgers, pies, luncheon meat, pates, breaded or battered foods or in sauce may contain lactose Ham and bacon (lactose may occasionally be used as a flavor enhancer – see ingredient label) Breaded/battered/creamed fish
Eggs	*Eggs* Egg custards, crème caramels
Non-meat alternatives Peas, beans, lentils (dried, canned, or frozen e.g., dhal or hummus); all nuts and seeds, tofu, peanut butter.	*Non-meat alternatives* Quorn based products, roasted nuts
Cereal, flour, pasta All grains; wheat, oats, corn, cornmeal, barley, maize, sago, rye, tapioca, bulgar, quinola, millet, arrowroot. Pasta, spaghetti, macaroni, dried noodles, cous cous, rice (plain, boil in the bag, rice noodles) Flour: plain, self-raising; cornflour, rice flour, soy flour Custard power (original), semolina Carob	*Cereal, flour, pasta* Tinned pasta such as spaghetti hoops may contain cheese. Dried pasta mixes. Instant custard powders Pancakes, doughnuts
Breakfast cereals Most are suitable, e.g., Weetabix, Cornflakes, Rice Krispies	*Breakfast cereals* A few cereals may contain chocolate derivatives or milk (muesli and chocolate flavored—particularly) Instant cereals
Bread and yeast products Most bread is suitable Pitta, chapatti, water-based breads (Italian [focaccia, ciabatta]), French, Jewish rye, bagels, wraps, plain rice cakes, Pareve-Jewish bakery products.	*Bread and yeast products* Brioche, naan breads, milk bread, bread rolls, sweet rolls, waffles, muffins, crumpets, teacakes, croissants, may not be suitable.
Cakes, biscuits, crackers Oatcakes, Rye crackers	*Cakes, biscuits, crackers* Many cakes, biscuits and crackers contain milk. Cheesy crackers, cheese thins, cheese oatcakes, cheese cakes, commercial cake

	and cookie mixes.
Desserts Sorbet, jelly, soy desserts, soy ice cream, soy yogurt Homemade soy milk custard or rice pudding	*Desserts* Most desserts contain milk in some form. Ice cream.
Fruit All fresh, frozen, tinned or dried fruit All fruit juices	*Fruit* Fruit bars Chocolate and yogurt dried fruit
Vegetables All fresh, frozen, tinned or dried vegetables (without added milk or milk products) Most dried and tinned pulses, e.g., red kidney beans, chick peas, lentils Tomato paste and puree All vegetable juices Olives	*Vegetables* Baked beans and ready-made vegetable dishes such as coleslaw, potato salad, vegetables in mayonnaise and salad cream. Potatoes or vegetables: creamed, scalloped, mashed with milk or breaded/battered vegetables, packaged dried potato mixes, sauces with milk or cheese sauce.
Savoury snacks Plain popcorn, plain pretzels, plain potato and tortilla chips	*Savoury snacks* Butter popcorn, toffee/chocolate popcorn, ranch-style or cheese-flavored snack pretzels, flavored crisps may not be suitable because they contain cheese or lactose as a filler in the flavoring Dry roasted nuts and popcorn may contain milk/butter
Seasonings Herbs, spices, salt, pepper, vinegar, mustard	*Seasonings*
Soups, gravies and sauces Bouillon, broth, meat, or vegetable stock soups; bisques and chowders made with water or lactose free milk. Stock cubes/concentrates, gravy granules, ketchup, sauces made without milk, salsa, relish, pickles. Sauces made without milk.	*Soups, gravies, and sauces* Tinned, packet, carton soups may contain lactose. Creamed soups. Gravy granules and stock cubes may contain milk Cook–in sauces, guacamole Mayonnaise and salad cream
Sugar, sweet spreads Sugar, honey, jams, jellies, maple and corn syrup, fruit based pouring syrup, molasses.	*Sugar, sweet spreads* Powdered and tablet artificial sweeteners may contain lactose
Confectionery Boiled or hard sweets/candy, most mints, marshmallow, plain fruit lollies, chewing gum, gum drops	*Confectionery* Milk chocolate, most plain chocolate, butterscotch and fudge, toffee Plain or carob chocolate
Drinks Soy milk	*Drinks* Drinking chocolate may contain milk

Milk shake syrup or powders Carbonated, squash, fruit juice Cocoa powder, tea, most coffee Flavored water	Instant milk drinks and malted milk drinks – avoid Instant iced tea, chocolate drink mixes, instant coffee (latte and cappuccino mixes) Milk based nutritional supplements Coffee whiteners
Miscellaneous – used in baking Baking powder, yeast, gelatine, marzipan Food essence and food coloring,	*Miscellaneous – used in baking* Sugar substitutes (sweeteners with added lactose) *Flavorings* Lactose may be used as a 'carrier' for flavorings particularly in crisps and similar snack foods. In sweets lactose is rarely used for this purpose except in some dairy flavors.
Food labels should always be checked because ingredients might change.	

COW'S MILK PROTEIN-FREE DIET

- Cow's milk allergy is the most common food allergy in infants and children. It usually presents in infancy when cow's milk formula or weaning solids containing cow's milk are introduced. The incidence in infants is approximately 2%–3% in developed countries.[56]
- Cow's milk allergy is also seen in 0.5% exclusively breast-fed infants.
- 45%–50% children will outgrow cow's milk allergy by 12 months of age, 60%–75% by 2 years, and 85%–90% by 3 years.[57]
- Approximately 50%–60% infants and children have cutaneous symptoms, 50%–60% have GI symptoms, and approximately 20%–30% respiratory symptoms.[56] Symptoms are diverse and can occur immediately (IgE mediated) or may be delayed (non-IgE mediated).
- Cow's milk allergy should not be confused with lactose intolerance which can present with similar GI symptoms.
- Due to the high possibility that children will grow out of cow's milk allergy, a formal challenge should be considered around 12 months of age or at least by 2 years.

TABLE 70-11 Lactose-Free Formula and Drinks

Lactose-Free Infant Formula Suitable for Infants	
Infant soy formula	Nutritionally complete soy infant formulas, e.g., InfaSoy and Wysoy, are lactose free. One potential disadvantage of soy infant formula is that it is a rich source of isoflavones, particularly genistein and daidzein, and this was identified as a concern by the UK Food Standards Agency.[53]
Infant low-lactose formula	There are a number of low lactose and casein/whey hydrolysate infant formula. All low lactose and protein hydrolysate formulas based on cow's milk protein will undoubtedly contain some residual lactose and the amounts vary. Manufacturers of low lactose casein based formula declare their lactose content at <10 mg/100 kcal (420 kJ) as defined by the European Food Safety Authority (EFSA).[54] Whey hydrolysate formulas are likely to contain a higher concentration of residual lactose and are unsuitable.
Infant amino acid–based formula	These are suitable providing lactose-free infant formulas are used. There are some products designed for children from the age of 6 months which are not nutritionally complete. Weaning products based on L-amino acids and pre-gelatinized rice starch are available from the age of 6 months. These provide a source of supplemental nutrition but are not designed to replace infant formula.
Lactose-free drinks suitable for children >1 year of age	
Vegetable/cereal milks for children over 1 year of age	There are a number of commercial nutritionally enriched plant-, cereal-, and nut-based drinks that can be purchased in supermarkets, specialist shops, or on the internet. Most of them contain approximately 120 mg/100 mL calcium and 0.8 µg/100 mL vitamin D but are low in energy content. There are also nutritionally enriched follow-on soy milks designed for children >1 year with added calcium, iron, vitamin D, some B vitamins and an energy profile similar to cow's milk. Generally, commercial soy drinks cook similarly to cows' milk and are a useful substitute in lactose intolerance. Oat drink is excellent for cereals and savoury dishes, but quite strongly flavored for desserts. Rice drink is thinner and sweetener than cow's milk. It is useful for desserts and sweet drinks. In the UK, it is not recommended for children under 5 years of age, as it contains a trace of arsenic. Coconut milk can be used in most cooked dishes but has a strong coconut flavor.
Milks suitable for lactose intolerance but not galactosemia	
Lactofree	Lactofree *(* Arla), low-lactose dairy drink. About half the lactose is removed by filtration and the remainder is removed enzymatically. It contains a significant quantity of galactose, rendering it unsuitable for galactosemia[55] and for those with cow's milk allergy. It may be useful for lactase deficiency. Lactase enzyme drops, which reduce the lactose content of cow's milk, are also unsuitable for galactosemia and cow's milk allergy but may be useful in lactose intolerance due to lactase

deficiency.

TABLE 70-12 Lactose-Free Sources of Calcium

	Calcium (mg/100 mL/100 g)	Vitamin D (µg/100 mL/100 g)
Calcium/vitamin D–enriched soy drinks	100–140	0.8
Soy dairy-free yogurts	120	Variable
Low-lactose hard cheese	730	0.3
Whitebait	860	Not available
Sardines in oil/brine	500	5
Pilchards	250	14
Sesame seeds	670	0
Red kidney beans	100	0
Bread	170	0
Spinach	170	0
Apricots	73	0

Source: *Roe MA, et al*[48]

Milk Substitutes (Table 70-13)

Removing cow's milk from the diet of an infant whose sole source of nutrition is provided by milk requires careful dietetic management.

Breast-fed infants can react to food allergens secreted in their mother's milk. These sensitive infants can continue to breast-feed if their mother eliminates all dairy foods in her diet. This requires great commitment from the mother and careful dietary management to ensure dietary adequacy, particularly calcium.

It is important that infants who are formula fed commence a suitable nutritionally adequate formula. The choice of milk substitute will depend on the clinical history, the age of the infant, palatability, cost, availability, and cultural restrictions and will vary between individuals.

Older children will continue to require a milk substitute although they remain sensitive to cow's milk protein. A range of products based on soy, rice, oats, almonds, hazelnuts, coconut, and hemp are available. Products fortified with calcium are more suitable for children. See section on lactose-free diets for more detail.

Goat's milk and sheep's milk are often tried as an alternative to cow's milk but because of the high cross-reactivity between cow's milk and other

mammalian milks, they frequently cause similar symptoms.

TABLE 70-13 Cow's Milk-Free Infant Formulas

Type of Formula	Example	Additional Information
Hydrolyzed whey	Pepti (Cow and Gate) Pepti-junior (Cow and gate) Althera (Vitaflo) Alfare (Nestle)	Contains lactose Clinically lactose free / 50% MCT Contains lactose Clinically lactose free / 50% MCT
Hydrolyzed casein	Nutramigen Lipil (Mead Johnson) Pregestimil (Mead Johnson) Similac Alimentum (Abbott)	Clinically lactose free Clinically lactose free / 55% MCT Clinically lactose free / 33% MCT
Amino acid	Nutramigen Puramino (Mead Johnson) Neocate LCP (Nutricia) Aptamil Pregomin AS (Milupa)	Clinically lactose free Clinically lactose free Clinically lactose free
Hydrolyzed soy and meat	Peptide (Nutricia) MCT Peptide (Nutricia)	Clinically lactose free Clinically lactose free/ 75% MCT
Soy	Infasoy (Cow and Gate) Wysoy (SMA) Enfamil Prosobee (Mead Johnson)	Due to the the potential risk associated with phytoestrogens soy formulas are not recommended for infants under 6 months with the exception of infants who refuse hydrolyzed protein formulae or if a vegan formula is required (Food Standards Agency 2003)[53]

	Similac Soy Isomil (Abbott)

Milk-Free Diet (Table 70-14)

All cow's milk, cow's milk products, and manufactured foods containing cow's milk must be removed from the diet. The list of ingredients on tins and packets of processed foods should always be checked. Some sources of milk may not be obvious, so parents and carers need to be taught to recognize the ingredients that may indicate the presence of cow's milk. EU[14] (Directive 2007/68/EC) and United States (Food Allergen Labeling and Consumer Protection Act, 2004[11]) food labeling legislation state that milk should be clearly labeled on food packaging. Any foods containing the following ingredients should be avoided:

Milk	Casein and caseinates
Milk solids	Buttermilk
Nonfat milk solids	Butter
Milk powder	Margarine
Skimmed milk powder	Shortening
Cream	Whey powder
Cheese and cheese powder	Hydrolyzed whey protein
Lactose	Hydrolyzed whey sugar
Yogurt	Whey syrup sweetener

Most major supermarkets will provide a list of their own products that are free from milk but ingredients sometimes change, so checking the labels is still necessary.

Parents and carers should be educated on the importance of strict adherence to the diet. Children should also receive age-appropriate education to aid understanding and compliance. Education should include possible sources of cross-contamination, meal planning, and advice for school and childcare. Many favorite children's foods contain milk, so advice on suitable alternatives and meal plans is helpful (Table 70-15).

TABLE 70-14 Cow's Milk-Free Diet

Foods Allowed	Foods to Avoid
Milk substitutes Infants: breast milk, hydrolysate formulas, amino acid formulae, soy formulas (over 6 months only)	*Milk, milk products, and milk derivatives* Normal infant formulae Cows, goat, sheep and other animal milks (including whole, skimmed, dried, evaporated

Over 12 months: Liquid soy milk–calcium enriched, rice milk, almond milk, hazelnut milk, hemp milk, coconut milk	and condensed milk) Yogurt, cream, sour cream, crème fraiche, fromage frais, cream cheese, whipped toppings
Cheese Dairy-free cheese	*Cheese* All cow's, goat and sheep cheeses
Fats and oils Milk-free margarine Vegetable oils Lard, dripping, suet Salad dressing without cheese or milk	*Fats and oils* Many margarines and low fat spreads contain milk
Meat and fish Plain beef, lamb, veal, pork, wild game, poultry, liver, kidney, offal, fish, shellfish (fresh or frozen)	*Meat and fish* Many processed meat or fish products such as sausages, salami, frankfurters, burgers, pies, luncheon meat, pates, breaded or battered foods or in sauce may contain milk Breaded/battered fish and fish in sauce
Eggs	*Eggs* Scrambled egg, omelette, scotch egg, egg custards, crème caramels
Non-meat alternatives Peas, beans, lentils (dried, canned, or frozen e.g., dhal or hummus); all nuts and seeds, tofu, peanut butter.	*Nonmeat alternatives* Quorn-based products, roasted nuts
Cereal, flour, pasta All grains; wheat, oats, corn, cornmeal, barley, maize, sago, rye, tapioca, bulgar, quinola, millet, arrowroot. Pasta, spaghetti, macaroni, dried noodles, cous cous, rice (plain, boil in the bag, rice noodles) Flour: plain, self-raising; cornflour, rice flour, soy flour Custard power (original), semolina Carob	*Cereal, flour, pasta* Tinned pasta such as spaghetti hoops may contain cheese. Macaroni cheese Dried pasta mixes. Instant custard powders Pancakes, doughnuts
Breakfast cereals Most are suitable, e.g., Weetabix, Cornflakes, Rice Krispies	*Breakfast cereals* A few cereals may contain chocolate derivatives or milk (muesli and chocolate flavored – particularly) Instant cereals
Bread and yeast products Most bread is suitable Pitta, chapatti, water-based breads (Italian [focaccia, ciabatta], French, Jewish rye, bagels, wraps, plain rice cakes, Pareve-Jewish bakery	*Bread and yeast products* Brioche, naan breads, bread sticks, garlic bread, milk bread, bagels, bread rolls, sweet rolls, waffles, muffins, crumpets, teacakes, croissants, may not be suitable.

products.

Cakes, biscuits, crackers Homemade biscuits and cakes made with cow's milk free ingredients Oatcakes, Rye crackers	*Cakes, biscuits, crackers* Many cakes, biscuits and crackers contain milk. Cheesy crackers, cheese thins, cheese oatcakes, cheese cakes, commercial cake and cookie mixes.
Desserts Sorbet, jelly, soy desserts, soy ice cream, soy yogurt Homemade soy milk custard or rice pudding	*Desserts* Yogurt, milk puddings, ice cream, fromage frais, mousse, instant desserts, blancmange, pies, crumbles and sponges
Fruit All fresh, frozen, tinned or dried fruit All fruit juices	*Fruit* Fruit bars Chocolate and yogurt dried fruit Fruit pie fillings
Vegetables All fresh, frozen, tinned or dried vegetables (without added milk or milk products) Most dried and tinned pulses, e.g., red kidney beans, chick peas, lentils Tomato paste and puree All vegetable juices Olives	*Vegetables* Baked beans and ready-made vegetable dishes such as coleslaw, potato salad, vegetables in mayonnaise and salad cream. Potatoes or vegetables: creamed, scalloped, mashed with milk or breaded/battered vegetables, waffles, potato shapes, wedges, packaged dried potato mixes, sauces with milk or cheese sauce.
Savory snacks Plain popcorn, plain pretzels, plain potato and tortilla chips	Savory snacks Butter popcorn, toffee/chocolate popcorn, ranch-style or cheese-flavored snack pretzels, flavored crisps may not be suitable because they contain cheese in the flavoring Dry roasted nuts and popcorn may contain milk/butter
Seasonings Herbs, individual spices, salt, pepper, vinegar, mustard	*Seasonings* Blended spices
Soups, gravies, and sauces Bouillon, broth, meat, or vegetable stock soups; bisques and chowders made with water. Stock cubes/concentrates, gravy granules, ketchup, sauces made without milk, salsa, relish, pickles.	*Soups, gravies, and sauces* Tinned, packet, carton soups may contain milk, creamed soups. Gravy granules and stock cubes may contain milk Cook-in sauces, guacamole Mayonnaise and salad cream
Sugar, sweet spreads Sugar, honey, jams, marmalade, jellies, treacle, maple and corn syrup, fruit-based pouring syrup, molasses.	*Sugar, sweet spreads* Lemon curd, chocolate spread, sandwich spread, peanut butter

Confectionery Boiled or hard sweets/candy, most mints, marshmallow, plain fruit lollies, chewing gum, gum drops Some plain chocolate, carob chocolate	*Confectionery* Milk chocolate, butterscotch, toffee and fudge
Drinks Milk substitute Milk shake syrup or powders Carbonated, squash, fruit juice Cocoa powder, tea, coffee Flavored water	*Drinks* Drinking chocolate may contain milk Instant milk drinks and malted milk drinks Instant iced tea, chocolate drink mixes, latte and cappuccino mixes Milk-based nutritional supplements Coffee whiteners
Miscellaneous – used in baking Baking powder, yeast, gelatin, marzipan Food essence and food coloring	*Miscellaneous – used in baking* Artificial sweeteners unless known to be milk free Blended spices

TABLE 70-15 Example of Meal Planning for a 5-Year-Old Child

Normal food intake	Milk-free menu
Breakfast	*Breakfast*
Weetabix with cow's milk	Weetabix with soy milk
Toast and butter with jam	Toast and milk free margarine with jam
Fruit juice	Fruit juice
Lunch	*Lunch*
Cheese sandwich	Chicken sandwich
Flavored crisps	Plain crisps
Chocolate bar	Milk-free cereal bar
Dinner	*Dinner*
Roast chicken	Roast chicken
Mashed potato with milk and butter	Mashed potato with soy milk and milk-free margarine
Vegetables	Vegetables
Yogurt and fruit	Soy yogurt and fruit

Nutritional Adequacy

Calcium: calcium supplements may be necessary if a calcium-enriched milk substitute is not taken or taken in adequate amounts. Other nondairy foods contain calcium but often cannot provide sufficient amounts to meet requirements. Table 70-12 illustrates other sources of calcium.[48]

Energy: a cow's milk-free diet can be low in energy. Dairy foods are major components of children's diets and provide a significant percentage of energy intakes. Elimination of milk, cheese, cream, milk-containing

desserts, snacks, and chocolate can lower energy intakes. Suitable alternatives need to be included and close monitoring of weight gain and growth is advisable.

Tablets and Medicines

Some tablets and medicines contain milk products. Parents and carers should be advised to check with their doctor and pharmacist to ensure medicines are suitable.

Cow's Milk Challenge

Due to the high possibility that children will grow out of cow's milk allergy, a formal challenge should be considered around 12 months of age or at least by 2 years. Informal challenges can sometimes occur at home before scheduled if children inadvertently or intentionally consume milk or a milk-containing food and this sometimes supersedes the need for a formal challenge. If there is a possibility of a severe reaction, the challenge should take place in hospital under medical supervision otherwise milk can be introduced gradually into the diet in incremental amounts to assess tolerance.

DIETS FOR GI DISEASE

In GI diseases, dietary manipulation and special diets are frequently the primary treatment. This may be a short course of treatment, a temporary requirement for a special diet, or sometimes a lifelong change.

CELIAC DISEASE

Celiac disease is an autoimmune condition triggered by a reaction to gluten. It is a relatively common condition with the prevalence estimated at 0.5%–1.0% in Europe. In children with celiac disease, ingestion of gluten damages the villi of the small intestine and the only treatment is a lifelong gluten-free diet.

Gluten-Free Diet (Table 70-16)

- Gluten is a protein found in certain cereal grains. All wheat, rye, and barley needs to be excluded from the diet, and in some cases oats. These grains form the base of bread, pasta, biscuits, cakes, and many breakfast cereals so excluding them from a child's diet

can be challenging. Oats contain a similar protein but are safe for most children with celiac disease but there remains controversy. Pediatric studies suggest no adverse effects but the quantity eaten may have been lower than prescribed.[57] Oats can generally be consumed but care must be taken to ensure oats are free from contamination with wheat, rye, or barley from the milling process.

- A gluten-free diet should be started as soon as possible after the diagnosis of celiac disease has been made. Until recently this diagnosis required endoscopic and histological confirmation. European and British guidelines suggest that endoscopy can be avoided in selected symptomatic children with raised tTG > 10 times upper limit of normal and either DQ2 or DQ8 HLA typing.[58,59]
- A gluten-free diet requires extensive dietary changes and parents and carers need careful education to ensure strict adherence. In turn, this leads to resolution of symptoms and normal growth and development.
- Many foods are naturally gluten free and are included in the diet freely. These include fresh meat, eggs, dairy products, fruit, vegetables, and other cereals including rice, maize, tapioca, and buckwheat.
- As well as the obvious sources (bread, pasta, biscuits, cakes, breakfast cereals), gluten may unexpectedly be found in processed foods where flours are used as thickening agents, fillers, and as carriers for flavorings. Other manufactured foods can become contaminated with gluten during production processes and may also need to be avoided as even the smallest amounts of gluten may cause symptoms and gut damage. As a result, all manufactured foods need checking to confirm they are gluten free. Similar foods made by different manufacturers may differ, so it is important that this is checked and the correct brands are used. Celiac UK produce a Food and Drink Directory annually for members that is regularly updated listing suitable gluten-free foods. This is available electronically and has recently been supplemented by a mobile phone app.
- Contamination can also occur during food preparation at home where gluten-containing foods are eaten by other members of the family. Advice needs to be given to avoid this including using separate bread boards for gluten-containing and gluten-free

breads, using a separate toaster or toaster bags, and avoiding contamination of butter and jam with knives and spoons. All surfaces and cooking utensils also need to be washed well to remove crumbs.

- A wide range of commercially produced gluten-free foods are available. These include staple food items such as breads, pasta, plain biscuits, crackers, and flour as well as other luxury items such as cakes, chocolate biscuits, pizza bases, cookies, muffins, cake mixes, and seasonal foods. In 2009, legislation reduced the permitted level of gluten in foods to be labeled as "gluten free" from 200 ppm (parts per million) to 20 ppm which came into effect in 2012.[60] In the United Kingdom, a range of gluten-free products are available free on prescription. The quantities available are often at the discretion of the prescribing doctor, but guidelines suggest the approximate monthly requirement for children of different ages.[61]

School and Nursery

The school should be made aware that a child has celiac disease, particularly for young children gluten-free school lunches can usually be arranged or packed lunches can be taken. Schools should ensure that children do not swap foods at mealtimes and particular care needs to be taken for any cooking lessons, school trips, or parties.

TABLE 70-16 Gluten-Free Diet

	Gluten Free	Check Ingredients	Gluten Containing
Milk and dairy products	Milk, cheese, cream, plain yogurt, plain fromage frais	Flavored yogurts and fromage frais, rice milk and soy milk, soft and processed cheeses	Yogurt with muesli or wholegrains
Meat and Poultry	Fresh meats and poultry, cured and smoked meats	Meat or poultry in sauce, processed meats eg sausages and burgers	Meat or poultry cooked in batter or breadcrumbs, breaded ham, faggots, rissoles, meat pie
Meat alternatives	Plain tofu	Marinated tofu, soy mince, Quorn products	

Fish and shellfish	Fresh fish and shellfish	Tinned fish, fish in sauce	Fish in breadcrumbs and batter
Eggs	Boiled, fried, smoked and scrambled egg, omelette		Scotch eggs
Fats and oils	Butter, cooking oil, ghee, margarine and low fat spreads		Suet
Fruit and nuts	Fresh, dried, canned and frozen fruit Plain or roasted salted nuts	Fruit pie fillings, dry roasted and coated nuts	Fruit pies
Vegetables	Fresh, frozen, dried and tinned vegetables	Baked beans, vegetables in sauce, cauliflower cheese, potato waffles and potato shapes, vegetable burgers,	Battered vegetables, samosas, vegetable pies, croquette potatoes
Flours and Grains	Buckwheat, corn, millet, rice, sago, tapioca, polenta, quinoa, Gluten free flour, gram flour, potato flour, chestnut flour, cornflour	Oats	Barley, wheat, rye, semolina, bulgar wheat, couscous, spelt, durum wheat
Bread, Cakes and Biscuits	Gluten free bread, rolls, pizza bases, muffins Gluten free cakes, biscuits and crackers Gluten free bread and cake mixes	Meringues, macaroons, rice cakes	Bread, cakes and biscuits made with wheat, rye or barley flour
Breakfast cereals	All cereals labeled as gluten free	Porridge oats, Corn and rice based cereals and those containing barley malt	Wheat-based cereals, muesli
Pasta and Noodles	Gluten free pasta and noodles, rice noodles and pasta		Fresh, dried, canned wheat pasta and noodles
Savoury snacks	Plain potato crisps, plain rice cakes, homemade popcorn	Flavored crisps, flavored popcorn and flavored rice cakes	Wheat rye and barley based snacks Pretzels
Soups and sauces	Homemade soups, mint sauce, Worcestershire sauce, tomato puree	Brown sauce, ketchup, ready-made mustard, mayonnaise, salad cream,	Soy sauce

		packet and jars of sauce, pickles, canned and packet soups, gravy, peanut butter, sandwich spread	
Desserts	Jelly, fresh and tinned fruit, homemade gluten free pies, puddings and desserts	Ice cream, mousse, instant desserts, custard powder, imitation cream	Sponge puddings, pies, semolina and macaroni pudding, ice-cream wafers and cones
Sweets and Chocolate		All sweets and chocolate, seaside rock	Chocolate bars containing biscuit
Beverages	Tea, coffee, milk, fruit juice, cordials, cocoa	Drinking chocolate, milkshake flavorings, vending machine hot drinks, fizzy drinks (especially if cloudy)	Horlicks, Ovaltine and other malted milk drinks, barley waters
Miscellaneous	Honey, jam, marmalade, golden syrup, treacle, yeast extract, vinegar, herbs, pure spices, cream of tartar, bicarbonate of soda, salt, freshly ground pepper	Mincemeat, Mustard powder, blended spices, baking powder, curry powder, ground pepper, stock cubes and powders, gravy granules	

Eating Out

Eating out at restaurants and takeaway food can be challenging for children with celiac disease. Popular children's foods such as pizzas, burgers, sausages, and breaded fish and meat products are unsuitable. Careful planning and discussion with the chef are needed to identify gluten-free menu choices and to ensure that there is no cross-contamination. Some restaurants offer the option to take in gluten-free pizza bases or gluten-free burger buns, but care must be taken that these are not contaminated by gluten-containing foods prepared and cooked in the same kitchen. Deep fat fryers used to cook gluten-containing foods should not be used to fry gluten-free options. Many chefs are happy to provide a gluten-free option if there is nothing suitable on the menu.

Adherence

Adherence to the diet is influenced by a number of factors including accurate information about the diet and the availability and cost of gluten-free foods. The dietitian needs to provide individual advice for children to aid compliance. If children are unwell at the time of their diagnosis, compliance is often helped when the gluten-free diet is associated with resolution of troublesome symptoms. Children who have less troublesome symptoms or who are identified through screening may find compliance harder, particularly if they feel no difference after commencing a gluten-free diet. Inadvertent or intentional exposure to gluten may cause abdominal pain and diarrhea, which acts as an aid to compliance. However, many children will have no symptoms after gluten consumption and find compliance harder to achieve. In these cases, discussions about the importance of compliance and the long-term risk of complications are needed. Education for the extended family may also be helpful.

Diabetes and Celiac Disease

Celiac disease is more common in children that have type 1 diabetes. Between 2% and 10% of people with celiac disease will also have type 1 diabetes. Usually, the diagnosis of diabetes is made first and later children are found to have celiac disease either through screening or investigation of GI symptoms. After diagnosis, absorption of nutrients improves, including carbohydrate, and so, blood glucose levels should be monitored closely and it may be necessary to adjust insulin doses.

Gluten Challenge

Routine rechallenge is not required if diagnosis is secure. If initial diagnosis is uncertain and the child is on a gluten-free diet, then further testing is recommended.[59] HLA testing can identify when a challenge may be required. If HLA-DQ2 or HLA-DQ8 is positive, a formal gluten challenge will be required for clarification of the diagnosis. A normal diet containing gluten should be consumed for three months, or a minimum of four to six weeks if symptoms are troublesome. It may be confusing for a young child to comprehend why they can temporarily eat gluten-containing foods when they usually have a gluten-free diet. This can potentially affect compliance if the restricted diet needs to be resumed after a challenge. For these children, gluten powder can be used for the challenge, with the child continuing to follow a gluten-free diet. The amount of gluten required will vary with age, but it is estimated that an intake of 10–15 g gluten a day is sufficient. Biopsy is repeated if serology

becomes positive or children are symptomatic.

Support Groups

There are a number of support groups worldwide. These groups provide educational materials, practical tips, and support and recipes for children and their families and also fund research into celiac disease.

United Kingdom: www.celiac.org.uk
United States: www.celiac.org
Worldwide: www.celiac.org/celiac-disease/resources/celiac-associations-around-the-world/

CARBOHYDRATE INTOLERANCE

Carbohydrate intolerance in infants and children may be a primary or secondary disorder. Malabsorbed carbohydrate results in diarrhea that may be mild or severe leading to dehydration.

LACTOSE INTOLERANCE

- Lactose is a disaccharide sugar present in milk. It is broken down by the enzyme lactase to glucose and galactose. Lactase enzyme is normally found in the intestinal brush border.
- Lactose intolerance is the result of a lactase deficiency and should not be confused with cow's milk allergy. Congenital lactase deficiency is rare and high levels of the enzyme are found in infants who rely on milk as a primary source of nutrition. After the age of 2, when breast-feeding or bottle-feeding has stopped, children are less reliant on milk and dairy products and lactase levels decline. In some population groups, particularly East and South East Asians, Africans, and native Americans, hypolactasia is significant enough to cause lactose intolerance. Secondary lactose intolerance can occur at any age and results from an injury or insult to the intestinal mucosa and is often transient. This commonly occurs after gastroenteritis but also in other GI disease states such as celiac disease and inflammatory bowel disease.
- Symptoms of lactose intolerance result from maldigestion and fermentation of lactose and include diarrhea, bloating, flatulence and acidic stool, and perianal excoriation.

- Dietary lactose restriction is required to manage the symptoms of lactose intolerance, although commercially produced lactase substitutes are available. Details of the lactose-free diet are covered earlier.
- Commercial lactase substitutes are available to help digest the lactose in food and drinks. The lactase substitute comes in liquid form and can be taken before a meal or added to milk. Both lactase enzyme drops and capsules are available from most health food shops.

SUCRASE ISOMALTASE DEFICIENCY

This is a rare autosomal recessive condition characterized by an absence of the disaccharidase sucrease and an absence or reduction in isomaltase activity. Malabsorption of carbohydrate results in diarrhea, distension, flatulence, and abdominal pain. Breast milk and normal infant formulae are tolerated well so presentation is usually at the time of weaning after the introduction of foods containing sucrose and starch. Diagnosis can be made with a positive hydrogen breath test or low-disaccharidase activity in intestinal biopsy specimens.

TABLE 70-17 Sucrose Content of Fruits and Vegetables

Low Sucrose (≤0.1 g/100 g)	Moderate Sucrose (0.2–0.5 g/100 g)	High Sucrose (>0.5 g/100 g)
Grapes, avocado pear, currants, raisins, sultanas,dates, pomegranate, lime, lemon, blackberries, blueberries, rhubarb, William's pear, kiwi, cranberries, lychees, gooseberries, Sharon fruit Asparagus, aubergine, baby sweetcorn, beansprouts, broccoli, cabbage, cauliflower, celeriac, cress, cucumber, endive, garlic, kale, lettuce, marrow, mushrooms, peppers, radish, frozen spinach, swede, tomato, watercress	blackcurrants, cherries, raw figs, grapefruit segments in juice, raspberries, strawberries, guava, Commice pear, fruit cocktail in juice Celery, courgettes, fresh and canned spinach, spring onions, turnip, pumpkin, yam, leeks, green beans, runner beans, lentils, fresh garden peas, mange tout, processed peas	Apple, banana, pear, orange, peach, nectarine, pineapple, honeydew melon, apricot, tangerine, Satsuma, plums, melon, dried figs Brussel sprouts, corn on the cob, frozen and canned corn, carrots, broad beans, onions, squash, frozen and canned peas, frozen mixed vegetables

Source: Holland B et al.[62]

- Dietary management usually requires a low-sucrose diet and a restriction in starch foods. The tolerance of starch is variable and starchy foods can be included by some children but restricted in others.
- Sucrose (white sugar) is added to many foods but is also found naturally in many fruits and vegetables. Sucrose is initially excluded from the diet to resolve symptoms then introduced to the level of tolerance. Added sugar should be avoided and sugary drinks, confectionary, cakes, biscuits, and desserts should also be excluded. Care should be taken to read the labels of manufactured foods, drinks, and medicines that may unexpectedly contain large amounts sucrose. Fruit and vegetables high in sucrose should be avoided and replaced with lower sucrose alternatives (Table 70-17).
- Starch restriction should be advised initially and then starch reintroduced to tolerance. An exchange system of measured amounts of starch-containing foods can be helpful to ensure tolerance is not exceeded. Tolerance increases with age and many older children consume a normal amount of dietary starch with no side effects.
- Patient noncompliance is common. Children may relax their diet in order to consume a wider variety of foods. They will tolerate some symptoms that are not dangerous but should be avoided if this practice leads to diarrhea and weight loss.
- Enzyme replacement therapy with Sucraid® offers a pharmacologic alternative to sucrose-free diets. Sucraid is an effective option for the management of sucrase deficiency that reduces the need for dietary restrictions and the symptoms of sucrose isomaltase deficiency.[63]

GLUCOSE–GALACTOSE MALABSORPTION

Glucose–galactose malabsorption is an inherited disorder characterized by the small intestine's inability to transport and absorb the monosaccharides, glucose and galactose. Infants fed breast milk or normal infant formulae containing lactose will present in the first few days of life with severe watery diarrhea, dehydration, and failure to thrive.

- The diarrhea stops when feeds are withheld, but unlike in other congenital and secondary carbohydrate intolerances, diarrhea will continue when the infant is given oral rehydration solutions

containing glucose.

- Dietary management requires the exclusion of glucose and galactose from the diet and all carbohydrate sources containing these monosaccharides (Table 70-18). This includes lactose, sucrose, dextrose, maltose, starch, and glucose polymer. Fructose is the only carbohydrate tolerated and infants should be swapped to the formula Galactomin 19 (Nutricia). Weaning should start at the usual time with very low carbohydrate foods. The diet is extremely restricted, although tolerance may increase slightly with age.

Oral rehydration solutions containing glucose cannot be given to infants and children with glucose–galactose malabsorption. If a child develops gastroenteritis and Galactomin 19 is not tolerated, it is important that either an oral solution containing fructose is substituted or intravenous fluids are given until feeds are tolerated.

All medicines should be free of carbohydrate. The ingredients should always be checked by the pharmacist before given.

CROHN'S DISEASE

Crohn's disease is a chronic incurable inflammatory disease of the GI tract. Approximately 25% of cases of Crohn's disease are diagnosed during childhood and adolescence.[64] Nutritional problems are common in pediatric Crohn's disease. Studies show weight loss in 88% and growth failure in 19% children at diagnosis.[65]

Treatment strategies in pediatric Crohn's disease are needed to manage acute exacerbations and for maintenance of remission and nutritional therapy has an important role both as a primary treatment and as an adjunct to medical and surgical management.

Exclusive Liquid Diet Therapy (LDT) is often the first-line treatment for children with active Crohn's disease (Table 70-19). Normal food is discontinued and replaced with a nutritional complete liquid formula usually for a period of six to eight weeks, but sometimes up to 12 weeks.[66] Full feeds are achieved in the first three to four days with an initial goal for volume based on expected energy requirements. This requires regular review and adjustment throughout the treatment to achieve timely nutritional rehabilitation and to satisfy hunger. A prospective study concluded that the median energy intake of children receiving an exclusive liquid diet for management of Crohn's disease was nearer 120% expected

requirements.[67]

TABLE 70-18 Low-Glucose/Galactose–Containing Foods

	Foods Allowed
Milk substitute	Galactomin 19 (Nutricia)
Protein Sources	Meat, poultry, fish, egg
Fruits	Avocado pear, rhubarb, lemon juice
Vegetables	Asparagus, bamboo shoots, beansprouts, broccoli, celery, cucumber, fennel, lettuce, mushrooms, marrow, spinach, watercress, spring greens
Miscellaneous	Fructose, sugar-free jelly, sugar free drinks, Marmite, Bovril, salt, pepper, herbs, spices

- All solid food should be stopped for the duration of the treatment to induce remission.[68] Some centers only permit additional water whereas others permit clear fluids, boiled sweets, chewing gum, and ice lollies.
- At the end of the course of treatment, food is reintroduced. There is no evidence to exclude single or multiple foods while reintroducing diet after LDT.
- Once in remission, it is important to maintain the nutritional benefits, prolong remission, and achieve prolonged linear growth. There is no evidence to suggest that eating or avoiding specific foods influences the severity of the disease or the frequency of relapses and a balanced diet with adequate calories, protein, fluid, and micronutrients is usually recommended.
- A high energy diet with food fortification, nutritional complete sip feeds, energy supplements, and homemade “milkshakes” can all be used to improve nutritional intakes. Overnight nasogastric feeding may also be necessary and has been shown to improve linear growth[69] and to maintain remission.[70]

INBORN ERRORS OF METABOLISM

Common IMDs treated by lifelong dietary management are responsible for a collection of diverse clinical conditions. Each condition may present at different ages with variable severity and outcome. Examples of dietary treatment orders will be given for the following disorders:

1. Avoidance/reduction of dietary precursors of toxic metabolites includes phenylketonuria (PKU), galactosemia (example of low-lactose diet given in section on lactose intolerance), and minimal fructose, sucrose, and sorbitol diet.
2. Modified low-fat diet, use of medium-chain triglyceride (MCT) supplements, and prevention of fasting for long-chain fatty acid disorders (very long chain acyl Co-A dehydrogenase deficiency, long chain acyl CoA dehydrogenase deficiency).

LOW-PHENYLALANINE DIET IN PKU

- Dietary treatment is essential in PKU (see Table 70-20).
- It aims to correct abnormal biochemistry by (1) decreasing the phenylalanine load on the affected pathway and (2) supplementation with tyrosine that is deficient due to the metabolic block.
- Diet may be the sole therapy or used in combination with the cofactor sapropterin.
- The diet is constructed from a combination of a phenylalanine-free L-amino acid supplements, a restricted and controlled amount of phenylalanine, and low protein foods (natural and specially manufactured foods), with appropriate vitamin and mineral supplementation. Application of dietary management varies between countries and centers.

TABLE 70-19 Suitable Products for Exclusive Liquid Diet

Type of Formula	Examples
Elemental formula:	Elemental 028 Extra (Nutricia)
Disease specific polymeric formula:	Modulen IBD (Nestles) Alicalm (Nutricia) *1 kCal/mL* Paediasure (Abbott) Fortini 1.0 (Nutricia) Frebini (Fresenius) Ensure (Abbott) Fresubin (Fresenius) Resource (nestles) *1.5 kCal/mL*
Standard polymeric sip feed:	Paediasure Plus (Abbot) Fortini (Nutricia)

Standard polymeric tube feed:	Fortisip (Nutricia) Resource Energy (Nestles) Ensure Plus (Abbot) *1 kCal/mL* Nutrini (Nutricia) Paediasure (Abbot) Frebinbi (Fresenius) Ensure (Abbot) Tentrini (Nutricia) Nutrison (Nutricia) Clinutren Junior powder (Nestles) *1.5 kCal/mL* Nutrini Energy (Nutricia) Paediasure Plus (Abbot) Frebini Energy (Fresenius) Tentrini Energy (Nutricia) Ensure Plus (Abbot) Nutrison Energy (Nutricia) Clinutren Junior powder (Nestles)

TABLE 70-20 Goals of Dietary Treatment in Phenylketonuria

To restrict foods high in natural protein to prevent excess accumulation of phenylalanine. Foods such as meat, fish, eggs, cheese, nuts and seeds are not permitted (except patients with a mild PKU phenotype).
To limit the amount of natural protein to maintain phenylalanine within the target treatment range. The types of natural food sources allocated for substrate amino acids are usually from cereal, potato and potato products, some vegetables and milk products.
Provision of phenylalanine-free L-amino acids to meet at least safe levels of protein/nitrogen requirements.
Provision of the conditionally indispensible amino acid tyrosine that may become deficient as a result of the enzyme block and dietary treatment.
To maintain a normal (but not excessive) energy intake by encouraging use of foods naturally low in protein and specially manufactured low protein foods such as bread and pasta.

Source: Adapted from MacDonald et al.[71]

High Protein/Phenylalanine Foods to be Avoided

Foods that are high in protein and so avoided include all kinds of meat, fish, eggs, cheese derived from animal milk (cow, goat, sheep), nuts, seeds, foods made from flour or Quorn (a meat substitute made from protein of fungal origin), and anything containing aspartame sweetener (aspartame consists of phenylalanine and aspartate linked together, and is metabolized to these separate amino acids). Sweets and jellies containing gelatin should also be avoided.

Phenylalanine Tolerance and Prescription

- Phenylalanine tolerance is the amount of phenylalanine that maintains the blood phenylalanine concentrations within a target treatment range. The amount of dietary phenylalanine tolerated from natural food sources will vary depending on the severity of the patient's phenotype, age, target blood phenylalanine concentrations, and adjunct drug therapy. The amount of phenylalanine from breast milk, infant formula, or food should be systematically titrated until phenylalanine is stable and within the target treatment range. Most patients with severe PKU tolerate between 4 and 10 g/d natural protein (150–500 mg/d phenylalanine).
- Systems for allocating phenylalanine vary throughout Europe.[72] Some countries allocate phenylalanine in daily amounts where all phenylalanine-containing foods are estimated in the diet.[72] In the United Kingdom, the amino acid is allocated in the form of a 50 mg phenylalanine exchange system (50 mg ≅1 g natural protein). The protein content of most foods contain 5% phenylalanine (vegetables and fruits are an exception, with the protein content in most vegetables containing 3%–4% phenylalanine and 3% in fruits).[73] Other countries may use a variation of the phenylalanine exchange system.[72]
- Exchange foods are generally sourced from low-biological protein foods such as potatoes, peas, sweet corn, rice, pasta, and breakfast cereals, although sometimes milk products such as yogurt and cream cheese may be used depending on phenylalanine tolerance. It is recommended that "counted" sources of phenylalanine are weighed.

TABLE 70-21 Foods That Can be Eaten Without Measurement/Restriction in PKU

Food Groups	Examples of Suitable Foods That Can be Eaten Without Measurement
Fruits and vegetables	Studies suggest the fruit and vegetables that can be given without measurement are those containing ≤ 50 mg/100 g phenylalanine[74,75,76] although practices vary from country to country.
Fats	Butter, margarine, ghee, lard, dripping and vegetable oils.
Starches	Cassava flour, arrowroot, cornflour, custard powder, potato starch,

	sago, tapioca, and tapioca starch. Some protein-free low protein cheese replacements based on food starches and oils are available.
Sugars	Sugar, glucose, jam, honey, marmalade, golden syrup, maple syrup, fruit sorbets, ice lollies, sweets containing <0.3 g protein/100 g.
Miscellaneous	Vegetarian jelly, agar-agar, salt, pepper, herbs, spices and vinegar; food essences and coloring; fruit/vegetable 'table top' sauces and 'cook in sauces' containing <1.0 g protein/100 g.
Drinks	Water, squash, lemonade, cola drinks and fruit juice, black tea, fruit tea, green tea, coffee, tonic water, soda water and mineral water, providing all are aspartame free. Rice drink is very low in protein but is unsuitable for children under 5 years of age due to trace amounts of arsenic. Other plant drinks e.g., oat milk, contain some phenylalanine which should be calculated in the diet.
Low-protein special foods	A selection of low protein breads, flour mixes, pizza bases, pasta, biscuits, egg replacers, and milk-replacements are available. Other low-protein specialist products contain some natural protein/amino acids e.g., low protein cheese substitutes, snack pots, burger mixes, milk replacements, but can be incorporated into the diet but as part of the natural protein exchange system. Any special food containing ≥0.5 g/100 g of protein should be calculated into the natural protein allowance.

Foods That Can Be Eaten Without Measurement

There are some foods that are naturally very low in protein so can be eaten without measurement (Table 70-21). It is a good practice to encourage five portions of low-phenylalanine fruits and vegetables daily. The range and availability of low-phenylalanine vegan foods (through supermarkets and the internet) is extending, thereby, increasing the choice of foods available.

Phenylalanine-Free L-Amino Acids

- The provision of additional phenylalanine-free L-amino acid supplement is essential to prevent protein deficiency and optimize metabolic control. In the majority of patients, *it is likely that precursor-free amino acids will supply >75% of the total protein intake.*[77] The optimal protein intake in PKU, accounting for inefficiency of precursor-free amino acids, is not agreed.
- It is best to give L-amino acids in small frequent doses, three to four times spread evenly throughout the day rather than only once or twice daily,[78] and ideally together with natural protein and a carbohydrate source.
- Most supplements contain additional vitamins, minerals, and

trace minerals. Many also contain essential fatty acids and omega-3 LC-PUFA's such as docosahexaenoic acid (DHA). If phenylalanine-free L-amino acids do not contain micronutrients, supplements should be administered separately, although adherence may be suboptimal.[79]

- Blood phenylalanine concentrations are monitored regularly to ensure they are maintained within target treatment range. Growth, clinical, and biochemical nutritional status is also monitored carefully.

VERY LOW-FRUCTOSE DIET FOR HEREDITARY FRUCTOSE INTOLERANCE

- Hereditary fructose intolerance (HFI) is a rare, inherited disorder of fructose metabolism, resulting from deficiency of aldolase B, the main enzyme responsible for hepatic metabolism of dietary fructose.[80] It is reported to affect 1 in 20,000 to 1 in 30,000 newborns in Europe.[80]
- Children with HFI develop symptoms after the introduction of fruits, vegetables, and particularly table sugar (the fructose–glucose disaccharide sucrose) to their diet. This is often between 4 and 8 months of age.[81] In HFI, accumulation of fructose-1-phosphate causes inhibition of glycogen breakdown and glucose synthesis, thereby, causing severe hypoglycemia following fructose ingestion. It can lead to hepatomegaly, jaundice, bleeding tendency, and renal tubular dysfunction.
- Some patients may not be diagnosed until later in life. Older patients may develop an aversion to fructose-containing foods after experiencing abdominal pain, nausea, and vomiting after fructose ingestion. Some may remain asymptomatic into adulthood, with significant absence of dental caries and a positive family history as only disease indicators.[82]
- It is treated by a very strict exclusion of dietary fructose, sucrose, and sorbitol but it is not possible to exclude these sugars completely (Tables 70-22 and 70-23).
- Different thresholds of fructose intake leading to the development of symptoms vary from 40 to 250 mg/kg/d.[81] Some have suggested it should be as little as 1500 mg/d.[83] The diet suggested in Table 70-24 provides less than 2 g/d.[84] The average

amount of fructose consumed in a “normal” Western society diet is 1–2 g/kg/d.

- Sucrose, sorbitol, and artificial sweeteners may be used in medicines, tablets and syrups, and toothpastes.
- Starch, glucose, and lactose can be included in the diet. Glucose can be used as an alternative sweetener, although many individuals with HFI dislike the taste and avoid sweet tasting foods.
- Cooking of vegetables causes a loss of free sugars, and they therefore have a lower fructose content than raw vegetables. Newer potatoes have higher fructose content than older potatoes.[84]
- Sucrose and fructose may be used as a carrier for flavorings.
- Individuals with HFI are at risk of vitamin C and possibly folic acid deficiency due to the exclusion of the major sources of these vitamins (fruits and vegetables). Therefore, suitable supplementation of these vitamins is important.
- With a careful diet, people with HFI should remain symptom free and be able to live normal lives.
- Intravenous fructose and sorbitol are potential lethal sources of fructose.[85]
- HFI is not the same condition as fructose malabsorption or dietary fructose intolerance, which is a cause of GI discomfort. This is an uncomfortable but benign condition.[87]

TABLE 70-22 Dietary Sources of Fructose

Fructose	Fructose or fruit sugar is a simple monosaccharide found in many plants. Commercially, fructose is frequently derived from sugar cane, sugar beets and corn.
Sucrose	Sucrose, is composed of fructose and glucose linked by a glycosidic bond.
Sorbitol	Fructose can be synthesized endogenously from sorbitol (a polyol synthesized from glucose). Hepatic sorbitol dehydrogenase converts sorbitol to fructose.
Isomalt and Lycasin	Isomalt is an artificial sweetener. Its hydrolysis yields glucose, sorbitol and mannitol. Lycasin is a trade name for hydrogenated glucose syrup. It contains maltitol, derived from the hydrogenation of maltose.

LONG-CHAIN FATTY ACID DISORDERS

- Mitochondrial long-chain fatty acid oxidation disorders (LCFAOD) are a rare group of inherited metabolic autosomal recessive disorders due to enzyme deficiencies in fatty acid oxidation and transport proteins.[87] They require attentive diet therapy.
- These disorders *demonstrate an abnormal response to fasting adaptation.*[88] *They* lead to accumulation of fatty acids and a decrease in cell energy metabolism. *They* are an important cause of disturbed energy homeostasis.
- Fatty acids are a major fuel source for most tissues of the body, especially during fasting, when glucose supply is limited. They are the principal energy source for cardiac muscle and skeletal muscle in the resting state and during prolonged exercise. Fatty acid oxidation is also important for the production of ketone bodies.
- LCFAOD conditions vary in severity and may present at any time from the neonatal period to adulthood. The organs affected are heart, liver, and skeletal muscles.
- *Several disorders of long*-chain fatty acid oxidation have been described including:
  – Carnitine transporter deficiency
  – Carnitine palmitoyl transferase 1 (CPT1) and 11 (CPT11) deficiency
  – Carnitine acylcarnitine translocase (CACT) deficiency
  – Very long-chain acyl CoA dehydrogenase deficiency (VLCADD)
  – Long-chain 3 hydroxyacyl CoA dehydrogenase deficiency (LCHADD)
- Clinically, patients may present in infancy with acute life-threatening hypoketotic hypoglycemia, hypotonia, hepatic dysfunction, hyperammonemia, and cerebral edema. Cardiomyopathy is common. Chronically, there may be muscle involvement with hypotonia or exercise intolerance with or without cardiomyopathy and rhabdomyolysis. In milder variants, skeletal myopathy with intermittent myoglobinuria may develop later in life.
- The aims of diet therapy are too:
  – To minimize fat lipolysis
  – To reduce accumulation of toxic intermediates
  – Ensure normal growth

– To help maximize exercise tolerance

- These aims are achieved by (1) adhering to a limited long-chain fatty acid intake, (2) avoiding prolonged fasting by regular daytime feeding and, usually, overnight tube feeds (3) replacing long-chain dietary fat with medium-chain triglyceride fat (MCT Oil), and (4) during illness, administering emergency, high carbohydrate feeds throughout 24 hours.
- A low-fat, high-carbohydrate diet with limited long-chain fatty acid intake (10% of total energy) is beneficial.
- Addition of MCT oil treatment (providing 10%–30% of energy requirements) is reported to be beneficial with improvement in dicarboxylic aciduria and a normalization of the plasma level of long-chain acylcarnitines.
- Coordinating MCT supplementation (0.5 g/kg of lean body mass) with periods of increased activity will help improve exercise tolerance.
- Supplementation with linoleic acid and linolenic acid to provide minimal essential fatty acid requirements. DHA supplementation (100 mg/d) may help visual function, particularly with LCHADD.
- Prevention of fasting with frequent feeds is crucial.
- A daily multivitamin and mineral supplement that includes all fat-soluble vitamins is required.

TABLE 70-23 E Numbers That are Sources of Fructose and Should Be Avoided

E420: Sorbitol
E473 and E474: Sugar esters
E491–E495: Sorbitol esters, some sorbitol may be liberated
E953: Isomalt

TABLE 70-24 Low-Fructose Diet (Containing Less Than 2 g/d Fructose)

Foods Allowed	Foods to Avoid
Sugars, sweeteners and preserves Glucose, glucose polymers, glucose syrup, dextrose, lactose, starch, maltodextrin, malt extract, saccharin, aspartame	*Sugars, sweeteners and preserves* Sugar or sucrose (cane or beet)- white, brown, caster, icing. Fruit sugar, fructose, laevulose Honey, treacle, molasses Sorbitol, Lycasin, Isomalt Golden syrup, corn syrup, invert syrup, high fructose or isoglucose syrups,,

	hydrogenated glucose syrup Jam, marmalade, lemon curd.
Fruit Avocado, rhubarb (occasionally)	*Fruit* All other fruit and fruit products
Vegetables Cook, boil and discard water Seaweed, vine leaves, allowed freely Fructose from vegetables should not exceed 1–1.5 g/daily. *Group 1 (<0.5 g fructose/100 g)** Celery, globe artichokes, mango tout, mushrooms, spinach, watercress Beans: haricot, mung, red kidney Dried split peas, lentils Potato: old potato, plain potato crisps (potato waffle, potato croquette-check food label) *Group 2 (0.5–1.0 g fructose/100 g)** Aubergine, asparagus, beansprouts, broccoli, cabbage, cauliflower, courgette, cucumber, fennel, artichoke, leeks, lettuce, marrow, new potato, pumpkin, radish, spring greens, swede, turnip. Beans: black-eye, broad, butter, soy Marrowfat peas, processed peas canned in water, chick peas.	*Vegetables* Beetroot, Brussels sprouts, carrots, gherkins, green beans, kohlrabi, okra, onion, parsnip, peas, pepper, plantain, shallots, spring onion, squash, sweetcorn, sweet potato, tomato, tomato puree. Beans: green, French, runner Baked beans, tinned vegetables with added sugar, mayonnaise or salad cream, coleslaw Flavored crisps Pickles, chutney
Milk Cow’s milk, Cream, cheese, plain cottage cheese Natural yogurt	*Milk* Flavored milk, condensed milk, milk shake powders/syrups Liquid soy milks Aerosol cream Cheese with added ingredients e.g., nuts, fruit Fruit and flavored yogurt, fromage frais Ice cream
Eggs Allowed	*Eggs*
Meat and poultry All fresh meat and poultry If processed read label to check for added sucrose, fructose, or honey	*Meat and poultry* Processed meats which have added sucrose e.g., meat pastes, frankfurters, salami, pate, sausages, tinned meat. Tender sweet meats, e.g., ham Honey cured meals Readymade meat meals- possible sources are the gravy, sauces, vegetables, breadcrumbs, batter, pastry.
Meat substitutes Soy meat, tofu, Quorn	*Meat substitutes*

Fish Fresh and frozen fish, shell fish, fish tinned in brine	Fish Fish tinned in tomato sauce, fish paste, fish cakes, fish oil or fish fingers Ready-made fish meals, possible sources are the sauce, vegetables, breadcrumbs, batter, pastry.
Flour and cereals Flour (white in preference to wholemeal), buckwheat, cornflour, custard powder, sago, semolina, tapioca, oatmeal, barley Flaky pastry, shortcrust pastry (not sweetened).	*Flour and cereals* Bran, wheatgerm
Pasta and rice Spaghetti, macaroni, other pasta (white in preference to whole meal) Noodles, egg noodles Rice (white in preference to brown)	*Pasta and rice* Pasta tinned in tomato sauce Pot Noodle
Breakfast cereals Porridge, puffed wheat, Original Ready Break, Shredded Wheat (Kallo) puffed rice cereal	*Breakfast cereals* Most manufactured breakfast cereals
Bread and crackers White bread(pre-packed) Baker's bread- check if sugar is added to dough mixture. Cream crackers, Matzo crackers, water crackers, original Ryvita (not sesame), plain rice cakes, crumpets	*Bread and crackers* Wholemeal bread, sweetened breads e.g., malt bread, soda bread, currant bread NB: Sucrose is present in wheat germ and bran. Savoury snack biscuits
Cakes, biscuits and pastries Home-made using permitted ingredients and sweeteners	*Cakes, biscuits and pastries* All cakes, biscuits, and pastries
Desserts Home-made using permitted ingredients and sweeteners	*Desserts* Most desserts, e.g., jelly, meringue, mousse, gateaux, fruit pie, or crumble, ice cream, yogurts
Fats and oils Butter, margarine, vegetable oils, lard, suet	*Fats and oils*
Drinks Soda water, mineral water (without fruit flavor) Some low energy drinks may be suitable (check the label for fructose, sucrose, sorbitol, polyols, or comminuted fruits). Tea, coffee, cocoa, herbal teas (no sugar)	*Drinks* Fruit juices, vegetable juices, fruit squash, fizzy drinks, diabetic squash containing sorbitol or fructose, low energy drinks, tonic water. Drinking chocolate, malted milk drinks. Instant tea mixes, coffee essence

Confectionary	*Confectionary* Sweets, chocolate, toffee, jelly. Ice lollies, chewing gum, diabetic sweets (sweetened with fructose, sorbitol, or other polyols). Glucose tablets, e.g., orange flavor Lucozade tablets.
Gravies, sauces and soups Marmite, Bovril White or cheese sauce, made with milk, flour, fat and cheese only	*Gravies, sauces and soups* Gravy granules, stock cubes Bottled sauces and dressings, e.g., tomato ketchup, horseradish sauce, mint sauce, soy sauce Sauce mixes e.g., sweet and sour, curry Mayonnaise, salad cream All soups (packet, tinned, or fresh)
Herbs, spices, nuts and seeds Pure herbs, mustard and spices, salt, pepper. Sesame seeds Pumpkin and sunflower seeds maximum of 10 g/day total	*Herbs, spices, nuts and seeds* Nuts, peanut butter, marzipan
Baking products Baking powder, bicarbonate of soda, yeast, arrowroot, food coloring, food essences, gelatin	*Baking products*

Source: Adapted from Dixon.[84]

TABLE 70-25 A Very Low-Fat Diet

Low-Fat Foods: Permitted	High-Fat Foods to Avoid
Milk and milk products Skimmed milk, very low fat yogurt, low fat cottage cheese, egg whites	Any other milk, yogurt, cheese, and egg yolks
Meat and fish Chicken, turkey (white breast meat) White fish, e.g., haddock, sole. Prawns	Fatty meat, duck, skin and dark meat from chicken and turkey, red meat, processed meats and meat products, meat pastries, meat in breadcrumbs and batter, meat paste or pate, ham Oily fish
Fats and oils MCT oil	Butter, margarine, vegetable oils, mayonnaise
Vegetables/fruit/nuts/seeds Pulses, fruit and vegetables, fat-free fruit crisps, potatoes mashed with skimmed milk, baked beans	Avocado pears, olives, potato fries/chips, processed potato, potato waffle, and crisps, nuts, seeds, vegetable burger, vegetable sausage
Cereals/breads White pasta, cous cous and rice, white	Naan bread, croissants, crackers, pastry, cakes, biscuits, Muesli, Cereal

flour, semolina and tapioca, white bread (in moderation), chapatti made without fat, 99% fat free crackers, rice cakes, Rice Krispies, Cornflakes.	containing added nuts
Sugar, desserts, confectionary. Jelly, meringue, marshmallows, ice lollies, sorbet, boiled sweets, jelly sweets, gummy sweets, fruit gums, jelly sweets, jam, honey, fruit dessert syrups, sugar	Chocolate, fudge, toffee, sweets containing oil, chewy sweets, chocolate spread, ice cream, milk puddings, cakes, biscuits/cookies made with ordinary fat/butter, sweet pastries
Drinks Fruit juice, squash, fizzy drinks, flavored water, tea, coffee	Hot chocolate, milk shakes, yogurt drinks
Soups, sauces, and spreads Clear soup/vegetable soup Low energy soups Stock cubes, ketchup, brown sauces Pickles in vinegar, vinegar Herbs and spices Cooking sauces (<0.5 g/100 g fat)	Cream of soups, meat soups, peanut butter, gravy made with meat sauce. Salad cream, mayonnaise Cream sauces, French dressing/vinaigrette, jar sauces.

Low-Fat Diet

- There is evidence that long- chain fat should be restricted to no more than 10% of total energy intake for LCHADD *and 10%–15% of energy intake for severe patients with VLCADD.* However, the degree of fat restriction varies according to the severity of the condition and age of presentation. By comparison, a normal diet contains on average approximately 35%–40% of its total energy from fat- containing foods.
- Foods that are permitted in the diet include skimmed milk, egg whites, chicken, turkey, white fish, fruit, vegetables, potatoes, pasta, rice, lentils, low fat yogurt, cottage cheese or minimal fat cheese (1% fat), jam, sugar, sweets, and fruit juices (Table 70-25).

MCT Oil

- Replacing long-chain fat with medium- chain fat is particularly beneficial for patients with LCHAD and VLCAD deficiency. It is thought that the metabolism of MCT bypasses the long- chain enzymes of the β-*oxidation spiral and only requires* β-oxidative enzymes with shorter chain-length for further breakdown and energy production. *MCT is converted to ketone bodies by the*

liver, which acts as an energy source for other organs including the brain and heart muscle. This may even help reduce the risk of heart complications. There is also evidence that MCT oil may reduce body fat breakdown and prevents the accumulation of potentially toxic chemicals.

- Between 10% and 30% of energy from MCT has been recommended for patients with LCHAD and VLCAD deficiency. MCT oil should be used in cooking. It may be used to make margarine, salad dressings, mayonnaise, cakes, biscuits, and chips helping make the diet far more palatable. However, this oil should not be heated to very high temperatures because it will oxidize and break down, affecting the taste and smell of food. Some children may take a special MCT supplement powder two or three times during the day.

MCT–Based Infant Formula

- Babies are usually given a nutritionally complete infant formula that is high in carbohydrate, low in long- chain fats, and supplemented with MCT oil. Some children may continue to take this product for many years either in their overnight tube feeds, bottles, or drink it via a cup.

Regular Feeds

- Children with LCFAOD deficiency require feeds at frequent intervals day and night. Frequency of feeding should be individually assessed according to severity of disorder and metabolic stability, but as a starting point, in children who present symptomatically in the first 1–2 years of life, feeds should be given three to four hourly during the day with continuous overnight tube feeding (either via a gastrostomy or nasogastric pump).

Exercise

- Exercise management also needs consideration. Fatty acid oxidation is increased during times of increased energy use (e.g., prolonged exercise). Some children will choose to limit their exercise activities but some who enjoy exercise may develop muscle pain and weakness following prolonged activities. MCT

supplements or glucose drinks before exercise in LCHAD patients is important.

Illness Management

- It is well recognized that poor food intake due to infection or energy losses due to vomiting or diarrhea causes rapid body fat breakdown, which can quickly cause metabolic instability. An emergency regimen should be started early in order to try and prevent or limit body fat breakdown. The first stage of any emergency plan consists of giving high carbohydrate drinks at least every three hours day and night or via a continuous tube feed over 24 hours. This ensures an adequate intake of carbohydrate and fluid. If high carbohydrate feeds are not tolerated, intravenous fluids providing 10% dextrose are administered immediately.

HIGH-ENERGY DIET IN GROWTH FAILURE

Growth failure and failure to thrive are terms that describe the condition of infants, children, and adolescents whose physical growth deviates significantly from standards or from their individual growth pattern. Nutritional support is the cornerstone of therapy for these individuals (Table 70-26). Oral nutritional support by a high-energy diet is indicated for promoting weight gain and catch up growth under the condition that the child can swallow safely and the GI tract is functional. Dietary advice to improve dietary intake from normal food can be accompanied or preceded by the use of oral nutritional supplements containing a balanced mixture of macro- and micronutrients. Problems such as severe loss of appetite, swallowing problems, or obstructive and motility problems of the GI tract may limit the use of oral nutritional support. Under these circumstances, more invasive nutritional support, in the form of enteral tube feeding or parenteral nutrition, should be considered. The patient should follow a well-balanced diet based on a wide variety of food from all the food groups. A high-energy intake could be achieved by increasing meal frequency and/or increasing energy density of the meals. Table 70-26 gives practical suggestions for increasing the energy content of the diet.

Practical Tips for Increasing Meal Frequency

- Eat small, frequent meals distributed throughout the whole day

- Add a small late meal before going to bed (in form of a snack or a high-energy beverage)
- Include snacks (e.g., chips, nuts, ice cream, chocolate bar)

Practical Tips for Increasing Energy Density

- Prefer foods with high energy content:
  – Full-fat milk and milk products (whole-milk yogurt, cheese)
  – High-fat meat or sausage, high-fat fish, for example, salmon, trout, mackerel, sardines
  – Peanut butter
  – High-energy beverages
- Add carbohydrate or fat to food
  – Maltodextrin or sugar in tea, fruit juice, lemonade, milk, or yogurt
  – Maltodextrin in soups, sauces
  – Cream in milk or milk products
  – Oil or butter to pasta, rice, vegetables

Recipes (Always Calculated for 500 mL)

Shake With Neutral Taste

100 mL cream (30% fat)
50 g maltodextrin
350 mL cow's milk (3.5 % fat)
Total kcal: 706

TABLE 70-26 High-Energy Diet

Food Group	Choose
Cereals, bread, pasta and other grain products, rice and potatoes	Breakfast cereals with cream and nuts Rice or pasta with butter or oil Potatoes as fried potatoes or chips Bread with generous butter or margarine
Vegetable and legumes	Vegetables with added butter, margarine, bacon or sauce, e.g., hollandaise. Dip sticks of carrot or pepper in mayonnaise or cream cheese.
Fruit and nuts	Prepare fruit salad with nuts and whipped cream. Add cream, full fat yogurt or vanilla sauce to fruit. Mix Fruit Smoothies with sugar and cream.

	Prepare avocado as a spread.
Beverage	Add sugar or honey to tea. Add maltodextrin to tea, fruit juice or lemonade.
Milk and milk products	High fat cheese or cream cheese Add cream to yogurt or curd cheese Full cream milk Milk shakes with cream, sugar or ice cream Use sour cream for cooking (e.g., soups, sauces) Cheese au gratin Add grated cheese to casseroles or pasta
Egg, meat, and fish	High fat meat Fried sausage, salami, bacon Fried breaded meat or fish Poultry with skin High fat fish (e.g., salmon, tuna, herring)
Fats and oils	Generous use of butter or margarine on bread. Generous use of vegetable oil for cooking and preparing dishes.
Sweets and snacks	Nuts and seeds Bars with or without chocolate Marzipan. Salty snacks like potato chips or peanut snacks Cakes with nuts, chocolate or cream Salty or sweet pastries
Formula/drinks	High-energy (≥ 1.5 kcal/mL), ready-to-use drinks based on milk protein in different flavors.

Chocolate Shake

100 mL cream (30% fat)
50 g maltodextrin
25 g cocoa
325 mL cow's milk (3.5 % fat)
Total kcal: 776

Fruit Shake

100 mL cream (30% fat)
50 g maltodextrin
120 g fruit (e.g., banana)
160 mL cow's milk (3.5% fat)
Total kcal: 735

CONGENITAL HEART DISEASE AND

HYPERLIPIDEMIA

Pediatric patients with congenital heart disease or hyperlipidemia require careful nutritional monitoring to ensure optimal growth and development. Growth failure in congenital heart disease is common and can be attributable to anorexia, hypermetabolism, frequent respiratory infections, decreased peripheral blood flow, malabsorption, or pulmonary hypertension, among other factors. Patients requiring dietary fat restriction because of hyperlipidemia are placed at greater risk for energy deficits, as well as deficits in calcium and iron. Nutrition education with frequent monitoring is necessary to prevent these deficiencies.

Fluid-restricted and sodium-restricted diets are often used in the management of congenital heart disease.

Fluid-Restricted Diet

The fluid-restricted diet is a modification of the normal diet in which all fluids are limited to a prescribed level. Individuals with renal, liver, or cardiac insufficiency might require it.

Noteworthy Points

- All foods contain water in varying amounts. All foods that are liquid at room temperature are considered to be fluids. These include water, tonic, coffee, tea, fruit and vegetable juices, milk, ice, ice cream, sherbet, Popsicles, fruit ices, gelatins, soups, gravies, sauces, cream, and alcohol.
- Fluid from solid foods is not routinely restricted in a fluid-restricted diet. Solid foods with the highest water content are fruits and vegetables. Excessive consumption of these foods should be avoided.
- Liquid medications and fluids consumed while ingesting medications must be accounted for in the fluid-restricted diet.
- The following measurements are useful in calculating daily fluid allowance:
  - 1 oz = 30 mL = two tbsp
  - 4 oz = 120 mL = half cup
  - 8 oz = 240 mL = one cup
  - 32 oz = 960 mL = four cups = 1 quart
  - 1L = 1,000 mL = 1kg

Sodium-Restricted Diet

The sodium-restricted diet is a modified, healthy, balanced diet. The sodium content is limited to a prescribed level. It may be indicated in children with hypertension, congestive heart failure, liver disease, renal disease, and corticosteroid therapy. Table 70-27 includes foods that should be preferred and avoided if a strict sodium restriction is recommended.

Practical Tips for Limiting Sodium Intake

- Do not use salt for cooking.
- Do not spread salt over food at the table.
- Prefer freshly prepared dishes instead of convenience foods that often contain a high salt content.
- Read labels carefully on all canned, frozen, and packaged foods. Avoid products with added sodium or salt. Ingredients are listed on the package by weight, from the largest amount to the smallest.
- Use low-sodium or sodium-free canned soups, broth, potato crisps/chips, crackers, and fresh nuts.
- Instead of salt try to flavor food with spices and flavorings such as herbs, onion powder, thyme, or lemon juice.
- Limit fast foods and restaurant foods that are high in sodium. They often contain added salt or salt-containing ingredients such as monosodium glutamate or soy sauce.

CHOLESTEROL-LOWERING DIET

Cardiovascular disease is associated with a variety of risk factors, including hypercholesterolemia.

Longitudinal studies report a reduction of low-density lipoprotein (LDL) cholesterol concentration of about 7%–15 % by dietary therapy.

In children and adolescents with hypercholesterolemia, from the age of 3 years dietary therapy including nutrition education by qualified dietetic experts should be the basis of treatment. No restriction of fat or cholesterol is recommended for children less than 2 years of age when rapid growth and development require high energy and nutrient intake per kilogram body weight.

Effects of Dietary Fats on Serum Lipoproteins

There are four types of fatty acids in the diet: saturated, polyunsaturated,

monounsaturated, and *trans* fatty acids.

Saturated fatty acids have the strongest influence on blood cholesterol. As they increase blood cholesterol the most, an important step is to limit the amount of saturated fat in the diet. Animal fats have a high content of saturated fatty acids. Examples are butter, shortening, meat fats (suet, lard, salt pork, bacon fat), chicken fat (including skin), whole milk, cheese, yogurt, and cream. But also some vegetarian fats have a high saturated fat content: for example, coconut oil or palm oil.

Polyunsaturated fatty acids lower total blood cholesterol, LDL, and high-density lipoprotein (HDL) cholesterol. A high content is found in corn, safflower, sunflower, soybean, cottonseed, walnut, and sesame oils, as well as fish, which contains long-chain omega-3 polyunsaturated fats.

Monounsaturated fatty acids lower serum LDL while maintaining or even increasing HDL cholesterol levels. Olive, canola, and peanut oil have a high content.

Trans **fatty acids** raise LDL and lower HDL cholesterol levels. They are found in fat of ruminant animals (e.g., milk fat, beef). They are also formed when liquid oils are hydrogenated into a solid form. So called partially hardened vegetable fats are found in margarine, cookies, crackers, pastries, baked goods, and many other processed foods, and snack foods.

Recommendations for Dietary Intake

The intake of saturated and *trans* fatty acids (from partially hardened vegetable fat or fat from ruminant animal) should not exceed 8%–12% of energy intake. The intake of fat should be mostly from monounsaturated fat (> 10% of energy); whereas the consumption of polyunsaturated fat should not exceed 10% of energy. In normal-weight children, total fat intake should not differ from the recommendation for healthy children (30%–35% of energy). A lower fat intake is usually not necessary and for some children and adolescents such a restriction may not provide sufficient energy for optimal growth and development.

Intake of dietary cholesterol should not exceed 150 mg/d in children and 250 mg/d in adolescents. Complex and slowly digested carbohydrates should be preferred. Soluble fiber (oat bran, psyllium), but not insoluble fiber (wheat bran), can contribute to reduced blood cholesterol. If triglyceride concentration is high, it is additionally necessary to decrease simple carbohydrate intake including glucose, saccharose, and fructose. However, it has to be considered that a diet high in fiber and low in sugars is less attractive for children and may lead to poor compliance with dietary therapy.

Due to the potential risk of insufficient energy intake, careful monitoring is necessary to ensure normal growth and development in all children. As no single food item provides all the essential nutrients in the amounts needed, choosing a wide variety of food from all the food groups will ensure an adequate diet.

Table 70-28 provides recommended intakes with the goal to prevent coronary heart disease. Table 70-29 includes foods that should be allowed and avoided in hyperlipidemia.

TABLE 70-27 Sodium-Restricted Diet

Foods Allowed	Foods to Avoid
Milk, milk products, and cheese Milk, fresh milk Yogurt Curd cheese (natural without salt)	*Milk, milk products, and cheese* Buttermilk Processed cheese Hard and soft cheese Curd cheese with added salt
Breads and cereals Low-sodium special breads, cereal products, bread mixes, rolls and crackers. Cooked cereals without added salt: pearl barley, cracked wheat, plain farina, rolled oats, and rice. Dry cereals: puffed rice, puffed wheat, shredded wheat; macaroni, spaghetti, noodles (cooked without added salt)	*Breads and cereals* Breads and rolls made with salt or commercial mixes; quick breads (pancakes, waffles, cakes, pastries, muffins) made from baking powder, baking soda, salt, monosodium glutamate or commercial mixes; quick cooking and enriched cereals that contain a sodium compound (read label); cornflakes and dry cereals (except those allowed); crackers (except low sodium)
Fruits and juices All fresh, frozen, canned, or dried to which salt, sodium benzoate, or sodium sulfite has not been added (read labels)	*Fruits and juices* Regular tomato and vegetable juices, maraschino cherries, any fruit or fruit product that contains salt or sodium benzoates, sodium-preserved dried fruits
Vegetables Fresh, frozen, or dietetic canned without salt: asparagus, green beans, dried lima beans, dried navy beans, brussels sprouts, cabbage, cauliflower, corn, cucumbers, parsley, parsnips, peas (fresh), peppers, potatoes, sweet potatoes, pumpkin, radishes, rutabaga, soybeans, squash (winter, summer), tomatoes, turnip greens	*Vegetables* All regular canned vegetables: beets, beet greens, carrots, celery, dandelion greens, hominy, kale, mustard greens, pickles, sauerkraut, spinach, white turnips, frozen peas or lima beans if processed with salt; commercially seasoned frozen vegetables, frozen mixed vegetables
Fat Lard, vegetable oil, shortening,	*Fat* Salted butter or margarine, bacon fat, salt

salt-free butter or margarine	pork, commercial mayonnaise or other salad dressing (except low sodium)
Sugar and sweets Sugar, honey, syrups, jams, marmalades, and jellies that contain no preservatives, marshmallows, chocolate marshmallow, gummy bear, ginger bread	*Sugar and sweets* Molasses, commercial jams containing sodium benzoate or other sodium preservatives (read labels)
Beverages Fruit juices, lemonade, milk (regular or low sodium as allowed), coffee, tea, coffee alternative, cocoa (made with allowed milk or water), mineral water with < 20 mg Na/L	*Beverages* Fountain beverages, “Dutch process” cocoa, instant cocoa mixes, powdered milk, prepared beverage, mixes including fruit-flavored powders, mineral waters with > 20 mg Na/L
Seasonings and condiments All spices, extracts, herbs, except those with sodium as an ingredient; lemon juice, vinegar, salt substitute (with doctor’s approval), low-sodium bouillon/stock cube	*Seasonings and condiments* Salt, celery salt, onion salt, garlic salt, prepared mustard, ketchup, horseradish prepared with salt, barbecue sauce, chilli sauce; meat extracts, gravies, and tenderizers; monosodium glutamate, soy sauce, Worcestershire sauce, regular bouillon/stock cubes, relishes, mixed pickles
Miscellaneous Unsalted nuts	*Miscellaneous* Canned, dehydrated, or frozen potato; spaghetti, macaroni, or noodle products (ready to eat); pre-prepared gravies; canned or dehydrated soups, except low sodium; salted popcorn, potato chips, pretzels, and other snack items such as corn chips or Doritos; party spreads and dips; regular baking powder, baking soda (sodium bicarbonate), rennet tablets, laxatives, Chinese/ Asian food, Italian food, fast foods; salted nuts, olives

Practical Tips for Lowering Cholesterol, Saturated and *Trans* Fat Intake

- Use only lean cuts of meat. Cut away any visible fat from meat before cooking.
- Remove skin from poultry.
- Do not fry foods.
- Eat no more than two egg yolks per week. Substitute two eggs whites for a whole egg in recipes. Use egg whites to batter meats and vegetables, coat with crumbs moistened with acceptable oil, and then bake. Use low-cholesterol commercial egg substitutes in

place of whole eggs.

- Read labels of commercially prepared products to identify hardened vegetable fats in cakes, spreads, and so on.
- Avoid fast fried foods that could be high in saturated fat (e.g., fried chicken, French fries).
- Increase fish intake, especially oily sea fish (herring, mackerel, salmon), to two servings per week.
- Try some meatless dishes, such as pasta with tomato sauce or vegetarian burgers.
- Use high-protein vegetables such as legumes (kidney, red, or pinto beans) and soy products (bean curd) to supply protein.
- Limit high-fat sauces such as cream, cheese, and Alfredo sauce.
- Eat low-fat dairy products daily.
- Encourage nuts or popcorn instead of chocolate or potato crisps as a snack.
- In case of high triglyceride levels:
  – Avoid sweetened drinks such as lemonade, fruit juices, and ice tea
  – Get used to drinking water, mineral water, or tea without sugar
  – Avoid large or frequent servings of foods with high sugar content such as cookies, candy, or ice cream

TABLE 70-28 Recommended Dietary Intakes of the European Society for Paediatric Gastroenterology, Hepatology, and Nutrition for Children (>2–3 y) and Adolescents

Nutrient	Recommendation
Total fat	30%–35% of energy intake
Saturated fatty acids	~8%–12% of energy or less
Trans Fatty acids	Low intake
Polyunsaturated fatty acids	~6%–10% of energy
Monosaturated fatty acids	No limitation
Cholesterol	~300 mg/d or less
Antioxidant vitamins	Generous intake
Sodium	Low intake
Complex carbohydrates	Generous intake

MINERAL-RICH DIETS

Children on any type of special diet (whether prescribed or self-imposed) may be at risk of deficiencies of vitamins and minerals due to restricted intake of certain foods or food groups. This in turn can contribute to delayed growth and development. As a result, nutrient intake should be monitored regularly by way of diet histories or food records and where appropriate nutritional biochemistry. A multivitamin and mineral supplement can be administered if the diet is lacking in several nutrients, or individual vitamins or minerals can be given to rectify specific deficits.

HIGH-CALCIUM DIET

Calcium is a mineral that is essential for the normal development and functioning of bones, teeth, muscles, and the nervous system. Growing children in particular have high requirements for calcium due to higher rates of bone formation. Most calcium in the body is stored in the bones and teeth with less than 1% circulating in the blood, muscles, and tissues.[89,90] Dietary deficiency of calcium does not usually result in obvious symptoms because the body maintains calcium levels in the blood by resorbing it from the bone. However, in the longer term, this may result in reduced bone density (osteopenia) and risk of bone fracture.

As milk and dairy products are the main source of calcium in the Western diet, any diet that omits or reduces the intake of these foods is likely to low in calcium. There is evidence to suggest that for diets habitually low in calcium, the body does adapt by improving the rate of absorption from foods. This is because calcium absorption decreases with increasing intakes. Other factors that aid absorption include Vitamin D (from sunlight and food sources), protein, and exercise. Conversely, phytic acid (found particularly in high-fiber breads and cereals and legumes) and oxalic acid (found in leafy green vegetables) may reduce absorption, although the effect in people eating a varied diet is minimal. High sodium intakes may also increase excretion of calcium.[91,92]

Dairy products, leafy green vegetables, and fish with soft bones (e.g., sardines, salmon) are the best food sources of calcium although bioavailability from vegetable sources is lower. The bioavailability of calcium from dairy products is about 30% compared with 5% for spinach due to the oxalic acid in the latter.[90] Many foods such as breakfast cereals are often fortified with calcium by the manufacturer and because they are consumed in relatively large amounts, they make a significant contribution

to calcium intake. For a list of foods high in calcium see Table 70-30. (See Appendix II for UK Dietary Reference Values, US Dietary Reference Intakes, and EU Reference Intakes).

Calcium supplements are best taken in divided doses as absorption of calcium decreases as intake increases.

Sources of calcium information:

[93]Dietary Reference Intakes for Calcium, Phosphorus, Magnesium, Vitamin D and Fluoride (1997). www.nap.edu

[94]British Nutrition Foundation. www.nutrition.org.uk

[95]Academy of Nutrition and Dietetics. www.eatright.org

[96]Food & Nutrition Board. Dietary Reference Intakes, 2001.

[97]Dietary Reference Values for Food, Energy and Nutrients for the United Kingdom, 1991.

[98]Nutrient and Energy Intakes for the European Community, 1993.

TABLE 70-29 Diet for Hyperlipidemia

Foods Allowed	Foods to Avoid
Fats and oils Rapeseed and olive oil are vegetable oils of the first choice (high amount of monounsaturated fatty acids). Soybean, wheat germ, peanut and walnut oil (ratio of n-6 to n-3 fatty acids: <7:1) in small amounts. Diet margarine (low in *trans* fatty and saturated fatty acids). Salad dressings made with vegetable oils listed above. Seeds and nuts—peanut butter, other nut butters	*Fats and oils* Coconut oil, palm kernel oil, palm oil Butter, lard, shortening, bacon fat Coconut Cocoa butter Dressings made with egg yolk, cheese, sour cream, whole milk
Breads and cereals Breads—whole grain (if accepted) Cereals—oat, wheat, corn, multigrain Pasta (100% Durum) Rice *Dry beans and peas* Low fat crackers Homemade baked goods using unsaturated oil, skim or 1% milk, and egg substitute e.g., quick breads, biscuits, cornbread muffins, bran muffins, pancakes, waffles	*Breads and cereals* Bread in which eggs are a major ingredient, croissants and other puff pastry Granola made with coconut Egg noodles and pasta containing egg yolk
Sauces	*Sauces*

Ketchup, barbecue sauce	Mayonnaise, cheese sauce
Fruits and Vegetables Fresh, frozen, canned or dried Fruit juice—fresh, frozen, or canned	*Fruits and Vegetables* Fruits or vegetables fried or prepared or served with butter, cheese, or cream sauce
Sweets, desserts and snacks (*if triglyceride level is normal*) Sugar, syrup, honey, jam, Candy made without fat (gumdrops, hard candy, marshmallow), fruit-flavored gelatin Frozen desserts - sherbet, sorbet, fruit ice, water ice. Cookies, cake, pie, pudding prepared with egg whites, low fat milk, and rapeseed oil or margarine. Popcorn, nuts	*Sweets, desserts and snacks* (*if triglyceride level is normal*) Candy made with chocolate, coconut oil, palm oil Ice cream and frozen treats made with ice cream Commercially baked pies, cakes, donuts, high-fat cookies, cream pies Chocolate, potato crisps/chips
Meat, poultry, and fish Beef, pork, lamb-lean cuts well trimmed before cooking Poultry without skin Processed meat - prepared from lean meat (e.g., turkey ham, tuna, wieners) Sea fish and low-fat freshwater fish	*Meat, poultry, and fish* Beef, pork, lamb - regular ground beef, fatty cuts, spare ribs, organ meats, sausage,bacon Poultry with skin, fried chicken Fried fish Regular luncheon meat (e.g., bologna, salami, sausage, wieners) High-fat freshwater fish Shellfish
Eggs Eggs whites (two whites equal one whole egg in recipes), cholesterol-free egg substitute	*Eggs* Egg yolks; includes egg used in cooking
Dairy products Low-fat milk (≤1.5% fat), skim milk (<0.1% fat) Buttermilk Yogurt /yogurt beverages - non-fat or low-fat Cheese - low-fat natural or processed cheese, e.g., part-skim mozzarella, ricotta Cottage cheese - low fat, non-fat, or dry curd (0%–2% fat) Frozen dairy dessert—low-fat or nonfat ice milk, frozen yogurt	*Dairy products* Whole milk (fluid, evaporated, condensed), Whole-milk yogurt, whole-milk yogurt beverages Regular cheese (American,Blue,Brie,cheddar, Colby, Edam, Monterey Jack, whole-milk mozzarella, Parmesan, Swiss), cream cheese, Neufchatel cheese Cottage cheese (> 4% fat) Ice cream, cream, half and half, whipping cream, nondairy creamer, whipped topping, sour cream

TABLE 70-30 Food Sources of Calcium

Calcium	Calcium

Food	mg/100 g
Milk and dairy	
Whole	118
Semi-skimmed (2% fat)	120
Skimmed/nonfat (<1% fat)	125
Buttermilk	120
Chocolate, whole milk	115
Soy milk, calcium enriched	89
Cream, double	49
Cream, single	91
Cheese	
Processed	610
Goats	133
Brie	256
Camembert	235
Cheddar	720
Cheddar, low fat Spread	840
Cottage	498
Cream	127
Edam	98
Blue, Stilton	795
Gouda	326
Mozzarella	773
Parmesan	362
Feta	1025
Ricotta	360
Soy	240
Frozen desserts	450
Ice cream, dairy	
Ice cream, non-dairy	100
Yogurt/dairy desserts	72
Plain, whole milk	
Fruit, whole milk	200
Plain, low fat	122
Fruit, low fat	160
Fromage fraise, plain	140
Fromage fais, fruit	110
Custard	86
Rice pudding	100
Fish	88
Anchovies, canned, drained	
Clams, canned, drained	300
	74

Food	mg/100 g
Meat and Eggs	
Eggs	57
Sausages, pork	110
Grain foods	
Crumpets	123
Pancakes, made with whole milk	118
English muffin	
Scones, plain	123
Waffles	186
Bread, white	267
Bread, granary	110
Biscuits, semi-sweet	209
Cereal[a]	120
Porridge (oats), made with milk	
Muesli, swiss style	128
Coco pops	110
Ready Brek	453
Baby rice cereal	1200
Vegetables/legumes	870
Broccoli	
Butternut squash	
Cabbage	56
Kale, boiled	41
Okra	54
Olives	150
Spinach, boiled	120
Spring greens, boiled	61
Turnip	160
Watercress	75
Fruits/juices	45
Apricots, dried	170
Blackberries	
Dates, dried	
Figs, dried	92
Orange	41
Raisins	45
Rhubarb	250
Sultanas	47
Currants	46
Other	93
Malted milk powder	64
Combination and fast foods	93
Macaroni cheese	
Lasagne	
Pizza, cheese and tomato	349

Crab, canned, drained	120	Quiche Lorraine	170
Cod, battered, fried	67	Cheese omelette	80
Haddock, battered, fried	180	Cheeseburger	230
Herrings, grilled	79	Sausage roll	231
Fish fingers, frozen	140	Quarter pounder with cheese	287
Oysters, raw	140	Chocolate mousse	85
Pilchards, in sauce	250	Chocolate, milk	100
Salmon, canned with bones	300	Molasses	110
Salmon, canned no bones	91	Treacle	97
Sardines, canned	540		220
Shrimp, canned, drained	110		550
Whitebait, in flour, fried	860		210
Nuts/legumes/seeds			
Baked beans	53		
Red kidney beans	71		
Haricot beans, boiled	65		
Soy beans, boiled	83		
Green/French beans	56		
Tofu, steamed	510		
Almonds	240		
Brazil nuts	170		
Hazelnuts	140		
Pistachio	110		
Walnuts	94		
Sesame seeds	670		

[a] Many breakfast cereals are enriched with calcium (check the label).

Source: Roe MA et al. [48]

IRON-RICH DIET

Iron is a mineral essential as a component of the protein hemoglobin in red blood cells, which transports oxygen to the muscles and tissues and takes carbon dioxide back to the lungs for excretion. Iron is also part of enzyme reactions that produce energy and has a role in the immune system.[94] About two-thirds of the body's iron is stored as hemoglobin, the remainder is found in the body organs and tissues in the form of ferritin.[99]

Insufficient dietary intake of iron leads to reduced levels in the blood and subsequently reduced levels of hemoglobin, which can lead to iron

deficiency anemia with symptoms of tiredness, lethargy, weakness, and decreased immunity. In children, it can affect cognitive function, concentration, and school performance. Frequent eating of nonnutritive substances such as ice, chalk, dirt, or sand (known as “pica”) can also be a sign of iron deficiency.

An iron-enriched diet may be recommended if there has been a diagnosis of anemia due to insufficient dietary intake of iron, excessive blood loss, or impaired absorption.

There are two forms of dietary iron: heme iron and nonheme iron. Heme iron is derived from hemoglobin and so is found in animal products. About half the iron in meat, poultry, fish, and seafood is heme iron and between 15% and 35% of this is absorbed.[100] Nonheme iron is derived from plant sources (cereals, vegetables, fruits) and makes up a larger portion of our iron intake but is less readily absorbed by the body (2%–20%)[100] and is more sensitive to inhibitors and enhancers of iron absorption. Nonheme iron absorption can be enhanced by consuming foods at the same time or with the same meal as foods containing Vitamin C (citrus fruits or juices, tomatoes, and green leafy vegetables) or heme iron (e.g., meat). Iron is also absorbed more readily when body stores are low, such that fussy eaters with poorer dietary intakes will absorb a higher percentage of available iron. Conversely, absorption may be impaired by phytic acid (found particularly in high-fiber breads and cereals and legumes), oxalic acid (found in leafy green vegetables), and the tannins found in tea. There is evidence to suggest that individuals may adapt to lower intakes of iron over time resulting in increased absorption and decreased losses.[101]

Full-term healthy babies are born with iron stores that last for the first 4–6 months of life. After this time, introduction of iron-enriched solid foods helps to ensure iron requirements are met. The iron in breast milk is well absorbed compared with that in formula milk; hence the iron content of the latter is much higher. Older infants, toddlers, and young children are particularly at risk of iron deficiency as requirements are high and fussy eating and a lack of variety in the diet can exacerbate the problem.

The main sources of iron in the Western diet are red meat, poultry, fish, cereal products, pulses and beans, green leafy vegetables, eggs, nuts, and dried fruit.

Iron supplements are available but the form of iron is less well absorbed than that in food sources, as a result, larger amounts need to be consumed and this can cause constipation and nausea in some people. As the amount of iron absorbed decreases with increasing intake, supplements

are best taken in divided doses.

For a list of foods high in iron see Table 70-31. (See Appendix II for UK Dietary Reference Values, US Dietary Reference Intakes, and EU Reference Intakes).

HIGH-FIBER DIET

Dietary fiber is a component of plant foods that is largely undigested by the acids and enzymes in the stomach or small intestine. Absorption of water by undigested contents increases the bulk and softens the stools, helping to prevent constipation. The breakdown of dietary fiber occurs mostly in the large intestine due to fermentation by bacteria, which produce short-chain fatty acids and gases (carbon dioxide, methane, and hydrogen). The bacterial mass and absorption of water contribute to stool bulk and consistency.

Different types of dietary fiber have different properties. Some absorb water in the intestines and are fermented by bacteria in the intestines, producing acids and gas. For example, pectins (found mostly in fruits, vegetables, and legumes), gums (mostly in oats, legumes, seeds and seaweed), some plant oligosaccharides (fructo- and galacto-oligosaccharides: found mostly in onions, Jerusalem artichokes, and chicory), and polysaccharides such as inulin (found mostly in vegetables, legumes, and fruits). These types of fermentable fiber help lower cholesterol by binding to bile acids and aiding their excretion and may also aid the absorption of some minerals, particularly calcium[102,103] in contrast to the effects of phytates.

Other types of less fermentable fiber absorb water and pass through the bowel largely undigested, providing satiety, contributing bulk to the stools, and speeding up transit time. For example, celluloses and lignin (found in grains, wheat bran, vegetables, legumes, fruits, nuts, and seeds). Resistant starch (found in legumes, whole grains, seeds, and potato) also passes through the small intestine undigested into the large intestine where it is fermented by bacteria. These types of dietary fiber have been associated with a reduced risk of diabetes.[104,105]

TABLE 70-31 Food Sources of Iron

Food	Iron mg/100 g	Food	Iron mg/100 g
Offal:		*Breads and cereals:*	

Heart, lamb[a]	6.0
Heart beef[a]	6.2
Heart pork[a]	5.3
Kidney, beef[a]	9.0
Kidney, lamb[a]	11.2
Kidney, pork[a]	9.2
Liver, beef[a]	7.8
Liver, veal[a]	12.2
Liver, chicken[a]	11.3
Liver, lamb[a]	7.7
Liver, pork[a]	17.0
Liverwurst[a]	6.0
Liver pate[a]	5.9
Meat:	
Beefburgers	
Beef, cooked	2.5
Chicken, dark meat	2.6
Chicken, light meat	0.8
Ham/bacon	0.4
Lamb, cooked	0.7
Pork, cooked	1.6
Turkey, dark meat	1.0
Turkey, light meat	1.2
Veal, cooked	0.5
Nuts and seeds:	0.9
Almonds	
Cashew nuts	
Coconut, desiccated	3.0
Hazelnuts	6.2
Peanuts	3.6
Peanut butter	3.2
Poppy seeds	2.5
Pumpkin seeds	2.1
Sesame seeds	11.1
Sunflower seeds	10.0
Tahini paste	10.4
Legumes/pulses, peas and beans:	6.4
Baked beans	10.6
Bean sprouts, mung, cooked	
Blackeye beans, cooked	
Butter beans, cooked	1.5
Broad beans, canned	2.2
Chickpeas, cooked	1.9
Haricot beans, cooked	1.7
Hummus	1.7
Lentils, cooked	

Bread, white	1.6
Bread, wholemeal	2.4
Bread, granary	1.9
All Bran	8.8
Bran Flakes	24.3
Cheerios	11.9
Coco Pops, Rice	7.9
Krispies	7.9
Cornflakes	23.3
Special K	18.2
Sultana Bran	11.9
Weetabix	1.4
Crumpets	1.3
Scones	6.9
Waffles	2.0
Soy flour	3.9
White flour	8.5
Wholemeal flour	28.0
Wheatgerm	20.0
Rusks	0.8
Baby rice cereal	1.4
Pasta, white, cooked	0.2
Pasta, wholemeal, boiled	0.5
Rice, white. boiled	
Rice, brown, boiled	4.1
Fish:	8.0
Anchovies	2.8
Clams	0.8
Crab, canned	5.7
Fish Fingers	0.8
Oysters	2.3
Salmon, canned	5.1
Sardines, canned	1.0
Shrimp, canned	5.1
Tuna, canned	
Whitebait	2.2
Other:	0.7
Egg	1.6
Infant formula, reconstituted	1.4
Chocolate mousse	10.5
Chocolate. milk	3.1
Cocoa powder	16.7
Malted milk drink, made up	1.4
Liquorice shapes	
Potato crisps	
Semi-sweet biscuits	
Molasses	
Treacle	

Peas, green, cooked	2.1	*Combination and fast foods:*	2.1
Red kidney beans	2.5	Hamburger	4.7
Soy beans, cooked	1.9	Big Mac	21.3
Split peas	3.5	Spaghetti bolognaise	
Tofu, steamed	1.5	Frankfurter with bun	1.2
Vegetables:		Chile con carne	0.9
Broccoli, boiled	2.5	Pizza, cheese and tomato	0.7
Chard, boiled	3.0	Quiche Lorraine	1.2
Kale, boiled	1.7	Chips	1.5
	1.2		1.1
	1.0		1.0
	2.3		0.8
	2.0		
Olives	1.0	Pea and ham soup	1.0
Spring greens, boiled	1.4	Lentil soup	2.0
Spinach, boiled	1.7		
Tomato, sun dried	1.9		
Watercress	2.2		
Fruit:			
Apricots	3.4		
Apricots, dried	4.1		
Dates, dried	1.3		
Figs, dried	4.2		
Peaches, dried	6.8		
Prune juice	0.7		
Prunes	2.2		
Mixed dried fruit	2.2		
Raisins	3.8		
Sultanas	2.2		

[a] Organ meats are high in cholesterol and should be eaten in moderation; liver and liver products should also be avoided.

Source: Roe MA et al.[48]

Foods high in dietary fiber also tend to have a lower energy density, may reduce hunger, improve satiety, improve glycemic control, and reduce the intake of other high fat foods, thereby reducing the risk of overweight and obesity. Dietary fiber that is fermented by bacteria in the bowel may also have beneficial effects on gut microflora and the short-chain fatty acids produced may help to regulate cholesterol and blood glucose levels and enhance the intestinal environment and immune function.[106]

Current fiber recommendations in the United States suggest for children an intake of 14 g/1000 kcal.[105] Whereas in the United Kingdom,

Dietary Reference Values [97] suggest children consume proportionately less than the 18–24 g/d recommended for adults. Williams et al[107,108] recommends for children over the age of 2 years an intake equivalent to the child's age in years plus 5 g/d. For example, a 5-year old should receive 10 g/d (5 g + 5 years).

Care does need to be taken that too much fiber is not consumed by children as the bulky nature of high-fiber foods can reduce appetite and subsequently reduce energy and nutrient intake.

During commercial processing, the fiber content of some foods, notably breads and cereals, can be significantly reduced. For this reason, wholegrain and wholemeal varieties are better sources of dietary fiber than white or refined. Raw and freshly cooked fruits and vegetables also tend to be higher in fiber than canned varieties.

Fiber supplements are available for those unable to take in sufficient fiber from food.

Tips for Increasing Dietary Fiber Intake in Children

- Include different types of dietary fiber including that from fruits and vegetables and wholegrain/wholemeal breads and cereals;
- Include raw fruit and vegetables with skins where possible;
- Increase fiber intake slowly to reduce the risk of excessive gas and bloating;
- Increase fluid intake, as an increase in fiber without extra fluid can contribute to constipation;
- If high-fiber foods are refused, continue to offer them, as children often need to be given the taste of new foods several times before they accept them;
- Give children choices about what to eat but make sure they are appropriate and healthy options;
- Add a few tablespoons of a high-fiber cereal to their favorite cereal;
- Add unprocessed bran to cereals, soups, pasta, casseroles, and desserts;

For a list of foods high in fiber see Table 70-32.

VEGETARIANISM

A vegetarian is generally defined as a person who does not eat meat or the flesh of any animal (including poultry and fish) and may include

abstinence from foods that are derived from animals such as eggs, milk, and dairy products. The choice to be vegetarian may be for cultural, religious, or ethical reasons such as the support of animal welfare. It may also be a lifestyle choice for health, environmental, or economic reasons. There are generally four types of vegetarians:

1. Vegans are vegetarians that eat no animal flesh or products derived from animals (including meat, poultry, fish, seafood, eggs, dairy products, and honey).
2. Lacto-ovo vegetarians consume eggs and dairy products but no meat, poultry, fish, or seafood.
3. Ovo-vegetarians consume eggs but no dairy products, meat, poultry, fish, or seafood.
4. Lacto-vegetarians consume dairy products but no eggs, meat, poultry, fish, or seafood.

TABLE 70-32 Food Sources of Dietary Fiber

Food	NSP g/100 g	Food	NSP g/100 g
Breads and grains:		*Vegetables:*	
Bran, wheat	36.4	Asparagus, canned	2.9
Bread, white	1.5	Beetroot, cooked	1.9
Bread, white, high fiber	3.1	Broccoli, cooked	2.3
Bread, wholemeal	5.0	Brussels sprouts	3.1
Bread, granary	3.3	Cabbage, raw	2.4
Bread, rye	4.4	Cabbage, boiled	1.8
Crackers, wholemeal	4.4	Carrots, raw	2.4
Crackers, oat based	3.5	Carrots, boiled	2.3
Cracker, rye	11.7	Carrots, canned	1.9
Flour, white	3.1	Cauliflower, raw	1.8
Flour, wholemeal	9.0	Cauliflower, boiled	1.6
Flour, rye	11.7	Celery, raw	1.1
Flour, soy	11.2	Celery, boiled	1.2
Pasta, white, cooked	1.9	Courgette, raw	1.2
Pasta, wholemeal, cooked	3.5	Courgette, boiled	0.9
Pastry, cooked	2.2	Cucumber	0.6
Pastry, wholemeal, cooked	6.3	Green beans	5.1
Rice, white, raw	0.5	Leeks, boiled	1.7
Rice, brown, raw	1.9	Lettuce	1.2
Cereals:		Mushrooms, cooked	1.5
All Bran		Mushrooms, canned	1.3
Bran Flakes		Onion, raw	1.4
Cheerios		Onion, fried	
Coco Pops, Rice Krispies	24.5	Parsnip, boiled	
Cornflakes	13.0	Peas, boiled	
Porridge (oats), cooked		Peppers, capsicum	

Shredded wheat	6.2	Potato, baked with skin	3.1
Special K	0.7	Potato, mashed	4.7
Sultana Bran	0.9	*Potato chips*	2.7
Swiss style muesli	0.9	Pumpkin, boiled	1.7
Weetabix	9.8	Spinach, boiled	1.1
Wheat germ	2.0	Spinach, canned	2.0
Cereal chewy bar	10.0	Spring greens, boiled	1.1
Legumes/pulses:	6.4	Sweet corn, boiled	2.1
Baked beans	9.7	Sweet corn, canned	1.6
Bean sprouts, mung, cooked	15.6	Sweet potato, boiled	2.6
Blackeye beans, cooked	3.2	Sweet potato, baked	2.2
Butter beans, cooked		Tomato, raw	1.5
Butterbeans, canned		Tomato, canned	2.3
Broad beans	3.7	Turnip, boiled	3.3
Chickpeas, cooked	1.3	*Fruit:*	1.0
Chickpeas, canned	3.5	Apple, raw	0.7
Haricot beans, cooked	5.2	Apple, stewed	1.9
Hummus	4.6	Apricots, dried	
Lentils, cooked	6.5	Apricots, raw	
Red kidney beans, cooked	4.3	Apricots, canned	1.8
Red kidney beans, canned	4.1	Avocado, raw	1.5
Soy beans, cooked	6.1	Banana	7.7
Nuts/seeds:	2.4	Blackberries, raw	1.7
Almonds	3.8	Dates, dried	0.9
Brazil nuts	6.7	Figs, dried	3.4
Cashews	6.2	Grapes	1.1
Coconut	6.1	Olives	3.1
Hazelnuts		Orange, raw	4.0
Peanuts	7.4	Mixed dried fruit	7.5
Pecans	4.3	Passion fruit	0.7
Peanut butter	3.2	Peaches, raw	2.9
Sesame seeds	13.7	Peaches, canned	1.7
Sunflower seed	6.5	Pears, raw	2.2
	6.2		3.3
	4.7		1.5
	5.4		0.8
	7.9		2.2
	6.0		
Other:		Pears, dried	8.3
Fish fingers	0.6	Pears, canned	1.4
Pancakes	0.8	Pineapple, raw	8.1
Pancakes, wholemeal	2.4	Pineapple, dried	1.2
Pizza, cheese and tomato	2.0	Pineapple, canned	0.5
Pizza, wholemeal cheese/tomato	3.2	Plums, raw	1.6
		Prunes	

Potato crisps	5.3	Raisins	6.5
Quiche Lorraine	0.7	Raspberries	
Quiche Lorraine, wholemeal	2.1	Rhubarb	2.4
Scones, plain	1.9	Strawberries	1.2
Scones, wholemeal, fruit	4.9	Sultanas	1.1
Semi-sweet biscuits	1.4		2.0
Shepherd's Pie	1.2		
Spaghetti bolognaise	0.9		

NSP, nonstarch polysaccharide

Source: Roe MA et al.[48]

However, vegetarian diets vary the world over and what is included or excluded from the diet is often based on individual preference. Some vegetarians, for example, may avoid red meat but will eat fish and seafood (pescetarians). A macrobiotic diet consists mostly of grains and beans and occasionally fish. Semi-vegetarians or flexitarians may eat meat but only occasionally. Many processed foods contain ingredients derived from animals, which some people may choose to avoid. For example, rennet in cheese (derived from stomach enzymes) and gelatin (derived from animal bones or tissues).

Initially, babies can be given breast milk or formula exclusively for the first 6 months and then as the main drink in addition to solid foods until 1 year. Alternatively, lacto-ovo and lacto-vegetarian infants can use standard cow's milk based-infant formula. Vegans and ovo-vegetarians will require a nondairy-based infant formula such as soy formula. Soy milk and rice milk are not appropriate before the age of 12 months as they contain insufficient protein and/or energy and micronutrients. After 12 months of age, infants can be weaned onto cow's milk or fortified (with calcium, Vitamins B12 and D) dairy-free alternatives such as soy milk. Solid foods can be introduced in much the same way as in nonvegetarian children, for example, starting with iron-fortified rice cereal. Substitute puree/mashed tofu or beans/lentils for meat and for vegans and ovo-vegetarians, soy-based milk, yogurt, and cheese.

If a wide variety of foods are eaten, a vegetarian diet should provide adequate nutrients for normal growth and development. However, there are some nutrients that require consideration.

Energy and Dietary Fiber

Vegetarian diets are often lower in energy than diets containing animal products and higher in dietary fiber. A high-fiber diet can be bulky and fill

children up before they have consumed sufficient energy. As children have high energy requirements, care needs to be taken to ensure adequate energy-containing foods are consumed for growth. This may mean using some refined grains, breads, and cereals rather than just wholegrain varieties. Fat-containing foods should not be limited (e.g., avocado, nut butters, nuts, vegetable oils, margarine) before the age of 2 years.

Protein

Vegetarian diets are often lower in protein and children have higher requirements per kilogram of bodyweight than adults. Although proteins from animal sources contain all the essential amino acids for growth, most plant foods (except soy beans, quinoa and hemp) are deficient or low in at least one essential amino acid. So eating a variety of plant foods is important to ensure all essential amino acids are consumed. Good sources of protein for vegetarians include soy beans and soy bean products (e.g., soy milk, tofu), legumes (e.g., lentils, baked beans, chickpeas), nuts (including peanut butter), seeds, grains (e.g., cereals, pasta, rice, and bread), manufactured meat substitutes (e.g., mycoprotein or Quorn, seitan) and if eaten, eggs, milk, and dairy products. Combining protein sources in a meal will maximize protein quality. For example: beans on toast, lentil soup with bread, peanut butter sandwich, cereal with milk, vegetable soup with lentils or barley, chilli con carne with rice, pasta and cheese, hummus with bread or crackers.

Calcium

Breast- and formula-fed infants will get sufficient calcium. However, after weaning, vegans and ovo-vegetarians may not receive sufficient calcium from foods due to the avoidance of dairy products. Vegetarians may also absorb less calcium due to the consumption of more plant products that contain oxalic and phytic acids. Calcium can be obtained from calcium fortified milk replacements such as soy milk and soy yogurt, tofu, rice or oat-based drinks, green leafy vegetables such as broccoli, bread, dried fruit, nuts, and seeds. Adequate intakes of protein and Vitamin D will aid absorption of calcium.[109]

Iron

Vegetarians are also at risk of iron deficiency as iron is less well absorbed from plant sources than it is from animal based sources. The lower bioavailability of iron in a vegetarian diet means that iron requirements

may be almost twice that of the average population.[111] However, a well-balanced diet should provide adequate iron from legumes, cereals, bread, leafy green vegetables, dried fruit, nuts, and seeds. Consuming these with a source of Vitamin C (e.g., orange juice, fruit, or vegetables) will help the body to absorb the iron from plant foods.

Zinc

Absorption of zinc, like iron, is impaired by phytates. Animal protein also aids absorption of zinc; therefore, the bioavailability of zinc on a vegetarian diet is lower than for nonvegetarians. However, a good intake of legumes, tofu, grains, fortified cereals, and nuts will ensure an adequate intake of zinc.

Vitamin B_{12}

Vitamin B_{12} may be deficient in the vegan diet as there is no reliable plant source of this vitamin. Eggs and/or dairy products will provide sufficient for other types of vegetarians, but vegans will need to rely on fortified foods such as soy milk and soy yogurt, breakfast cereals, rice and oat drinks, meat substitutes, or yeast extract. Infants will get sufficient from breast milk or formula. However, a vegetarian breast-feeding mother should ensure she eats foods (or a supplement) containing Vitamin B_{12}; if not, the baby will require a Vitamin B_{12} supplement. A deficiency of Vitamin B_{12} can result in physical and mental developmental delays and poor growth in children.

Vitamin D

Vegans, in particular, may be at risk of Vitamin D deficiency. Useful dietary sources include oily fish (salmon, sardines, mackerel), eggs, fortified margarines and spreads, fortified breakfast cereals, and fortified dairy and soy products. Vitamin D in fortified foods may be of animal origin (cholecalciferol) and the alternative (ergocalciferol) has a lower bioavailability so supplements may be required.[109]

Omega-3 Fatty Acids

Long-chain omega-3 fats (DHA and eicosapentaenoic acid [EPA]) are found mostly in oily fish and eggs. Due to the health benefits of these fatty acids, it may be beneficial for vegetarians to take a supplement (e.g., DHA derived from algae).

Health Benefits

Despite risks of nutrient deficiencies, there are also potential health benefits associated with a vegetarian diet due to it being lower in saturated fat and cholesterol and higher in dietary fiber, some minerals (e.g., potassium, magnesium), some vitamins (e.g., folate), and antioxidants (e.g., Vitamins C and E).[109] This in turn may reduce the risk of overweight and obesity, hypertension, hypercholesterolemia, heart disease, type 2 diabetes, and some cancers (e.g., prostate, colon, breast) and lengthen life expectancy. However, some studies are conflicting largely due to the variable definition of a vegetarian diet and the fact that vegetarians may choose other healthy lifestyle options more frequently, for example, exercise and nonsmoking.

Sources of Information on Vegetarian Diets

[94] British Nutrition Foundation www.nutrition.org.uk

[95] Academy of Nutrition and Dietetics www.eatright.org

[110] Vegetarian Nutrition Dietetic Practice Group www.vegetariannutrition.net

[111] British Dietetic Association www.bda.uk.com

[112] The Vegetarian Society (UK) www.vegsoc.org

[113] The Vegetarian Resource Group www.vrg.org

WEIGHT CONTROL

The terms “overweight” and “obesity” refer to body weight that is greater than what is considered healthy for a certain height. It is usually measured as body mass index (BMI) (weight in kg/height in m^2). For children, age and gender are taken into account as body fat changes during growth. BMI-for-age percentile charts are then used to identify overweight or obesity. The International Obesity Task Force (IOTF) [114] gives age- and gender-specific BMI cutoffs that correspond to the adult cutoffs of BMI 25 and 30 for defining overweight and obesity, respectively, based on data from six countries.[115] Two large surveys: the Health Examination Survey (European) and the Health and Nutrition Examination Survey (US) provided the basis for the World Health Organization (WHO) growth references that define overweight as a BMI between the 85th and 95th percentile and obesity >95th percentile.[116,117] In the United Kingdom, a BMI between the 91st and 98th percentile is considered overweight and

>98th percentile is obese.[118]

Overweight and obesity are becoming more prevalent in most affluent countries largely due to a sedentary lifestyle and an abundant supply of energy-dense foods. It may also be associated with genetics, psychological disorders (including eating disorders such as binge eating), stress, social and economic deprivation, and ethnic minorities. In the United States just over, and in the United Kingdom just under, one-third of children are overweight or obese.[119,120] However, there are suggestions that the rate of increase in overweight and obesity in children may be slowing.

Obesity in childhood has been associated with increased risk of cardiovascular disease (due to raised blood lipids, hypertension, and insulin resistance) and some cancers later in life. Type 2 diabetes used to be associated with adults but is increasingly seen in overweight adolescents and children. There is also an association between obesity and incidence of asthma, sleep apnea, and orthopedic problems.

Early introduction of solid foods (before 6 months) has been associated with an increased risk of obesity. Conversely, breast-feeding has been shown to protect against obesity in later life; the longer the duration, the smaller the risk.

In the shorter term, obesity can have a big impact on social and psychological well-being and has been linked with early puberty, low self-esteem, low self-confidence, and depression.

Overweight is essentially an imbalance between energy intake and energy expenditure. There does not appear to be any one dietary or lifestyle factor contributing to the rapid rise we have seen in obesity but rather a combination of many interlinking variables. One of the first steps is to try and identify what factors might be contributing to overweight. This will vary from individual to individual. Below are some of the possible contributing factors and suggestions for intervention.

Goal 1: Family focus

Before any change can happen, both the family and the child must be able to identify obesity as a problem and be willing to attempt lifestyle changes.

Focus on the whole family not just the child when discussing healthy eating and physical activity.

Encourage a long-term healthy lifestyle for the whole family. Children learn by example, if they see their parents and siblings eating healthy they are more likely to follow.

Just like anything we learn, repetition is the key; the more often healthy eating and physical activity is encouraged, the more likely it is to

become habit. And if the whole family is doing it, it is more likely to be maintained.

Parents need to take responsibility for ensuring that healthy foods are purchased and offered and older children need to take responsibility for how much food they choose to eat.

The family should aim to eat together as a family at least five times a week.

Involve the peer group if appropriate. Education in schools can play a big part here.

Goal 2: Eat Regular Planned Meals

Avoid skipping meals as this frequently leads to binge eating on energy-dense snacks.

Always eat breakfast. Studies have shown that breakfast improves concentration and learning, improves the quality of the diet, and reduces the risk of binge eating on energy-dense snack foods.

Try to eat regular, evenly spaced meals and snacks at the same times every day to avoid excessive hunger and the temptation to snack on energy-dense foods.

Planning meals helps to avoid impulse eating. Plan the shopping list in advance to avoid impulse buying and read food labels to choose low-fat, low-sugar, low-energy products.

Goal 3: Variety

Choose from the four main food groups: breads and cereals, vegetables and fruit, meat/protein foods, and dairy foods.

Fruits and Vegetables: these should make up at least one third to one half of every main meal. Fresh, frozen, and canned varieties are all good. Choose canned varieties with no added sugar/salt and juices with no added sugar. Avoid adding butter/margarine, sauces, and dressings.

Breads and Cereals: choose whole grain/wholemeal varieties of breads and cereals as these contain more dietary fiber which will keep children feeling full for longer. Spreads and sauces should be low fat.

Meat Group: recommend lean meat, removing all visible fat and skin from poultry, canned fish in brine/water rather than in oil, and limit fatty meats such as sausages, bacon, and processed sandwich meats.

Milk Group: low-fat or fat-free varieties of milk, yogurt and dairy desserts, and low-fat cheese should be encouraged.

Goal 4: Low Fat, Low Sugar, Low Energy

Reduce intake of energy-dense foods and drinks by reducing their availability.

A range of healthy snacks should be available, for example, fresh fruit, raw vegetables (e.g., carrot, cucumber, peppers, cherry tomato), low-fat yogurt, low-fat/sugar dairy desserts, popcorn, bread/toast.

Limit sugar sweetened drinks to no more than one per day (e.g., soft drinks, fruit juices drinks, sports drinks, flavored milk, squash).

Limit high-fat/sugar snacks that are energy dense but low in other nutrients, for example, crisps, chips, biscuits, pastries, sweets, chocolate.

Choose water as a drink or sugar-free drinks.

Choose low-fat, low-sugar varieties of packaged foods.

Choose low-fat cooking methods such as baking, steaming, boiling, microwave, or stir fry and avoid adding any fats during cooking (butter/margarine, oil).

Choose low-fat or fat-free dressings and sauces.

Skim the fat from soups, stews, and gravies.

Use low-fat or fat-free milk, yogurt, or fromage frais/crème fraiche instead of cream in soups, puddings, sauces, and baked products.

Sugar may also be listed as fructose, glucose, honey, or syrup. If sugar is one of the first few ingredients listed, the product is likely to be high in sugar.

Goal 5: Portion Size

Watch portion sizes. There is evidence that presenting children over the age of 3 years with larger portions of food leads to an increased intake of that food and in food and energy intake in general.[121]

Follow national plans for healthy eating such as “the eatwell plate” in the United Kingdom and “MyPlate” in the United States as a guide to the amount of different food groups that should make up a plate of food at each meal.

Remember that children need less than adults.

Avoid second helpings, particularly of meats, desserts, and other high-fat foods.

Slow down to eat, it should take about 20 minutes to eat a meal. This allows time for the stomach to register that it has had enough. If the child is still hungry at the end of a meal, wait 20 minutes and if still hungry offer low-fat items such as vegetables or fruits.

Goal 6: Eating Out

Limit takeaway food to no more than once a week.

When out and about, take food with you or look for low-energy choices.

Avoid fried foods and choose salads, fruits, and sandwiches with low-fat fillings.

Avoid adding extra sauces, gravies, and dressings or ask for low-fat varieties if available.

Drink water at meal times.

Avoid dishes that are cream or cheese based.

If portion sizes are large, it's okay to leave some, or ask for a container to take the rest home.

Check that the school offers appropriate choices, if not, provide a healthy packed lunch.

Allow a little bit of flexibility for parties and special occasions. Try to encourage healthy choices and avoid overeating; but accept that it is what you eat on most days that matters.

Goal 7: Physical Activity

Exercise is a vital part of any weight control program. It must go hand in hand with dietary changes.

Reduce sedentary behavior and increase lifestyle physical activity.

All children should aim for 60 minutes of moderate intensity physical exercise per day. Twice a week this should include more intense activities that improve bone health, muscle strength, and flexibility such as running, cycling, or swimming.

Limit TV to two hours a day and avoid eating in front of the TV.

Additional Guides

Aiming for weight maintenance (whilst height increases) rather than weight loss is generally recommended for children who are still growing. However, this depends on the severity of the problem and the age of the child. If a child is obese and is experiencing other health problems such as type 2 diabetes or hypertension, then carefully controlled and supervised (by a doctor and/or dietitian) weight loss may be advisable. For obese adolescents, weight loss and/or use of drugs may be considered in addition to lifestyle changes.

Avoid frequent weighing as this can be disheartening (every one to two weeks is more than enough). If a doctor or dietitian is monitoring their

weight, parents may not need to weigh them at all. This can help them to focus more on a healthy lifestyle rather than weight loss. Changing behavior is more important than focusing on one weight measurement.

Avoid using food as a reward or punishment.

Set goals that are specific and achievable, taking into account time, money, family routines, skills, and knowledge base.

Work on one or two changes at a time and use nonfood-based rewards to mark achievements, for example, a trip to the cinema.

Emphasize small gradual changes. If the aim is for weight loss, it should not be more than 1-2 pounds (1/2–1 kg) a week.

Remain positive and accept that there will be setbacks. Focus on what has been achieved rather than on moments of indiscretion.

Sources of information on weight control:

[94] British Nutrition Foundation www.nutrition.org.uk

[95] Academy of Nutrition and Dietetics www.eatright.org

[111] British Dietetic Association (BDA) www.bda.uk.com

[122] Shield J, Mullen MC. (2012) Healthy Eating, Healthy Weight for Kids and Teens. Academy of Nutrition and Dietetics, United States.

ACKNOWLEDGMENTS

The authors would like to thank Melanie V. Connolly for the preparation of the first *Special Diets* chapter for a previous edition of this textbook.

APPENDIX I

Allergy Organizations, Support groups, and Resources

Allergy UK

Email info@allergyuk.org

www.allergyuk.org

The Anaphylaxis campaign

Email info@anaphylaxis.org.uk

www.anaphylaxis.org.uk

British Society for Allergy and Clinical Immunology,

www.bsaci.org

APPENDIX II

UK Daily Reference Nutrient Intakes (RNIs)

Age	Thiamin (mg)	Riboflavin (mg)	Niacin equiv (mg)	Vitamin B6 (mg)	Vitamin B_{12} (µg)	Folate (µg)	Vitamin C (mg)	Vitamin A (µg)	Vitamin D (µg)	Calcium (mg)	Phosphorus (mg)	Magnesium (mg)	Sodium (mg)	Potassium (mg)	Chloride (mg)	Iron (mg)	Zinc (mg)	Copper (mg)	Selenium (µg)	Iodine (µg)
0–3 m	0.2	0.4	3	0.2	0.3	50	25	350	8.5	525	400	55	210	800	320	1.7	4.0	0.2	10	50
4–6 m	0.2	0.4	3	0.2	0.3	50	25	350	8.5	525	400	60	280	850	400	4.3	4.0	0.3	13	60
7–9 m	0.2	0.4	4	0.3	0.4	50	25	350	7	525	400	75	320	700	500	7.8	5.0	0.3	10	60
10–12 m	0.3	0.4	5	0.4	0.4	50	25	350	7	525	400	80	350	700	500	7.8	5.0	0.3	10	60
1–3 y	0.5	0.6	8	0.7	0.5	70	30	400	7	350	270	85	500	800	800	6.9	5.0	0.4	15	70
4–6 y	0.7	0.8	11	0.9	0.8	100	30	400	–	450	350	120	700	1100	1100	6.1	6.5	0.6	20	100
7–10 y	0.7	1.0	12	1.0	1.0	150	30	500	–	550	450	200	1200	2000	1800	8.7	7.0	0.7	30	110
Males																				
11–14 y	0.9	1.2	15	1.2	1.2	200	35	600	–	1000	775	280	1600	3100	2500	11.3	9.0	0.8	45	130
15–18 y	1.1	1.3	18	1.5	1.5	200	40	700	–	1000	775	300	1600	3500	2500	11.3	9.5	1.0	70	140
Females																				
11–14 y	0.7	1.1	12	1.0	1.2	200	35	600	–	800	625	280	1600	3100	2500	14.8**	9.0	0.8	45	130
15–18 y	0.8	1.1	14	1.2	1.5	200	40	600	–	800	625	300	1600	3500	2500	14.8**	7.0	1.0	60	140
Pregnancy	+0.1*	+0.3	–	–	–	+100	+10*	+100	10	–	–	–	–	–	–	–	–	–	–	–
Lactation																				
0–4 m	+0.2	+0.5	+2	–	+0.5	+60	+30	+350	10	+550	+440	+50	–	–	–	–	+6.0	+0.3	+15	–
4 + m	+0.2	+0.5	+2	–	+0.5	+60	+30	+350	10	+550	+440	+50	–	–	–	–	+2.5	+0.3	+15	–

*Last trimester only ** Insufficient for females with high menstrual losses
Source: Department of Health.[97]

US Daily Dietary Reference Intakes (DRIs): Recommended Dietary Allowances (RDAs) and Adequate Intakes (AIs)

Age	Thiamin (mg)	Riboflavin (mg)	Niacin equiv (mg)	Vitamin B6 (mg)	Vitamin B12 (µg)	Folate (µg)	Vitamin C (mg)	Vitamin A (µg)	Vitamin D (µg)	Calcium (mg)	Phosphorus (mg)	Magnesium (mg)	Sodium (g)	Potassium (g)	Chloride (g)	Iron (mg)	Zinc (mg)	Copper (µg)	Selenium (µg)	Iodine (µg)
0–6 m	0.2*	0.3*	2.0*	0.1*	0.4*	65*	40*	400*	10	200*	100*	30*	0.12*	0.4*	0.18*	0.27*	2*	200*	15*	110*
7–12 m	0.3*	0.4*	4.0*	0.3*	0.5*	80*	50*	500*	10	260*	275*	75*	0.37*	0.7*	0.57*	11	3	220*	20*	130*
1–3 y	0.5	0.5	6.0	0.5	0.9	150	15	300	15	700	460	80	1.0*	3.0*	1.5	7	3	340	20	90
4–8 y	0.6	0.6	8.0	0.6	1.2	200	25	400	15	1000	500	130	1.2*	3.8*	1.9	10	5	440	30	90
Males																				
9–13 y	0.9	0.9	12.0	1.0	1.8	300	45	600	15	1300	1250	240	1.5*	4.5*	2.3	8	8	700	40	120
14–18 y	1.2	1.3	16.0	1.3	2.4	400	75	900	15	1300	1250	410	1.5*	4.7*	2.3	11	11	890	55	150
Females																				
9–13 y	0.9	0.9	12.0	1.0	1.8	300	45	600	15	1300	1250	240	1.5*	4.5*	2.3	8	8	700	40	120
14–18 y	1.0	1.0	14.0	1.2	2.4	400	65	700	15	1300	1250	360	1.5*	4.7*	2.3	15	9	890	55	150
Pregnancy																				
≤18 y	1.4	1.4	18.0	1.9	2.6	600	80	750	15	1300	1250	400	1.5*	4.7*	2.3	27	12	1000	60	220
Lactation																				
≤18 y	1.4	1.6	17.0	2.0	2.8	500	115	1200	15	1300	1250	360	1.5*	5.1*	2.3	10	13	1000	70	290

Data is presented as Recommended Dietary Allowances (RDAs), the average daily dietary intake that will meet the requirements of 97–98% of healthy individuals.
*Adequate Intakes (AIs); used when there is insufficient evidence to establish an RDA but it is an amount that is believed to cover the needs of all healthy individuals.
Sources: Food and Nutrition Board, Institute of Medicine. [93,99,123,124,125,126]

EU Daily Reference Intakes

Age	Thiamin (mg/MJ)	Riboflavin (mg)	Niacin (mg/MJ)	Vitamin B6 (µg/g protein)	Vitamin B12 (µg)	Folate (µg)	Vitamin C (mg)	Vitamin A (µg)	Vitamin D (µg)	Calcium (mg)	Phosphorus (mg)	Potassium (mg)	Iron (mg)	Zinc (mg)	Copper (mg)	Selenium (µg)	Iodine (µg)
6–11 m	100	0.4	1.6	15	0.5	50	20	350	–	400	300	800	6.0	4.0	0.3	8	50
1–3 y	100	0.8	1.6	15	0.7	100	25	400	–	400	300	800	4.0	4.0	0.4	10	70
4–6 y	100	1.0	1.6	15	0.9	130	25	400	–	450	350	1100	4.0	6.0	0.6	15	90
7–10 y	100	1.2	1.6	15	1.0	150	30	500	–	550	450	2000	6.0	7.0	0.7	25	100
Males																	
11–14 y	100	1.4	1.6	15	1.3	180	35	600	–	1000	775	3100	10.0	9.0	0.8	35	120
15–17 y	100	1.6	1.6	15	1.4	200	40	700	–	1000	775	3100	13.0	9.0	1.0	45	130
Females																	
11–14 y	100	1.2	1.6	15	1.3	180	35	600	–	800	625	3100	18–22	9.0	0.8	35	120
15–17 y	100	1.3	1.6	15	1.4	200	40	600	–	800	625	3100	17–21	7.0	1.0	45	130
Pregnancy	100	1.6	1.6	15	1.6	400	55	700	–	700	550	3100	*	7.0	1.1	55	130
Lactation	100	1.7	1.6 (+2mg)	15	1.9	350	70	950	–	1200	950	3100		12.0	1.4	70	160

*Supplements required
Source: Reports of the Scientific Committee for food (thirty first series). [127]

REFERENCES

1. Kim JS, Sicherer SH. Living with food allergy: allergen avoidance. *Pediatr Clin North Am.* 2011;58:459–470.
2. Carlisle SK, Vargas PA, Noone S, et al. Food allergy education for school nurses: a needs assessment survey by the consortium of food allergy research. *J Sch Nurs.* 2010;26:360–367.
3. Skripak JM, Wood RA. Peanut and tree nut allergy in childhood. *Pediatr Allergy Immunol.* 2008;9:368–373.

4. Venter C, Laitinen K, Vlieg-Boerstra B. Nutritional aspects in diagnosis and management of food hypersensitivity: the dietitians role. *J Allergy (Cairo).* 2012;269–376.
5. Cummings AJ, Knibb RC, Erlewyn-Lajeunesse M, et al. Management of nut allergy influences quality of life and anxiety in children and their mothers. *Pediatr Allergy Immunol.* 2010;21(4 pt 1):586–594.
6. Longo G, Berti I, Burks AW, et al. IgE-mediated food allergy in children. *Lancet.* 2013;382(9905):1656–1664.
7. Cummings AJ, Knibb RC, King RM, Lucas JS. The psychosocial impact of food allergy and food hypersensitivity in children, adolescents and their families: a review. *Allergy.* 2010;65:933–945.
8. Sicherer SH, Noone SA, Muñoz-Furlong A. The impact of childhood food allergy on quality of life. *Ann. Allergy Asthma Immunol.* 2001;87:461–64.
9. Directive 2003/89/EC amending Directive 2000/13/EC of the European Parliament and of the Council of 20 March on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs.
10. Directive 2006/142/EC amending Annex 11A Directive 2000/13/EC of the European Parliament and of the Council of 20 March on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs.
11. Food Allergen Labelling and Consumer Protection Act of 2004 (Public Law 108-282, Title II)
12. Hefle SL, Nordlee JA, Taylor SL. Allergenic foods. *Crit Rev Food Sci Nutr.* 1996;36(suppl):S69–S89.
13. Parker SL, Leznoff A, Sussman GL, et al. Characteristics of patients with food-related complaints. *J Allergy Clin Immunol.* 1990;86(4 pt 1):503–511.
14. Commission Directive 2007/68/EC amending Annex11a Directive 200/13/EC of the European Parliament and of the Council as regards certain food ingredients.
15. Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the provision of food information to consumers, amending Regulations (EC) No 1924/2006 and (EC) No 1925/2006 of the European Parliament and of the Council, and repealing Commission Directive 87/250/EEC, Council Directive 90/496/EEC, Commission Directive 1999/10/EC, Directive 2000/13/EC of the European Parliament and of the Council, Commission Directives 2002/67/EC and 2008/5/EC and Commission Regulation (EC) No 608/2004
16. BRC Guidance on Allergen Labelling and the Requirements in Regulation 1169/2011. British Retail Consortium 2013.
17. Clark AT, Skypala I, Leech SC, et al; British society for Allergy and Clinical Immunology. British Society for Allergy and Clinical Immunology guidelines for the management of egg allergy. *Clin Exp Allergy.* 2010;40:1116–1129.
18. Palmer DJ, Gold MS, Makrides M. Effect of maternal egg consumption on breast milk ovalbumin concentration. *Clin Exp Allergy.* 2008;38:1186–1191.
19. Awazuhara H, Kawai H, Baba M, et al. Antigenicity of the proteins in soy lecithin and soy oil in soybean allergy. *Clin Exp Allergy.* 1998;28:1559–1564.
20. Sugiura K, Sugiura M. Soy sauce allergy. *J Eur Acad Dermatol Venereol.* 2010;24:852–855.
21. Neuman-Sunshine DL, Eckman JA, Keet CA, et al. The natural history of persistent peanut allergy. *Ann Allergy Asthma Immunol.* 2012;108:326–331.
22. Flinn A, Hourihane JO. Allergic reaction to peanuts: can we predict reaction

severity in the wild? *Curr Allergy Asthma Rep.* 2013; 13(6):645–650.
23. Lee LA, Burks AW. New insights into diagnosis and treatment of peanut food allergy. *Front Biosci (Landmark Ed).* 2009;14:3361–3371.
24. Hourihane JO, Bedwani SJ, Dean TP, Warner JO. Randomised, double blind, crossover challenge study of allergenicity of peanut oils in subjects allergic to peanuts. *BMJ.* 1997;314(7087):1084–1088.
25. Maleki SJ, Chung SY, Champagne ET, Raufman JP. The effects of roasting on the allergenic properties of peanut proteins. *J Allergy Clin Immunol.* 2000;106:763–768.
26. Peeters KA, Koppelman SJ, Penninks AH, et al. Clinical relevance of sensitization to lupine in peanut-sensitized adults. *Allergy.* 2009;64:549–555.
27. Food Standards Agency. Testing of Draft Revised Government Advice on Peanut Consumption During Early Life. 2009. FSA/1311/0809
28. Noimark L, Gardner J, Warner JO. Parents' attitudes when purchasing products for children with nut allergy: a UK perspective. *Pediatr Allergy Immunol.* 2009;20:500–504.
29. Monks H, Gowland MH, MacKenzie H, et al. How do teenagers manage their food allergies? C*lin Exp Allergy.* 2010;40:1533–1540.
30. Watson WT, Woodrow A, Stadnyk AW. Persistence of peanut allergen on a table surface. Allergy Asthma. *Clin Immunol.* 2013;9:7.
31. Bailey S, Albardiaz R, Frew AJ, Smith H. Restaurant staff's knowledge of anaphylaxis and dietary care of people with allergies. *Clin Exp Allergy.* 2011;41:713–717.
32. Leitch IS, Walker MJ, Davey R. Food allergy: gambling your life on a take-away meal. *Int J Environ Health Res.* 2005;15:79–87.
33. Hallett R, Haapanen LA, Teuber SS. Food allergies and kissing. *N Engl J Med.* 2002;346(23):1833–1834.
34. Maloney JM, Chapman MD, Sicherer SH. Peanut allergen exposure through saliva: assessment and interventions to reduce exposure. *J Allergy Clin Immunol.* 2006;118:719–724.
35. Byrne AM, Malka-Rais J, Burks AW, Fleischer DM. How do we know when peanut and tree nut allergy have resolved, and how do we keep it resolved? *Clin Exp Allergy.* 2010;40:1303–1311.
36. Bock SA, Muñoz-Furlong A, Sampson HA. Further fatalities caused by anaphylactic reactions to food, 2001-2006. *J Allergy Clin Immunol.* 2007;119:1016–1018.
37. Sicherer SH, Furlong TJ, Muñoz-Furlong A, et al. A voluntary registry for peanut and tree nut allergy: characteristics of the first 5149 registrants. *J Allergy Clin Immunol.* 2001;108:128–132.
38. Ewan PW, Clark AT. Efficacy of a management plan based on severity assessment in longitudinal and case-controlled studies of 747 children with nut allergy: proposal for good practice. *Clin Exp Allergy.* 2005;35:751–756.
39. Skolnick HS, Conover-Walker MK, Koerner CB, et al. The natural history of peanut allergy. *J Allergy Clin Immunol.* 2001;107:367–374.
40. Abramovitch JB, Kamath S, Varese N, et al. IgE reactivity of blue swimmer crab (Portunus pelagicus) tropomyosin, Por p 1, and other allergens; cross-reactivity with black tiger prawn and effects of heating. *PLoS One.* 2013;8:e67487.
41. Pascual CY, Reche M, Fiandor A, et al. Fish allergy in childhood. *Pediatr Allergy Immunol.* 2008;19:573–579.
42. Cianferoni A, Muraro A. Food-induced anaphylaxis. *Immunol Allergy Clin North Am.* 2012;32:165–195.

43. Turner P, Ng I, Kemp A, Campbell D. Seafood allergy in children: a descriptive study. *Ann Allergy Asthma Immunol.* 2011;106:494–501.
44. Vally H, Misso NL, Madan V. Clinical effects of sulfite additives. *Clin Exp Allergy*. 2009;39:1643–1651.
45. Jarvis JK, Miller GD. Overcoming the barrier of lactose intolerance to reduce health disparities. *J Natl Med Assoc.* 2002;94:55–66.
46. Heyman MB. Committee on Nutrition. Lactose intolerance in infants, children, and adolescents. *Pediatrics.* 2006;118:1279–1286.
47. Casein and Caseinate Regulations. Subsidiary Regulations 231.50. Legal notice 113 of 2002. 1 July, 2002.
48. Roe MA, Finglas PM, Church SM. *McCance and Widdowson's 'The Composition of Foods'* Sixth summary edition. Cambridge, ID: Royal Society of Chemistry; 2002.
49. Portnoi PA, MacDonald A. Determination of the lactose and galactose content of cheese for use in the galactosemia diet. *J Hum Nutr Diet*. 2009;22:400–408.
50. Kaufman FR, Loro ML, Azen C, *et al*. Effect of hypogonadism and deficient calcium intake on bone density in patients with galactosemia. *J Pediatr*. 1993;123:365–370.
51. MacDonald A. *Calcium and Galactosemia. A Compilation Of Papers Presented At The Second Dietitians' Meeting*. Gothenburg, Sweden: Society for Study of Inborn Errors of Metabolism; 1997: 2–6.
52. Rutherford PJ, Davidson DC, Matthai SM. Dietary calcium in galactosemia. *J Hum Nutr Diet.* 2002;15:39–42.
53. The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment (COT). Phytoestrogens and health. Food Standards Agency, 2003. www.foodstandards.gov.uk/multimedia/pdfs/phtoreport0503 accessed 30/10/12.
54. EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA). Scientific opinion on lactose thresholds in lactose intolerance and galactosemia. *EFSA 2010;8(9):1777*.
55. Galactosemia Support Group. Unpublished lactose and galactose data on file, 2012.
56. Host A. Frequency of Cow's milk protein allergy in childhood. *Ann Allergy Asthma Immunol.* 2002;89(6 suppl 1):33–37.
57. Hogberg L, Laurin P, Fälth-Magnusson K, et al. Oats to children with newly diagnosed celiac disease: a randomised double blind study *Gut*. 2004;53:649–654.
58. Husby S, Koletzko S, Korponay-Szábó IR, et al. ESPGHAN guidelines for the diagnosis of celiac disease in children and adolescents. An evidence-based approach. *JPGN* 2012;54:136–160.
59. Murch S,Jenkins H, Auth M et al. Joint BSPGHAN and Celiac UK guidelines for the diagnosis and management of celiac disease in children. *Arch Dis Child.* 2013;98:806–811.
60. Codex Alimentarius Commission regulation (EC) No 41/2009.
61. Celiac UK 2011 Gluten free foods a revised prescribing guide www.celiac.org.uk
62. Holland B, Unwin ID, Buss DH. *Fruit and Nuts. First supplement to 5th edition of McCance and Widdowson's The Composition of Foods*. Cambridge, ID: Royal Society of Chemistry; 1992.
63. Robayo-Torres CC, Opekun AR, Quezada-Calvillo R, et al. 13C-breath tests for sucrose digestion in congenital sucrase isomaltase-deficient and sacrosidase-supplemented patients. *J Pediatr Gastroenterol Nutr*. 2009;48:412–418.
64. Sawczenko A, Sandhu BK, Logan RFA, et al. Prospective survey of childhood inflammatory bowel disease in the British Isles. *Lancet*. 2001;357:1093–1094.

65. Spray C, Debelle GD, Murphy MS. Current diagnosis, management and morbidity in paediatric inflammatory bowel disease. Acta Paediatr. 2001;90(4):400–405, 2001;0803–5253.
66. Otley AR, Murray A, Christensen B, et al. Primary enteral nutrition therapy induces and maintains remission, and reduces steroid exposure in a pediatric Crohn's disease population. *Gastroenterology.* 2005;128(4)(suppl 2):W1053.
67. Gavin J, Anderson CE, Bremner AR, Beattie RM. Energy intakes of children with Crohn's disease treated with enteral nutrition as primary therapy. *J Hum Nutr Diet.* 2005;18(5):337–342, 0952–3871.
68. Johnson T, MacDonald S, Hill SM, et al. Treatment of active Crohn's disease in children using partial enteral nutrition with liquid formula: a randomised controlled trial. *Gut.* 2006;66:356–361.
69. Aiges H, Markowitz J, Rosa J, Daum F. Home nocturnal supplemental nasogastric feedings in growth-retarded adolescents with Crohn's disease. Gastroenterology. 1989; 97(4):905–910.
70. Wilschanski M, Sherman P, Pencharz P, et al. Supplementary enteral nutrition maintains remission in paediatric Crohn's disease. *Gut.* 1996;38(4):543–548.
71. MacDonald A, White F. Amino acid disorders. In Shaw V, ed. *Clinical Paediatric Dietetics.* West Sussex, UK: Wiley-Blackwell; 2014: 381–456.
72. Ahring K, Bélanger-Quintana A, Dokoupil K, et al. Dietary management practices in phenylketonuria across European centres. *Clin Nutr.* 2009;28:231–236.
73. Paul AA, Southgate DAT, Russell J. *First Supplement to McCance and Widdowson's The Composition of Foods*. London, England: HMSO; 1980.
74. MacDonald A, Rylance G, Davies P, et al. Free use of fruits and vegetables in phenylketonuria. *J Inherit Metab Dis.* 2003;26:327–338.
75. Rohde C, Mütze U, Weigel JF, et al. Unrestricted consumption of fruits and vegetables in phenylketonuria: no major impact on metabolic control. *Eur J Clin Nutr.* 2012;66:633–638.
76. Zimmermann M, Jacobs P, Fingerhut R, et al. Positive effect of a simplified diet on blood phenylalanine control in different phenylketonuria variants, characterized by newborn BH4 loading test and PAH analysis. *Mol Gen Metab.* 2012;106:264–268. CMO's Update 37. Department of Health, 2003. www.dh.gov.uk/assetRoot/04/07/01/76/04070176.pdf. Accessed October 30, 2012.
77. Hennermann JB, Roloff S, Gellermann J, et al. Chronic kidney disease in adolescent and adult patients with phenylketonuria. *J Inherit Metab Dis.* 2013;36(5):747–756.
78. MacDonald A, Rylance G, Davies P, et al. Administration of protein substitute and quality of control in phenylketonuria: a randomized study. *J Inherit Metab Dis.* 2003;26:319–326.
79. MacDonald A, Chakrapani A, Hendriksz C, et al. Protein substitute dosage in PKU: how much do young patients need? *Arch Dis Child.* 2006;91:588–593.
80. Bouteldja N, Timson DJ. The biochemical basis of hereditary fructose intolerance. *J Inherit Metab Dis.* 2010;33:105–112.
81. Steinmann B, Santer R. Disorders of Fructose Metabolism. In: Saudubray JM, van den Berghe G, Walter JH, eds. *Inborn Metabolic Diseases: Diagnosis and Treatment*. 5th ed. Berlin, Germany: Springer-Verlag; 2012:157–165.
82. Wong D. Hereditary fructose intolerance. *Mol Genet Metab.* 2005;85:165–167.
83. Smith W. Disorders of fructose metabolism. In: Ekvall SW, Ekvall VK, eds. *Pediatric Nutrition in Chronic Diseases and Developmental Disorders*. New York, NY: Oxford University Press; 2005:346–349.

84. Dixon M. Disorders of fructose metabolism. In: Shaw V, Lawson M, eds. *Clinical Paediatric Dietetics*. Oxford, ID: Blackwell Publishing; 2007:410–420.
85. Yasawy MI, Folsch UR, Schmidt WE, Schwend M. Adult hereditary fructose intolerance. *World J Gastroenterol.* 2009;15:2412–2413.
86. Marcason W. Is medical nutrition therapy (MNT) the same for hereditary vs dietary fructose intolerance? *J Am Diet Assoc.* 2010;110:1128.
87. Gregersen N, Bross P, Andresen BS. Genetic defects in fatty acid beta-oxidation and acyl-CoA dehydrogenases. Molecular pathogenesis and genotype-phenotype relationships. *Eur J Biochem.* 2004;271:470–482.
88. Bennett MJ, Rinaldo P, Strauss AW. Inborn errors of mitochondrial fatty acid oxidation. *Crit Rev Clin Lab Sci.* 2000;37:1–44.
89. Food and Nutrition Board, Institute of Medicine. Committee to Review Dietary Reference Intakes for Vitamin D and Calcium. *Dietary Reference Intakes for Calcium and Vitamin D*. Washington, DC: National Academy Press; 2010.
90. Theobald HE. British Nutrition Foundation Briefing Paper. Dietary calcium and health. *Nutr Bull*. 2005;30:237–277.
91. Weaver CM, Proulx WR, Heaney RP. Choices for achieving adequate dietary calcium with a vegetarian diet. *Am J Clin Nutr*. 1999;70:543S–548S.
92. Heaney RP. Bone mass, nutrition and other lifestyle factors. *Nutr Rev.*1996;54:S3–S10.
93. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Calcium, Phosphorus, Magnesium, Vitamin D and Fluoride.* Standing Committee on the Scientific Evaluation of Dietary Reference Intakes; 1997. www.nap.edu
94. British Nutrition Foundation. www.nutrition.org.uk. Accessed October 1, 2013.
95. Academy of Nutrition and Dietetics. www.eatright.org. Accessed October 1, 2013.
96. Food & Nutrition Board. *Dietary Reference Intakes*. Washington DC: National Academy Press; 2001.
97. Department of Health. Dietary reference values for food, energy and nutrients for the United Kingdom. Report of the Panel on Dietary Reference Values of the Committee on Medical Aspects of Food Policy. *Rep Health Soc Subj (Lond).*, 1991;41:1–210.
98. Nutrient and Energy Intakes for the European Community. *Report of the Scientific Committee for Food (thirty first series)*. Luxembourg: Office for Official Publications of the European Communities; 1993.
99. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Vitamin A, Vitamin K, Arsenic, Boron, Chromium, Copper, Iodine, Iron, Manganese, Molybdenum, Nickel, Silicon, Vanadium and Zinc*. Washington, DC: National Academy Press; 2001.
100. Monsen ER. Iron nutrition and absorption: dietary factors which impact iron bioavailability. *J am Diet Assoc.* 1988;88:786–790.
101. Hunt JR, Roughead ZK. Adaptation of iron absorption in men consuming diets with high or low iron bioavailability. *Am J Clin Nutr.* 1999;71:94–102.
102. Scholz-Azrens KE, Schrezenmeir J. Inulin and oligofructose and mineral metabolism: the evidence from animal trials. *J Nutr.* 2007;137:2513S–2523S.
103. Greger JL. Nondigestible carbohydrates and mineral bioavailability. *J Nutr.* 1999;129:1434S–1435S.
104. Weickert O, Pfeiffer AF. Metabolic effects of dietary fiber consumption and prevention of diabetes. *J Nutr.* 2008;138:439–442.
105. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty acids, Cholesterol, Protein and Amino Acids (Macronutrients)*. Washington, DC: National Academy Press; 2005.

106. Roy CC, Kien CL, Bouthillier L, Levy E. Short-chain fatty acids: ready for prime time? *Nutr Clin Prac.* 2006;21:351–366.
107. Williams C. Dietary fiber in childhood. *J Pediatr*. 2006:149:S121–S130.
108. Williams C, Bodella M, Wynder E. A new recommendation for dietary fiber in childhood. *Pediatr*. 1995;96:985–988.
109. American Dietetic Association (ADA). Position of the American Dietetic Association and Dietitians of Canada: vegetarian diets. *J Am Diet Assoc.* 2003;103:748–765.
110. Vegetarian Nutrition Dietetic Practice Group www.vegetariannutrition.net
111. British Dietetic Association www.bda.uk.com
112. The Vegetarian Society (UK). www.vegsoc.org
113. The Vegetarian Resource. Group www.vrg.org
114. International Obesity. Taskforce www.iaso.org. Accessed October 1, 2013.
115. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition for child overweight and obesity worldwide: international survey. *BMJ.* 2000;320:1240–1243.
116. Kuczmarski RJ, Ogden CL, Grummer-Strawn LM, et al. CDC Growth Charts: United States Advance Data from Vital and Health Statistics, no. 314. 2000. National Centre for Health Statistics: Atlanta.
117. De Onis M, Onyango AW, Borghi E, et al. Development of a WHO growth reference for school-aged children and adolescents. Bull World Health Organ. 2007;85:660–667.
118. Cole TJ, Freeman JV, Preece MA. Body mass index reference curves for the UK, 1990. *Arch Dis Child.* 1995;73:25–29.
119. Health and Social Care Information Centre (HSCIC). Health Survey for England 2011. HSCIC, Leeds, 2012.
120. Ogden CL, Carroll MD, Kit BK, Flegal KM. Prevalence of obesity and trends in body mass index among US children and adolescents, 199-2010. *JAMA.* 2012;307:483–490.
121. Benelam B. Satiation, satiety and their effects on eating behaviour. *Nutrition Bulletin.* 2009;34:126–173.
122. Shield J, Mullen MC. *Healthy Eating, Healthy Weight for Kids and Teens*. Chicago, IL: Academy of Nutrition and Dietetics; 2012.
123. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline*. Washington, DC: National Academy Press; 1998.
124. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Vitamin C, Vitamin E, Selenium, and Carotenoids*. Washington, DC: National Academy Press; 2000.
125. Food and Nutrition Board, Institute of Medicine. *Dietary Reference Intakes for Water, Potassium, Sodium, Chloride, and Sulphate*. Washington, DC: National Academy Press; 2005.
126. Dietary Reference Intakes for Calcium and Vitamin D (2011). http://fnic.nal.usda.gov/dietary-guidance/dietary-reference-intakes/dri-tables. Accessed October 18, 2013.
127. Reports of the Scientific Committee for Food (Thirty-first series). *Nutrient and Energy Intakes for the European Community*. Luxembourg: Commission of the European Communities; 1993.

Nutrition Support Teams

71

Virginie Colomb, MD, PhD and Cécile Lambe, MD

INTRODUCTION

Major insufficiencies in nutrition care have been reported in the hospital setting in developed countries. The reported prevalence of malnutrition in hospitalized children ranges from 15% to 30%, with an impact on growth, morbidity, and mortality. The implementation of nutrition support teams (NSTs) has been suggested to improve nutritional management for hospitalized patients. The main objectives of the NSTs include screening for nutritional risk, identification of patients who require nutritional support, provision of adequate nutritional management, education and training of hospital staff, and audit of practices including cost-effectiveness. The NST should be multidisciplinary, with expertise in all aspects of clinical nutrition care. The funds needed for NSTs should be raised from the health-care system whenever possible. This chapter reviews the disease related-mechanisms causing malnutrition, the consequences of malnutrition, and provides the frame for implementation of NSTs in pediatric units. Further research is needed to evaluate the impact of NSTs in prevention and management of pediatric nutritional disorders, including cost-effectiveness in different settings.

Malnutrition is common among hospitalized children, but hardly recognized and, therefore, not prevented or treated often enough. In spite of major advances in the quality of care in many other areas of pediatric medicine, the prevalence of malnutrition in hospitalized children has not changed apparently over the last three decades, ranging from 15% to 30% of patients, depending on the criteria used to define “malnutrition,” the patient population, and their disease spectrum.[1–10] The detrimental effects of malnutrition on growth, morbidity, and mortality in hospitalized

children are often underestimated. An expert report of the Council of Europe has highlighted major insufficiencies in nutritional care in European hospitals and provided recommendations for improving the situation, including the implementation of nutritional support teams (NSTs) in hospitals.[11] The main goal of NSTs should be to provide optimal nutrition to all the patients,[12] especially in case of need for enteral nutrition (EN) or parenteral nutrition (PN) supplementation.[13,14]

In 2005, the Committee on Nutrition of the European Society of Pediatric Gastroenterology, Hepatology, and Nutrition published a comment which underlined the need for NSTs in pediatric hospital settings.[15]

Mechanisms of Malnutrition in Sick People

Malnutrition starts with inadequate nutrient intake to meet physiological requirements, followed by metabolic and functional alterations, all of which accompanied by impairment of body composition. Regardless of primary disease and age, malnutrition is always the consequence of imbalance between (reduced) nutritional supplies and (increased) protein–energy needs. The primary disease often induces anorexia limiting food intake. In addition, the acute disease (e.g., sepsis, trauma, burn, cancer),[16,17] chronic illness (e.g., inflammatory bowel disease, congenital heart disease, chronic renal failure,[18] cystic fibrosis, severe neuromuscular impairment), or medications increase imbalance between protein and/or energy demands and supply. An increase in resting energy expenditure, protein turnover, and protein breakdown has been shown in children after blunt trauma.[16] Sepsis and fever also induce protein breakdown and increases resting energy expenditure.[19,20] However, in pathologic situations, a decrease in physical activity-related energy expenditure may compensate for the latter.

In children, linear growth depends on a permanent increase in fat and lean body mass, which requires positive energy and nitrogen balance. Therefore, any increase in nutritional demands competes with the specific needs of growth, especially during periods of high-speed growth, like infancy and adolescence or catch-up growth. Consequently, sustained nutritional imbalance induces growth retardation, with a decrease in height-for-age ratio (height divided by the expected height, taken as 50th centile for age and gender).

Assessment of Nutritional Risk and Malnutrition

In clinical practice, nutritional risk and malnutrition are difficult to assess due to lack of simple, sensitive assessment tools and due to the difficulty in separating the impact of malnutrition and of the disease state on some biological markers of malnutrition (e.g., hypoalbuminemia is a marker of both malnutrition and also a marker of severe inflammation or protein-loosing enteropathy) and on outcome.

Nutritional Risk

Children admitted to hospital are at risk for developing malnutrition, even when nutritional status is normal at the time of admission. The size of the so-called "reservoir of malnutrition" among children who are not measurably malnourished but are at risk to become malnourished is hard to determine.[4,10] Several pediatric nutritional risk scores have been proposed over the last 10 years. One of them, specifically designed for children with cancer, takes into account anorexia and gastrointestinal symptoms.[21] Another one, designed for all types of pediatric illnesses, emphasizes the role of severity of the underlying disease, pain, and reduced nutritional intake.[22] Systematic utilization of such scores could help for timely implementation of adequate nutritional support, thus preventing the occurrence or worsening of malnutrition. However, some tools mix criteria of malnutrition and criteria of nutritional risk,[23,24] leading to confusion in the concept of "risk,"[25] and none has been universally accepted and used.[10]

Nutritional Status

The nutritional status is not easier to determine in routine.[26,27] Either subjective[28] or objective, tool-based,[23,24] nutritional screening should lead to quick and reliable triage of children in order to identify those who need more detailed assessment and intervention. The most obvious and earliest clinical sign of malnutrition in children is the absence of linear weight gain, followed by weight loss with visible decrease in fat and muscle mass. The weight-for-height ratio (weight divided by the expected weight taken as 50th centile for height and gender) is widely accepted as a criterion for children malnutrition, but its calculation is a source of errors[26,27,29] and appropriate reference standards for weight-for-height are not available. Apparently, in western countries, not all the expert pediatric societies have developed any policy on the problem of identifying malnutrition in hospitalized children.[10] Whatever the screening method used, careful measurements and use of appropriate growth charts are essential for the

diagnosis of malnutrition in children.[29] One of the factors contributing to the nonrecognition of malnutrition is obviously the failure to measure routinely weight and height in hospitalized children at admission and during hospital stay. Recently, the Nutrition Committee of the French Pediatric Society again recommended weighing and measuring any child when hospitalized or seen in outpatient clinic.[30] These experts recommend to calculate the body mass index (BMI) when growth kinetics cannot be analyzed and state that any child with a BMI below the third centile, or minus 2 standard deviations for age and gender, needs further nutritional assessment.[30]

Consequences of Hospital Malnutrition

Malnutrition is associated with an increase in morbidity and mortality in hospitalized patients. For instance, it was demonstrated that the nutritional status of the children hospitalized in a pediatric intensive care unit with respiratory syncytial virus infection was related significantly to outcomes, such as length of stay, number of days on ventilation, and duration of PN.[31] Data obtained in adult patients have also shown that malnutrition increases length of stay, and therefore hospital costs.[32,33] The increasing prevalence of children with chronic diseases amplifies the problem both during hospital stays and in outpatient care.

The Impact of Hospital Practices on Nutrition Support

In the beginning of 2000s, the Council of Europe provided a review of the current practices in Europe regarding food provision and nutritional care and support of hospitalized patients, focusing mostly on adults.[11] This report highlights major deficiencies related to hospital organization of nutritional care. It states that *«routine nutritional risk screening and nutritional assessment is generally not performed (…);… «the use of energy and protein- dense menu is not widespread, meal times are often inflexible and/or unadapted to patients' food habits, food intake is infrequently recorded, the use of nutritional support for undernourished patients and nutritionally at risk patients is sparse, inconsistent and often performed in special patients or patients who are severely undernourished and that responsibilities and tasks of each personal involved in the food service and the nutritional care of the patient are not clearly assigned…».* The report also pointed out the lack of appropriate education and training of all professionals involved regarding nutrition, as a major cause of malnutrition in hospitalized patients and therefore, advocated the

implementation of NSTs in hospitals. The American Society of Parenteral and Enteral Nutrition also emphasizes that "*organized nutrition support services or teams are associated with improved patient outcomes, decreased length of hospitalization, and improved cost-effectiveness*" and recommends "*if an institution does not have a defined nutrition support services or team, it is recommended that an interdisciplinary team provides specialized nutrition support.*"[34,35]

The Concept of the Nutrition Support Team

The first NSTs were born in the early 1980s.[36–38] The concept of NST was based on the acknowledgement of the fact that most physicians are poorly teached and trained in the domain of nutrition and the idea that the nutrition management of the hospitalized patient would be best addressed through the intervention of a well-organized and coordinated multidisciplinary group, knowledgeable in clinical nutrition, working together as a team.

The Tasks of Nutrition Support Teams

As for any hospital service, the tasks of the NST might be divided into three major categories: patient's care, education/training of physicians and other professionals in the area of nutrition, and research. The main roles of the NST are to implement patient screening for nutritional risk and malnutrition; to identify patients who require further nutritional assessment and nutritional support; to ensure provision of adequate nutritional management to patients; to plan and organize optimal home nutrition after discharge; to educate hospital staff with respect to identification and management of nutritional problems; and to provide standardized protocols and algorithms. The NSTs also have to monitor and evaluate practices, to document and audit its own work, and to provide results to hospital management. The NST members should also participate in training activities by which their own level of practice is raised, and where possible in research.

Composition and Qualification of the Nutrition Support Team

The composition of NSTs varies according to the type of institution, the tasks of the team, and availability of staff and funding. In addition to a senior physician knowledgeable in nutrition, the NST should include a dietician/nutritionist, a nurse, and, if possible, a pharmacist, with some

team members contributing only part of their work time.

In pediatric hospitals, a pediatrician—whose specific competence should be recognized by other physicians and other members of the NST—usually assumes the role of the NST leader. Thus, the leader of the NST has to combine nutrition knowledge, teaching, and management skills enabling successful interactions with all involved disciplines.

Dieticians/nutritionists evaluate the nutritional intakes, advise on improving oral, EN and PN, and may participate in the patient's and family training at the time of planning home EN or PN. Nurses also play a key role in the education and training of hospital staff, especially regarding the techniques of EN and PN, including care for gastrostomies and central venous lines.[39] Nurses also participate in patient's and family training prior to discharge on home nutrition. Depending on the size of the hospital and the amount of tasks covered by the NST, one or more further pediatricians may be necessary, as well as secretarial support and a data coordinator.

Impact of the NST on Patients' Outcome and Cost of Care

Several studies in adults and children in hospitals have shown benefits of the presence of an NST. Patients' outcome is improved by early nutritional support, as assessed by a study in adult patients which showed that a 2-day-earlier nutrition intervention saved 1 day of hospital stay.[32] The NST can improve timing of nutrition intervention through a staff education plan, as shown in 1998 in a pediatric intensive care unit.[40] The implementation of an NST has appeared of particular importance to improve the quality and safety of PN prescription and follow-up, since this form of nutrition is at high risk and high costs.[37,38,41–48] The NST advises physicians on the indication for PN, the optimal composition of PN intakes and thus the choice of standardized or individual PN formulations.[49–51] In hospitals (units) where PN bags should be prepared "on demand," the NST, in cooperation with pharmacists,[51] should ensure that PN compounding and additions (of micronutrients or other components) are performed under sterile conditions according to agreed quality assurance guidelines. Along with the closer supervision of PN, implementation of NSTs has shown to induce a relative increase in the use of enteral feeding and finally helps cost savings.[13,41,53] In 1990, a study conducted in a 500 bed-institution in the United States demonstrated that the NST action, by close supervision of PN indications and procedures, could lead to savings of about 100,000 US $ in 1 year.[41] For all these reasons, the German

Association for Nutritional Medicine, in the Guidelines on Parenteral Nutrition published in 2009, stated that interdisciplinary NSTs should be established in all hospitals.[38]

Obstacles to Establishing a Nutrition Support Team

Although the benefits of NSTs have been confirmed in terms of patients' outcome and cost of care,[54] better recognition of malnutrition issues and initiation of adequate nutritional support as a priority by both physicians and hospital managers are still needed. Therefore, a new NST has often to demonstrate that malnutrition exists at admission and/or appears during hospitalization.

Measuring the Performance of an NST

The NST should show the deleterious medical and financial consequences of malnutrition and should perform audits on its own activity and results, and also to demonstrate the benefits of the intervention of nutrition specialists.[39,41,52,55] A business plan is needed to demonstrate the importance of the NST to the hospital management both medically (diagnosis and intervention) and financially (decreasing hospital costs). NSTs may justify their existence by specifying the most beneficial therapy for the patient at the most cost-effective method for the hospital. For example: the NST will improve the indications for nutrition support so that no patient receives expensive—and sometimes risky treatment unless it is clearly needed; the team will improve the choice of nutrition route so that 100% of the patients who can be fed enterally are fed via this route.[54] In view of the considerably potential for cost savings, the funds needed for running an NST should be raised from the health-care system whenever possible. At the start, it might be advisable to select a small field for a concentrated effort, for example, a pediatric cancer unit or a cystic fibrosis clinic, with a highest chance to demonstrate success.

The Weaknesses and Limitations of NSTs

NSTs still suffer from several weaknesses which limit their impact of the quality of nutritional care of hospitalized patients. Most of them are related to the poor recognition/acceptance of the nutrition therapy by both the health-care professionals and the hospital operating institutions:

- **Lack** of fundings and therefore lack of means devoted to nutrition care. This explains why an European survey showed that very

few of the physicians working in an NST are exclusively devoted to it, when most of the NST managers could spend a mean of 5.5 hours per week only for the NST.[56]

- **Lack** of faculty curriculum and approval of the qualification in Nutrition Medicine by the Health Board.[15,56]
- **Lack** of "decision autonomy" in nutrition care, lack of systematic screening of all patients admitted to the hospital… Due to its consultative function, the impact of the NST on patient's care and outcome is limited by the degree of acceptance/acknowledgment of expertise of the NST by the primary physicians who are in charge of the patients. A study demonstrated 30 years ago, that the incidence of PN complications was greater when the NST was purely consultative than during the period when the NST increased its involvement in patients' care.[37] More recently, an European survey of structure and organization of NSTs showed that none of them were deciding independently on the nutrition therapy and 52% were consulting.[56] Most of the requests to the NSTs were in case of questions or problems while the NSTs were called in less than 30% at admission of the patient.[56]

The Prevalence of NSTs in Pediatric Hospitals in the Twentieth Century

In 1995, nutrition support pharmacists in the United States provided a survey, which showed that >65% of them practiced in hospitals had an NST, but by 2002, this number had dropped to 49.5%.[57,58] In Germany, a survey was conducted in 2000 in the aim to investigate the number, structure, and performance of NSTs in more than 800 acute care hospitals of more than 250 beds. It showed a prevalence of NSTs of 5.6%, out of which the number of specific pediatric NSTs was not specified.[59] The majority of NSTs were informed only when specific questions or problems regarding clinical nutrition were raised and decision making in nutritional support was mainly done by health-care professionals outside the NST. Four years later, a survey screened more than 3000 hospitals in Germany, Austria, and Switzerland, and found NSTs, defined by a minimum one physician plus one staff, in only 3.2% of them. Most NSTs were located in university/academic teaching hospitals with a high number of beds and a large spectrum of specialties with a high requirement for nutrition-therapeutical interventions.[56] Ten years ago, a study described hospital pharmacists' practices in Belgium, France, and Switzerland and reported

that NSTs were present in 32%–45% of the hospitals surveyed.[60]

However, over the last 10 years, few data have been available in scrutinizing the prevalence and structure of NSTs in western countries, particularly in pediatric hospitals.

Key Messages

1. Implementation of NSTs in hospitals is recommended to improve nutritional management of sick children.
2. The main tasks of NSTs include patient screening for nutritional risk, identification of patients who require nutritional support, provision of adequate nutritional management for patients, education and training of hospital staff, and audit of practice.
3. The NST should be independent, multidisciplinary, with expertise in all major aspects of clinical nutrition care.
4. The funds needed for NSTs should be raised from the health-care system.
5. Further research is needed to evaluate the effects of NSTs in prevention and management of pediatric nutritional disorders, including cost effectiveness in different settings.

REFERENCES

1. Merritt RJ, Suskind RM. Nutritional survey of hospitalized children. *Am J Clin Nutr*. 1979;32:1320–1325.
2. Parsons HG, Francoeur TE, Howland P, et al. The nutritional status of hospitalized children. *Am J Clin Nutr*. 1980;33:1140–1146.
3. Cooper A, Jakobowski D, Spiker J, et al. Nutritional assessment. An integral part of the preoperative pediatric surgical evaluation. *J Pediatr Surg*. 1981;16:554–560.
4. Moy RJ, Smallman S, Booth IW. Malnutrition in a UK children's hospital. *J Hum Nutr Dietetics*. 1990;3:93–100.
5. Smith DE, Stevens MCG, Booth IW. Malnutrition at diagnosis of malignancy in childhood: common but mostly missed. *Eur J Pediatr*. 1991;150:318–322.
6. Hendriks KM, Duggan C, Gallagher L, et al. Malnutrition in hospitalized pediatric patients. *Arch Pediatr Adolesc Med*. 1995;149:1118–1122.
7. Hendrikse WH, Reilly J, Weaver LT. Malnutrition in a UK children's hospital. *Clin Nutr*. 1997;16:13–18.
8. Pawellek I, Dokoupil K, Koletzko B. Prevalence of malnutrition in paediatric hospital patients. *Clin Nutr*. 2008;27:72–76.
9. Joosten KF, Zwart H, Hop WC, et al. National malnutrition screening days in hospitalised children in the Netherlands. *Arch Dis Child*. 2010;95:141–145.
10. Sullivan PB. Malnutrition in hospitalized children. *Arch Dis Child*. 2010;95:489–490.
11. Beck AM, Balknäs UN, Camilo ME, et al. (ad hoc group on Nutrition

Programmes in Hospitals, Council of Europe). Practices in relation to nutritional care and support-report from the Council of Europe. *Clin Nutr*. 2002;21:351–354.
12. Jonkers CF, Prins F, Van Kempen A, et al. Towards implementation of optimum nutrition and better clinical nutrition support. *Clin Nutr*. 2001;20:361–366.
13. Brown RO, Carlson SD, Cowan GS Jr, et al. Enteral nutritional support management in university teaching hospitals: team vs non team. *JPEN J Parenteral Enteral Nutr*. 1987;11:52–56.
14. Planas M, Camilo ME. Artificial nutrition: dilemnas in decision-making. *Clin Nutr*. 2002;21:355–361.
15. Agostoni C, Axelson I, Colomb V, et al. ESPGHAN Committee on Nutrition. The need of nutrition teams for pediatric units. A commentary by the ESPGHAN Committee on Nutrition. *J Pediatr Gastroenterol Nutr*. 2005;10:41:8–11. Erratum in: *J Pediatr Gastroenterol Nutr*. 2005;41:267–271.
16. Winthrop AL, Wesson DE, Pencharz PB, et al. Injury severity, whole body protein turnover and energy expenditure in pediatric trauma. *J Pediatr Surg*. 1987;22:534–537.
17. Skillman HE, Mehta NM. Nutrition therapy in the critically ill child. *Curr Opin Crit Care*. 2012;18:192–198.
18. Castillo A, Santiago MJ, López-Herce J, et al. Nutritional status and clinical outcome of children on continuous renal replacement therapy: a prospective observational study. *BMC Nephrol*. 2012;13:125.
19. Stettler N, Schutz Y, Whitehead R, Jequier E. Effect of malaria and fever on energy metabolism in Gambian children. *Pediatr Res*. 1992;32:102–106.
20. Ben Hariz M, Goulet O, Salas J, et al. Energy cost of fever in children on total parenteral nutrition. *Clin Nutr*. 1997;1:251–255.
21. Attard-Montalto SP, Hadley J, Kingston JE, et al. Ongoing assessment of nutritional status in children with malignant disease. *Pediatr Hematol Oncol*. 1998;15:393–403.
22. Sermet-Gaudelus I, Poisson-Salomon AS, Colomb V, et al. Simple pediatric nutritional risk score to identify children at risk of malnutrition. *Am J Clin Nutr*. 2000;72:64–70.
23. Gerasimidis K, Mcleaod I, Maclean A, et al. Performance of the novel Paediatric Yorkhill Malnutrition Score (PYMS) in hospital practice. *Clin Nutr*. 2011;30:430–435.
24. McCarthy H, Dixon M, Crabtree I, et al. The development and evaluation of the Screening Tool for the Assessment of Malnutrition in Paediatrics (STAMP) for use by healthcare staff. *J Hum Nutr Diet*. 2012;25:311–318.
25. Wiskin AE, Owens DR, Cornelius VR, et al. Paediatric nutrition risk scores in clinical practice: children with inflammatory bowel disease. *J Hum Nutr Diet*. 2012;25:319–322.
26. Cross JH, Holden C, McDonald A, et al. Clinical examination compared with anthropometry in evaluating nutritional status. *Arch Dis Child*. 1995;72:60–61.
27. Smith DE, Booth IW. Nutritional assessment in children: guidelines on collecting and interpreting anthropometric data. *J Hum Nutr Diet*. 1989;2:217–224.
28. Secker DJ, Jeejeebhoy KN. How to perform Subjective Global Nutritional assessment in children. *J Acad Nutr Diet*. 2012;112:424–431.
29. Poustie VJ, Watling RM, Ashby D, et al. Reliability of the percentage ideal weight for height. *Arch Dis Child*. 2000;83:183–184.
30. Hankard R, Colomb V, Piloquet H, et al. Malnutrition screening in clinical practice. *Arch Pediatr*. 2012;19:1110–1117.
31. Mezoff A, Gamm L, Konek S, et al. Validation of a nutritional screen in children

with respiratory syncitial virus admitted to an intensive care complex. *Pediatrics*. 1996;97:543–546.
32. Tucker HN, Miguel SG. Cost containment through nutrition intervention. *Nutr Rev*. 1996;54:111–121.
33. Del Savio GC, Zelicof SB, Wexler LM, et al. Preoperative nutritional status and outcome of elective total hip replacement. *Clin Orthop Relat Res*. 1996;326:153–161.
34. Russell MK, Andrews MR, Brewer CK, et al. Standards of practice for specialized nutrition support: adult hospitalized patients. *Nutr Clin Pract*. 2002;17:384–391.
35. A.S.P.E.N. Board of Directors. *Nutrition Support Teams: An Anthology*. Silver Spring, MD: American Society for Parenteral and Enteral Nutrition; 2004.
36. Nehme AE. Nutritional support of the hospitalized patient: the team concept. *JAMA*. 1980;24:1906–1908.
37. Dalton MJ, Schepers G, Gee JP, et al. Consultative total parenteral nutrition support teams: the effect on the incidence of total parenteral nutrition-related complications. *JPEN J Parenteral Enteral Nutr*. 1984;8:146–152.
38. Bischoff SC, Kester L, Meier R, et al; Working group for developing the guidelines for parenteral nutrition of the German Association for Nutritional Medicine. Organisation, regulations, preparation and logistics of parenteral nutrition, in hospitals and homes; the role of the nutrition support team-Guidelines on parenteral nutrition, Chapter 8. *Ger Med Sci*. 2009;7:doc 20.
39. Goldstein M, Braitman LE, Levine GM. The medical and financial costs associated with termination of a nutrition support nurse. *JPEN J Parenter Enteral Nutr*. 2000;24:323–337.
40. Abad-Sinden AR, Vittone SB, Willson DF, Zaglul HF. Improving timeliness of nutrition intervention through a collaborative quality improvement program in a pediatric intensive care unit. *Nutr Clin Pract*. 1998;13:235–240.
41. Roberts MF, Levine GM. Nutritional support team recommendations can reduce hospital costs. *Nutr Clin Pract*. 1992;7:227–230.
42. Chrisanderson D, Heimburger DC, Morgan SL, et al. Metabolic complications of total parenteral nutrition: effects of a nutrition support service. *JPEN J Parenteral Enteral Nutr*. 1996;20:206–210.
43. Puntis JWL, Holden C, Smallman S, et al. Staff training-a key factor in reducing catheter sepsis. *Arch Dis Child*. 1991;66:335–337.
44. Gianino MS, Brunt LM, Eisenberg PG. The impact of a nutritional support team on the cost and management of multilumen central venous catheters. *J Intraven Nurs*. 1992;15:327–332.
45. Fisher GG, Opper FH. An interdisciplinary nutrition support team improves quality of care in a teaching hospital. *J Am Diet Assoc*. 1996;96:176–178.
46. Gales BJ, Riley DG. Improved total parenteral nutrition therapy management by a nutritional support team. *Hosp Pharm*. 1994;29:469–470.
47. Fettes SB, Lough M. An audit of the provision of parenteral nutrition in two acute hospitals: team versus non-team. *Scott Med J*. 2000;45:121–125.
48. Lopez-Martin C, Abilés J, Garrido Siles M, Faus Felipe V. Impact of the creation of a nutritional support team on the quality, safety and effectiveness of total parenteral nutrition. *Nutr Hosp*. 2012;27:871–878.
49. Rigo J, Marlowe ML, Bonnot D, et al. Benefits of a new pediatric triple-chamber bag for parenteral nutrition in preterm infants. *J Pediatr Gastroenterol Nutr*. 2012;54:210–217.
50. Colomb V, Marlowe ML, Bonnot D, Rigo J. Practical use of a new three-chamber bag for parenteral nutrition in pediatric patients. *e-SPEN*. 2012;7:e93–e99.

51. Gervasio J. Compounding vs standardized commercial parenteral product: pros and cons. *JPEN J Parenter Enteral Nutr*. 2012;36(2 suppl):40S–41S.
52. Howard P. Organizational aspects of starting and running an effective nutritional support service. *Clin Nutr*. 2001;20:367–374.
53. Ochoa JB, Magnuson B, Swintowsky M, et al. Long-term reduction in the cost of nutritional intervention achieved by a nutrition support service. *Nutr Clin Pract*. 2000;15:174–180.
54. Schneider PJ. Nutrition support teams: an evidence-based practice. *Nutr Clin Pract*. 2006;21:62–67.
55. Payne-James JJ. Cost-effectiveness of nutrition support-teams. Are they necessary? *Nutrition*. 1997;13:928–930.
56. Shang E, Hasenberg T, Schlegel B, et al. An European Survey of structure and organisation of nutrition support teams in Germany, Austria and Switzerland. *Clin Nutr*. 2005;24:1005–1013.
57. Holcombe BJ, Thorn DB, Strausburg MS. Analysis of the practice of nutrition support pharmacy specialists. *Pharmacotherapy*. 1995; 15:806–813.
58. Ebiasah RP, Schneider PJ, Pedersen CA, Mirtallo JM. Evaluation of board certification in nutrition support pharmacy. *JPEN J Parenteral Enteral Nutr*. 2002;26:239–247.
59. Senkal M, Dormann A, Stehle P, et al. Survey on structure and performance of nutrition-support teams in Germany. *Clin Nutr*. 2002;21:329–335.
60. Maisonneuve N, Raguso CA, Paolini-Giagobino A. Parenteral nutrition practices in hospital pharmacies in Switzerland, France and Belgium. *Nutrition*. 2004;20:528–535.

APPENDIX I

Nutritional Assessment

Melanie Connolly, MSc, RD

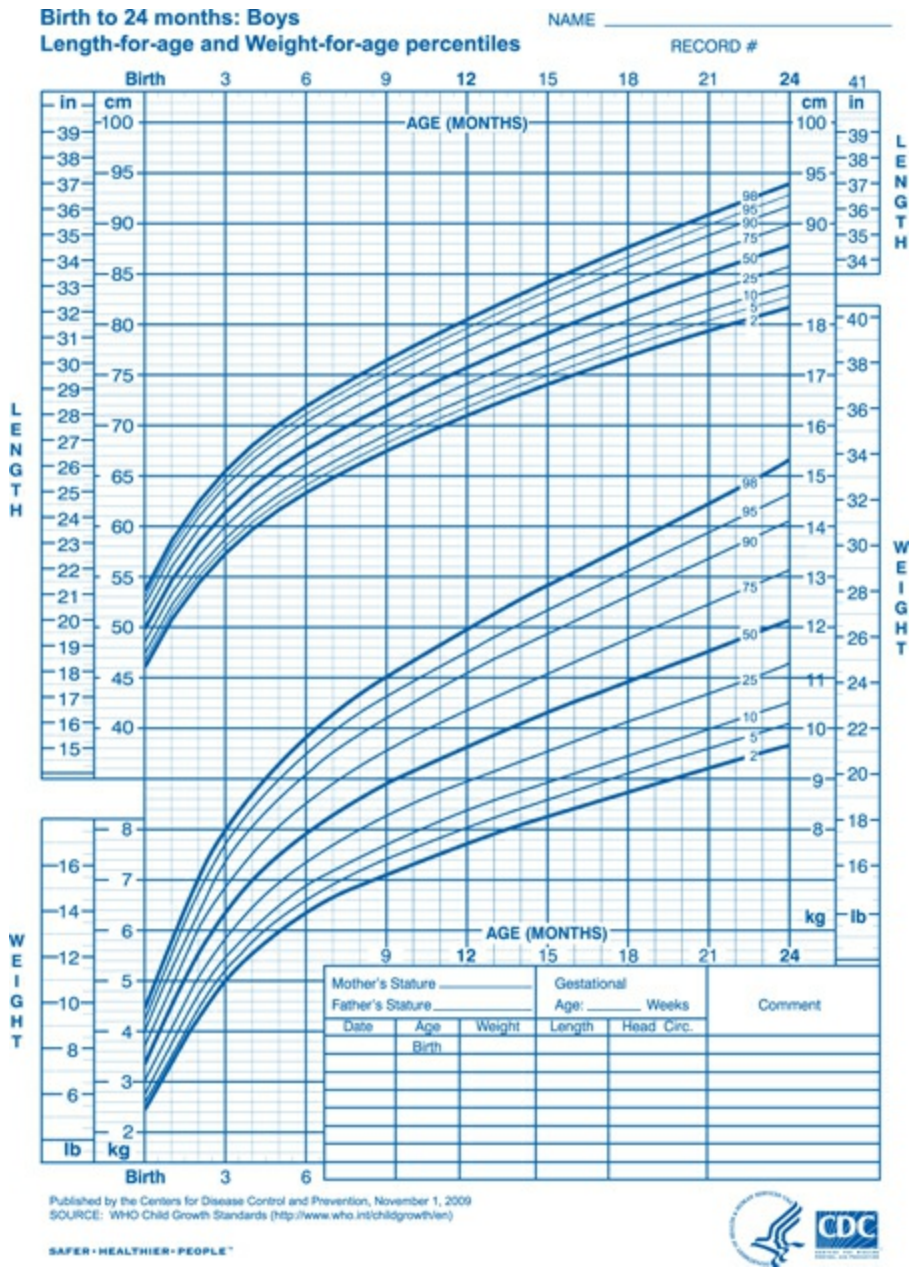


Figure I-1 Boys: length-for-age and weight-for-age percentiles (from birth to 24 months). (Developed by the World Health Organization [2006], www.cdc.gov/growthcharts/who_charts.htm.)

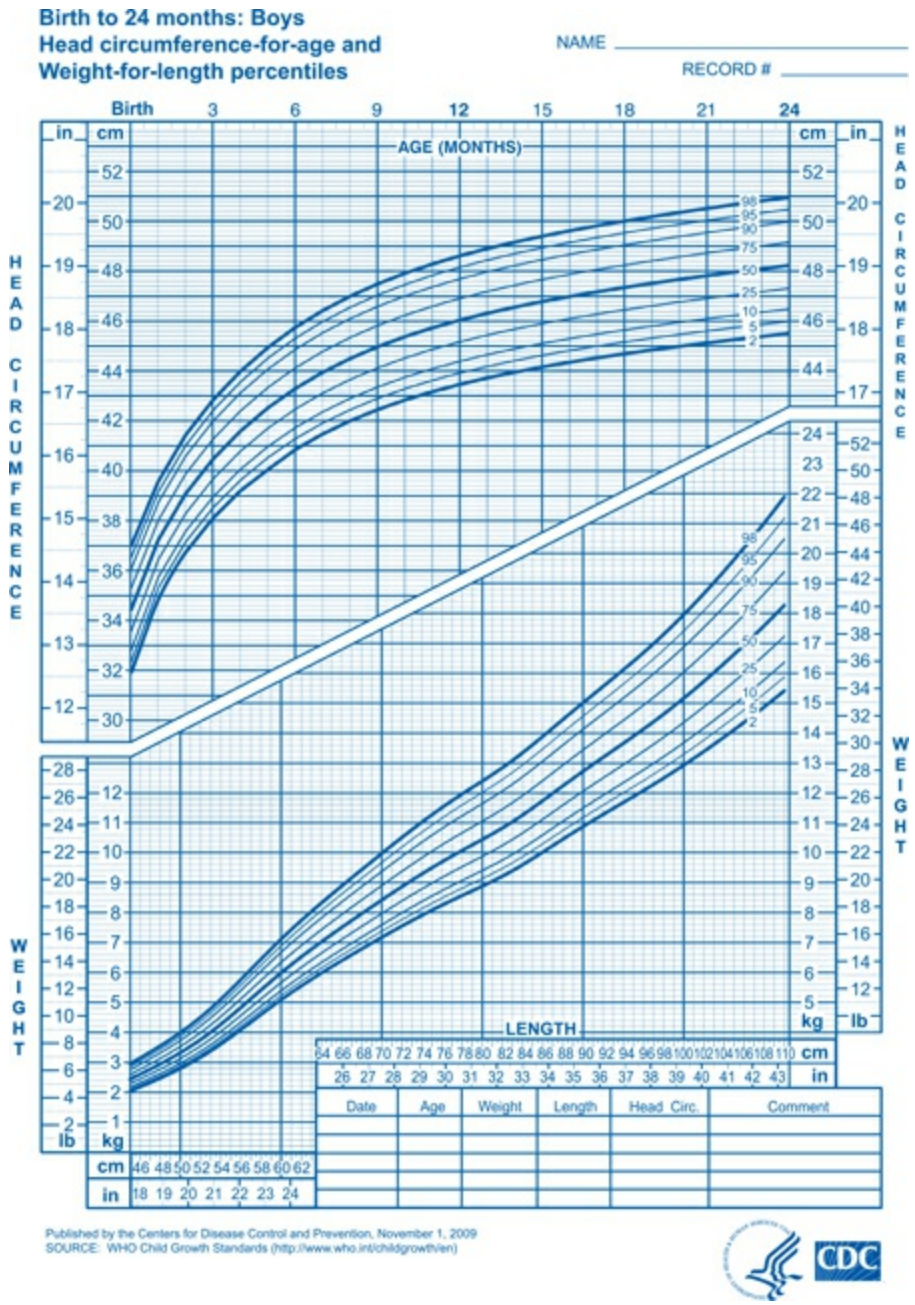


Figure I-2 Boys: weight-for-length and head circumference-for-age percentiles (from birth to 24 months). (Developed by the World Health Organization [2006], www.cdc.gov/growthcharts/who_charts.htm.)

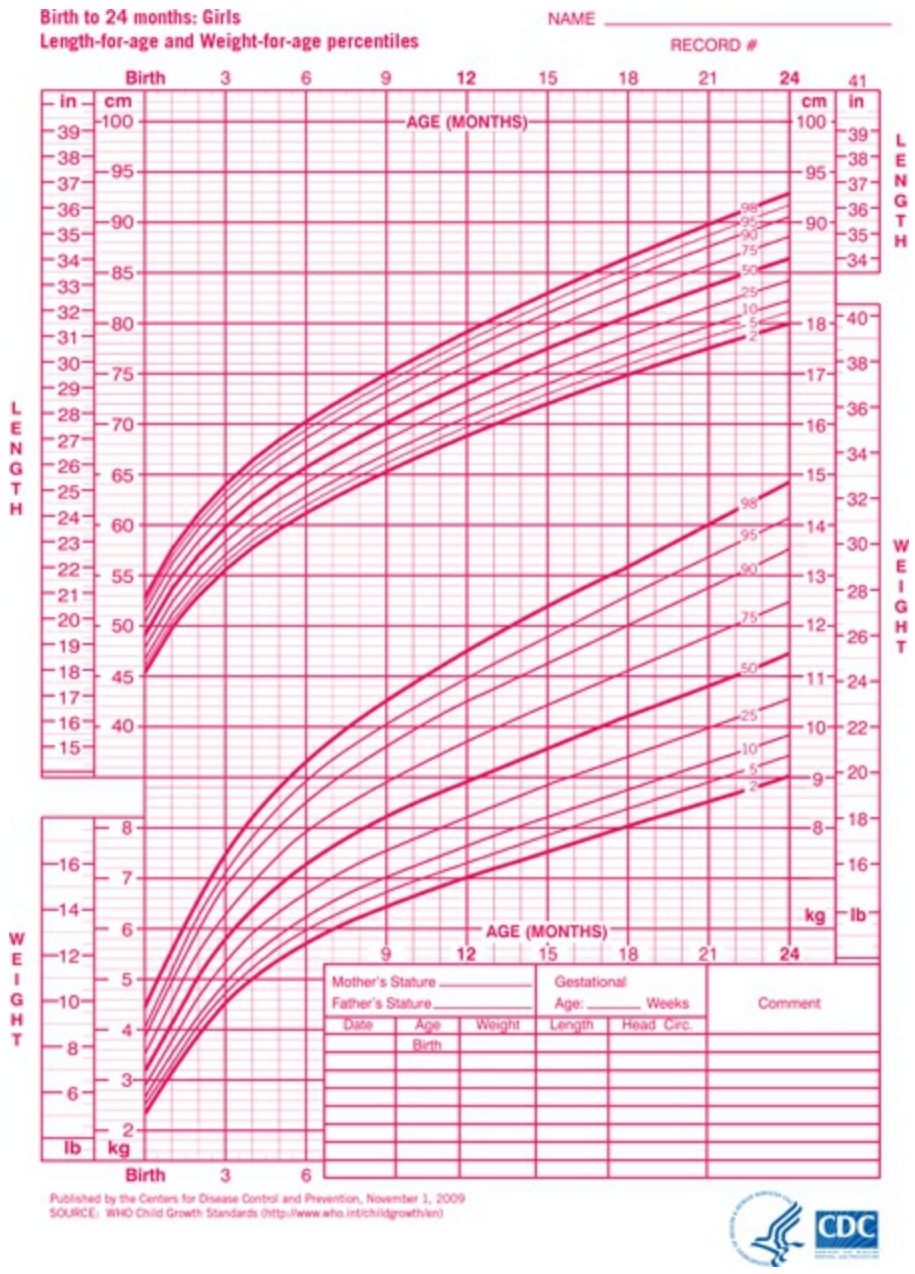


Figure I-3 Girls: length-for-age and weight-for-age percentiles (from birth to 24 months). (Developed by the World Health Organization [2006], www.cdc.gov/growthcharts/who_charts.htm.)

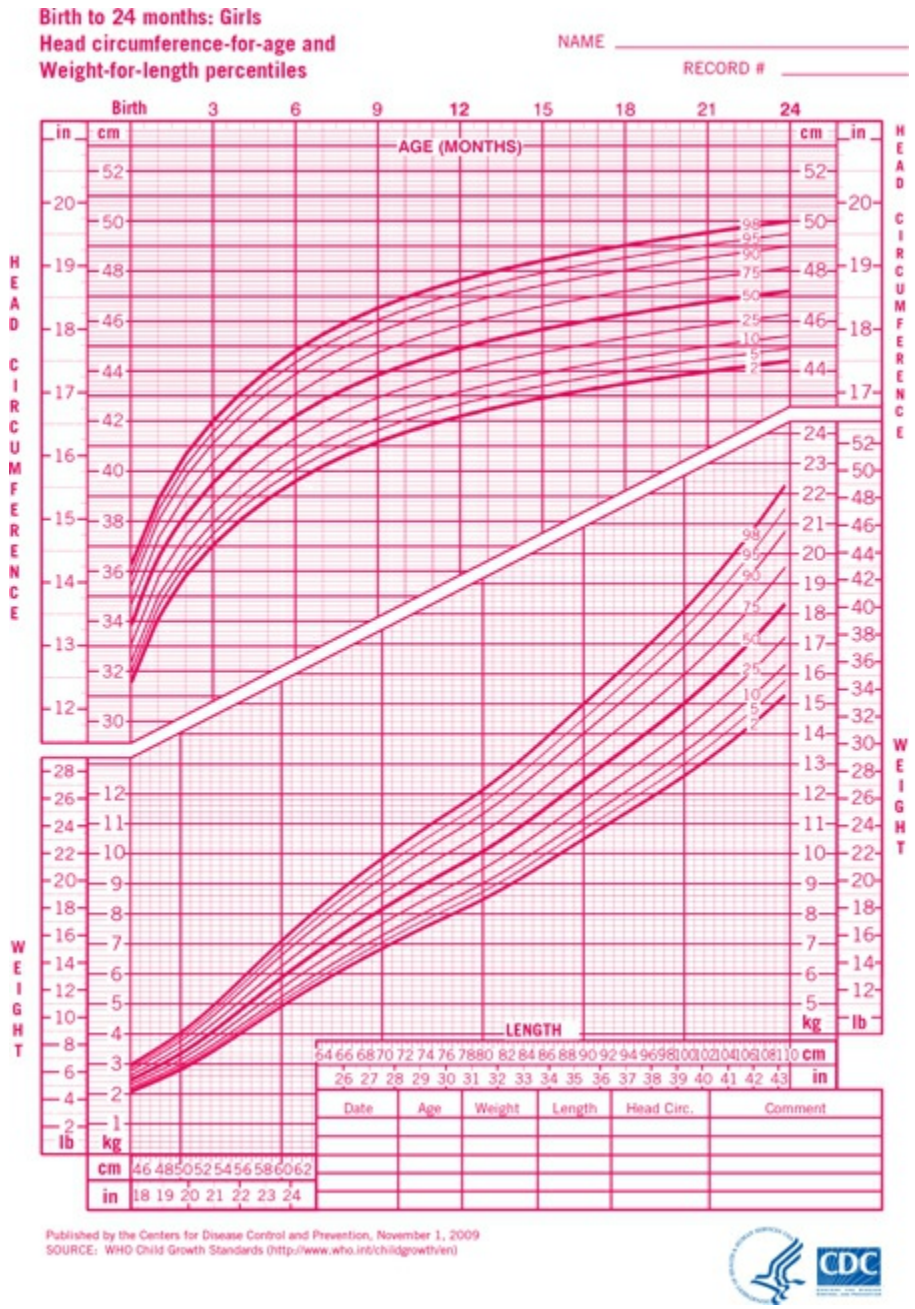


Figure I-4 Girls: weight-for-length and head circumference-for-age percentiles (from birth to 24 months). (Developed by the World Health Organization [2006], www.cdc.gov/growthcharts/who_charts.htm.)

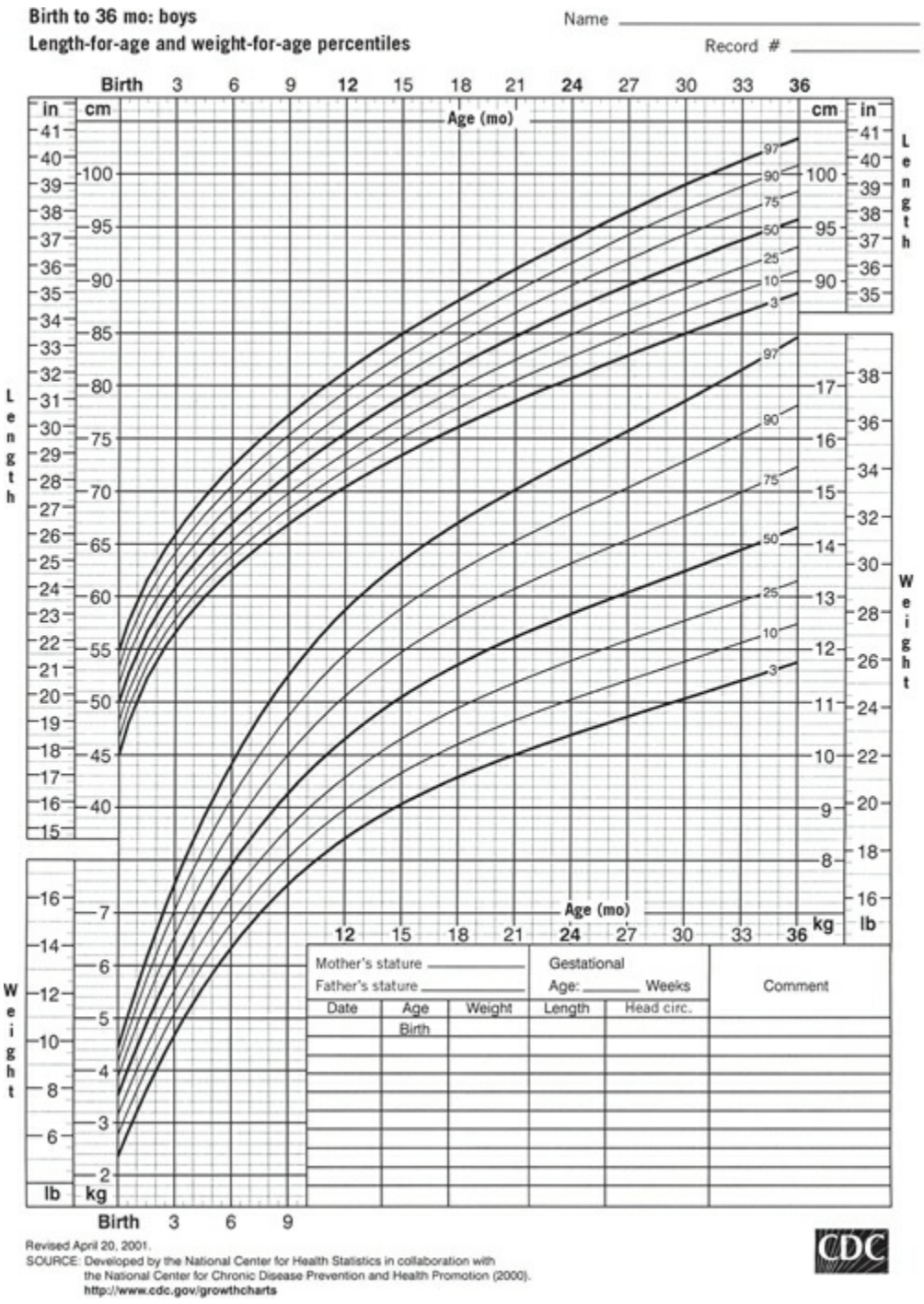


Figure I-5 Birth to 36 months: boys, length-for-age and weight-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000], http://www.cdc.gov/growthcharts.)

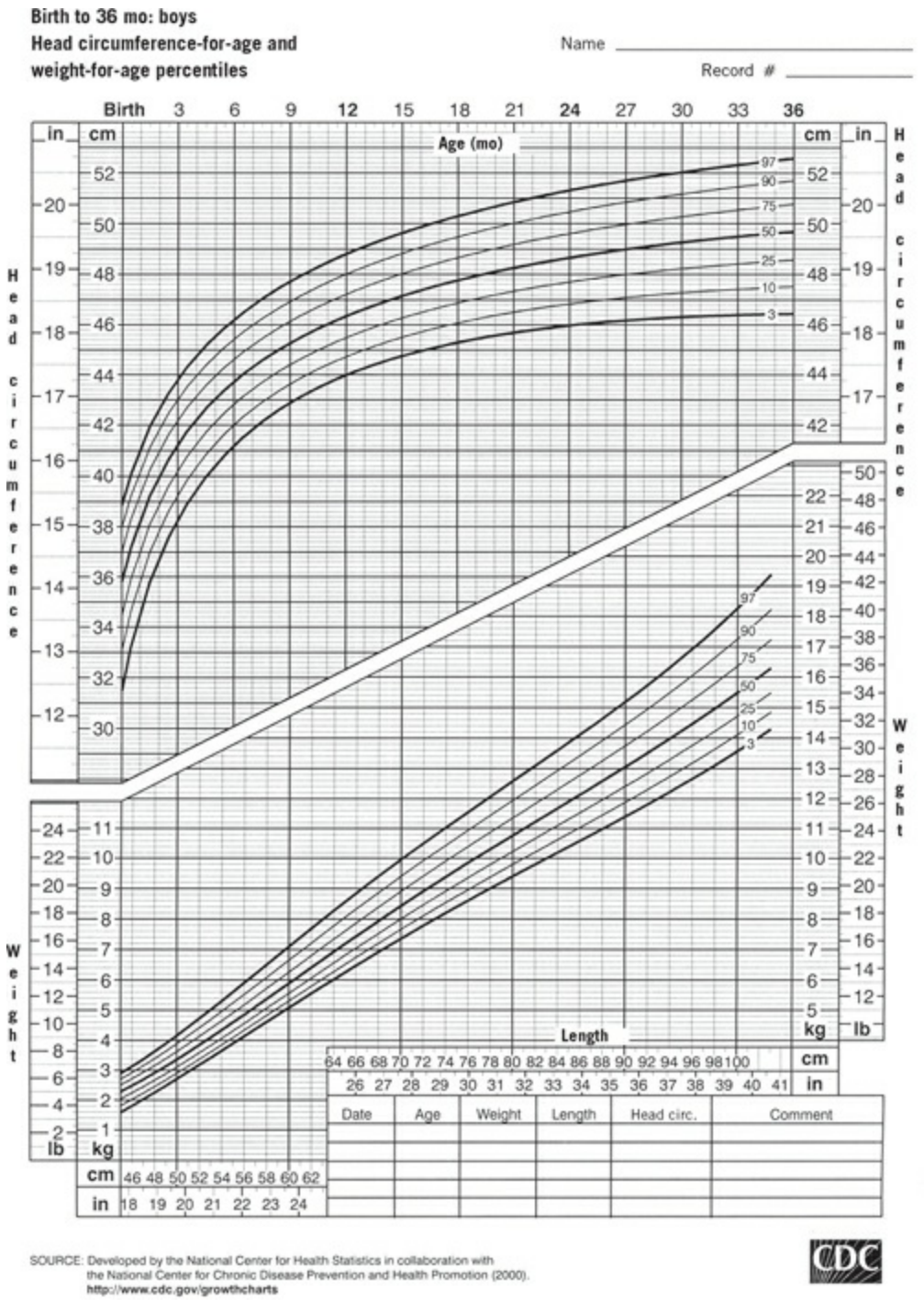


Figure I-6 Birth to 36 months: boys, head circumference-for-age and weight-for-length percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000], http://www.cdc.gov/growthcharts.)

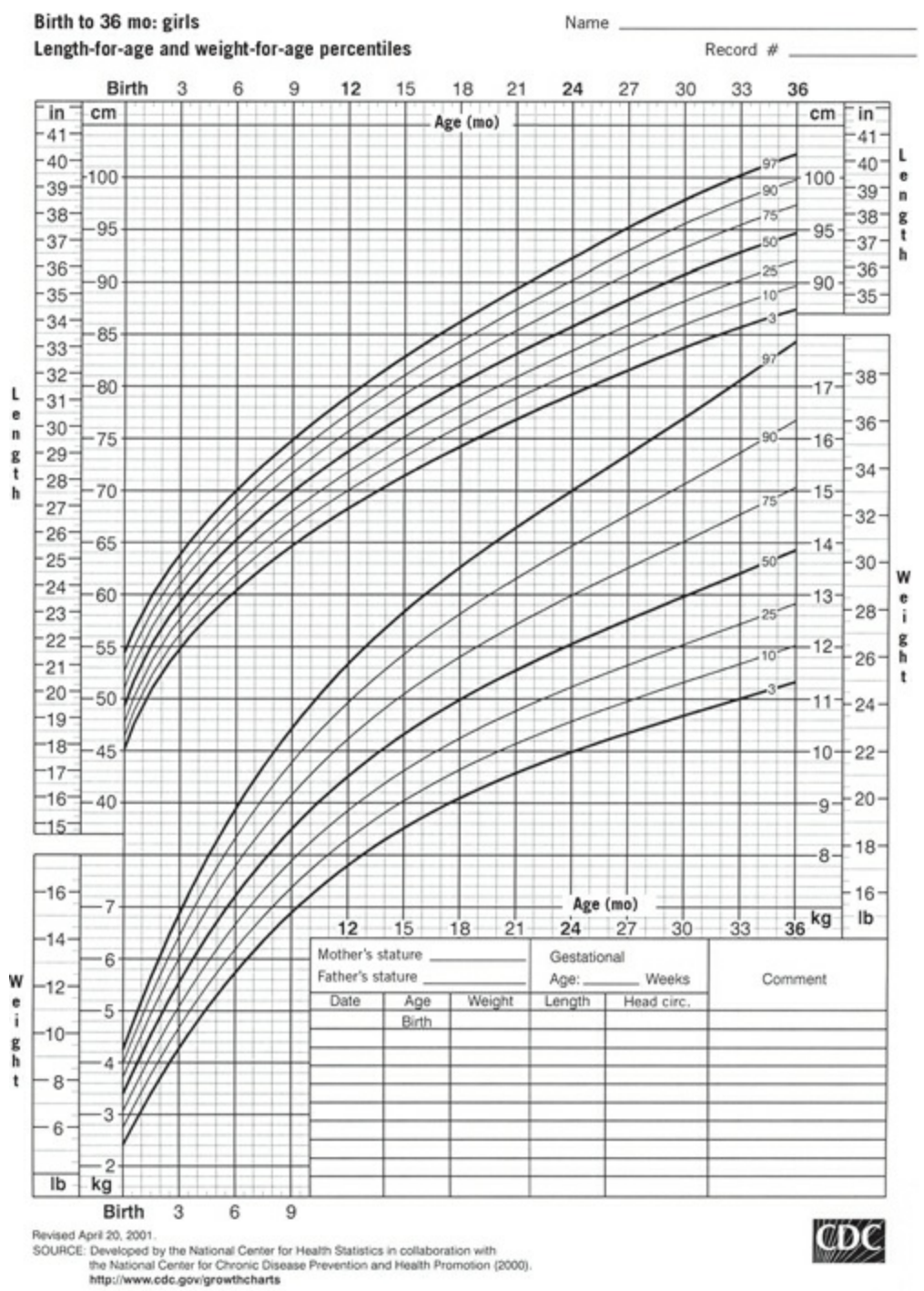


Figure I-7 Birth to 36 months: girls, length-for-age and weight-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000], http://www.cdc.gov/growthcharts.)

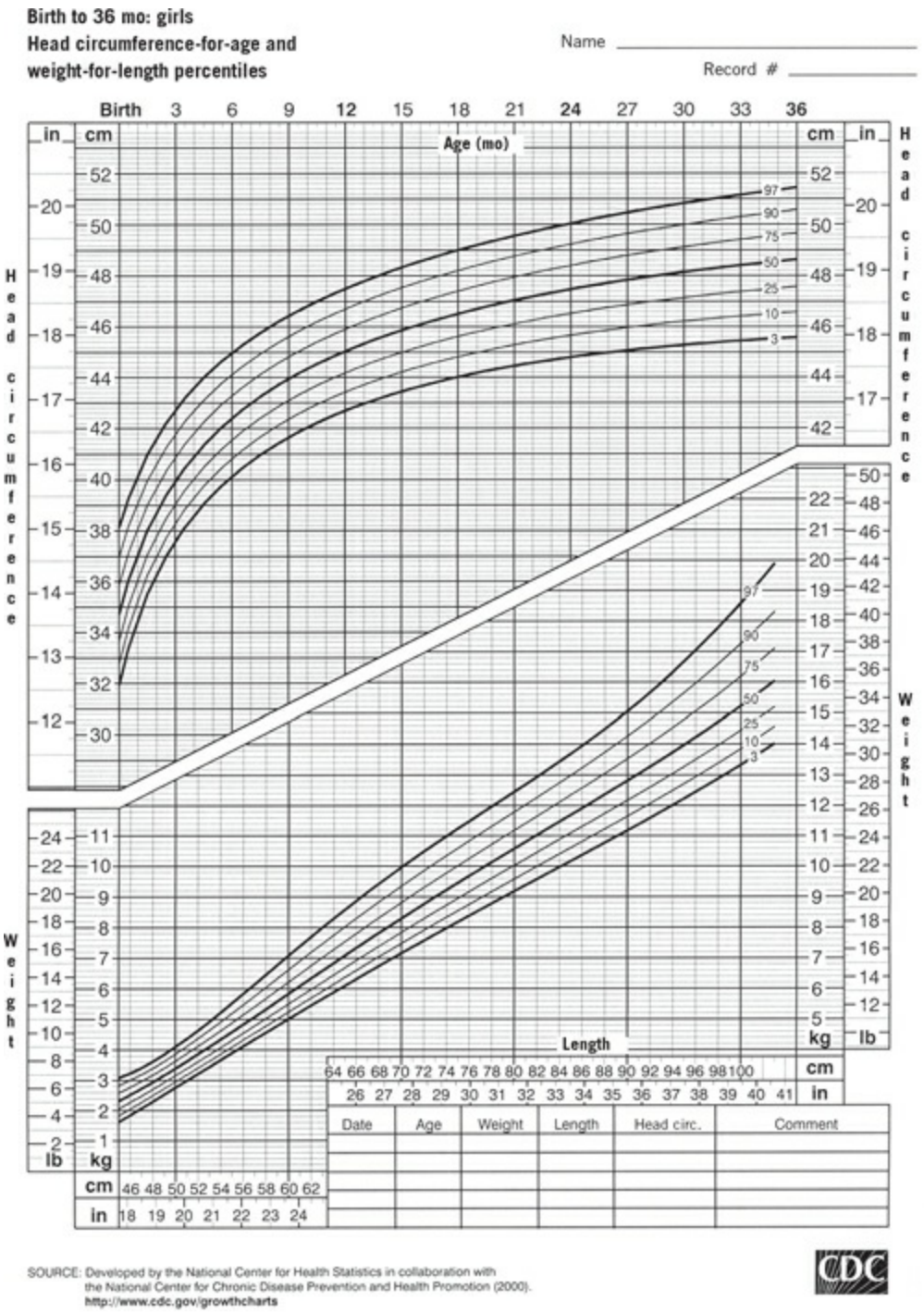


Figure I-8 Birth to 36 months: girls, head circumference-for-age and weight-for-length percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000], http://www.cdc.gov/growthcharts.)

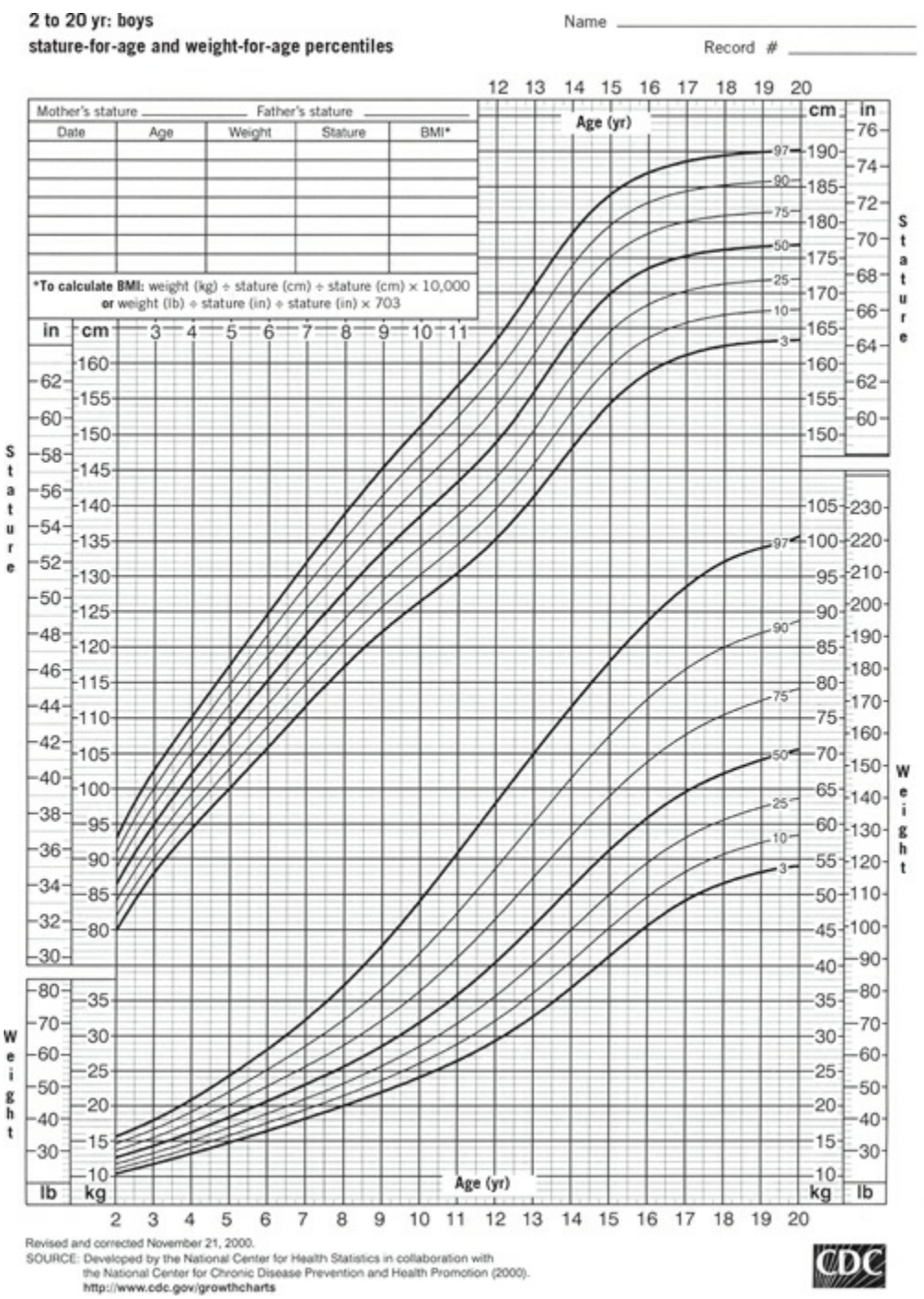


Figure I-9 Two to 20 years: boys, stature-for-age and weight-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].)

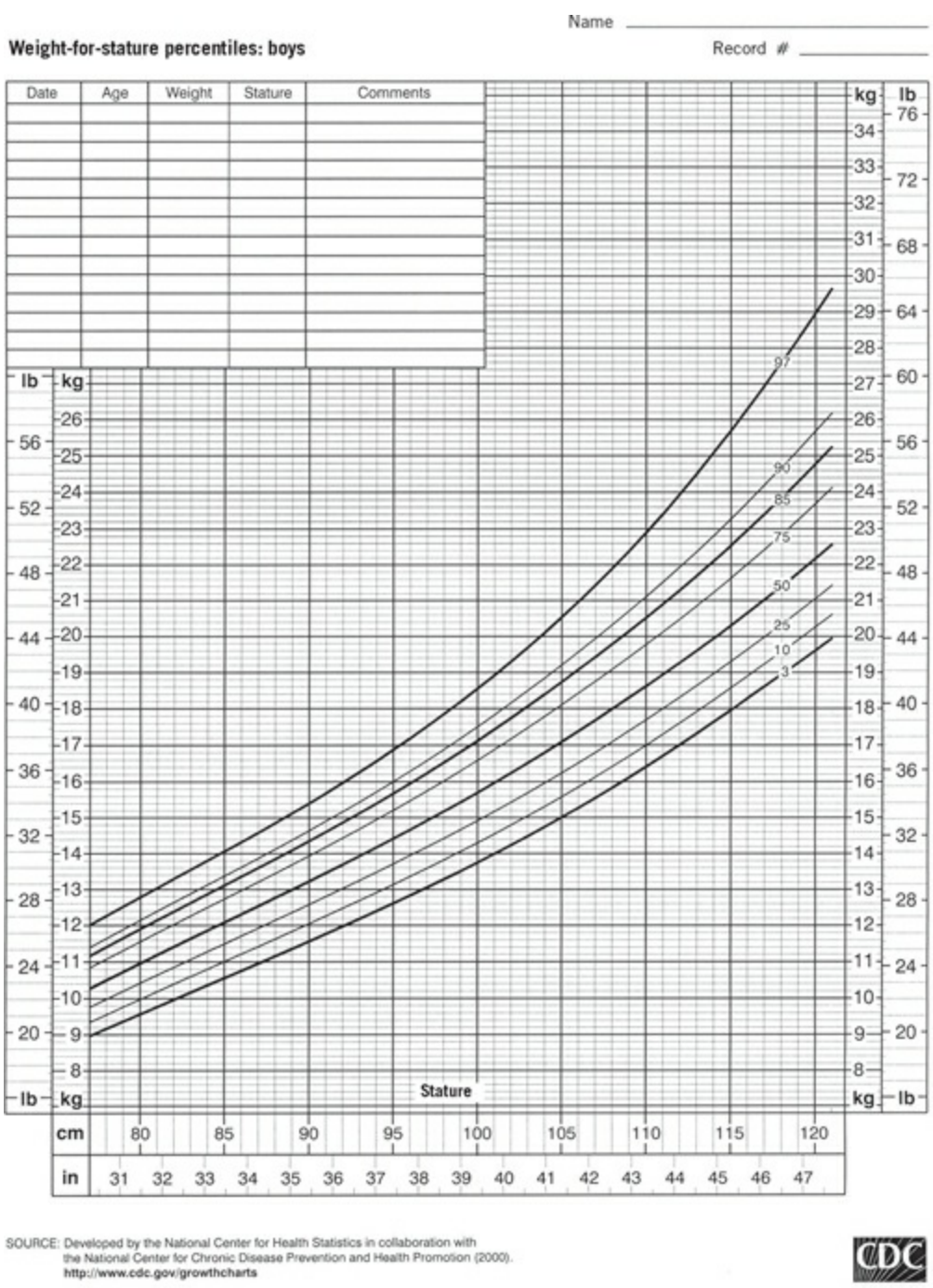


Figure I-10 Boys: weight-for-stature percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].)

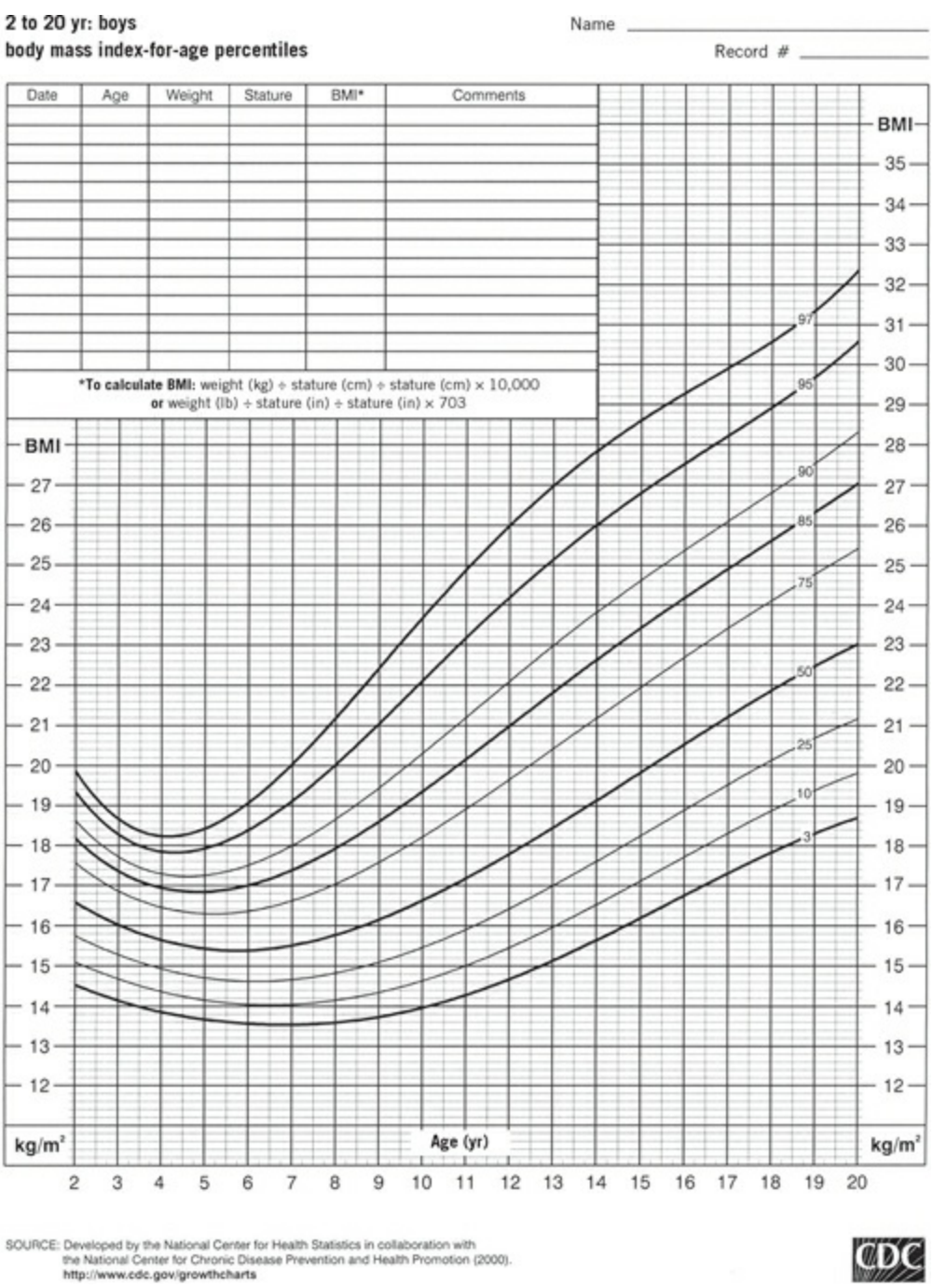


Figure I-11 Two to 20 years: boys, body mass index-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].)

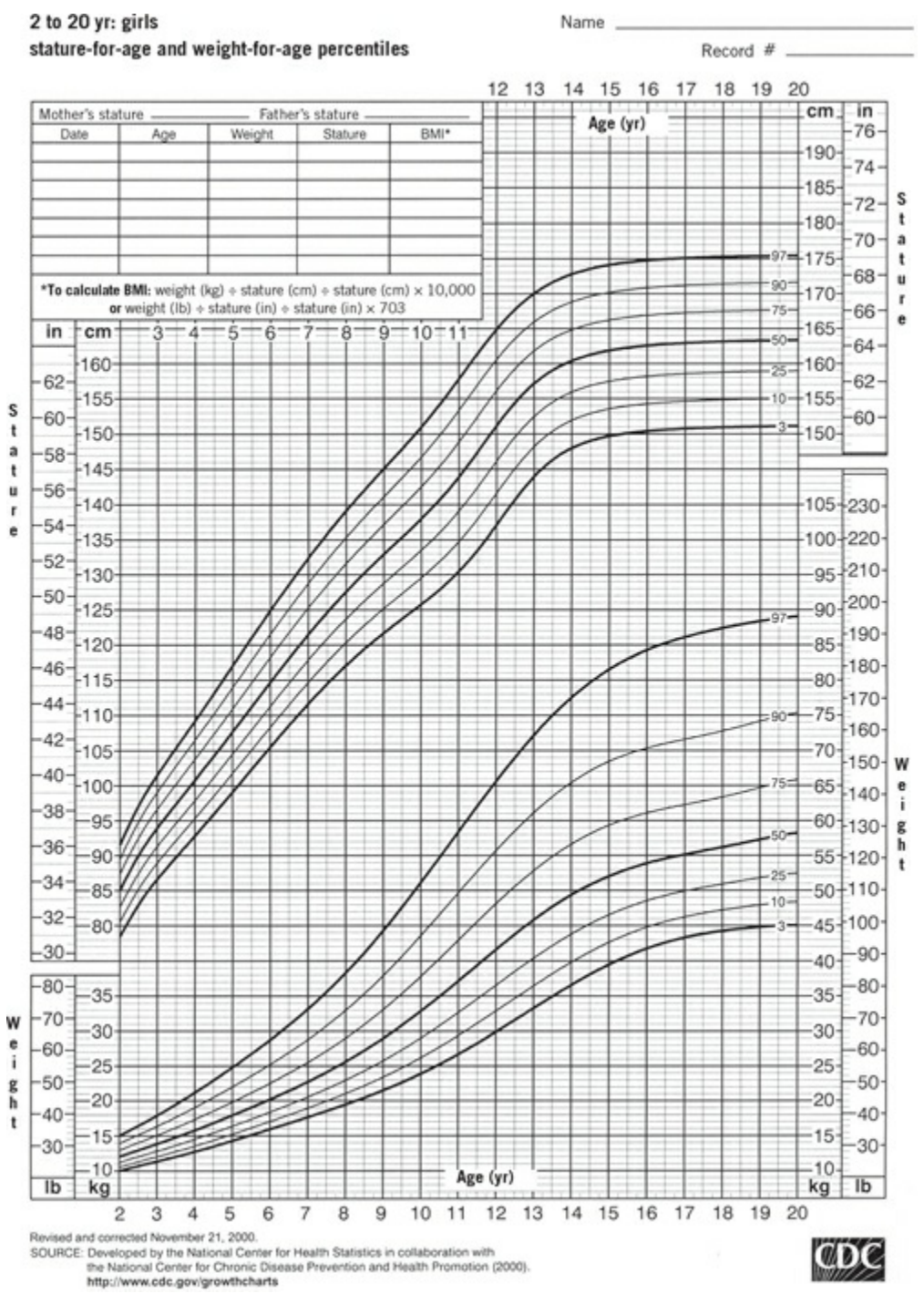


Figure I-12 Two to 20 years: girls, stature-for-age and weight-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].)

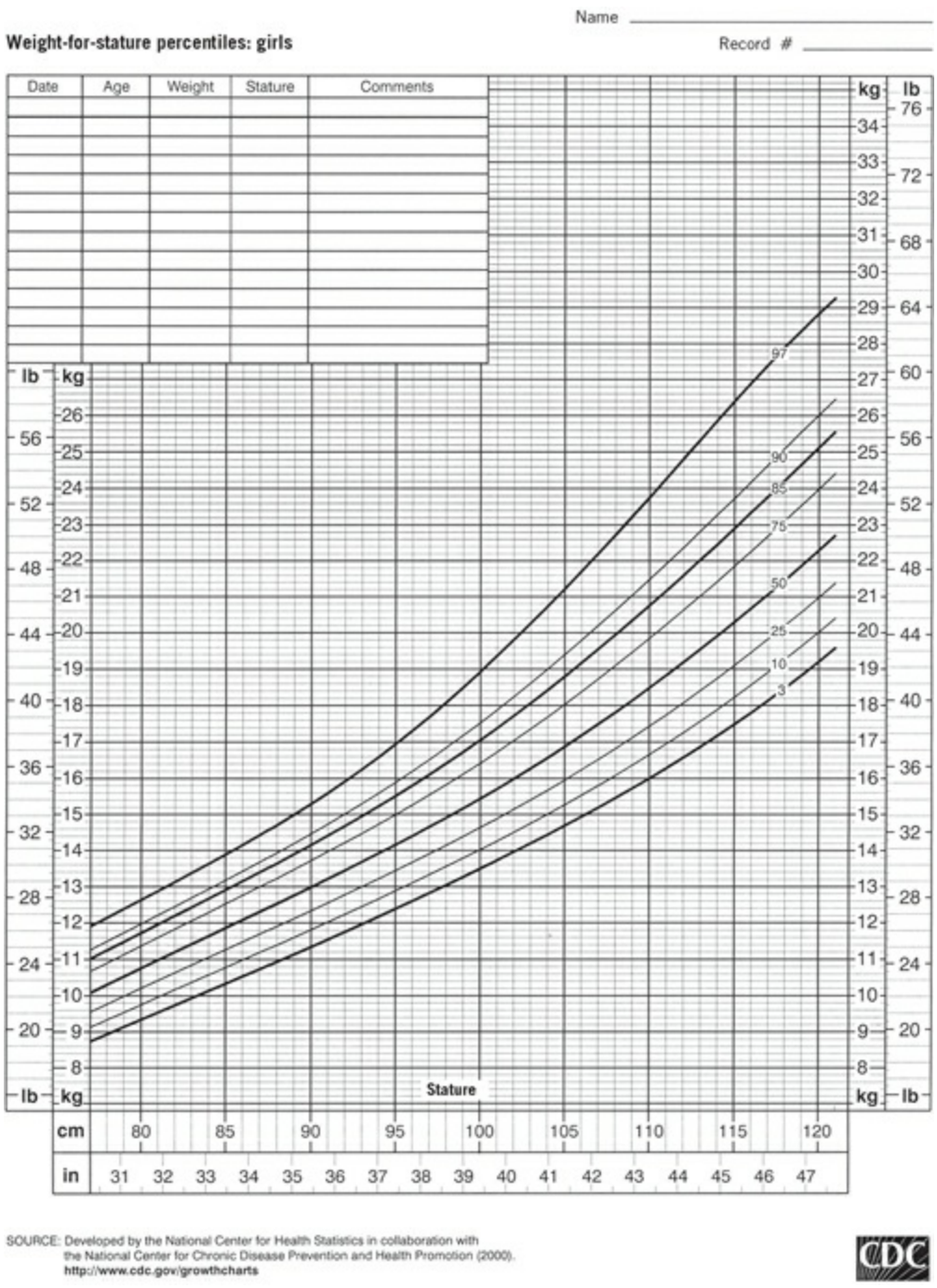


Figure I-13 Girls: weight-for-stature percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].).

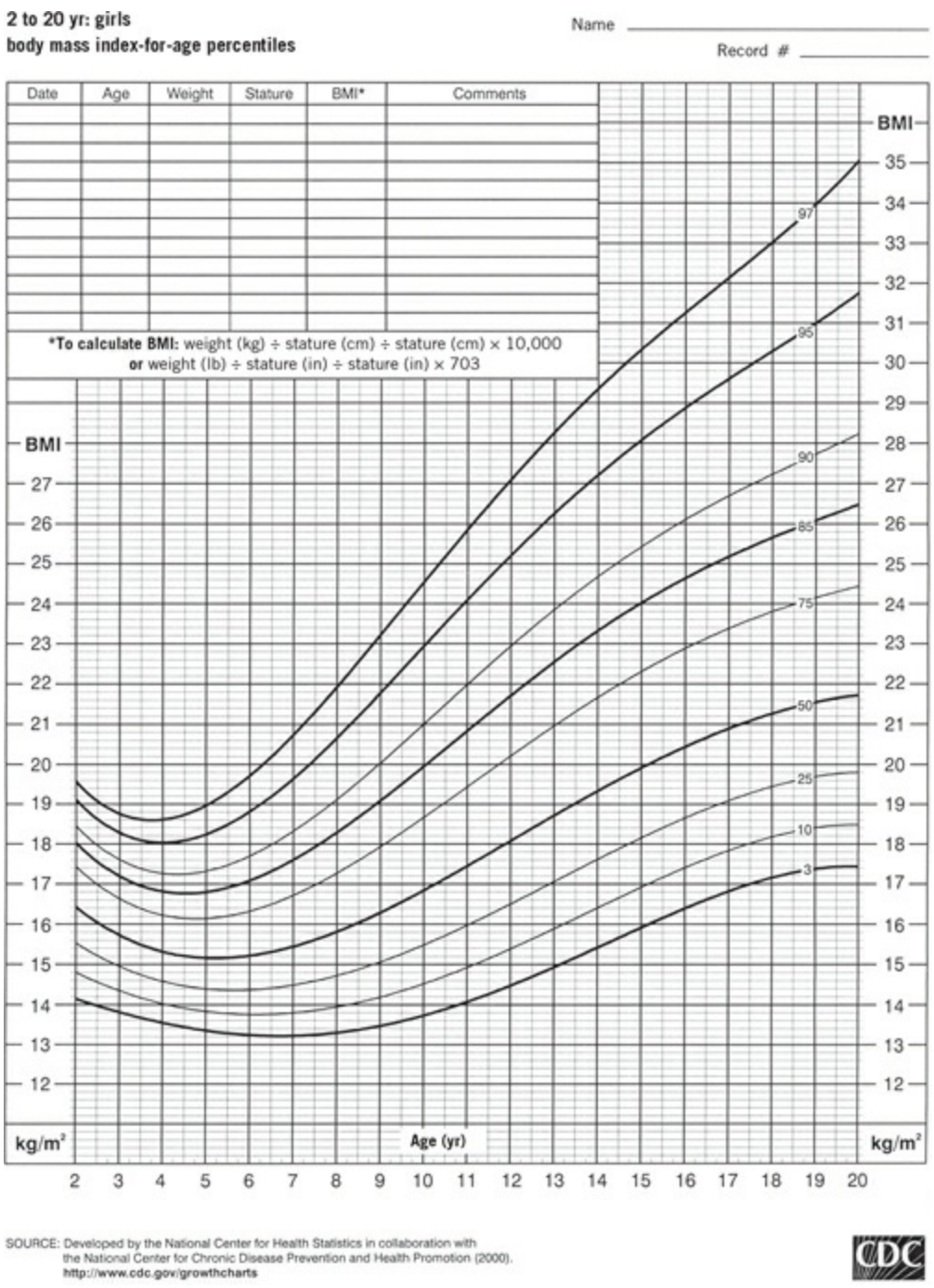


Figure I-14 Two to 20 years: girls, body mass index-for-age percentiles. (Developed by the National Center for Health Statistics in collaboration with the National Center for Chronic Disease Prevention and Health Promotion [2000].)

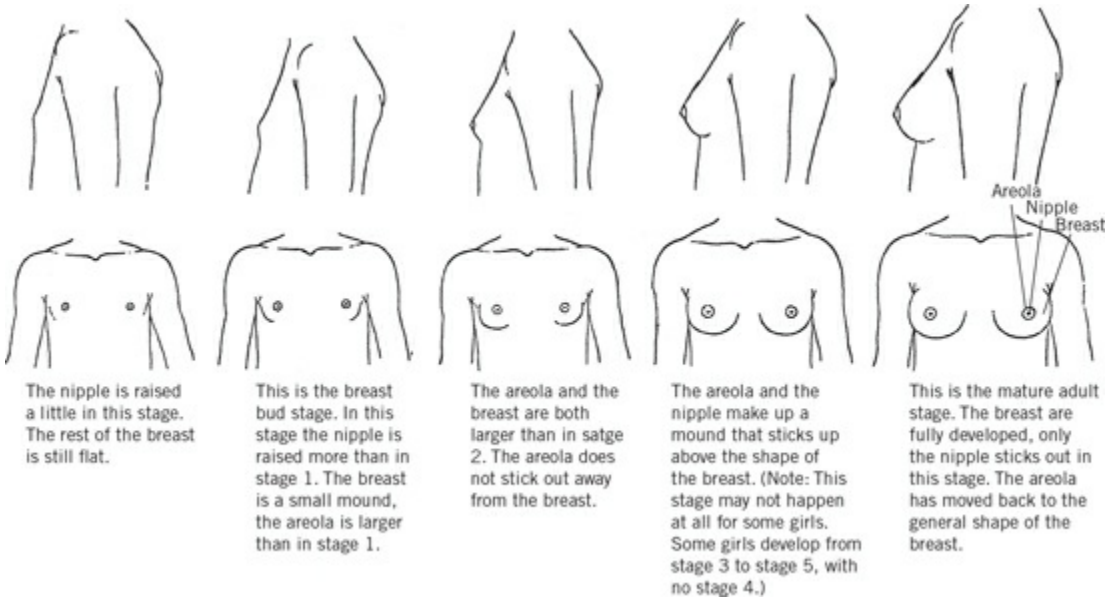


Figure I-15 Typical progression of female pubertal development. (Reproduced with permission from Springer Science and Business Media. Morris NM, Udry JR. Validation of a self-administered instrument to assess stage of adolescent development. *J Youth Adolesc.* 1980;9(3):271–280.)



Figure I-16 Typical progression of female pubertal development. (Reproduced with permission from Springer Science and Business Media. Morris NM, Udry JR. Validation of a self-administered instrument to assess stage of adolescent development. *J Youth Adolesc.* 1980;9(3):271–280.)

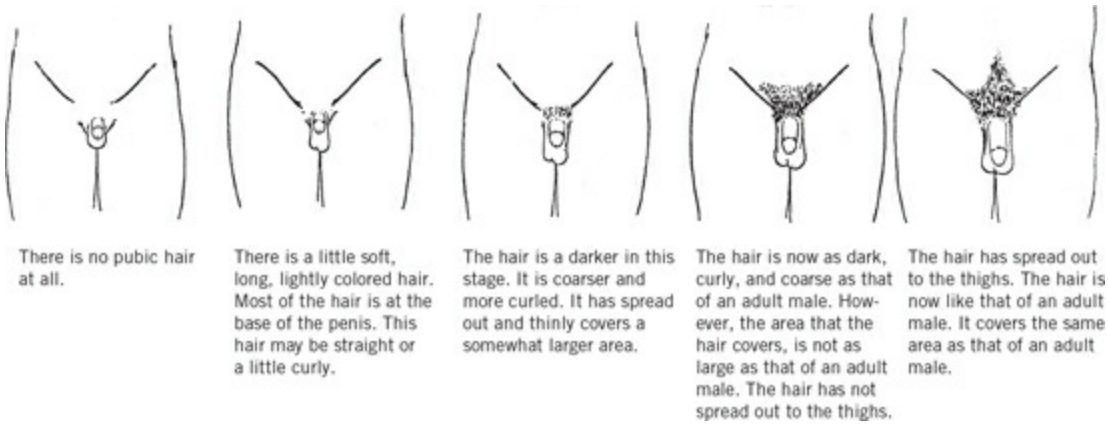


Figure I-17 Typical progression of male pubertal development. (Reproduced with permission from Springer Science and Business Media. Morris NM, Udry JR. Validation of a self-administered instrument to assess stage of adolescent development. *J Youth Adolesc.* 1980;9(3):271–280.)

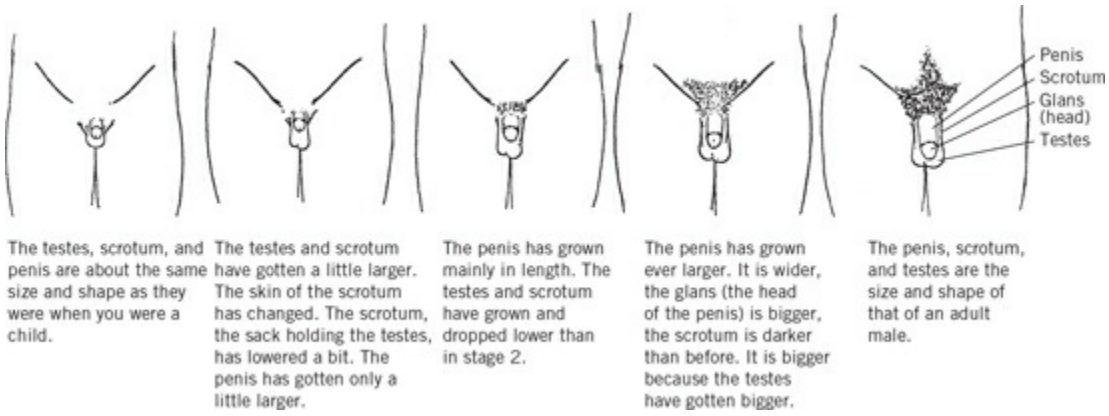


Figure I-18 Typical progression of male pubertal development. (Reproduced with permission from Springer Science and Business Media. Morris NM, Udry JR. Validation of a self-administered instrument to assess stage of adolescent development. *J Youth Adolesc.* 1980;9(3):271–280.)

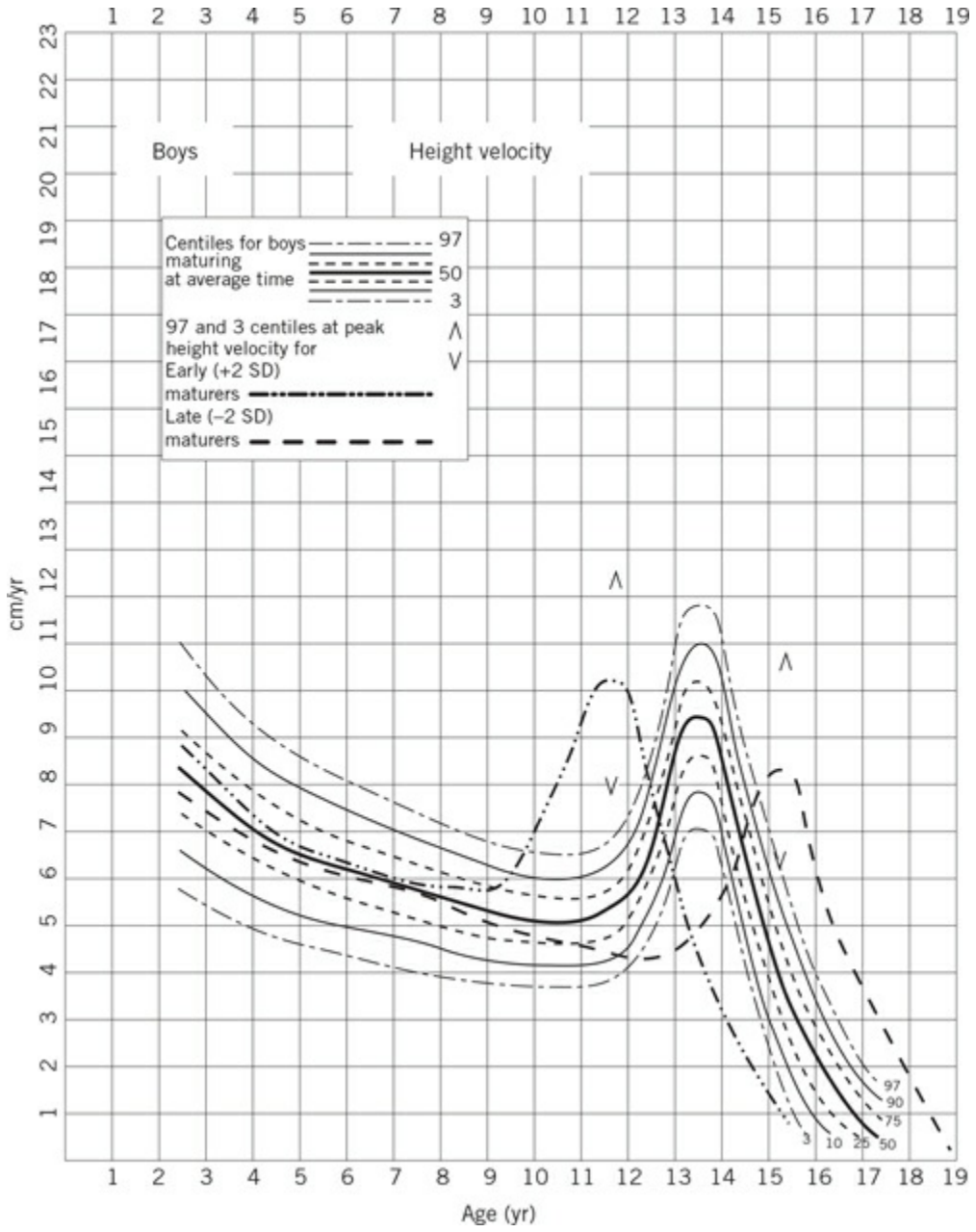


Figure I-19 Height velocity for American boys. (Reproduced from Tanner JM, Davies PSW. Clinical longitudinal standards for height and height velocity for North American children. *J Pediatr*. 1985;107(3):317–329. With permission from Elsevier.)

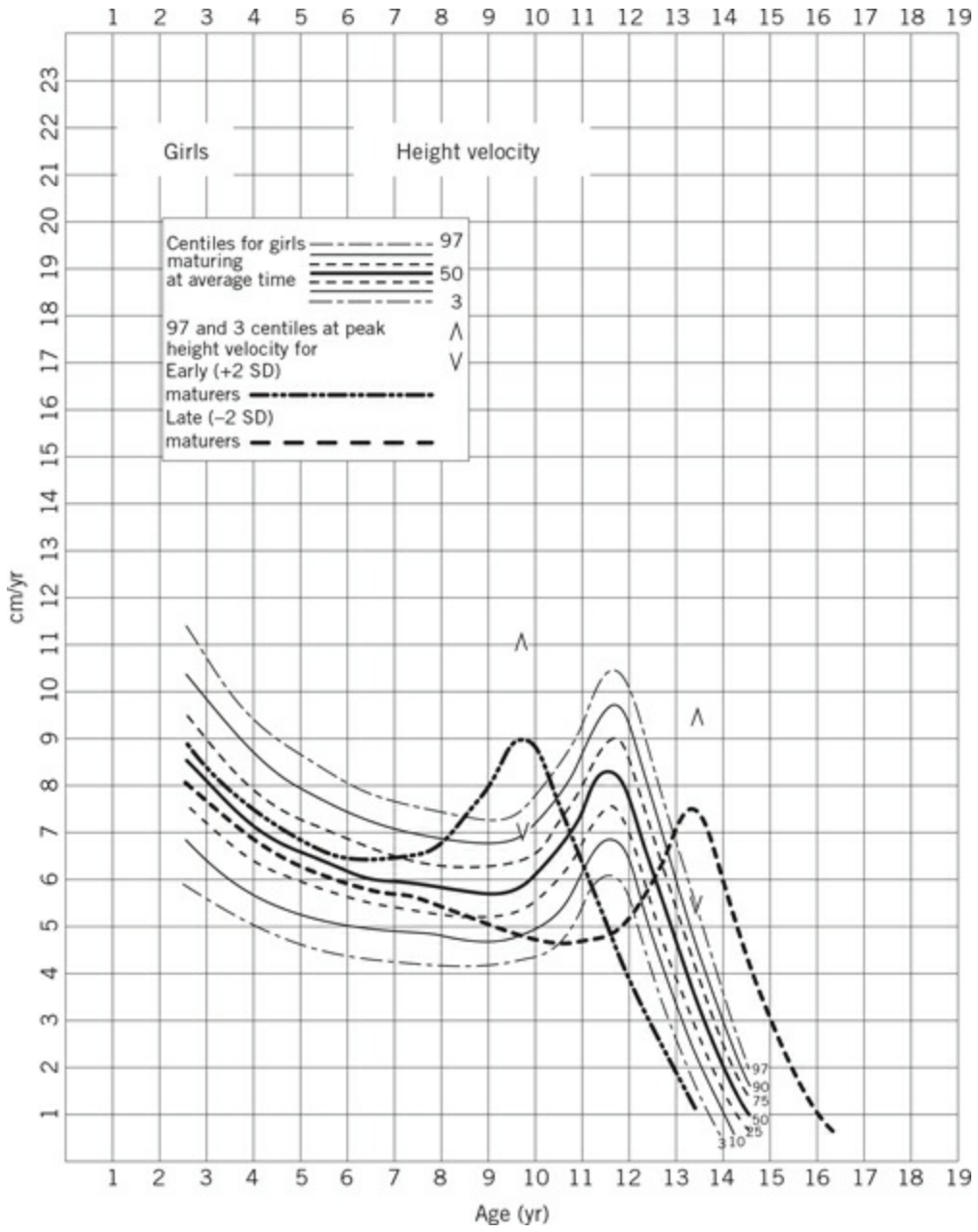


Figure I-20 Height velocity for American girls. (Reproduced from Tanner JM, Davies PSW. Clinical longitudinal standards for height and height velocity for North American children. *J Pediatr.* 1985;107:317–329. With permission from Elsevier.)

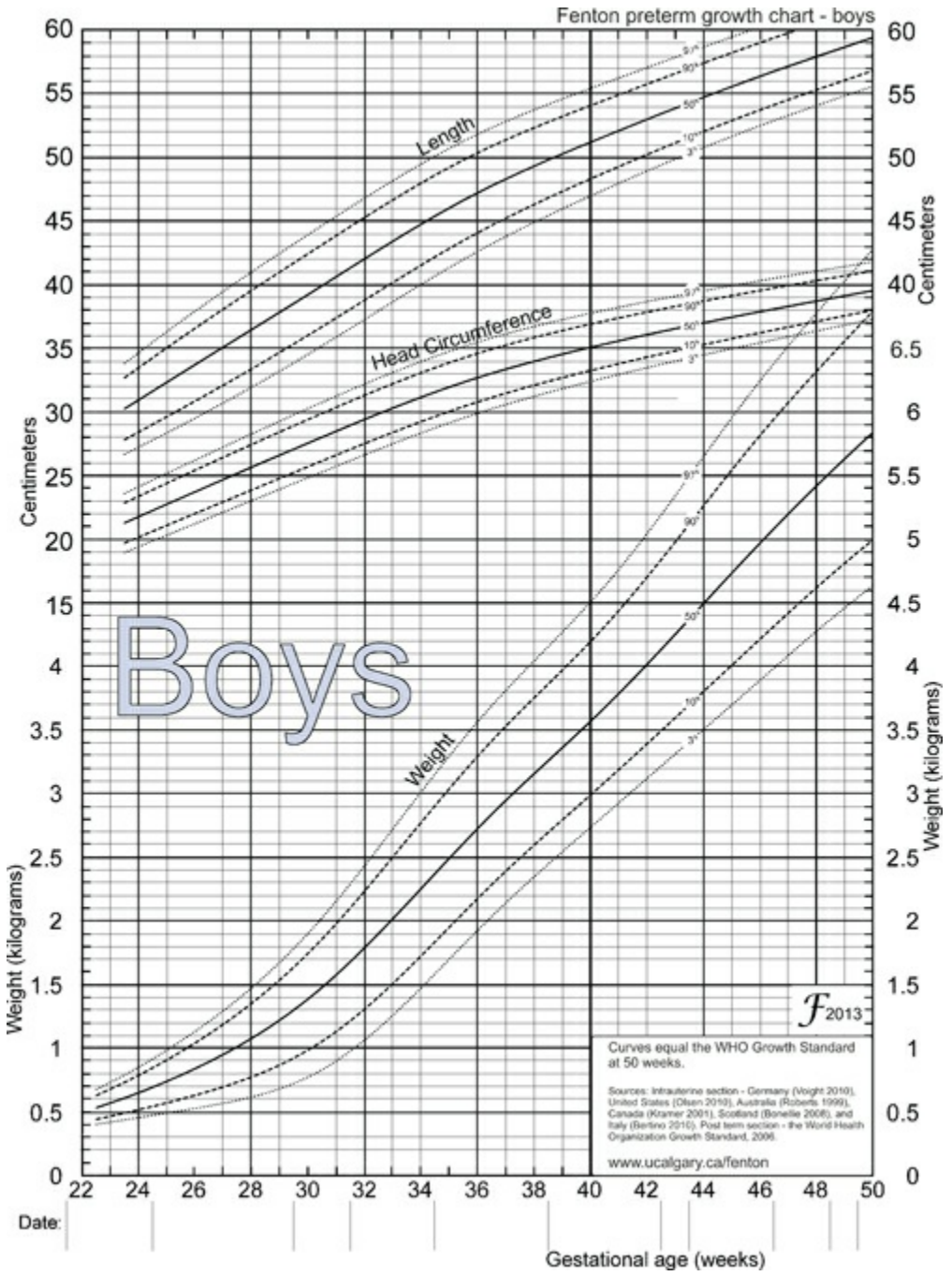


Figure I-21 Length-for-age, head circumference-for-age, and weight-for-age curves for premature boys 22–50 weeks gestational age. (Reproduced from Fenton TR, Kim JH. A systematic review and meta-analysis to revise the Fenton growth chart for preterm infants. *BMC Pediatrics*. 2013;13:59. Copyright 2013 Fenton and Kim.)

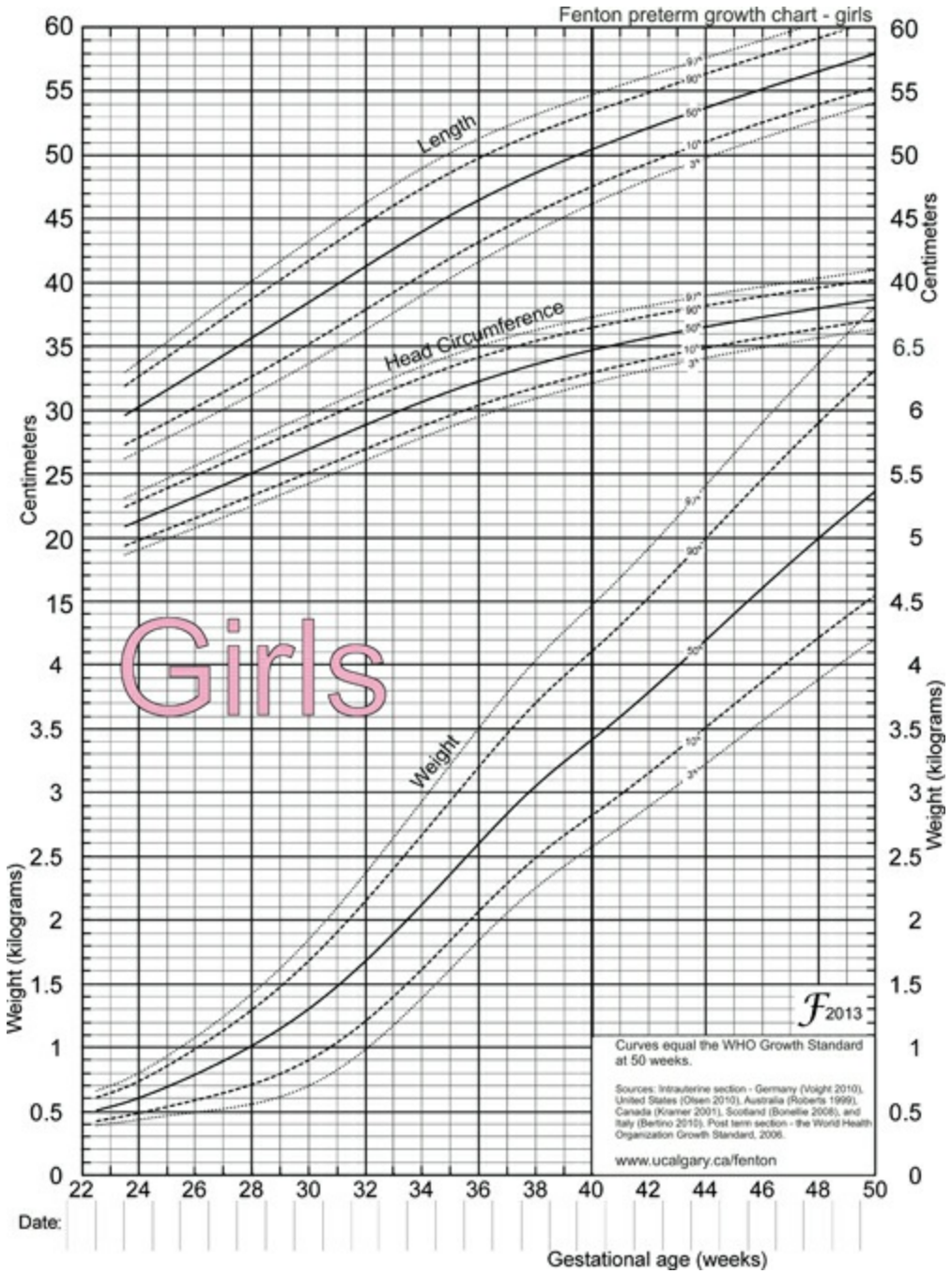


Figure I-22 Length-for-age, head circumference-for-age, and weight-for-age curves for premature girls, 22–50 weeks gestational age. (Reproduced from Fenton TR, Kim JH. A systematic review and meta-analysis to revise the Fenton growth chart for preterm infants. *BMC Pediatrics*. 2013;13:59. Copyright 2013 Fenton and Kim.)

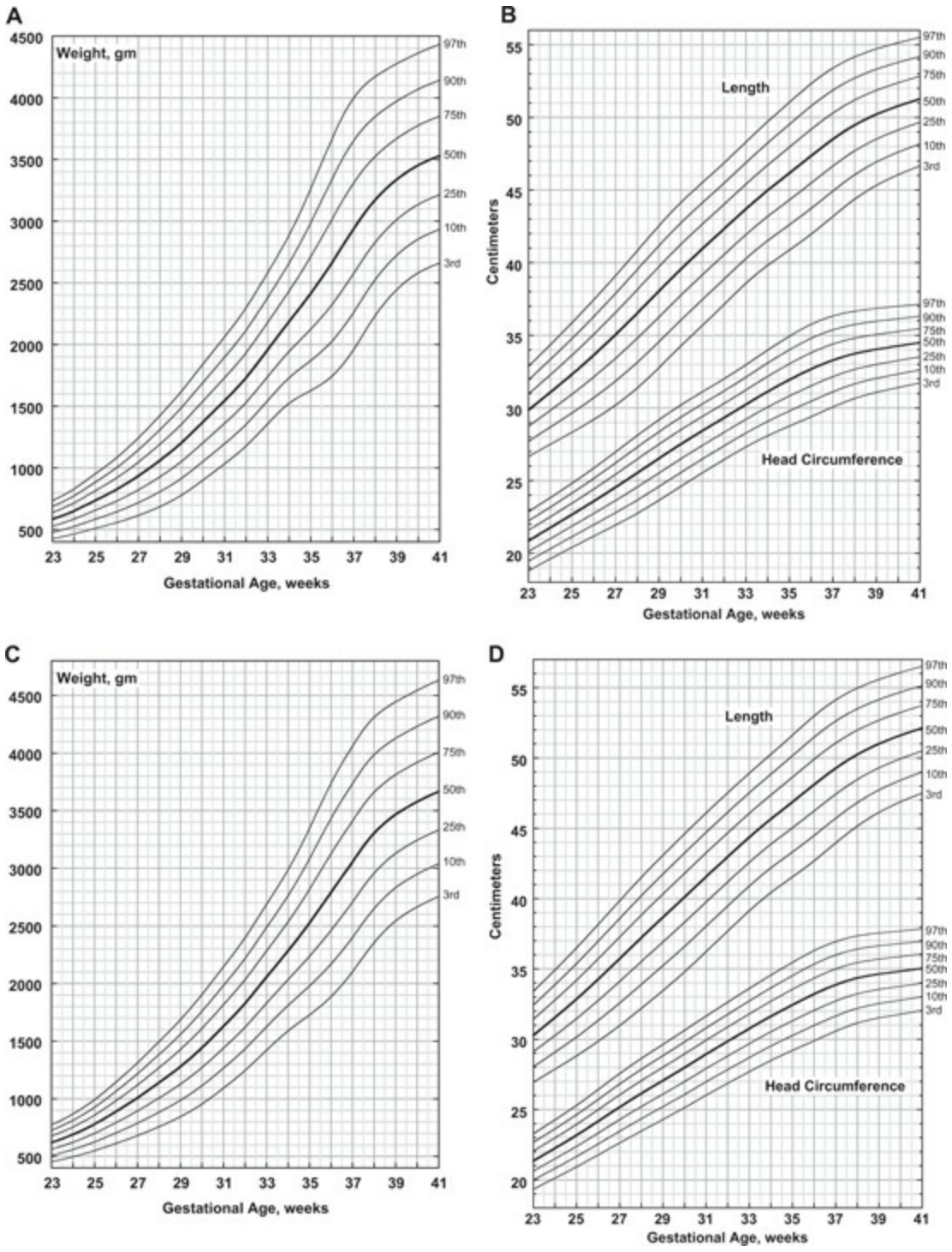


Figure I-23 Length-for-age, weight-for-age, and head circumference-for-age curves for girls (A, B) and boys (C, D), 23–41 weeks gestational age. (Reproduced with permission from Olsen IE, Groveman SA, Lawson L, et al. New intrauterine growth curves based on United States data. *Pediatrics*. 2010;125:e214–e224. Copyright 2010 by the AAP.)

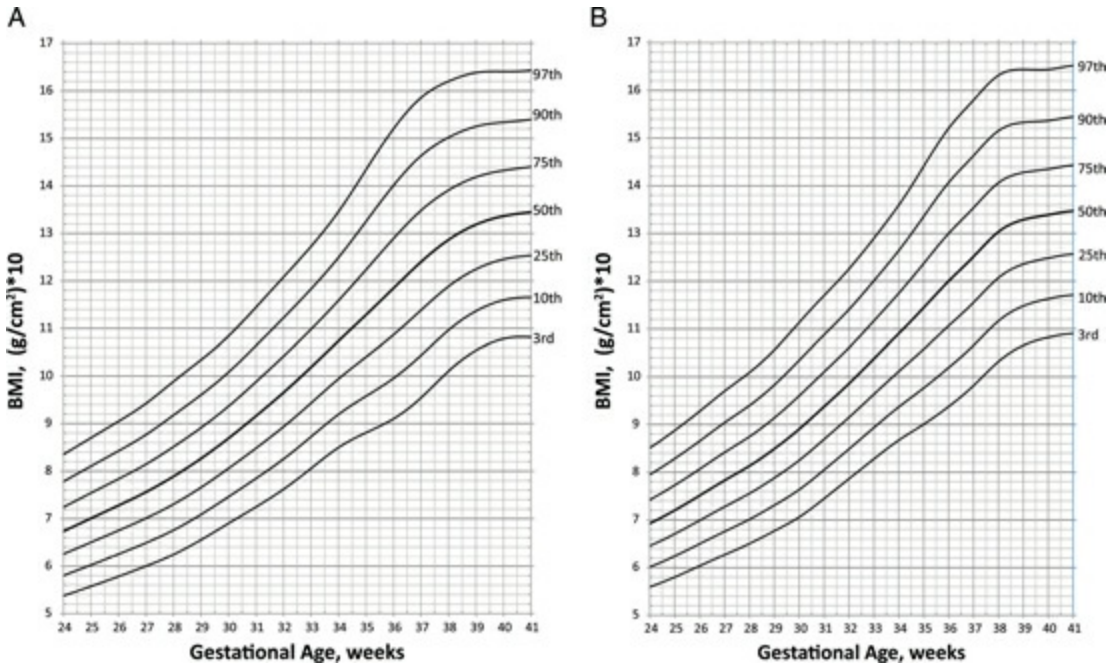


Figure I-24 BMI-for-age curves for premature girls (A) and boys (B), 24–41 weeks gestational age. (Reproduced with permission from Olsen IE, Lawson L, Ferguson N, et al. BMI curves for preterm infants. *Pediatrics*. 2015; 135(3): e572–e581. Copyright 2015 by the AAP.)

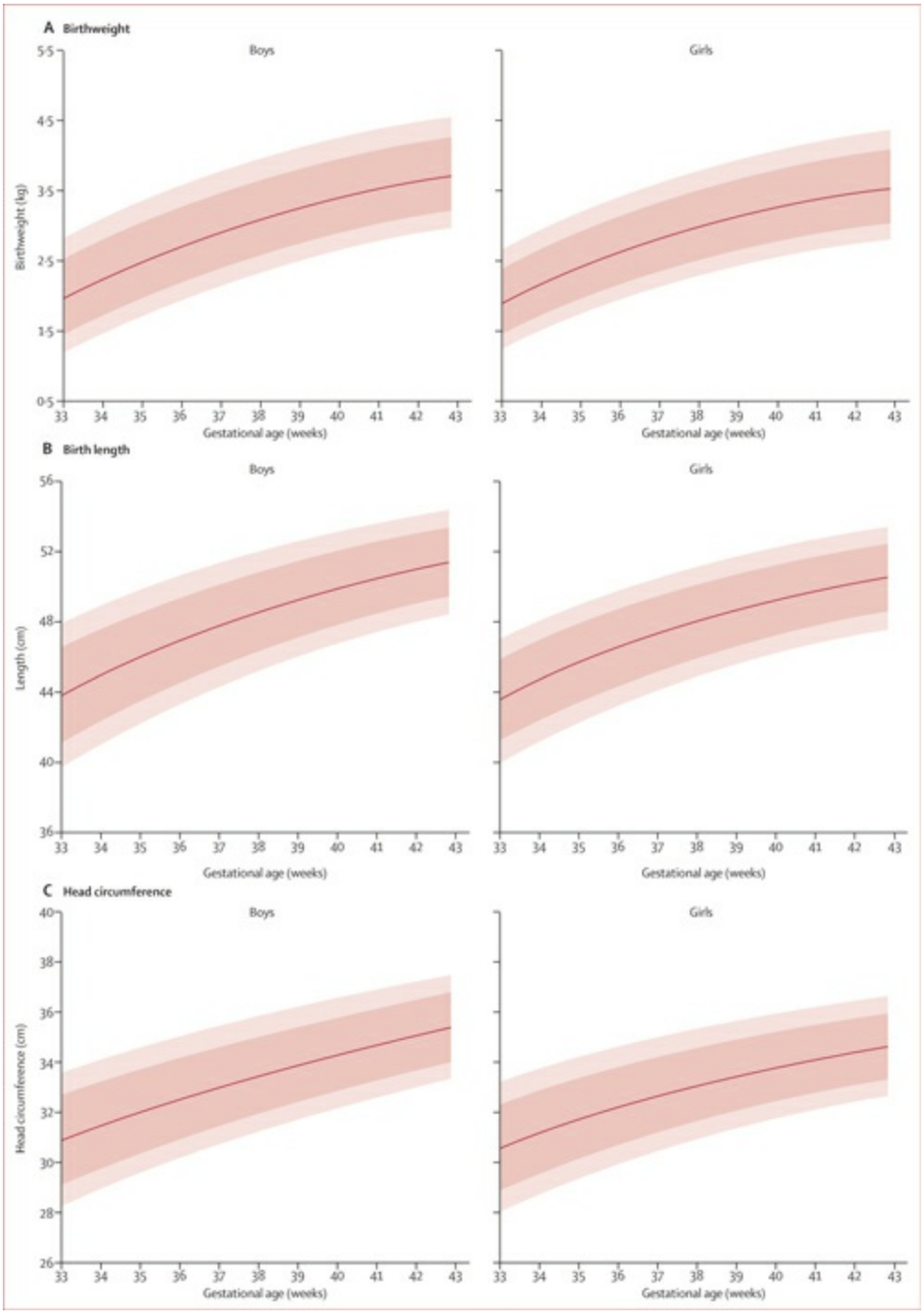


Figure I-25 Birthweight, birth length, and head circumference curves for 33–43 weeks gestational age. (From Villar J, Ismail LC, Victora CG, et al. International standards for newborn weight, length, and head circumference by gestational age and sex: the Newborn Cross-Sectional Study of the INTERGROWTH-21st Project. *Lancet*. 2014;384:857–68.)

TABLE I-1 Smoothed Centiles for Birthweight of Boys and Girls According to Gestational Age, 33–42 Weeks

	Boys						Girls					
	Number of observations	Centiles for birthweight (kg)					Number of observations	Centiles for birthweight (kg)				
		3rd	10th	50th	90th	97th		3rd	10th	50th	90th	97th
33 weeks	34	1·18	1·43	1·95	2·52	2·82	17	1·20	1·41	1·86	2·35	2·61
34 weeks	48	1·45	1·71	2·22	2·79	3·08	65	1·47	1·68	2·13	2·64	2·90
35 weeks	128	1·70	1·95	2·47	3·03	3·32	114	1·71	1·92	2·38	2·89	3·16
36 weeks	323	1·93	2·18	2·69	3·25	3·54	293	1·92	2·14	2·60	3·12	3·39
37 weeks	857	2·13	2·38	2·89	3·45	3·74	803	2·11	2·33	2·80	3·32	3·60
38 weeks	2045	2·32	2·57	3·07	3·63	3·92	1802	2·28	2·50	2·97	3·51	3·78
39 weeks	3009	2·49	2·73	3·24	3·79	4·08	2869	2·42	2·65	3·13	3·66	3·94
40 weeks	2568	2·63	2·88	3·38	3·94	4·22	2523	2·55	2·78	3·26	3·80	4·08
41 weeks	1179	2·76	3·01	3·51	4·06	4·35	1195	2·65	2·89	3·37	3·92	4·20
42 weeks	206	2·88	3·12	3·62	4·17	4·46	224	2·74	2·98	3·46	4·01	4·30
Total	10 397	..	..	..	..	..	9905	..	..	..	..	..

Source: Reprinted from Villar J, Ismail LC, Victora CG, et al. International standards for newborn weight, length, and head circumference by gestational age and sex: the Newborn Cross-Sectional Study of the INTERGROWTH-21st Project. *Lancet*. 2014;384:857–68, with permission from Elsevier.

TABLE I-2 Smooth Centiles for Birth Length of Boys and Girls According to Gestational Age, 33–42 Weeks

	Boys						Girls					
	Number of observations	Centiles for length (cm)					Number of observations	Centiles for length (cm)				
		3rd	10th	50th	90th	97th		3rd	10th	50th	90th	97th
33 weeks	33	39·69	41·09	43·81	46·55	47·97	17	39·79	41·01	43·39	45·70	46·85
34 weeks	48	41·05	42·38	44·98	47·59	48·94	65	41·04	42·22	44·55	46·79	47·92
35 weeks	128	42·26	43·54	46·03	48·53	49·82	111	42·14	43·30	45·57	47·76	48·86
36 weeks	320	43·36	44·58	46·97	49·38	50·62	292	43·13	44·26	46·48	48·62	49·69
37 weeks	849	44·34	45·52	47·82	50·14	51·34	799	44·01	45·11	47·29	49·39	50·44
38 weeks	2031	45·22	46·37	48·59	50·83	51·99	1786	44·79	45·88	48·01	50·07	51·10
39 weeks	2983	46·02	47·13	49·29	51·46	52·59	2846	45·49	46·56	48·65	50·68	51·69
40 weeks	2531	46·75	47·83	49·92	52·03	53·13	2486	46·12	47·17	49·23	51·23	52·22
41 weeks	1146	47·41	48·46	50·50	52·56	53·62	1180	46·68	47·72	49·75	51·72	52·70
42 weeks	202	48·01	49·04	51·03	53·03	54·07	218	47·19	48·21	50·22	52·15	53·12
Total	10 271	..	..	..	..	..	9800	..	..	..	..	..

Source: Reprinted from Villar J, Ismail LC, Victora CG, et al. International standards for newborn weight, length, and head circumference by gestational age and sex: the Newborn Cross-Sectional Study of the INTERGROWTH-21st Project. *Lancet*. 2014;384:857–68, with permission from Elsevier.

TABLE I-3 Smoothed Centiles for Head

Circumference of Boys and Girls According to Gestational Age, 33–42 Weeks

	Boys						Girls					
	Number of observations	Centiles for head circumference (cm)					Number of observations	Centiles for head circumference (cm)				
		3rd	10th	50th	90th	97th		3rd	10th	50th	90th	97th
33 weeks	33	28·25	29·11	30·88	32·71	33·62	17	27·92	28·76	30·46	32·24	33·14
34 weeks	48	28·93	29·76	31·47	33·23	34·11	65	28·64	29·44	31·08	32·78	33·65
35 weeks	127	29·56	30·37	32·02	33·73	34·58	111	29·28	30·06	31·64	33·28	34·12
36 weeks	322	30·15	30·93	32·53	34·19	35·02	293	29·87	30·62	32·14	33·74	34·55
37 weeks	848	30·69	31·46	33·02	34·63	35·43	798	30·40	31·13	32·61	34·15	34·94
38 weeks	2032	31·21	31·95	33·47	35·04	35·83	1783	30·88	31·59	33·03	34·53	35·30
39 weeks	2985	31·69	32·42	33·90	35·44	36·20	2849	31·32	32·01	33·41	34·88	35·62
40 weeks	2532	32·15	32·86	34·31	35·81	36·56	2486	31·72	32·39	33·76	35·19	35·92
41 weeks	1147	32·58	33·28	34·70	36·17	36·91	1180	32·08	32·74	34·08	35·48	36·19
42 weeks	204	32·99	33·68	35·07	36·52	37·24	218	32·41	33·06	34·37	35·74	36·44
Total	10 278	..	..	..	..	..	9800	..	..	..	..	..

Source: Reprinted from Villar J, Ismail LC, Victora CG, et al. International standards for newborn weight, length, and head circumference by gestational age and sex: the Newborn Cross-Sectional Study of the INTERGROWTH-21st Project. *Lancet*. 2014;384:857–68, with permission from Elsevier.

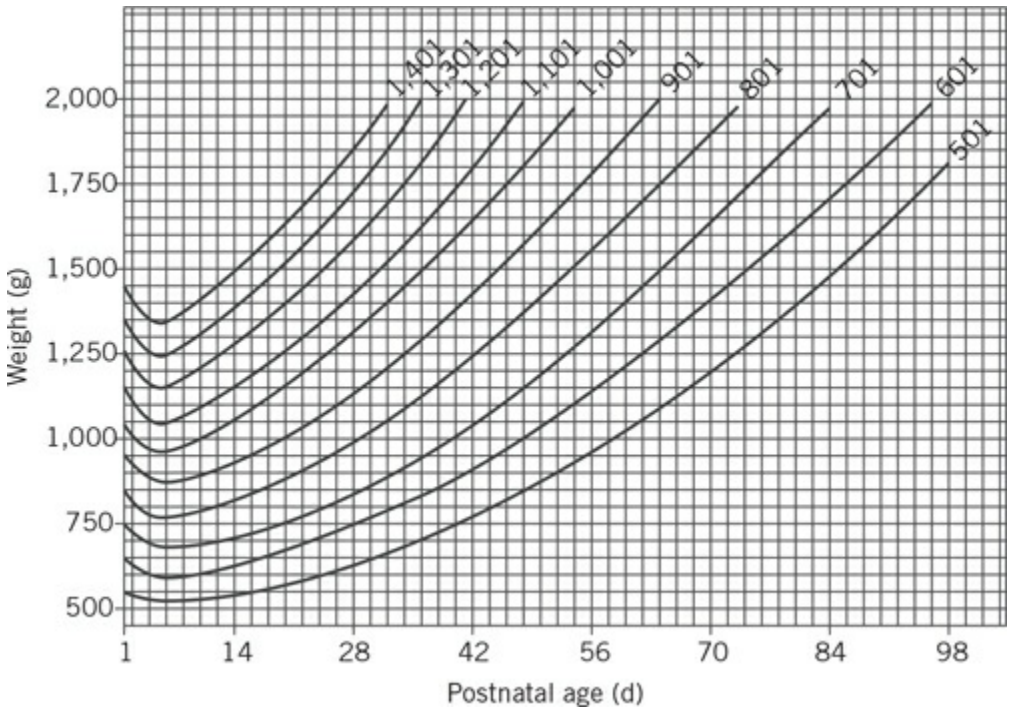


Figure I-26 Average daily body weight versus postnatal age in days for infants stratified by 100-g birth weight intervals. (Reproduced from

Ehrenkranz R, Younes N, Lemons JA, et al. Longitudinal growth of hospitalized very low birth weight infants. *Pediatrics*. 1999;104(2):280–289. With permission from the American Academy of Pediatrics.)

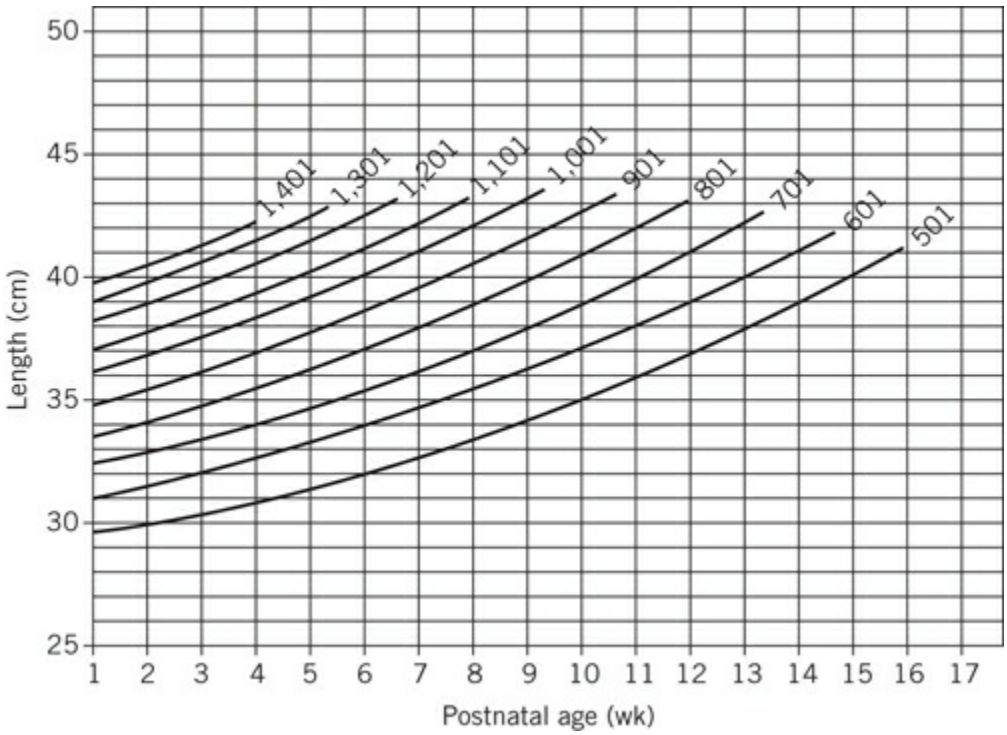


Figure I-27 Average weekly length versus postnatal age in weeks for infants stratified by 100-g birth weight intervals. (Reproduced from Ehrenkranz R, Younes N, Lemons JA, et al. Longitudinal growth of hospitalized very low birth weight infants. *Pediatrics*. 1999;104(2):280–289. With permission from the American Academy of Pediatrics.)

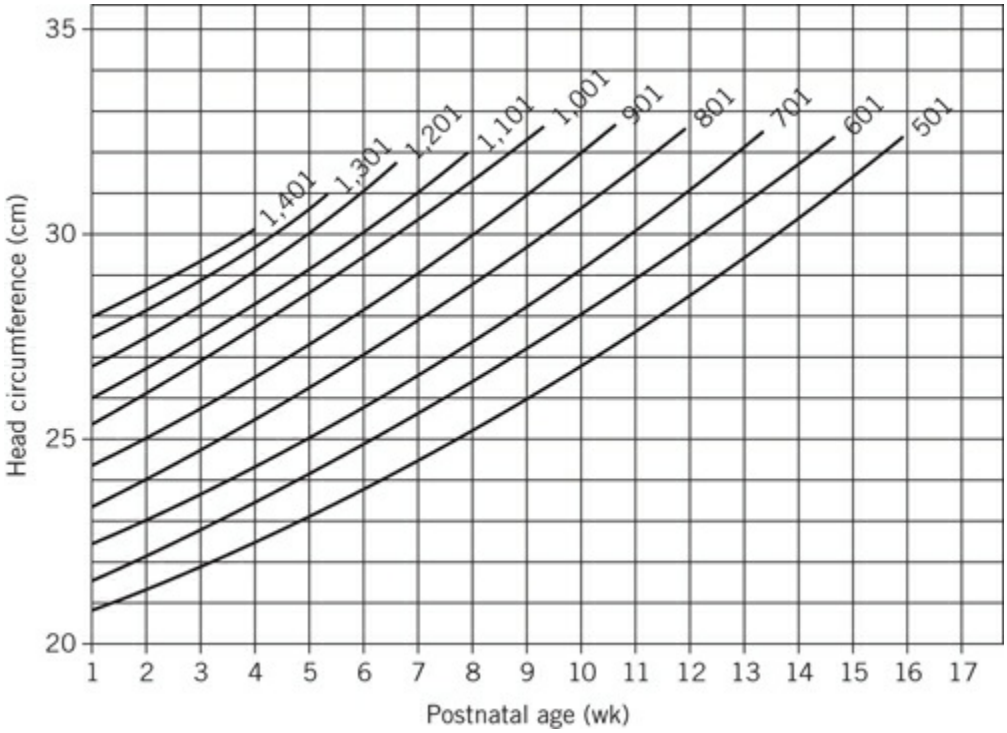


Figure I-28 Average weekly head circumference versus postnatal age in weeks for infants stratified by 100-g birth weight intervals. (Reproduced from Ehrenkranz R, Younes N, Lemons JA, et al. Longitudinal growth of hospitalized very low birth weight infants. *Pediatrics*. 1999;104(2):280–289. With permission from the American Academy of Pediatrics.)

Turner girls: 2–19 yr
Physical growth

Name _____ Record # _____

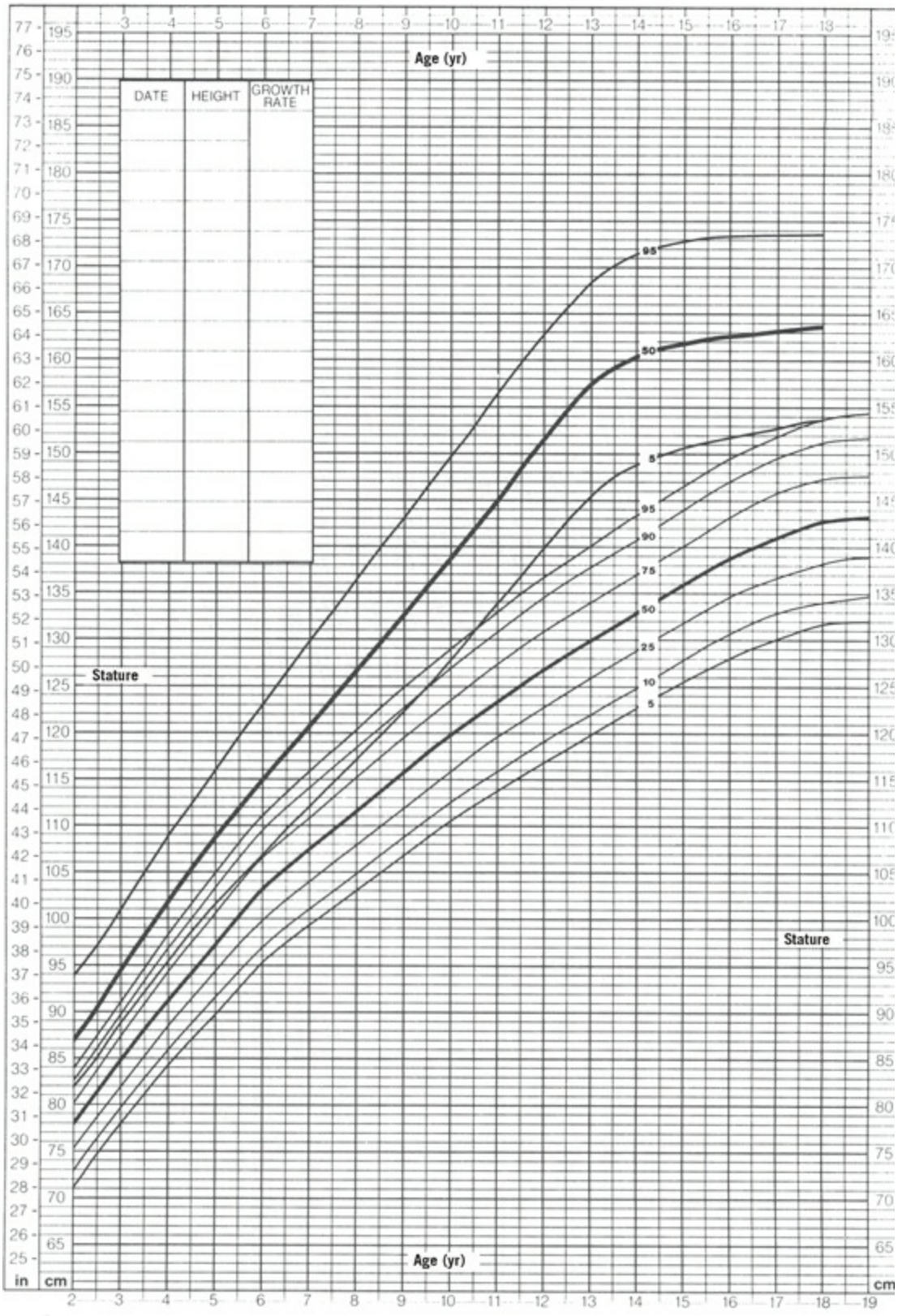


Figure I-29 Physical growth of girls with Turner syndrome (2–19 years). (Normal girls' (top three lines) percentiles derived from National Center for Health Statistics. Untreated Turner syndrome growth rates derived from Lyon AJ, Preece MA, Grant DB. Growth curve for girls with Turner syndrome. *Arch Dis Child*. 1985;60:932–935. With permission from the BMJ Publishing Group.)

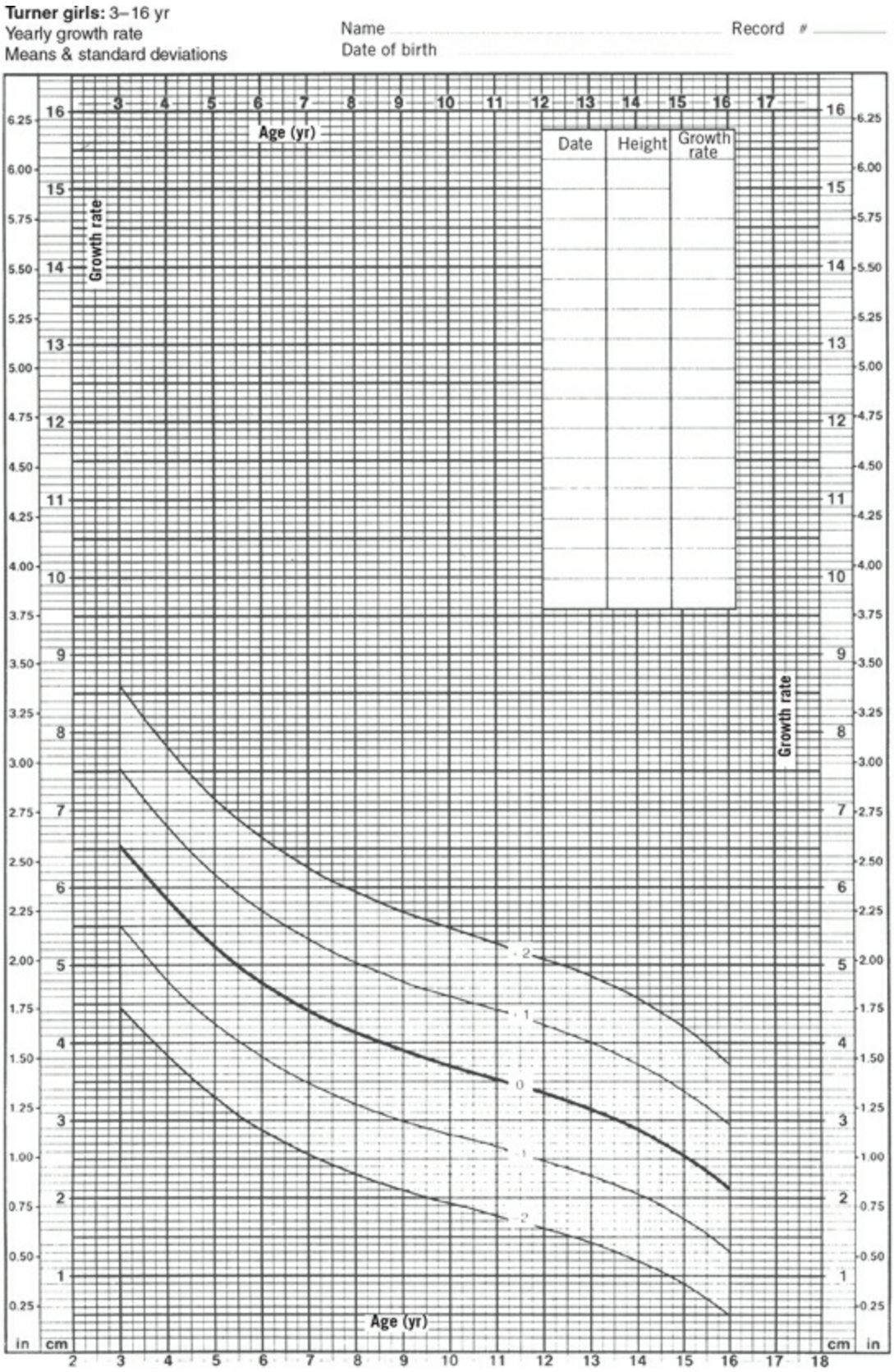


Figure I-30 Yearly growth rate means and standard deviations for girls

with Turner syndrome (3–16 years). (Turner syndrome growth rates derived from Ranke MB, Pfluger H, Rosendahl W, et al. Turner syndrome: spontaneous growth in 150 cases and review of the literature. *Eur J Pediatr*. 1983;141:81–88. With permission from Springer Science and Business Media.)

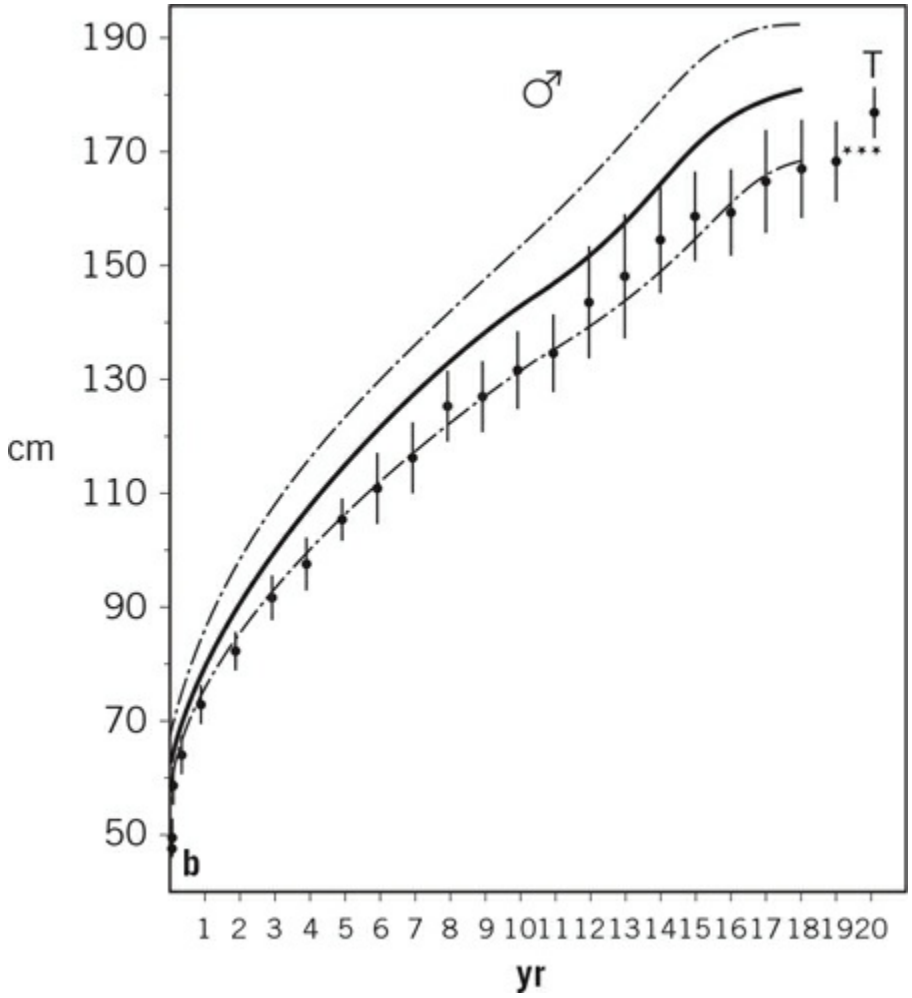


Figure I-31 Mean (+1 SD) height-for-age percentile curves for boys with Williams–Beuren syndrome (6 months to 20 years). T, target height. (Reproduced from Pankau R, Partsch C-J, Gosch A, et al. Statural growth in Williams-Beuren syndrome. *Eur J Pediatr.* 1992;151(10):751755. With permission from Springer Science and Business Media.)

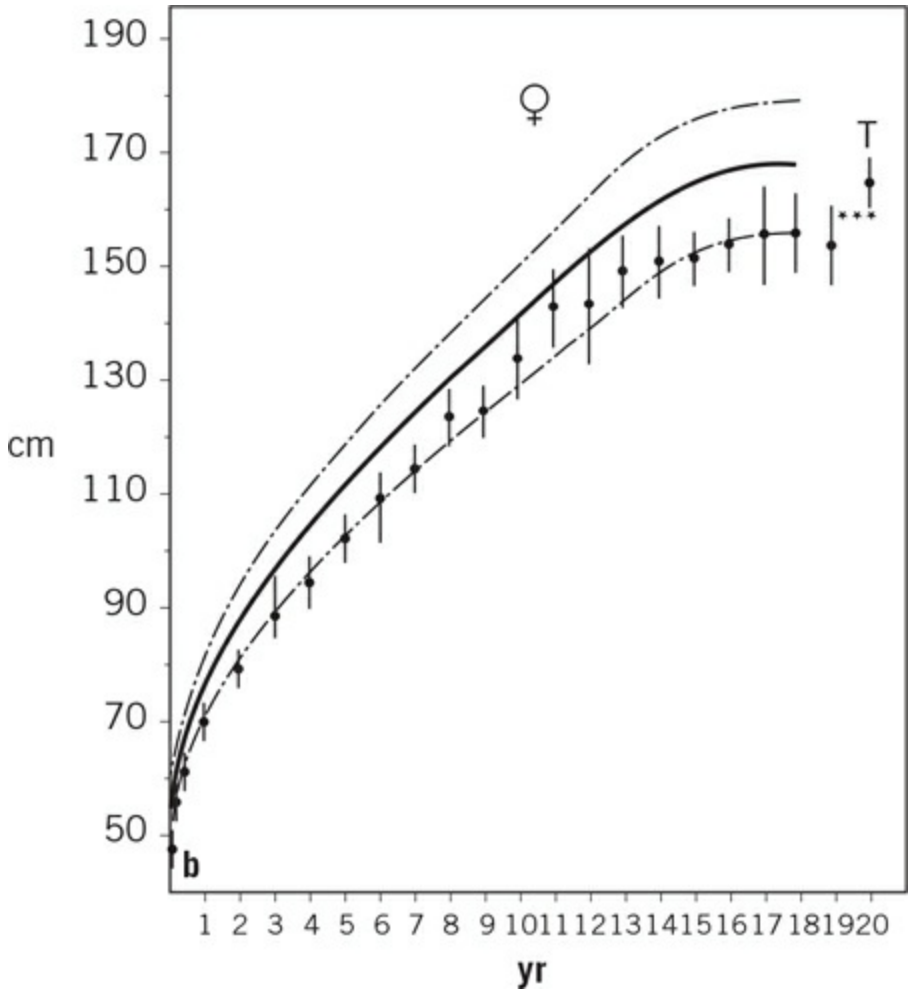


Figure I-32 Mean (+1 SD) height-for-age percentile curves for girls with Williams–Beuren syndrome (6 months to 20 years). T, target height. (Reproduced from Pankau R, Partsch C-J, Gosch A, et al. Statural growth in Williams-Beuren syndrome. *Eur J Pediatr*. 1992;151(10):751755. With permission from Springer Science and Business Media.)

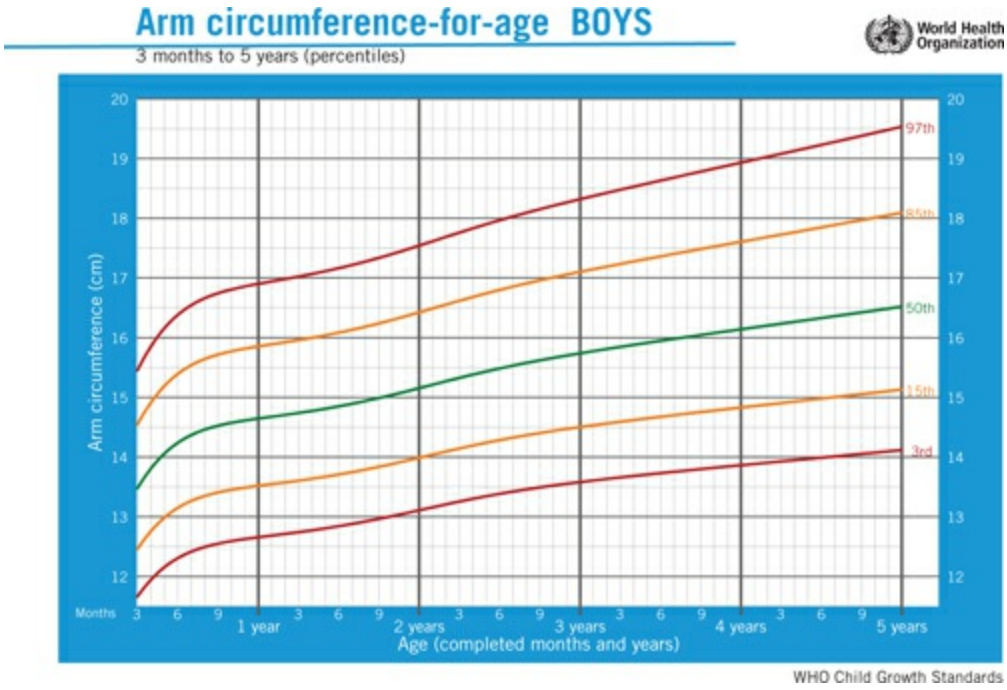


Figure I-33 Boys: arm circumference-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

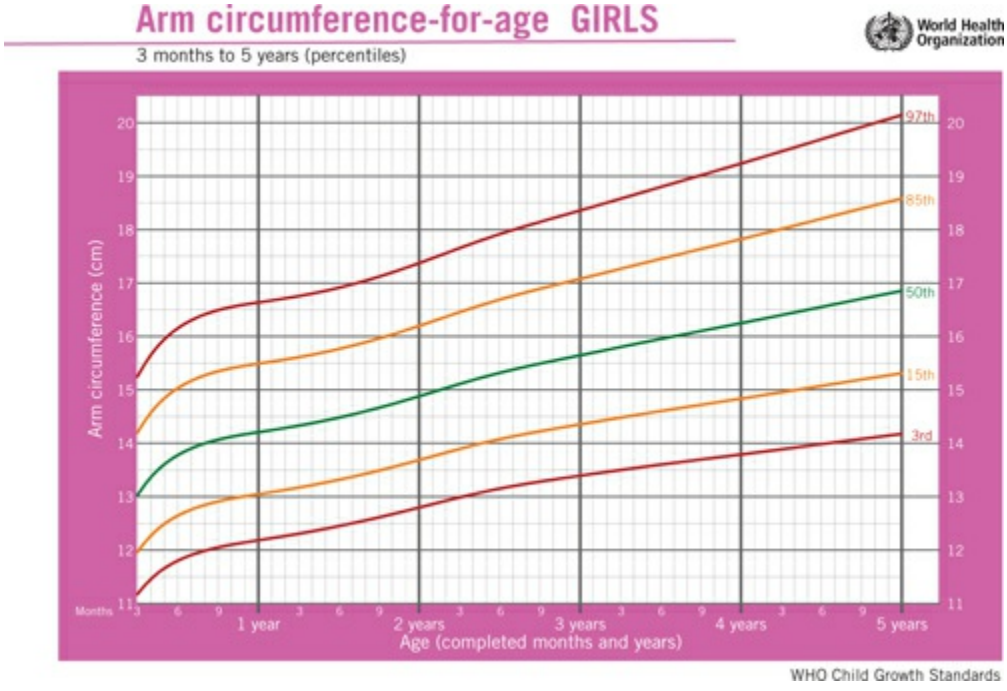


Figure I-34 Girls: arm circumference-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

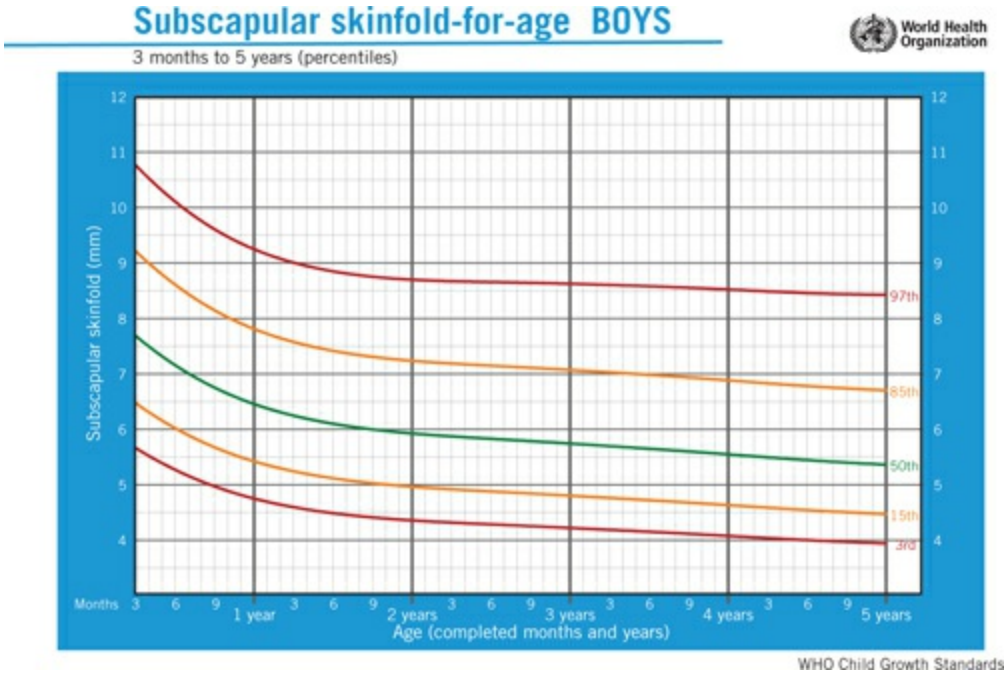


Figure I-35 Boys: subscapular skinfold-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

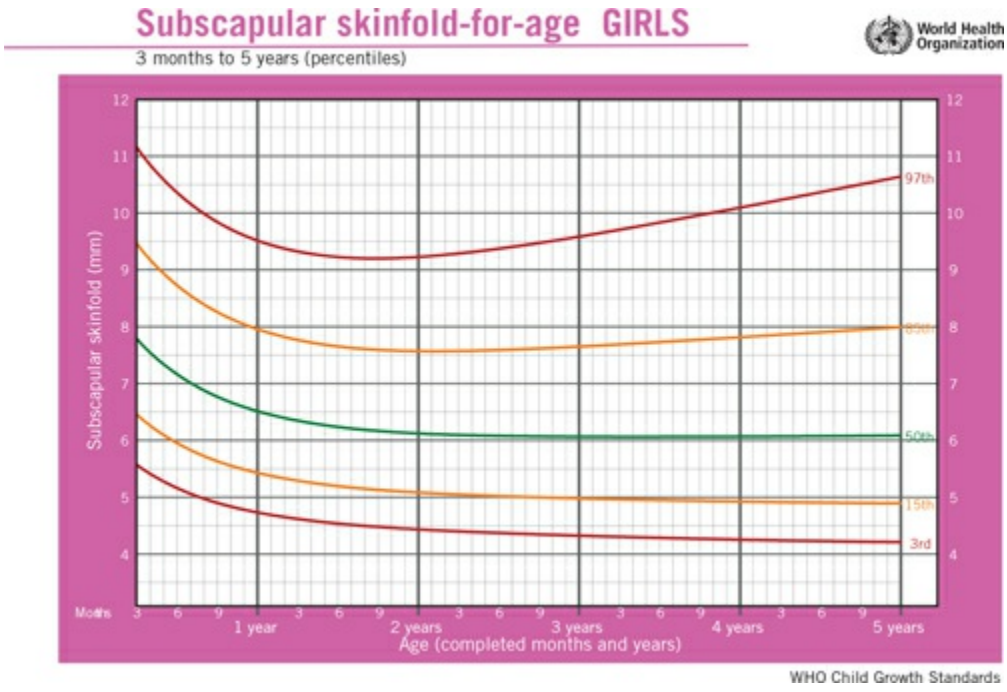


Figure I-36 Girls: subscapular skinfold-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

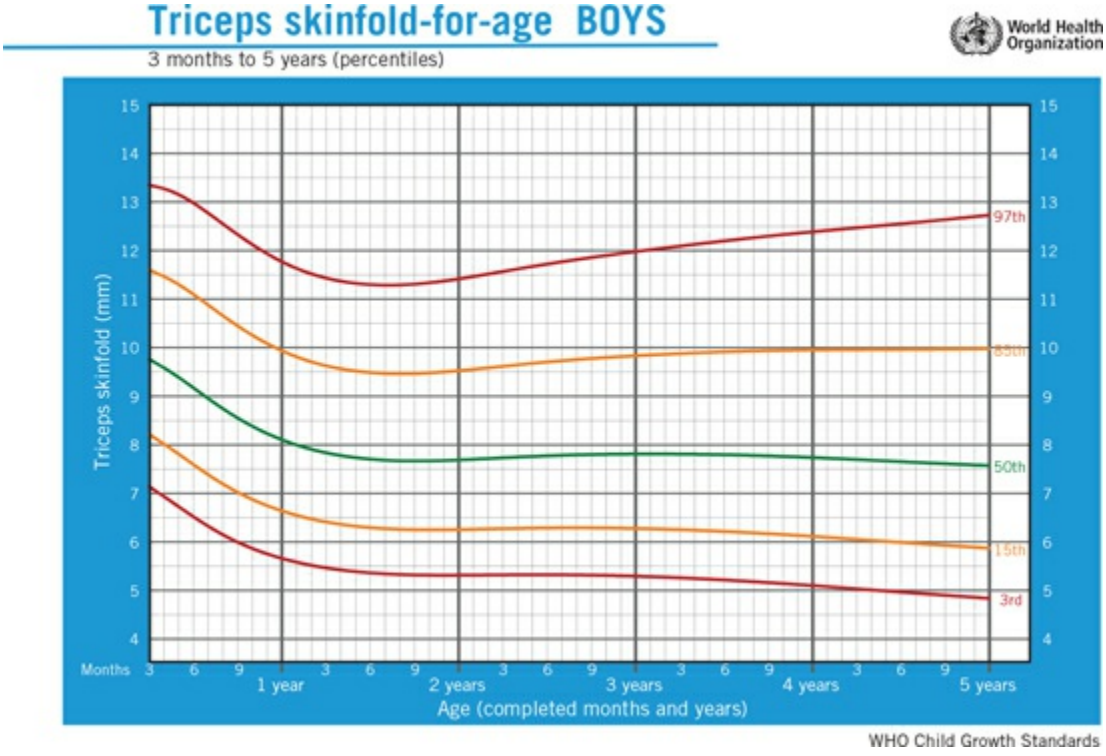


Figure I-37 Boys: triceps skinfold-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

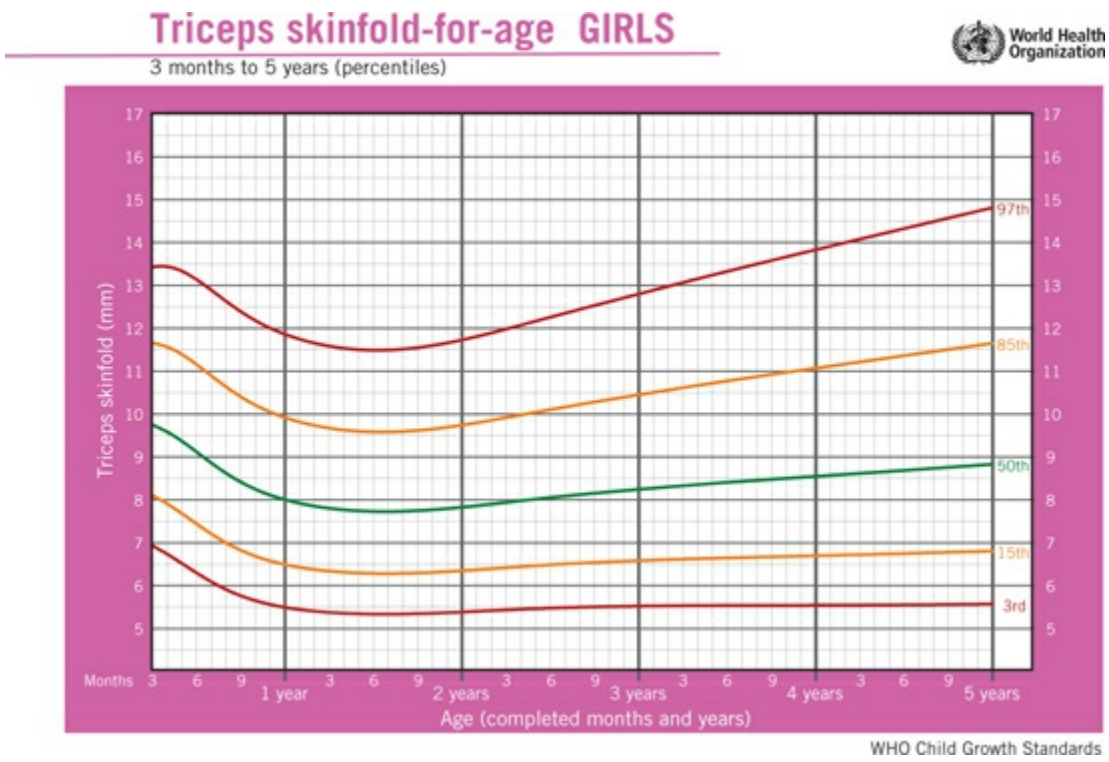


Figure I-38 Girls: triceps skinfold-for-age (3 months to 5 years). (Developed by the World Health Organization [2006], http://www.who.int/childgrowth/standards/en/.)

TABLE I-4 Percentiles of Upper Arm Circumference and Estimated Upper Arm Muscle Circumference

Age Group (years)	Arm Circumference (mm)							Arm Muscle Circumference (mm)						
	5th	10th	25th	50th	75th	90th	95th	5th	10th	25th	50th	75th	90th	95th
Males														
0.0–0.4	113			134			153							
0.5–1.4	128			152			175							
1–1.9	142	146	150	159	170	176	183	110	113	119	127	135	144	147
2–2.9	141	145	153	162	170	178	185	111	114	122	130	140	146	150
3–3.9	150	153	160	167	175	184	190	117	123	131	137	143	148	153
4–4.9	149	154	162	171	180	186	192	123	126	133	141	148	156	159
5–5.9	153	160	167	175	185	195	204	128	133	140	147	154	162	169
6–6.9	155	159	167	179	188	209	228	131	135	142	151	161	170	177
7–7.9	162	167	177	187	201	223	230	137	139	151	160	168	177	190
8–8.9	162	170	177	190	202	220	245	140	145	154	162	170	182	187
9–9.9	175	178	187	200	217	249	257	151	154	161	170	183	196	202
10–10.9	181	184	196	210	231	262	274	156	160	166	180	191	209	221
11–11.9	186	190	202	223	244	261	280	159	165	173	183	195	205	230
12–12.9	193	200	214	232	254	282	303	167	171	182	195	210	223	241
13–13.9	194	211	228	247	263	286	301	172	179	196	211	226	239	245
14–14.9	220	226	237	253	283	303	322	189	199	212	223	240	260	264
15–15.9	222	229	244	264	284	311	320	199	204	218	237	254	266	272
16–16.9	244	248	262	278	303	324	343	213	225	234	249	269	287	296
17–17.9	246	253	267	285	308	336	347	224	231	245	258	273	294	312
18–18.9	245	260	276	297	321	353	379	226	237	252	264	283	298	324
19–24.9	262	272	288	308	331	355	372	238	245	257	273	289	309	321
25–34.9	271	282	300	319	342	362	375	243	250	264	279	298	314	326
35–44.9	278	287	305	326	345	363	374	247	255	269	286	302	318	327
45–54.9	267	281	301	322	342	362	376	239	249	265	281	300	315	326
55–64.9	258	273	296	317	336	355	369	236	245	260	278	295	310	320
65–74.9	248	263	285	307	325	344	355	223	235	251	268	284	298	306
Females														
0.0–0.4	107			127			150							
0.5–1.4	125			146			170							
1–1.9	138	142	148	156	164	172	177	105	111	117	124	132	139	143
2–2.9	142	145	152	160	167	176	184	111	114	119	126	133	142	147
3–3.9	143	150	158	167	175	183	189	113	119	124	132	140	146	152
4–4.9	149	154	160	169	177	184	191	115	121	128	136	144	152	157
5–5.9	153	157	165	175	185	203	211	125	128	134	142	151	159	165
6–6.9	156	162	170	176	187	204	211	130	133	138	145	154	166	171
7–7.9	164	167	174	183	199	216	231	129	135	142	151	160	171	176
8–8.9	168	172	183	195	214	247	261	138	140	151	160	171	183	194
9–9.9	178	182	194	211	224	251	260	147	150	158	167	180	194	198
10–10.9	174	182	193	210	228	251	265	148	150	159	170	180	190	197
11–11.9	185	194	208	224	248	276	303	150	158	171	181	196	217	223
12–12.9	194	203	216	237	256	282	294	162	166	180	191	201	214	220
13–13.9	202	211	223	243	271	301	338	169	175	183	198	211	226	240
14–14.9	214	223	237	252	272	304	322	174	179	190	201	216	232	247
15–15.9	208	221	239	254	279	300	322	175	178	189	202	215	228	244
16–16.9	218	224	241	258	283	318	334	170	180	190	202	216	234	249
17–17.9	220	227	241	264	295	324	350	175	183	194	205	221	239	257
18–18.9	222	227	241	258	281	312	325	174	179	191	202	215	237	245
19–24.9	221	230	247	265	290	319	345	179	185	195	207	221	236	249
25–34.9	233	240	256	277	304	342	368	183	188	199	212	228	246	264
35–44.9	241	251	267	290	317	356	378	186	192	205	218	236	257	272
45–54.9	242	256	274	299	328	362	384	187	193	206	220	238	260	274
55–64.9	243	257	280	303	335	367	385	187	196	209	225	244	266	280
65–74.9	240	252	274	299	326	356	373	185	195	208	225	244	264	279

Data collected from Whites in the United States Health and Nutrition Examination Survey 1 (1971–

1974).

Source: Adapted from Frisancho AR. Triceps skin fold and upper arm muscle size norms for assessment of nutritional status. *Am J Clin Nutr*. 1974;27(10):1052–1058; Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr.* 1981(11);34:2540–2545. With permission from the American Society for Nutrition.

TABLE I-5 Percentiles of Triceps Skinfolds

Age Group (years)	5th	10th	25th	50th	75th	90th	95th
Males							
1–1.9	6	7	8	10	12	14	16
2–2.9	6	7	8	10	12	14	15
3–3.9	6	7	8	10	11	14	15
4–4.9	6	6	8	9	11	12	14
5–5.9	6	6	8	9	11	14	15
6–6.9	5	6	7	8	10	13	16
7–7.9	5	6	7	9	12	15	17
8–8.9	5	6	7	8	10	13	16
9–9.9	6	6	7	10	13	17	18
10–10.9	6	6	8	10	14	18	21
11–11.9	6	6	8	11	16	20	24
12–12.9	6	6	8	11	14	22	28
13–13.9	5	5	7	10	14	22	26
14–14.9	4	5	7	9	14	21	24
15–15.9	4	5	6	8	11	18	24
16–16.9	4	5	6	8	12	16	22
17–17.9	5	5	6	8	12	16	19
18–18.9	4	5	6	9	13	20	24
19–24.9	4	5	7	10	15	20	22
25–34.9	5	6	8	12	16	20	24
35–44.9	5	6	8	12	16	20	23
45–54.9	6	6	8	12	15	20	25
55–64.9	5	6	8	11	14	19	22
65–74.9	4	6	8	11	15	19	22
Females							
1–1.9	6	7	8	10	12	14	16
2–2.9	6	8	9	10	12	15	16
3–3.9	7	8	9	11	12	14	15
4–4.9	7	8	8	10	12	14	16
5–5.9	6	7	8	10	12	15	18
6–6.9	6	6	8	10	12	14	16
7–7.9	6	7	9	11	13	16	18
8–8.9	6	8	9	12	15	18	24
9–9.9	8	8	10	13	16	20	22
10–10.9	7	8	10	12	17	23	27
11–11.9	7	8	10	13	18	24	28
12–12.9	8	9	11	14	18	23	27
13–13.9	8	8	12	15	21	26	30
14–14.9	9	10	13	16	21	26	28
15–15.9	8	10	12	17	21	25	32

16–16.9	10	12	15	18	22	26	31
17–17.9	10	12	13	19	24	30	37
18–18.9	10	12	15	18	22	26	30
19–24.9	10	11	14	18	24	30	34
25–34.9	10	12	16	21	27	34	37
35–44.9	12	14	18	23	29	35	38
45–54.9	12	16	20	25	30	36	40
55–64.9	12	16	20	25	31	36	38
65–74.9	12	14	18	24	29	34	36

Source: Adapted from Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr.* 1981;34:2540–2545. With permission from the American Society for Nutrition.

TABLE I-6 Percentiles for Estimates of Upper Arm Muscle Area and Upper Arm Fat Area

Age group (years)	Arm Muscle Area (mm)							Arm Fat Area (mm)						
	5	10	25	50	75	90	95	5	10	25	50	75	90	95
Males														
1–1.9	956	1014	1133	1278	1447	1644	1720	452	486	590	741	895	1036	1176
2–2.9	973	1040	1190	1345	1557	1690	1787	434	504	578	737	871	1044	1148
3–3.9	1095	1201	1357	1484	1618	1750	1853	464	519	590	736	868	1071	1151
4–4.9	1207	1264	1408	1579	1747	1926	2008	428	494	598	722	859	989	1085
5–5.9	1298	1411	1550	1720	1884	2089	2285	446	488	582	713	914	1176	1299
6–6.9	1360	1447	1605	1815	2056	2297	2493	371	446	539	678	896	1115	1519
7–7.9	1497	1548	1808	2027	2246	2494	2886	423	473	574	758	1011	1393	1511
8–8.9	1550	1664	1895	2089	2296	2628	2788	410	460	588	725	1003	1248	1558
9–9.9	1811	1884	2067	2288	2657	3053	3257	485	527	635	859	1252	1864	2081
10–10.9	1930	2027	2182	2575	2903	3486	3882	523	543	738	982	1376	1906	2609
11–11.9	2016	2156	2382	2670	3022	3359	4226	536	595	754	1148	1710	2348	2574
12–12.9	2216	2339	2649	3022	3496	3968	4640	554	650	874	1172	1558	2536	3580
13–13.9	2363	2546	3044	3553	4081	4502	4794	475	570	812	1096	1702	2744	3322
14–14.9	2830	3147	3586	3963	4575	5368	5530	453	563	786	1082	1608	2746	3508
15–15.9	3138	3317	3788	4481	5134	5631	5900	521	595	690	931	1423	2434	3100
16–16.9	3625	4044	4352	4951	5753	6576	6980	542	593	844	1078	1746	2280	3041
17–17.9	3998	4252	4777	5286	5950	6886	7726	598	698	827	1096	1636	2407	2888
18–18.9	4070	4481	5066	5552	6374	7067	8355	560	665	860	1264	1947	3302	3928

19–24.9	4508	4777	5274	5913	6660	7606	8200	594	743	963	1406	2231	3098	3652
25–34.9	4694	4963	5541	6214	7067	7847	8436	675	831	1174	1752	2459	3246	3786
35–44.9	4844	5181	5740	6490	7265	8034	8488	703	851	1310	1792	2463	3098	3624
45–54.9	4546	4946	5589	6297	7142	7918	8458	749	922	1254	1741	2359	3245	3928
55–64.9	4422	4783	5381	6144	6919	7670	8149	658	839	1166	1645	2236	2976	3466
65–74.9	3973	4411	5031	5716	6432	7074	7453	573	753	1122	1621	2199	2876	3327
Females														
1–1.9	885	973	1084	1221	1378	1535	1621	401	466	578	706	847	1022	1140
2–2.9	973	1029	1119	1269	1405	1595	1727	469	526	642	747	894	1061	1173
3–3.9	1014	1133	1227	1396	1563	1690	1846	473	529	656	822	967	1106	1158
4–4.9	1058	1171	1313	1475	1644	1832	1958	490	541	654	766	907	1109	1236
5–5.9	1238	1301	1423	1598	1825	2012	2159	470	529	647	812	991	1330	1536
6–6.9	1354	1414	1513	1683	1877	2182	2323	464	508	638	827	1009	1263	1436
7–7.9	1330	1441	1602	1815	2045	2332	2469	491	560	706	920	1135	1407	1644
8–8.9	1513	1566	1808	2034	2327	2657	2996	527	634	769	1042	1383	1872	2482
9–9.9	1723	1788	1976	2227	2571	2987	3112	642	690	933	1219	1584	2171	2524
10–10.9	1740	1784	2019	2296	2583	2873	3093	616	702	842	1141	1608	2500	3005
11–11.9	1784	1987	2316	2612	3071	3739	3953	707	802	1015	1301	1942	2730	3690
12–12.9	2092	2182	2579	2904	3225	3655	3847	782	854	1090	1511	2056	2666	3369
13–13.9	2269	2426	2657	3130	3529	4081	4568	726	838	1219	1625	2374	3272	4150
14–14.9	2418	2562	2874	3220	3704	4294	4850	981	1043	1423	1818	2403	3250	3765
15–15.9	2426	2518	2847	3248	3689	4123	4756	839	1126	1396	1886	2544	3093	4195
16–16.9	2308	2567	2865	3248	3718	4353	4946	1126	1351	1663	2006	2598	3374	4236
17–17.9	2442	2674	2996	3336	3883	4552	5251	1042	1267	1463	2104	2977	3864	5159
18–18.9	2398	2538	2917	3243	3694	4461	4767	1003	1230	1616	2104	2617	3508	3733
19–24.9	2538	2728	3026	3406	3877	4439	4940	1046	1198	1596	2166	2959	4050	4896
25–34.9	2661	2826	3148	3573	4138	4806	5541	1173	1399	1841	2548	3512	4690	5560
35–44.9	2750	2948	3359	3783	4428	5240	5877	1336	1619	2158	2898	3932	5093	5847
45–54.9	2784	2956	3378	3858	4520	5375	5964	1459	1803	2447	3244	4229	5416	6140
55–64.9	2784	3063	3477	4045	4750	5632	6247	1345	1879	2520	3369	4360	5276	6152
65–74.9	2737	3018	3444	4019	4739	5566	6214	1363	1681	2266	3063	3943	4914	5530

Source: Adapted from Frisancho AR. New norms of upper limb fat and muscle areas for assessment of nutritional status. *Am J Clin Nutr.* 1981;34:2540–2545. With permission from the American Society for Nutrition.

TABLE I-7 Percentiles for Estimates of Upper Arm Muscle Area and Upper Arm Fat Area

Age (mo)	Infants (n)	Mean (g/d)	SD (g/d)	Percentiles (g/d)						
				5th	10th	25th	50th	75th	90th	95th
Males										
Birth to 2	580	33	7.0	21	24	28	32	38	42	44
1–3	580	31	6.9	20	22	27	31	35	39	43
2–4	65	23	4.7	–	17	19	23	26	29	–
3–5	298	19	3.2	14	15	17	18	20	22	24
4–6	298	16	2.9	12	13	14	16	18	20	21
5–7	233	15	2.4	11	12	13	15	16	18	18
6–8	233	13	2.4	10	11	12	13	15	16	17
7–9	233	12	2.4	9	10	11	12	14	15	16
8–10	233	12	2.4	9	9	10	11	13	15	15
9–11	233	11	2.3	8	8	9	11	12	14	14
10–12	233	10	2.3	7	8	9	10	12	13	14
Females										
Birth to 2	562	28	6.5	17	20	23	28	32	36	38
1–3	562	26	6.3	16	19	22	26	30	34	37
2–4	72	22	5.4	–	16	19	21	24	27	–
3–5	298	18	4.7	13	14	16	17	19	21	22
4–6	298	15	4.6	11	12	14	15	17	18	19
5–7	224	14	4.7	11	11	13	14	15	17	17
6–8	224	13	4.6	10	10	12	13	14	16	16
7–9	224	12	4.5	9	10	11	12	13	15	15
8–10	224	12	4.5	8	9	10	11	13	14	14
9–11	224	11	4.4	8	8	9	10	12	13	14
10–12	224	10	4.3	7	8	9	10	11	13	13

Source: A dapted from Guo S, Roche AF, Fomon SJ, et al. Reference data on gains in weight and length during the first two years of life. *J Pediatr*. 1991;119:355–362. With permission from Elsevier.

TABLE I-8 Selected Percentiles for three-Month Increments in Length (From Birth to 14 Months)

Age (mo)	Infants (n)	Mean (mm/d)	SD (mm/d)	Percentile						
				5th	10th	25th	50th	75th	90th	95th
Males										
Birth to 3	580	1.07	0.11	0.89	0.92	0.99	1.06	1.14	1.21	1.26
1–4	65	1.00	0.08	–	0.94	1.01	1.06	1.09	–	
2–5	65	0.84	0.09	–	0.74	0.79	0.84	0.91	0.95	–
3–6	255	0.69	0.08	0.56	0.60	0.64	0.68	0.73	0.79	0.82
4–7	190	0.62	0.06	0.54	0.55	0.58	0.61	0.65	0.73	0.72

5–8	190	0.56	0.05	0.49	0.50	0.53	0.56	0.59	0.69	0.65
6–9	190	0.52	0.05	0.46	0.46	0.49	0.52	0.54	0.58	0.60
7–10	190	0.48	0.05	0.42	0.43	0.45	0.48	0.51	0.54	0.57
8–11	190	0.45	0.04	0.39	0.40	0.43	0.45	0.48	0.51	0.53
9–12	190	0.43	0.04	0.36	0.38	0.40	0.43	0.45	0.48	0.51
10–13	190	0.41	0.04	0.34	0.36	0.38	0.41	0.43	0.46	0.49
11–14	190	0.39	0.04	0.33	0.34	0.36	0.39	0.41	0.44	0.47
Females										
Birth to 3	562	0.99	0.10	0.82	0.86	0.93	0.99	1.06	1.11	1.15
1–4	74	0.95	0.10	–	0.84	0.87	0.95	1.02	1.07	–
2–5	74	0.80	0.10	–	0.67	0.73	0.81	0.87	0.92	–
3–6	241	0.67	0.08	0.55	0.58	0.63	0.67	0.72	0.77	0.79
4–7	167	0.60	0.06	0.53	0.54	0.57	0.61	0.64	0.67	0.69
5–8	167	0.56	0.05	0.49	0.50	0.52	0.56	0.59	0.62	0.63
6–9	167	0.52	0.05	0.45	0.46	0.48	0.52	0.55	0.57	0.58
7–10	167	0.48	0.04	0.42	0.43	0.45	0.49	0.52	0.54	0.55
8–11	167	0.46	0.04	0.39	0.41	0.43	0.46	0.49	0.51	0.52
9–12	167	0.44	0.04	0.37	0.38	0.41	0.44	0.46	0.48	0.49
10–13	167	0.42	0.04	0.35	0.37	0.39	0.42	0.45	0.46	0.48
11–14	167	0.40	0.04	0.34	0.35	0.37	0.40	0.43	0.44	0.46

Adapted from Guo S, Roche AF, Fomon SJ, et al. Reference data on gains in weight and length during the first two years of life. *J Pediatr.* 1991;119:355–362. With permission from Elsevier.

TABLE I-9 Most Common Classifications of Protein–Energy Malnutrition

	Normal	Mild	Moderate	Severe
Weight for height[a]	>90	80–90	70–80	<70
Weight for height[b]			−3 < SD-score[c] < −2	SD-score[c] < −3
Height for age[a]	>95	87–98	80–87	<80
Height for age[b]			−3 < SD-score[c] < −2	SD-score[c] < −3

Sources: [a]Adapted with permission from the BMJ Publishing Group. Waterlow JC. Classification and definition of protein-calorie malnutrition. *BMJ*. 1972;3:566–569.

[b]Adapted from World Health Organization. Management of severe malnutrition: A manual for physicians and other senior health care workers. Geneva, Switzerland: WHO; 1999. p.4.

[c]SD-score = (observed value) – (median reference value from NCHS/WHO reference)/standard deviation of reference population.

TABLE I-10 Growth Charts for Specific Syndromes

Condition	References
Achondroplasia	Hagenäs L, Hertel T. Skeletal dysplasia, growth hormone treatment and body proportion: comparison with other syndromic and non-syndromic short children. *Horm Res*. 2003;60(suppl 3): 65–70. Height for boys and girls, 0–20 y; sitting height to total height

	ratio in girls, 0–20 y Horton WA, Rotter JI, Rimoin DL, et al. Standard growth curves for achondroplasia. *J Pediatr*. 1978;93(3):435–438. Stature, growth velocity, head circumference, upper and lower segments
Brachmann (Cornelia)-de Lange syndrome	Kline AD, Barr M, Jackson LG. Growth manifestations in the Brachmann-de Lange syndrome. *Am J Med Genet*. 1993;47:1042–1049. Length- and weight-for-age, from birth to 36 mo; height- and weight-for age, 2–18 y; head circumference-for-age, birth to18 y
Cerebral palsy (quadriplegia)	Krick J, Murphy-Miller P, Zeger S, Wright E. Pattern of growth in children with cerebral palsy. *J Am Diet Assoc*. 1996;96(7):680–685. Stature- and weight-for-age and weight-for-stature, birth to 10 y Stevenson RD, Conaway M, Chumlea WC, et al. Growth and health in children with moderate-to-severe cerebral palsy. *Pediatrics*. 2006;118(3):1010–1018. Knee height- and weight-for-age, 2–18 y Day SM, Strauss DJ, Vachon PJ, et al. Growth patterns in a population of children and adolescents with cerebral palsy. *Dev Med Child Neurol*. 2007;49;167–171. Available at www.lifeexpectancy.org/articles/growthcharts/all.pdf Weight-, height-, BMI-for-age, 2–20 y
Down syndrome	Cronk CE. Growth of children with Down's syndrome: birth to age 3 years. *Pediatrics*. 1978;61:564–568 and *Pediatrics*. 1988;81:102–110. Length- and weight-for-age, birth to 36 mo; stature- and weight-for-age, 2–18 y Palmer CGS, Cronk C, Pueschel SM, et al. Head circumference of children with Down syndrome (0–36 months). *Am J Med Genet* 1992;42:61–67. Head circumference-for-age, 0–36 mo Styles ME, Cole TJ, Dennis J, et al. New cross sectional stature, weight, and head circumference references for Down's syndrome in the UK and Republic of Ireland. *Arch Dis Child*. 2002;87:104–108. Weight and height, 0–18 y; head circumference, 0–5 y
Marfan's syndrome	Pyeritz RE. Marfan syndrome and other disorders of fibrillin. In: Rimoin DL, Connor JM, Pyeritz RE, eds. *Principles and Practice of Medical Genetics*. New York, NY: Churchill Livingstone;1997. Pyeritz RE, Murphy EA, Lin SJ, et al. Growth and anthropometrics in the Marfan syndrome. In: Papadatos CJ, Bartsocas CS, eds. *Endocrine Genetics and Genetics of Growth (Prog Clin Biol Res v200)*. New York, NY: Alan R. Liss, Inc.; 1985. Stature- and weight-for-age, 2–18 y, 20–24 y, and >24 y; upper and lower segment ratios, 2–20 y and adult
Myelomeningocele	Appendix 2, Growth grids for special conditions. In: Ekvall SW,

	Ekvall VK, eds. *Pediatric Nutrition in Chronic Diseases and Developmental Disorders: Prevention, Assessment, and Treatment*. New York, NY: Oxford University Press; 2005. Preliminary charts, height- and weight-for-age, 2–18 y Duval-Beaupere G, Kaci M, Lougovoy J, et al. Growth of trunk and legs of children with myelomeningocele. Dev *Med Child Neur.* 1987;29:225–231. Height, upper segment, lower segment measurements, 1–16 y Trollmann R, Dorr HG, Strehl E, et al. Growth and pubertal development in patients with meningomyelocele: a retrospective analysis. *Acta Paediatr*. 1996;85:76–80. Arm span Shurtleff DB, Walker WO, Duguay S, et al. Obesity and myelomeningocele: anthropometric measurements. *J Spinal Cord Med* 2010;33(4):410–419. Using arm span as substitute for height in published BMI growth curves
Noonan syndrome	Witt DR, Biedermann B, Hall JG, Allanson JE. Growth curves for height in Noonan syndrome. *Clin Genet* 1986;30(3):150–153. Stature-for-age, birth to 18 y Ranke MB, Heidemann P, Knupfer C, et al. Noonan syndrome: growth and clinical manifestations in 144 cases. *Eur J Pediatr*. 1988;148(3):220–227. Height-for-age
Prader–Willi syndrome	Holm VA. Appendix A. In: Greenswag LR, Alexander RC, eds. *Management of Prader-Willi Syndrome*. New York, NY: Springer Verlag; 1998. p. 317. Height-for-age, 3–25 y Butler MG, Meaney FJ. Standards for selected anthropometric measurements in Prader-Willi syndrome. *Pediatrics*. 1991;88(4):853–860. Weight, height, sitting height, head circumference, triceps and subscapular skinfold, and other measures, 2–22 y Butler MG, Sturich J, Lee J, et al. Growth standards of infants with Prader-Willi syndrome. *Pediatrics*. 2011;127(4):687–695. Weight-, length-, head circumference-for-age, 0–36 mo; BMI and weight-for-length, 0–36 mo
Sickle-cell disease	Phebus CK, Gloninger MG, Maciak BJ. Growth patterns by age and sex in children with sickle cell disease. *J Pediatr*. 1984;105(1):28–33. Height- and weight-for-age, birth to 18 y. Tanner JM, Davies PS. Clinical longitudinal standards for height and height velocity for North American children. *J Pediatr*. 1985;107(3):317–329. Height velocity (cm/y), 2½–19 y Stevens MCG, Maude GH, Cupidore L, et al. Prepubertal growth and skeletal maturation in children with sickle cell disease. *Pediatrics*. 1986;78(1):124–132. Weight, height, height velocity, bone age, 0–9 y

Silver–Russell syndrome	Tanner JM, Lejarraga H, Cameron N. The natural history of the Silver-Russell syndrome: a longitudinal study of thirty-nine cases. *Pediatr Res*. 1975;9(8):611–623. Height- and height velocity-for-age, 2–19 y (includes periods of treatment with human growth hormone)
Turner syndrome	Lyon AJ, Preece MA, Grant DB. Growth curve for girls with Turner syndrome. *Arch Dis Child*. 1985:60:932–935.[a] Height-for-age, 2–19 y (girls) Ranke MB, Pflüger H, Rosendahl W, et al. Turner syndrome: spontaneous growth in 150 cases and review of the literature. *Eur J Pediatr*. 1983;141:81–88.[a] Height- and height velocity-for-age, bone age
Williams–Beuren syndrome	Morris CA, Demsey SA, Leonard CO, et al. Natural history of Williams syndrome: physical characteristics. *J Pediatr*. 1988;113(2):318–326. Stature-for-age, birth to 24 mo and birth to 18 y; weight-for-age, birth to 18 y; head circumference-for-age, birth to 36 mo and 2–18 y Pankau R, Partsch CJ, Gosch A, et al. Statural growth in Williams-Beuren syndrome. *Eur J Pediatr*. 1992;151;751–755.[a] Height-for-age, head-circumference-for-age Pankau R, Partsch CJ, Neblung A, et al. Natural history of body mass index in Williams-Beuren syndrome. *Am J Med Genet*. 1994;52(1):51–54. BMI-for-age, both genders, 0–18 y Martin NDT, Smith WR, Cole TJ, Preece MA. New height, weight, and head circumference charts for British children with Williams syndrome. *Arch Dis Child*. 2007;92(7):598–601. Height-, weight-, head circumference-, and BMI-for-age

[a] A copy of the growth chart is included in this appendix.

TABLE I-11 Clinical Examination in Nutritional Deficiencies and Excesses

	Major Physiologic Status Functions	Deficiency Signs	Excess Signs	Important Food Sources	Potential Causes of Deficiency	Status Assessment
Nutrient						
Carbohydrate	Supplies energy at an average of 4 kcal/g (sparing protein) and is the major energy source for CNS function; unrefined, complex carbohydrates supply fiber that aids in normal bowel function	Ketosis	May cause diarrhea, obesity	Bread, cereals, crackers, potatoes, corn, simple sugar (sugar, honey), fruits and vegetables, milk, breast milk, infant formula	Malabsorption	Blood sugar, OGTT
Fat	Concentrated calorie source at an average of 9 kcal/g; constitutes part of the membrane structure of every cell; supplies essential fatty acids; provides and carries fat-soluble vitamins (A, D, E, K)	Essential fatty acid deficiency (dry, scaly skin, poor weight gain, hair loss); requirements are increased by cell turnover	Atherosclerosis may be affected by excessive intakes of certain dietary fats; altered blood lipid levels	Shortening, oil, butter, margarine, protein-rich foods (meat, dairy, nuts), breast milk, infant formula	Cystic fibrosis, biliary disease, short bowel syndrome, hereditary lipoprotein disorders	Total cholesterol, LDL, HDL
Protein	Supplies energy at an average of 4 kcal/g; regulates body processes as part of enzymes, hormones, body fluids, and antibodies, which increase resistance to infection; provides nitrogen; constitutes part of the membrane structure of every cell	Dry, depigmented, easily pluckable hair; bilateral, dependent edema, cirrhosis, fatty liver, decreased visceral proteins; skin is dry with pellagroid dermatosis in severe cases	Azotemia, acidosis, hyperammonemia	Meat, poultry, fish, legumes, eggs, cheese, milk and other dairy products, nuts, breast milk, infant formula	Protein-losing enteropathy, liver disease, gastrointestinal disease, renal disease	Albumin, retinol-binding protein, prealbumin
Fat-soluble vitamins						
Vitamin A	Formation and maintenance of skin and mucous membranes; necessary for the formation of rhodopsin (the photosensitive pigment of the rods governing vision in dim light); regulation of membrane structure and function; necessary for growth and normal immune function	Night blindness, degeneration of the retina, xerophthalmia, follicular hyperkeratosis, poor growth, keratomalacia, Bitot's spots	Fatigue, malaise, lethargy, abdominal pain, hepatomegaly, alopecia, headache with increased intracranial pressure, vomiting	Carrots, liver, green vegetables, sweet potatoes, butter, margarine, apricots, melons, peaches, broccoli, cod liver oil, breast milk, infant formula	Liver disease, cystic fibrosis, short bowel syndrome, protein deficiency (alters transport)	Plasma retinol (HPLC), plasma retinol-binding protein, relative dose response, dark adaptation test, liver biopsy concentration

Vitamin D	Promotes intestinal absorption of calcium and phosphate and renal conservation of calcium and phosphorus	Rickets, osteomalacia, costochondral beading, epiphyseal enlargement, cranial bossing, bowed legs, persistently open anterior fontanel	Hypercalcemia, vomiting, anorexia, diarrhea, convulsions	Cod liver oil, fish, eggs, liver, butter, fortified milk, sunlight (activation of 7-dehydrocholesterol in the skin), infant formula	Liver disease, cystic fibrosis, short bowel syndrome, renal disease	Plasma 25-hydrooxyvitamin D (HPLC), serum alkaline phosphatase, calcium and phosphate, radiography, bone densitometry
Vitamin E	Acts as an antioxidant and free radical scavenger to prevent peroxidation of polyunsaturated fatty acids in the body; neuromuscular function	Hemolytic anemia in the premature and newborn, enhanced fragility of red blood cells, increased peroxidative hemolysis	In anemia, suppresses the normal hematologic response to iron	Oils high in polyunsaturated fatty acids, milk, eggs, breast milk, infant formula	Cystic fibrosis, short bowel syndrome, liver disease	Plasma tocopherol (HPLC) (corrected for total or LDL cholesterol), hydrogen peroxide hemolysis
Vitamin K	Necessary for prothrombin and the three blood-clotting factors VII, IX, and X; half of the vitamin K in humans is of intestinal origin, synthesized by gut flora; necessary for bone mineralization	Hemorrhagic manifestations (especially in newborns), prolonged clotting	Hemolytic anemia, nerve palsy	Green leafy vegetables, fruits, cereals, dairy products, soybeans, breast milk, infant formula	Liver disease, antibiotic therapy	Prothrombin time (prolonged), plasma phylloquinone, clotting factor levels, proteins induced by vitamin K absence or antagonists-II (PIVKA-II)
Water-soluble vitamins						
Ascorbic acid (vitamin C)	Forms collagen cross-linkage of proline hydroxylase, thus strengthening tissue and improving wound healing and resistance to infection; aids absorption of iron	Joint tenderness, scurvy (capillary hemorrhaging), impaired wound healing, acute periodontal gingivitis, petechiae, purpura, anemia	Documentation of a chronic high intake may result in "rebound" deficiency symptoms	Heat labile; broccoli, papaya, orange, mango, grapefruit, strawberries, tomatoes, potatoes, leafy vegetables, breast milk, infant formula	Stress	Plasma level (enzyme assay/HPLC), leukocyte concentration (longer-term), whole blood concentration, urine concentration
Biotin	Component of several carboxylating enzymes; plays an important role in the metabolism of fat and carbohydrate	Anorexia, nausea, vomiting; glossitis; depression; dry, scaly dermatitis; thin hair; loss of eyebrows	None known	Liver, kidney, egg yolk, breast milk, infant formula	Certain inborn errors of metabolism	Plasma (microbiologic assay), plasma lactate, urine organic acids, lymphocyte carboxylase

Cobalamin (B_{12} intrinsic factor required)	Cobalamin-containing co enzymes function in the degradation of certain odd chain fatty acids and in the recycling of tetrahydrofolate	Megaloblastic anemia, neurologic deterioration	None known	Animal products, breast milk, infant formula	Ileal disease, strict vegetarian, lack of intrinsic factor	Plasma level (RIA or microbiologic), Schilling test, plasma homocysteine, deoxyuridine suppression test
Folacin	Utilized in carbon transfer and nucleotide synthesis	Megaloblastic anemia, stomatitis, glossitis, neural tube defects in pregnancy	None known	Liver, leafy vegetables, fruit, yeast, breast milk, infant formula	Liver disease, alcoholism, celiac disease, inflammatory bowel disease	Plasma level (RIA or microbiologic), red cell level
Niacin	Aids in energy utilization as part of a coenzyme (NAD^+ and $NADP^+$) in fat synthesis, tissue respiration, and carbohydrate utilization; aids digestion and fosters normal appetite; synthesized from the amino acid tryptophan	Pellagra (dermatitis, diarrhea, dementia, death), cheilosis, angular stomatic inflammation of mucous membranes, weakness	Dilation of the capillaries, vasomotor instability, "flushing" (utilization of muscle glycogen, serum lipids, mobilization of fatty acids during exercise)	Liver, meat, fish, poultry, peanuts, fortified cereal products, yeast, breast milk, infant formula	B_6 deficiency (impairs conversion of tryptophan to niacin)	Urine ratio of metabolites (*N*-methyl-nicotinamide: 2-pyridone), tryptophan load, red cell NAD or NAD:NADP ratio
Pantothenic acid	Component of coenzyme A; plays a role in release of energy from carbohydrates and in synthesis and degradation of fatty acids	Infertility, abortion, slow growth, depression, vomiting, malaise, abdominal stress	Diarrhea, water retention	Meat, fish, poultry, whole grains, legumes, breast milk, infant formula	Severe malnutrition	Urine excretion, whole blood level (RIA/ microbiologic)
Pyridoxine (B_6)	Coenzyme component for many of the enzymes of amino acid metabolism; all compounds implicated as neuro-transmitters are synthesized and/or metabolized in B_6-dependent reactions	Convulsions, weight loss, ab-dominal distress, vomiting, hyperir-ritability, depres-sion, confusion, hypochromic and macrocytic anemia	Neuropathy	Fish, poultry, meat, wheat, breast milk, infant formula	Elderly, high protein intake	Red cell amino-transferase activity, plasma pyridoxal phosphate (HPLC), tryptophan load test, urine 4-pyridoxic acid

Riboflavin (B_2)	Functions primarily as the reactive portion of flavoproteins concerned with biologic oxidations (cellular metabolism)	Cheilosis, glossitis, photophobia, angular stomatitis, corneal vascularization, scrotal skin changes, seborrhea, magenta tongue	None known	Dairy products, liver, almonds, lamb, pork, breast milk, infant formula	Alcoholism, starvation, chronic diarrhea, malabsorption	Red cell glutathione reductase activity (EGR), red cell flavin adenine dinucleotide, urine riboflavin: creatinine ratio
Thiamine (B_1)	Aids in energy utilization as part of coenzyme component to promote the utilization of carbohydrate; promotes normal functioning of the nervous system; coenzyme for oxidative carboxylation of 2-keto acids	Beriberi, neuritis, cardiac failure, anorexia, restlessness, confusion, loss of vibration sense and deep tendon reflexes, calf tenderness, edema	None known	Lean pork, nuts, whole grain and fortified cereal products, breast milk, infant formula	Alcoholism, refeeding syndrome, prolonged dialysis	Red cell transketolase activity, whole blood level (HPLC), urine thiamine: creatinine ratio
Minerals						
Calcium	Essential for calcification of bone (matrix formation); assists in blood clotting; functions in normal muscle contraction and relaxation and in normal nerve transmission	Osteomalacia, osteoporosis	Hypercalcemia, vomiting, anorexia, lethargy	Dairy products (milk, cheese), sardines, oysters, salmon, herring, greens, breast milk, infant formula	Renal disease, liver disease	Plasma total calcium, plasma free calcium in altered protein binding (e.g., hypoalbuminemia), acidosis, radiographs, CT, photon densitometry
Magnesium	Essential part of many enzyme systems; important for maintaining electrical potential in nerves and muscle membranes and for energy turnover	Tremor, convulsions, hyperexcitability (hypocalcemia tetany)	Diarrhea, transient hypocalcemia	Widely distributed, especially in food of vegetable origin; breast milk, infant formula	Protein calorie malnutrition, refeeding syndrome	Plasma total or free magnesium, magnesium loading test
Phosphorus	Important intracellular anion; involved in many chemical reactions within the body; necessary for energy turnover (ATP)	Weakness, anorexia, malaise, bone pain, growth arrest	Hypocalcemia (when parathyroid gland not fully functioning)	Dairy products, fish, legumes, pork, breast milk, infant formula	Renal disease, liver disease, refeeding syndrome	Plasma concentration, alkaline phosphatase activity, radiography, densitometry, renal tubular excretion threshold

Trace elements						
Chromium	Maintenance of normal glucose metabolism, cofactor for insulin	Disturbed glucose metabolism (lower glucose tolerance caused by insulin resistance)	None known	Brewer's yeast, meat products, cheeses	Protein calorie malnutrition, elderly	Plasma chromium, glucose tolerance
Copper	Constituent of proteins and enzymes, some of which are essential for the proper utilization of iron; immunity; skeletal development	Anemia (hemolytic), neutropenia, bone disease	Excess accumulation in the liver, brain, kidney, cornea; anemia, diarrhea	Oysters, nuts, liver, kidney, corn oil margarine, dried legumes	Menke's kinky hair syndrome. Excess: Wilson's disease	Plasma copper, plasma ceruloplasmin (ferrochelatase), liver biopsy concentration, superoxide dismutase activity
Fluoride	The main target organs of fluoride in man are the enamel of teeth and bones, where fluoride is incorporated into the crystalline structure of hydroxyapatite and produces increased carie resistance	Poor dentition, caries, osteoporosis	Mottling, brown staining of teeth (in excess of 4 ppm); fluorosis occurs after prolonged (10–20 y) ingestion of 20–80 mg/d	Fluoridated water; depends on the geochemical environment and therefore amount in food varies widely	Unfluoridated water, bottled water	
Iodine	Component of thyroid hormones triiodothyronine and thyroxine; important in regulation of cellular oxidation and growth	Goiter, depressed thyroid function, cretinism	Thyroid suppression (thyrotoxicosis)	Iodized table salt, salt water, fish, shellfish (content of most other foods geographically dependent), breast milk, infant formula	Endemic goiter in low-iodine areas	Thyroid hormones, TSH, urinary iodide/creatinine ratio
Iron	Part of hemoglobin molecule; prevents nutritional anemia and fatigue; increases resistance to infection; functions as part of enzymes involved in tissue respiration	Anemia, malabsorption, irritability, anorexia, pallor, lethargy	Hemosiderosis, hemochromatosis	Red meats, liver, dried beans and peas, enriched farina, breast milk, infant formula, infant cereal	Protein-losing enteropathy, malabsorption, acute or chronic blood loss. Excess: hemochromatosis	Plasma iron and ferritin, total iron-binding capacity, hemoglobin/hematocrit, red cell indices, RBC zinc protoporphyrin: heme ratio, bone marrow aspirate stain

Manganese	Essential part of several enzyme systems involved in protein and energy metabolism and in the formation of mucopolysac charide	Impaired growth, skeletal abnormalities, lowered reproductive function, neonatal ataxia	In extremely high exposure of contamination, severe psychiatric and neurologic disorders	Nuts, whole grains, dried fruits, fruits, leafy vegetables		Plasma level, whole blood level, mitochondrial superoxide dismutase
Molybdenum	Essential for the function of flavin-dependent enzymes involved in the production of uric acid and in the oxidation of aldehydes and sulfites	Not described in man	Acts as an antagonist to the essential element copper; gout-like syndrome associated with elevated levels of molybdenum, uric acid, and xanthine oxidase	Varies considerably, depending on growing environment; main contributions come from meat, legumes		
Selenium	Functions as a part of the enzyme glutathione peroxidase, which protects cellular component from oxidative damage	Cardiomyo-pathy probably secondary to oxidative damage	In animals, blindness, abdominal pain	Seafoods, kidney, liver meat, grains (depending on growing environment)	Cystic fibrosis	Plasma concentration, glutathione peroxidase activity, nail/ hair selenium
Zinc	Constituent of enzymes involved in most major metabolic pathways (specifically nucleic acid synthesis for growth and repair)	Growth failure, skin changes, delayed wound healing, hypogeusia, sexual immaturity, hair loss, diarrhea	Acute gastrointestinal upset, vomiting, sweating, dizziness, copper deficiency	Whole grains, legumes, beef, lamb, pork, poultry, nuts, seeds shellfish, eggs, some cheeses, breast milk, infant formula	Malabsorp-tion, chronic diarrhea, liver disease, sickle cell disease	Plasma concentration, alkaline phosphatase activity, urinary excretion, leukocyte concentration

APPENDIX II

Nutritional Requirements

Melanie V. Connolly, MSc, RD



Figure II-1 Choose my plate. (From the US Department of Agriculture. www.choosemyplate.gov.)

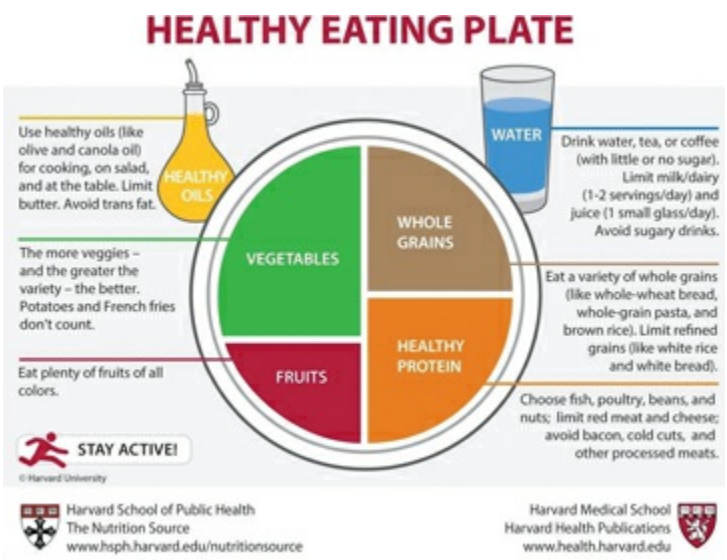


Figure II-2 Healthy eating plate. (Copyright © 2011 Harvard University. For more information about The Healthy Eating Plate, please see The Nutrition Source, Department of Nutrition, Harvard School of Public Health, www.thenutritionsource.org and Harvard Health Publications, www.health.harvard.edu.)

TABLE II-1 Dietary Reference Intakes (DRIs): Recommended Intakes for Individuals and Tolerable Upper Intake Levels, Vitamins

		Infants		Children		Males						Females						Pregnancy		Lactation	
		0–6 mo	7–12 mo	1–3 y	4–8 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	<18 y	19–50 y	<18 y	19–50 y
Biotin (µg/d)	RDA/AI*	5*	6*	8*	12*	20*	25*	30*	30*	30*	30*	20*	25*	30*	30*	30*	30*	30*	30*	35*	35*
	UL	ND[a]	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Choline (mg/d)	RDA/AI*	125*	150*	200*	250*	375*	550*	550*	550*	550*	550*	375*	400*	425*	425*	425*	425*	450*	450*	550*	550*
	UL	ND	ND	1000	1000	2000	3000	3500	3500	3500	3500	2000	3000	3500	3500	3500	3500	3000	3500	3000	3500
Folate (µg/d)	RDA/AI*	65*	80*	150	200	300	400	400	400	400	400	300	400	400	400	400	400	600	600	500	500
	UL	ND	ND	300	400	600	800	1000	1000	1000	1000	600	800	1000	1000	1000	1000	800	1000	800	1000
Niacin (mg/d)	RDA/AI*	2*	4*	6	8	12	16	16	16	16	16	12	14	14	14	14	14	18	18	17	17
	UL	ND	ND	10	15	20	30	35	35	35	35	20	30	35	35	35	35	30	35	30	35
Pantothenic Acid (mg/d)	RDA/AI*	1.7*	1.8*	2*	3*	4*	5*	5*	5*	5*	5*	4*	5*	5*	5*	5*	5*	6*	6*	7*	7*
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Riboflavin (mg/d)	RDA/AI*	0.3*	0.4*	0.5	0.6	0.9	1.3	1.3	1.3	1.3	1.3	0.9	1.0	1.1	1.1	1.1	1.1	1.4	1.4	1.6	1.6
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Thiamine (mg/d)	RDA/AI*	0.2*	0.3*	0.5	0.6	0.9	1.2	1.2	1.2	1.2	1.2	0.9	1.0	1.1	1.1	1.1	1.1	1.4	1.4	1.4	1.4
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Vitamin A (µg/d)	RDA/AI*	400*	500*	300	400	600	900	900	900	900	900	600	700	700	700	700	700	750	770	1200	1300
	UL	600	600	600	900	1700	2800	3000	3000	3000	3000	1700	2800	3000	3000	3000	3000	2800	3000	2800	3000
Vitamin B_6 (mg/d)	RDA/AI*	0.1*	0.3*	0.5	0.6	1.0	1.3	1.3	1.3	1.7	1.7	1.0	1.2	1.3	1.3	1.5	1.5	1.9	1.9	2.0	2.0
	UL	ND	ND	30	40	60	80	100	100	100	100	60	80	100	100	100	100	80	100	80	100
Vitamin B_{12} (µg/d)	RDA/AI*	0.4*	0.5*	0.9	1.2	1.8	2.4	2.4	2.4	2.4	2.4	1.8	2.4	2.4	2.4	2.4	2.4	2.6	2.6	2.8	2.8
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Vitamin C (mg/d)	RDA/AI*	40*	50*	15	25	45	75	90	90	90	90	45	65	75	75	75	75	80	85	115	120
	UL	ND	ND	400	650	1200	1800	2000	2000	2000	2000	1200	1800	2000	2000	2000	2000	1800	2000	1800	2000
Vitamin D (IU/d)	RDA/AI*	400*	400*	600	600	600	600	600	600	600	800	600	600	600	600	600	800	600	600	600	600
	UL	1000	1500	2500	3000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000	4000
Vitamin E (mg/d)	RDA/AI*	4*	5*	6	7	11	15	15	15	15	15	11	15	15	15	15	15	15	15	19	19
	UL	ND	ND	200	300	600	800	1000	1000	1000	1000	600	800	1000	1000	1000	1000	800	1000	800	1000
Vitamin K (µg/d)	RDA/AI*	2.0*	2.5*	30*	55*	60*	75*	120*	120*	120*	120*	60*	75*	90*	90*	90*	90*	75*	90*	75*	90*
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

RDA, recommended dietary allowance; UL, upper intake levels.

Adequate intakes (AIs) are followed by an asterisk (*).

[a]ND = not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; The Institute of Medicine. *Dietary Reference Intakes for Calcium and Vitamin D*. Washington, DC: National Academy of Sciences; 2010; Reprinted with permission from the National Academies Press, Copyright 2006 and 2010, National Academy of Sciences. Complete reports can be accessed at www.nap.edu and www.iom.edu.

TABLE II-2 Dietary Reference Intakes (DRIs): Recommended Intakes for Individuals and Tolerable Upper Intake Levels, Elements

		Infants		Children		Males						Females						Pregnancy			Lactation		
		0–6 mo	7–12 mo	1–3 y	4–8 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	<18 y	19–30 y	31–50 y	<18 y	19–50 y	31–50 y
Arsenic (mg/d)	RDA/AI*	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Boron (mg/d)	RDA/AI*	ND[a]	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
	UL	ND	ND	3	6	11	17	20	20	20	20	11	17	20	20	20	20	17	20	20	17	20	20
Calcium (mg/d)	RDA/AI*	200*	260*	700	1000	1300	1300	1000	1000	1000	1200	1300	1300	1000	1000	1200	1200	1300	1000	1000	1300	1000	1000
	UL	1000	1500	2500	2500	3000	3000	2500	2500	2000	2000	3000	3000	2500	2500	2000	2000	3000	2500	2500	3000	2500	2500
Chromium (µg/d)	RDA/AI*	0.2*	5.5*	11*	15*	25*	35*	35*	35*	30*	30*	21*	24*	25*	25*	20*	20*	29*	30*	30*	44*	45*	45*
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Copper (µg/d)	RDA/AI*	200*	220*	340	440	700	890	900	900	900	900	700	890	900	900	900	900	1000	1000	1000	1300	1300	1300
	UL	ND	ND	1000	3000	5000	8000	10,000	10,000	10,000	10,000	5000	8000	10,000	10,000	10,000	10,000	8000	10,000	10,000	8000	10,000	10,000
Fluoride (mg/d)	RDA/AI*	0.01*	0.5*	0.7*	1*	2*	3*	4*	4*	4*	4*	2*	3*	3*	3*	3*	3*	3*	3*	3*	3*	3*	3*
	UL	0.7	0.9	1.3	2.2	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
Iodine (µg/d)	RDA/AI*	110*	130*	90	90	120	150	150	150	150	150	120	150	150	150	150	150	220	220	220	290	290	290
	UL	ND	ND	200	300	600	900	1100	1100	1100	1100	600	900	1100	1100	1100	1100	900	1100	1100	900	1100	1100
Iron (mg/d)	RDA/AI*	0.27*	11	7	10	8	11	8	8	8	8	8	15	18	18	8	8	27	27	27	10	9	9
	UL	40	40	40	40	40	45	45	45	45	45	40	45	45	45	45	45	45	45	45	45	45	45
Magnesium (mg/d)	RDA/AI*	30*	75*	80	130	240	410	400	420	420	420	240	360	310	320	320	320	400	350	360	360	310	320
	UL	ND	ND	65	110	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350	350
Manganese (mg/d)	RDA/AI*	0.003*	0.6*	1.2*	1.5*	1.9*	2.2*	2.3*	2.3*	2.3*	2.3*	1.6*	1.6*	1.8*	1.8*	1.8*	1.8*	2.0*	2.0*	2.0*	2.6*	2.6*	2.6*
	UL	ND	ND	2	3	6	9	11	11	11	11	6	9	11	11	11	11	9	11	11	9	11	11
Molybdenum (µg/d)	RDA/AI*	2*	3*	17	22	34	43	45	45	45	45	34	43	45	45	45	45	50	50	50	50	50	50
	UL	ND	ND	300	600	1100	1700	2000	2000	2000	2000	1100	1700	2000	2000	2000	2000	1700	2000	2000	1700	2000	2000
Nickel (mg/d)	RDA/AI*	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
	UL	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	...
Phosphorus (mg/d)	RDA/AI*	100*	275*	460	500	1250	1250	700	700	700	700	1250	1250	700	700	700	700	1250	700	700	1250	700	700
	UL	ND	ND	3000	3000	4000	4000	4000	4000	4000	3000	4000	4000	4000	4000	4000	3000	3500	3500	3500	4000	4000	4000
Selenium (µg/d)	RDA/AI*	15*	20*	20	30	40	55	55	55	55	55	40	55	55	55	55	55	60	60	60	70	70	70
	UL	45	60	90	150	280	400	400	400	400	400	280	400	400	400	400	400	400	400	400	400	400	400
Silicon	RDA/AI*	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Vanadium (mg/d)	RDA/AI*	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
	UL	ND	ND	ND	ND	ND	ND	1.8	1.8	1.8	1.8	ND	ND	1.8	1.8	1.8	1.8	ND	ND	ND	ND	ND	ND
Zinc (mg/d)	RDA/AI*	2*	3	3	5	8	11	11	11	11	11	8	9	8	8	8	8	12	11	11	13	12	12
	UL	4	5	7	12	23	34	40	40	40	40	23	34	40	40	40	40	34	40	40	34	40	40

RDA, recommended dietary allowance; UL, upper intake levels.

Adequate intakes (AIs) are followed by an asterisk (*).

[a]ND = not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; The Institute of Medicine. *Dietary Reference Intakes for Calcium and Vitamin D*. Washington, DC: National Academy of Sciences; 2010; Reprinted with permission from the National Academies Press, Copyright 2006 and 2010, National Academy of Sciences. Complete reports can be accessed at www.nap.edu and www.iom.edu.

TABLE II-3 Dietary Reference Intakes (DRIs): Recommended Intakes for Individuals and Acceptable Macronutrient Distribution Ranges, Macronutrients

		Infants		Children		Males						Females						Pregnancy	Lactation
		0–6 mo	7–12 mo	1–3 y	4–8 y	9–13 y	14–18 y	19–30 y	31–50 y	>50 y		9–13 y	14–18 y	19–30 y	31–50 y	>50 y		<18–50 y	<18–50 y
Carbohydrate (g/d)	RDA/AI*	60*	95*	130	130	130	130	130	130	130		130	130	130	130	130		175	210
	AMDR	ND[a]	ND	45–65	45–65	45–65	45–65	45–65	45–65	45–65		45–65	45–65	45–65	45–65	45–65		45–65	45–65
Total fiber (g/d)	RDA/AI*	ND	ND	19*	25*	31*	38*	38*	38*	30*		26*	26*	25*	25*	21*		28*	29*
Fat (g/d)	RDA/AI*	31*	30*	ND	ND	ND	ND	ND	ND	ND		ND	ND	ND	ND	ND		ND	ND
	AMDR	ND	ND	30–40	25–35	25–35	25–35	20–35	20–35	20–35		25–35	25–35	20–35	20–35	20–35		20–35	20–35
Linoleic acid (g/d)	RDA/AI*	4.4*	4.6*	7*	10*	12*	16*	17*	17*	14*		10*	11*	12*	12*	11*		13*	13*
	AMDR	ND	ND	5–10	5–10	5–10	5–10	5–10	5–10	5–10		5–10	5–10	5–10	5–10	5–10		5–10	5–10
α-Linolenic acid (g/d)	RDA/AI*	0.5*	0.5*	0.7*	0.9*	1.2*	1.6*	1.6*	1.6*	1.6*		1.0*	1.1*	1.1*	1.1*	1.1*		1.4*	1.3*
	AMDR	ND	ND	0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2		0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2	0.6–1.2		0.6–1.2	0.6–1.2
Protein (g/d)b	RDA/AI*	9.1*	13.5	13	19	34	52	56	56	56		34	46	46	46	46		71	71
	AMDR	ND	ND	5–20	10–30	10–30	10–30	10–35	10–35	10–35		10–30	10–30	10–35	10–35	10–35		10–35	10–35

RDA, recommended dietary allowance; UL, upper intake levels.

Adequate intakes (AIs) are followed by an asterisk (*).

[a] ND = not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts.

[b] Using reference body weight, based on 1.5 g/kg/d for infants, 1.1 g/kg/d for 1–3 year, 0.95 g/kg/d for 4–13 year, 0.85 g/kg/d for 14–18 year, 0.8 g/kg/d for adults, and 1.1 g/kg/d for pregnant (using prepregnancy weight) and lactating women.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; Reprinted with permission from the National Academies Press, Copyright 2006, National Academy of Sciences. Complete report can be accessed at www.nap.edu.

TABLE II-4 Dietary Reference Intakes (DRIs): Recommended Intakes for Individuals and Tolerable Upper Intake Levels, Macronutrients, Fluids, and Electrolytes

		Infants		Children		Males						Females						Pregnancy		Lactation	
		0–6 mo	7–12 mo	1–3 y	4–8 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	9–13 y	14–18 y	19–30 y	31–50 y	51–70 y	>70 y	<18 y	19–50 y	<18 y	19–50 y
Sodium (g/d)	AI	0.12	0.37	1.0	1.2	1.5	1.5	1.5	1.5	1.3	1.2	1.5	1.5	1.5	1.5	1.3	1.2	1.5	1.5	1.5	1.5
	UL	ND[a]	ND	1.5	1.9	2.2	2.3	2.3	2.3	2.3	2.3	2.2	2.3	2.3	2.3	2.3	2.3	2.3	2.3	2.3	2.3
Chloride (g/d)	AI	0.18	0.57	1.5	1.9	2.3	2.3	2.3	2.3	2.0	1.8	2.3	2.3	2.3	2.3	2.0	1.8	2.3	2.3	2.3	2.3
	UL	ND	ND	2.3	2.9	3.4	3.6	3.6	3.6	3.6	3.6	3.4	3.6	3.6	3.6	3.6	3.6	3.6	3.6	3.6	3.6
Potassium (g/d)	AI	0.4	0.7	3.0	3.8	4.5	4.7	4.7	4.7	4.7	4.7	4.5	4.7	4.7	4.7	4.7	4.7	4.7	4.7	5.1	5.1
	UL	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Water[b] (L/d)	AI	0.7	0.8	1.3	1.7	2.4	3.3	3.7	3.7	3.7	3.7	2.1	2.3	2.7	2.7	2.7	2.7	3.0	3.0	3.8	3.8
	UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL
Carbohydrate (total digestible, g/d)	RDA/AI	60	95	130	130	130	130	130	130	130	130	130	130	130	130	130	130	175	175	210	210
Fat (g/d)	AI	31	30	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Fiber (g/d)	AI	ND	ND	19	25	31	38	38	38	30	30	26	26	25	25	21	21	28	28	29	29
	UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL
Protein (g/d)[c]	AI	9.1	11.0	13	19	34	52	56	56	56	56	34	46	46	46	46	46	71	71	71	71
	UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL	No UL

AI, adequate intake; RDA, recommended dietary allowance; UL, upper intake levels.

RDAs are indicated in bold type.

[a]ND = not determinable due to lack of data of adverse effects in this age group and concern with regard to lack of ability to handle excess amounts.

[b]Total water includes all water in food and beverages.

[c]Based on 0.8g/kg for the reference body weight.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; Reprinted with permission from the National Academies Press, Copyright 2006, National Academy of Sciences. Complete report can be accessed at www.nap.edu.

TABLE II-5 Dietary Reference Intakes (DRIs): Estimated Energy Requirements

Category	Estimated Energy Requirements (kilocalories)
Infants and children (0–2 y)	
0–3 mo	(89 × wt [kg] – 100) + 175
4–6 mo	(89 × wt [kg] – 100) + 56
7–12 mo	(89 × wt [kg] – 100) + 22
13–35 mo	(89 × wt [kg] – 100) + 20
Males	
3–8 y	88.5 – 61.9 × age [y] + PA[a] × (26.7 × wt [kg] + 903 × ht [m]) + 20
9–18 y	88.5 – 61.9 × age [y] + PA × (26.7 × wt [kg] + 903 × ht [m]) + 25
>19 y	662 – 9.53 × age [y] + PA × (15.91 × wt [kg] + 539.6 × ht [m])
Females	
3–8 y	135.3 – 30.8 × age [y] + PA × (10.0 × wt [kg] + 934 × ht [m]) + 20
9–18 y	135.3 – 30.8 × age [y] + PA × (10.0 × wt [kg] + 934 × ht [m]) + 25
>19 y	354 – 6.91 × age [y] + PA × (9.36 × wt [kg] + 726 × ht [m])
Pregnancy	
1st trimester	Nonpregnant EER
2nd trimester	Nonpregnant EER + 340
3rd trimester	Nonpregnant EER + 452
Lactation	
1st 6 mo	Nonpregnant EER + 330
2nd 6 mo	Nonpregnant EER + 400

EER, estimated energy requirement

[a]PA = physical activity coefficient based on physical activity level (PAL); PA = 1.00 if PAL is estimated to be 1.0–1.39 (sedentary); PA = 1.12 if PAL is estimated to be 1.4–1.59 (low active); PA = 1.24 if PAL is estimated to be 1.6–1.89 (active); PA = 1.45 if PAL is estimated to be 1.9–2.5 (very active)

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; Reprinted with permission from the National Academies Press, Copyright 2006, National Academy of Sciences. Complete report can be accessed at www.nap.edu.

TABLE II-6 Criteria and Dietary Reference Intake Values for Energy by Active Individuals by Life Stage Group*

Life Stage Group	Criterion	Active PAL[a] EER (kcal/d)	
		Male	Female
0–6 mo	Energy expenditure plus energy deposition	570	520 (3 mo)
7–12 mo	Energy expenditure plus energy deposition	743	676 (9 mo)
1–2 y	Energy expenditure plus energy deposition	1046	992 (24 mo)
3–8 y	Energy expenditure plus energy deposition	1742	1642 (6 y)
9–13 y	Energy expenditure plus energy deposition	2279	2071 (11 y)
14–18 y	Energy expenditure plus energy deposition	3152	2368 (16 y)
>18 y	Energy expenditure	3067[b]	2403[b] (19 y)
Pregnancy			
14–18 y	Adolescent female EER plus change in TEE plus pregnancy energy deposition		
1st trimester	2368 (16 y)		
2nd trimester	2708 (16 y)		
3rd trimester	2820 (16 y)		
19–50 y	Adult female EER plus change in TEE plus pregnancy energy deposition		
1st trimester	2403 (19 y)[b]		
2nd trimester	2743 (19 y)[b]		
3rd trimester	2855 (19 y)[b]		
Lactation			
14–18 y	Adolescent female EER plus milk energy output minus weight loss		
1st 6 mo	2698 (16 y)		

2nd 6 mo	2768 (16 y)

[a] EER, estimated energy requirement; PAL, physical activity level; TEE, total energy expenditure, the intake which meets the average energy expenditure of individuals at the reference height, weight, and age.

[b] Subtract 10 kcal/d for males and 7 kcal/d for females for each year above 19 years.

For healthy, active Americans and Canadians.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006. Complete report can be accessed at www.nap.edu.

TABLE II-7 Criteria and Dietary Reference Intake Values for Carbohydrate by Life Stage Group

Life Stage Group	Criterion	EAR (g/kg/d)[a]		RDA (g/kg/d)[b]		AI (g/kg/d)[c]
		Male	Female	Male	Female	
0–6 mo	Average content of human milk					60
7–12 mo	Average intake from human milk plus complementary foods					95
1–3 y	Extrapolation from adult data	100	100	130	130	
4–8 y	Extrapolation from adult data	100	100	130	130	
9–13 y	Extrapolation from adult data	100	100	130	130	
14–18 y	Extrapolation from adult data	100	100	130	130	
> 18 y	Brain glucose use	100	100	0.80	130	
Pregnancy						
14–18 y	Adolescent female EAR plus fetal brain glucose use	135	175			
19–50 y	Adult female EAR plus fetal brain	135	175			
Lactation						
14–18 y	Adolescent female EAR plus average human milk content of carbohydrate	160	210			
19–50 y	Adult female EAR plus average human milk content of carbohydrate	160	210			

[a]EAR, estimated average requirement. The intake that meets the estimated nutrient needs of half of the individuals in a group.

[b]RDA, recommended dietary allowance. The intake that meets the nutrient needs of almost all (97%–98%) the individuals in a group.

[c]AI, adequate intake. The observed average or experimentally determined intake by a defined population or subgroup that appears to sustain a defined nutritional status, such as growth rate, normal circulatory nutrient values, or other functional indicators of health. The AI is used if sufficient scientific evidence is not available to derive an EAR. For healthy infants receiving human milk, the AI is the mean intake. The AI is not equivalent to an RDA.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential*

Guide to Nutrient Requirements. Washington, DC: The National Academies Press; 2006; Reprinted with permission from the National Academies Press, Copyright 2006, National Academy of Sciences. Complete report can be accessed at www.nap.edu.

TABLE II-8 Criteria and Dietary Reference Intake Values for Protein by Life Stage Group

Life Stage Group	Criterion	AI or RDA for Reference Individual (g/d)		EAR (g/kg/d)[a]		RDA (g/kg/d)[b]		AI (g/kg/d)[c]
		Males	Females	Males	Females	Males	Females	
0–6 mo	Average consumption of protein from human milk	9.1 (AI)	9.1 (AI)					1.52
7–12 mo	Nitrogen equilibrium plus protein deposition	13.5	13.5	1.0	1.0	1.2	1.2	
1–3 y	Nitrogen equilibrium plus protein deposition	13	13	0.87	0.87	1.05	1.05	
4–8 y	Nitrogen equilibrium plus protein deposition	19	19	0.76	0.76	0.95	0.95	
9–13 y	Nitrogen equilibrium plus protein deposition	34	34	0.76	0.76	0.95	0.95	
14–18 y	Nitrogen equilibrium plus protein deposition	52	46	0.73	0.71	0.85	0.85	
>18 y	Nitrogen equilibrium	56	46	0.66	0.66	0.80	0.80	
Pregnancy							0.88	1.1
Lactation							1.05	1.3

[a]EAR, estimated average requirement. The intake that meets the estimated nutrient needs of half of the individuals in a group.

[b]RDA, recommended dietary allowance. The intake that meets the nutrient needs of almost all (97%–98%) the individuals in a group.

[c]AI, adequate intake. The observed average or experimentally determined intake by a defined population or subgroup that appears to sustain a defined nutritional status, such as growth rate, normal circulatory nutrient values, or other functional indicators of health. The AI is used if sufficient scientific evidence is not available to derive an EAR. For healthy infants receiving human milk, the AI is the mean intake. The AI is not equivalent to an RDA.

The EAR and RDA for pregnancy are only for the second half of pregnancy. For the first half of pregnancy, the protein requirements are the same as those of the nonpregnant woman.

Sources: Adapted from Otten JJ, Pitzi HJ, Meyers LD, eds. *DRI: The Essential Guide to Nutrient Requirements*. Washington, DC: The National Academies Press; 2006; Reprinted with permission from the National Academies Press, Copyright 2006, National Academy of Sciences. Complete report can be accessed at www.nap.edu.

TABLE II-9 Normal Electrolyte Requirements

Electrolyte	Daily Requirement
Sodium	2–4 mEq/kg
Potassium	2–3 mEq/kg
Chloride	2–3 mEq/kg
Magnesium	0.25–0.5 mEq/kg
Calcium	0.5–2.5 mEq/kg
Phosphorus	1–2 mmol/kg

Source: Adapted from Collier S, Gura KM, DeLoid L, et al. Parenteral nutrition. In: Sonneville K, Duggan C, eds. *Manual of Pediatric Nutrition*. 5th ed. Shelton, CT: People's Medical Publishing House USA; 2013.

TABLE II-10 Fluid Requirements

Holliday-Segar Method	
0–10 kg	100 mL/kg
10–20 kg	1000 mL + 50 mL/kg > 10 kg
Over 20 kg	1500 mL + 20 mL/kg > 20 kg
Body Surface Area Method	
	1500 mL/m2 24 h

Source: Adapted from Collier S, Gura KM, DeLoid L, et al. Parenteral nutrition. In: Sonneville K, Duggan C, eds. *Manual of Pediatric Nutrition*. 5th ed. Shelton, CT: People's Medical Publishing House USA; 2013.

TABLE II-11 Milliequivalent–Milligram Conversion Table

Mineral Element	Chemical Symbol	Atomic Weight	Valence
Bicarbonate	HCO_3	61	1
Calcium	Ca	40	2
Chlorine	Cl	35.4	1
Magnesium	Mg	24.3	2
Phosphate	PO_4	95	3
	HPO_4	96	2
	H_2PO_4	97	3
Potassium	K	39	1
Sodium	Na	23	1
Sulfur	S	32	2
Sulfate	SO_4	96	2

Zinc	Zn	65.4	2

Source: Adapted from Gura KM. Appendix 4.2 conversion tables. In: Sonneville K, Duggan C, eds. *Manual of Pediatric Nutrition*. 5th ed. Shelton, CT: People's Medical Publishing House USA; 2013.

TABLE II-12 Recommended Daily Allowances (RDA) for Energy and Protein

Age	kcals/kg		g protein/kg	
0–6 mo	108		2.2	
6–12 mo	98		1.6	
1–3 y	102		1.2	
4–6 y	90		1.1	
7–10 y	70		1.0	
	Males	Females	Males	Females
11–14 y	55	47	1.0	1.0
15–18 y	45	40	0.9	0.8
19–24 y	40	38	0.8	0.8
25–50 y	37	36	0.8	0.8

Source: Adapted from National Academy of Sciences. *Recommended Daily Allowances*. 10th ed. Washington, DC: National Academies Press; 1989; Reprinted with permission from the National Academies Press, Copyright 1989, National Academy of Sciences.

TABLE II-13 Schofield Equations for Calculating Basal Metabolic Rate in Children

Males	
0–3 y	REE = 0.167W + 15.174H – 617.6
3–10 y	REE = 19.59W + 1.303H + 414.9
10–18 y	REE = 16.25W + 1.372H + 515.5
>18 y	REE = 15.057W + 1.004H + 705.8
Females	
0–3 y	REE = 16.252W + 10.232H – 413.5
3–10 y	REE = 16.969W + 1.618H + 371.2
10–18 y	REE = 8.365W + 4.65H + 200
>18 y	REE = 13.623W + 23.8H + 98.2

H, height (cm); REE, resting energy expenditure (kcal/d); W, weight (kg).

Source: Adapted from Schofield W. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr.* 1985;39(suppl 1):5–41.

TABLE II-14 Harris-Benedict Equation for Estimating Basal Energy Expenditure (kcal/d)

Men	66.5 (13.8 × W) + (5 × H) – (6.8 × A)

Women	655 + (9.6 × W) + (1.8 × H) – (4 × A)

A, age; H, height (cm); W, weight (kg).

Source: Adapted from Harris J, Benedict F. *A Biometric Study of Basal Metabolism in Man*. Washington, DC: Carnegie Institute of Washington; 1919. Publication no. 279.

TABLE II-15 Equations for Predicting Basal Metabolic Rate from Body Weight in Kilograms

Age Range (y)	kcal/kg/d
Males	
0–3	60.9 W – 54
3–10	22.7 W + 495
10–18	17.5 W + 651
18–30	15.3 W + 679
30–60	11.6 W + 879
>60	13.5 W + 487
Females	
0–3	61.0 W – 51
3–10	22.5 W + 499
10–18	22.2 W + 746
18–30	14.7 W + 496
30–60	8.7 W + 829
>60	10.5 W + 596

Source: Adapted from World Health Organization. *Energy and Protein Requirements*. Geneva, Switzerland: WHO; 1985. Report of a Joint FAO/WHO/UNU Expert Consultation, Technical Report Series 724.

TABLE II-16 Assessment of Energy Requirements in Hospitalized Pediatric Patients: Step 1. Estimating Basal Metabolic Requirements

Body Wt (kg)	kcal/d		Body Wt (kg)	kcal/d	
	Male	Female		Male	Female
3.0	120	144	36.0	1270	1173
4.0	191	191	38.0	1305	1207
5.0	270	274	40.0	1340	1241
6.0	330	336	42.0	1370	1274
7.0	390	395	44.0	1400	1306
8.0	445	448	46.0	1430	1338
9.0	495	496	48.0	1460	1369
10.0	545	541	50.0	1485	1399
11.0	590	582	52.0	1505	1429
12.0	625	620	54.0	1555	1458
13.0	665	655	56.0	1580	1487
14.0	700	687	58.0	1600	1516
15.0	725	718	60.0	1630	1544
16.0	750	747	62.0	1660	1572
17.0	780	775	64.0	1690	1599
18.0	810	802	66.0	1725	1626
19.0	840	827	68.0	1765	1653
20.0	870	852	70.0	1785	1679
22.0	910	898	72.0	1815	1705
24.0	980	942	74.0	1845	1731
26.0	1070	984	76.0	1870	1756
28.0	1100	1025	78.0	1900	1781
30.0	1140	1063	80.0	1935	1805
32.0	1190	1101	82.0	1970	1830
34.0	1230	1137	84.0	2000	1855

Source: Adapted from Schofield W. Predicting basal metabolic rate, new standards and review of previous work. *Hum Nutr Clin Nutr.* 1985;39(suppl 1):5–41.

TABLE II-17 Assessment of Energy Requirements in Hospitalized Pediatric Patients: Step 2. Factors Used in the Calculation of Total Energy Expenditure

Factors	
Activity	
Bedridden	1.20
Ambulatory	1.30
Fever	
Per degree Fahrenheit over normal	1.07
Injury	
Starvation	0.70
Minor surgery	1.00–1.20
Peritonitis	1.20–1.50

Soft tissue trauma	1.14–1.37
Skeletal trauma	1.35
Major sepsis	1.40–1.60
Severe thermal injury	2.00

Source: Adapted from Long C, Schaffel N, Geiger J, et al. Metabolic response to injury and illness: estimation of energy and protein needs from indirect calorimetric and nitrogen balance. *J Parenter Enteral Nutr.* 1979;3(6):452; Reprinted by permission of SAGE Publications.

Appendix III

Enteral Products

Melanie V. Connolly, MSc, RD

TABLE III-1 Enteral Product References: Infant (per 100 mL)

Product (Manufacturer)	kcal/mL	Protein			Fat			Carbohydrate			Na mEq	K mEq	Ca mg	Fe mg	P mg	Water mOsm/kg	General Comments
		g	% kcal	Source	g	% kcal	Source	g	% kcal	Source							
Human milk, term, mature	0.67	1.0	5	Whey and casein	3.90	55	Long chain fatty acids high in palmitic, oleic, linoleic and linolenic	7.2	38	Lactose	0.78	1.35	28	0.04	14	255	Preferred nutrition in human infants
EleCare (Abbott)	0.67	2.0	15	Free L-amino acids	3.2	42	High oleic safflower oil, MCT oil, soy oil	7.2	43	Corn syrup solids	1.3	2.6	78	1.2	57	335	100% Free amino acids 33% fat as MCT oil
Enfamil A.R. (Mead Johnson)	0.67	1.7	10	Nonfat milk	3.4	46	Palm olein, soy oil, coconut oil, high oleic sunflower oil	7.3	44	Lactose, rice starch, malto-dextrin	1.17	1.87	53	1.2	36	240	Thickened with rice cereal
Enfamil Enfacare 22 cal/oz (Mead Johnson)	0.74	2.1	11	Whey protein concentrate and nonfat milk	3.9	46	High oleic vegetable oil, soy oil, MCT oil, coconut oil	7.7	43	Liquid: maltodex-trin, lactosepow-der: lactose, corn syrup solids	1.13	2	89	1.3	49	310	Designed for former premature infants now >2 kg or 40 weeks corrected age. Additional Ca, P, vit A, vit D, folate, Zn
Enfamil Gentlease (Mead Johnson)	0.67	1.53	9	Partially hydrolyzed nonfat milk and whey protein concentrate solids	3.6	48	Palm olein, soy, coconut, and high oleic sunflower oils	7.3	43	Corn syrup solids	1	1.87	55	1.2	31	230	
Enfamil 24 (Mead Johnson)	0.81	1.7	8.5	Reduced minerals whey, nonfat milk	4.3	48	Palm olein, soy, coconut and high oleic sunflower oils, DHA, ARA	8.8	43.5	Lactose	.95	2.25	63	1.46	43	360	
Enfamil Premature (Mead Johnson)	0.67	2.0	12	Whey protein concentrate nonfat milk	3.4	44	MCT, soy and high oleic vegetable oils, single cell oil blend rich in DHA and ARA	7.4	44	Corn syrup solids, lactose	1.69	1.69	112	1.22	56	240	Designed for former premature infants Additional Ca, P, vit A, vit D, folate, Zn, Ca:P ratio of 2:1 High in protein

Enfamil Premature 24 cal/ oz (Mead Johnson)	0.81	2.4	12	Whey protein concentrate nonfat milk	4.1	44	MCT, soy and high oleic vegetable oils, single cell oil blend rich in DHA and ARA	8.9	44	Corn syrup solids, lactose	2.04	2.05	134	1.46	67	300	Designed for former premature infants additional Ca, P, vit A, vit D, folate, Zn, Ca:P ratio of 2:1 High in protein
Enfamil Infant (Mead Johnson)	0.67	1.3	8.5	Whey protein concentrate, nonfat milk	3.5	48	Palm olein oil, soy oil, coconut oil, high oleic sunflower oil	7.5	43	lactose, galactooligo-saccharides, polydextrose	0.78	1.8	52	1.2	29	300	
Enfamil Newborn (Mead Johnson)	0.67	1.4	8.5	Nonfat milk, whey protein concentrate	3.6	48	Palm olein, coconut, soy, and high oleic sunflower oils	7.5	43.5	Lactose, galactooligo-saccharides, polydextrose	0.8	1.9	53	1.22	29	300	Newborn to 3 months Higher in vitamin D (400 IU in 27 oz.)
Enfamil Prosobee (Mead Johnson)	0.67	1.69	10	Soy protein isolate	3.6	48	Palm olein, soy, coconut, and high oleic sunflower oils, single-cell oil blend rich in DHA and ARA	7.2	42	Corn syrup solids	1.05	2.1	70	1.22	46	200	Lactose free
Enfaport (Mead Johnson)	1.01	3.6	14	Calcium caseinate, sodium caseinate	5.5	45	MCT oil, soy oil, DHA and ARA	10.3	41	Corn syrup solids	1.3	3	95	1.83	53	280	Ready to feed 30 cal/oz. formula that can be modified to as low as 20 cal/oz. 84% fat as MCT oil Includes all essential fatty acids
Gerber Good Start Soy (Nestle)	0.67	1.7	10	Soy protein isolate	3.4	46	Palm olein, soy, coconut, high oleic safflower or sunflower oils	7.5	44	Corn maltodex-trin, sucrose	1.1	2.0	70	1.2	42	180	
Gerber Good Start Gentle (Nestle)	0.67	1.5	9	Hydrolyzed whey protein concentrate	3.4	46	Palm olein, soy, coconut and high oleic safflower or sunflower oils	7.8	45	Lactose, corn maltodextrin	0.79	1.9	45	1.0	26	250	100 % whey protein
Monogen (Nutricia)	0.73	2	11	Whey protein concentrate	2	25	Oil blend (including coconut, walnut)	11.9	64	Corn syrup solids	1.52	1.62	48	0.74	35	250	90% fat as MCT oil Contains complete vitamin and mineral profile (including trace elements)
Neocate infant DHA and ARA (Nutricia)	0.67	1.9 pro eq.	12	Free amino acids	4.5	41	High oleic safflower oil, refined vegetable oil (coconut oil and/or palm kernel oil), MCT	11.7	47	Corn syrup solids	1.1	1.9	78	1.0	55	375	33% MCT oil Severe protein allergy Lactose free
Nutramigen with Enflora LGG (Mead Johnson)	0.67	1.89	11	Casein hydrolysate, L-cystine, L-tyrosine, L-tryptophan, taurine, L-carnitine	3.6	48	Palm olein, soy, coconut, and high oleic sunflower oils, DHA, ARA	7	41	Corn syrup solids, modified corn starch	1.39	1.89	64	1.22	43	270	Lactose free
Nutramigen AA (Mead Johnson)	0.67	1.89	11	Amino acids	3.6	48	Palm olein, soy, coconut, and high oleic sunflower oils	7	41	Corn syrup solids, tapioca starch	1.4	1.9	64	1.44	35	350	
Pregestimil 20 (Mead Johnson)	0.67	1.87	11	Casein hydrolysate, L-cystine, L-tyrosine, L-trytophan, taurine, L-carnitine	3.7	48	Liquid: MCT, soy oil, high oleic safflower oils powder: MCT, soy oil, corn oil, high oleic oils	6.8	41	Liquid: corn syrup solids, modified corn starch powder: corn syrup solids, dextrose, modified corn starch	1.4	1.9	63	1.2	35	320	Lactose free
Pregestimil 24 (Mead Johnson)	0.80	2.22	12	Casein hydrolysate L-cystine L-tyrosine L-trytophantaurine L-carnitine	4.5	48	Liquid: MCT, soy oil, high oleic safflower oils	8.1	41	Liquid: corn syrup solids, modified corn starch	1.6	2.3	75	1.4	42	320	Lactose free
PurAmino (Mead Johnson)	0.67	1.88	11	Free amino acid	3.6	48	Palm olein, soy oil and high oleic sunflower oil	6.9	41	Corn syrup solids	1.37	1.2	63	1.89	35	350	Severe protein allergy Lactose free

Similac Advance (Abbott)	0.67	1.4	8	Nonfat milk, whey protein concentrate	3.7	49	High oleic safflower oil, coconut oil, soy oil	7.3	43	Lactose, galactooligo-saccharides (GOS)	0.71	1.8	76	1.22	41	270	Prebiotic
Similac Expert Care Alimentum (Abbott)	0.67	1.86	11	Casein hydrolysate, L-cystine, L-tryptophan, L-tyrosine	3.74	48	Safflower oil, MCT oil, soy oil, DHA, ARA	6.9	41	Corn maltodex-trin, sucrose	1.29	2.04	71	1.22	51	370	33% MCT oil Lactose free
Similac Expert Care 24 with iron (Abbott)	0.80	2.19	11	Nonfat milk	4.25	49	Soy oil, coconut oil	8.5	42	Lactose	1.5	2.6	73	1.45	57	380	
Similac Expert Care Neosure (Abbott)	0.74	2.08	11	Nonfat milk, whey protein concentrate	4.1	49	Soy oil, high-oleic safflower oil, MCT oil, coconut oil	7.5	40	Lactose, corn syrup solids	1.0	2.4	70	1.2	42	250	Designed for former premature infants >2 kg or >40 weeks corrected age Additional Ca, P, vit A, vit D, folate, Zn
Similac PM 60/40 (Abbott)	0.67	1.5	9	Whey protein concentrate, sodium caseinate	3.78	50	High oleic safflower, soy, and coconut oils	6.90	41	Lactose	0.71	1.4	38	0.5	19	280	Low iron Lower in Ca, P, maintaining 2:1 ratio Lower in K
Similac Sensitive (Abbott)	0.64	1.45	9	Milk protein isolate	3.65	49	High oleic safflower, soy, and coconut oil	7.23	43	Corn maltodex-trin, sucrose, galactooligo-saccharides (GOS)	0.88	1.85	57	1.22	38	180	Lactose free
Similac Soy Isomil (Abbott)	0.67	1.65	10	Soy protein isolate, L-methionine	3.69	49	High oleic safflower oil, coconut oil, soy oil, DHA, ARA	6.97	41	Corn syrup, sucrose, fructooligo-saccharides (FOS)	1.29	1.87	71	1.22	51	200	Lactose free
Similac Special Care 20 with iron (Abbott)	0.67	2.03	12	Nonfat milk, whey protein concentrate	3.67	47	MCT oil, soy oil, coconut oil	7.0	41	Corn syrup solids, lactose	1.26	2.2	121	1.22	68	235	Designed for growing low birth weight and premature infants Additional Ca, P, vit A, vit D, folate, Zn, Ca:P ratio of 2:1 High in protein
Similac Special Care 24 with iron (Abbott)	0.81	2.43	12	Nonfat milk, whey protein concentrate	4.41	47	MCT oil, soy oil, coconut oil	8.4	41	Corn syrup solids, lactose	1.51	2.66	145	1.45	81	280	Designed for growing low birth weight and premature infants Additional Ca, P, vit A, vit D, folate, Zn, Ca:P ratio of 2:1 High in protein
Similac Special Care 24 High Protein (Abbott)	0.81	2.7	13	Nonfat milk, whey protein concentrate	4.4	47	MCT oil, soy oil, coconut oil	8.1	40	Corn syrup solids, lactose	1.52	2.7	146	1.46	81	280	
Similac Special Care 30 with iron (Abbott)	1	3	12	Nonfat milk, whey protein concentrate	6.7	57	MCT oil, soy oil, coconut oil	7.8	31	Corn syrup solids, lactose	1.9	3.4	183	1.8	101	325	

Abbreviations: ARA, arachidonic acid, DHA, docosahexaenoic acid; FOS, fructooligosaccharides; GOS, galactooligosaccharides; MCT, medium-chain triglycerides.

At the time of publication, every effort was made to ensure that all product information was accurate. The most current information can always be obtained by referring to the package label.

Source: Adapted with permission from Grunko A, Keenan E, Kinne J. Enteral nutrition. In: Sonneville K, Duggan C, editors. *Manual of Pediatric Nutrition*, 5th edition. Shelton, CT: People's Medical Publishing House USA; 2013.

TABLE III-2 Enteral Products: Non-infant (1000 mL)

Product (Manufacturer)	kcal/mL	Protein			Fat			Carbohydrate			mEq	mg	mEq	mg	mOsm/kg	General Comments
		g	% kcal	Source	g	% kcal	Source	g	% kcal	Source	Na	Ca	K	P	Water	
Boost (Nestle)	1.01	40	17	Milk protein concentrate	16	16	Canola, high oleic sunflower and corn oils	173	67	Corn syrup solids and sugar	28	1390	63	1310	610–670	85 % Free water Gluten and lactose free, low residue, and kosher *Chocolate malt flavor contains gluten
Boost Breeze (Nestle)	1.05	38	14	Whey protein isolate	0			228	86	Sugar, corn syrup	15	0	0	633	900–930	83% Free water Clear liquid, protein containing oral supplement
Boost Kid Essentials (Nestle)	1.0	30	12	Sodium and calcium caseinates, whey protein concentrates	38	34	High-oleic sunflower oil, soybean oil, MCT oil	135	54	Sugar, maltodextrin	24	1260	29	840	550–600	84% free water 20% fat as MCT oil Multiple flavors and fiber containing versions available Lactose and gluten free, low residue, kosher
Boost Kid Essentials 1.5 (Nestle)	1.5	42	11	Sodium and calcium caseinates, whey protein concentrates	75	45	High-oleic sunflower oil, soybean oil, MCT oil	165	44	Sugar, maltodextrin	30	1304	33	992	390	72% free water Fiber-containing versions available Lactose and gluten free, low residue, kosher
Boost Plus (Nestle)	1.52	59	16	Milk protein concentrate, calcium and sodium caseinates	58	34	Canola, high oleic, sunflower and corn oils	200	50	Corn syrup solids, sugar	31	1390	41	1310	720	78% free water Gluten and lactose free, kosher and low residue
EleCare Jr (Abbott)	1.0	30	15	Free L-amino acids	48	42	High oleic safflower oil, medium chain triglycerides, soy oil	109	43	Corn syrup solids	20	1096	39	822	551	100% free amino acid 33% fat as MCT oil Unflavored Gluten, lactose, fructose, galactose, milk and soy protein free
Ensure (Abbott)	1.06	37	14	Calcium caseinates, soy protein isolate, whey protein concentrate	26	22	High-oleic safflower oil, canola oil, corn oil	169	64	Corn syrup, sucrose, maltodextrin	37	1266	40	1055	590	84% free water High protein and high calcium versions available Gluten and lactose free Kosher
Ensure Fiber with FOS (Abbott)	1.06	37	14	Soy protein isolate, sodium and calcium caseinates	26	22	Corn oil, canola oil, high-oleic safflower oil	177	64	Maltodextrin, sucrose, oat fiber, soy fiber, FOS	37	1477	40	1266	500	82% free water Contains fructooligosaccharides 11.8 g fiber/L Gluten and lactose free Kosher
Ensure Plus (Abbott)	1.5	54	17	Sodium and calcium caseinates, soy protein isolates	49	29	Corn oil, canola oil, high-oleic safflower oil	211	56	Corn syrup, malto dextrin, sucrose	44	844	48	844	680	76% free water Gluten and lactose free Kosher
Glucerna (Abbott)	1.0	42	17	Sodium and calcium caseinates	54	49	High-oleic safflower oil, canola oil, soy lecithin	96	34	Corn maltodextrin, fructose, soy fiber	40	705	40	705	355	85% free water Low-carbohydrate content 14 g fiber/L Gluten and lactose free Kosher Comes in 1.2 and 1.5 kcal/mL
Hi-Cal (Abbott)	2.0	84	17	Sodium and calcium caseinates	89	40	Corn oil, soy lecithin	215	43	Corn syrup solids, corn, maltodextrin, sucrose	63	844	62	844	705	70% free water Gluten and lactose free Low residue Kosher
IsoSource HN (Nestle)	1.2	53	18	Soy protein isolate	39	29	Canola oil, MCT oil	160	53	Corn syrup	48	1200	49	1200	490	82% free water 50% fat as MCT oil Higher calorie version available (1.5) Gluten and lactose free
Jevity (Abbott)	1.06	44	17	Sodium and calcium caseinates	35	29	High-oleic safflower oil, canola oil, MCT oil, soy lecithin	154	54	Maltodextrin, corn syrup, soy fiber	40	910	40	760	300	84% free water 20% fat as MCT oil 14.4 g fiber/L Higher calorie versions available (1.2, 1.5) Gluten and lactose free Kosher
Neocate Splash (Nutricia)	1.0	25	10	Free amino acids	35	32	Canola oil, high oleic safflower oil, coconut oil	146	58	Maltodextrin, sucrose, corn syrup solids	8.5	620	24	620	820	3.4 g Gln/L 2.3g Arg/L Flavored version available
Neocate Junior (unflavored) (Nutricia)	1.0	30	13	Free amino acids	50	45	Fractionated coconut oil, MCT oil, canola oil, high oleic safflower oil	104	42	Corn syrup solids	22	790	35	540	590–700	Free amino acid Severe protein allergy Various flavors With or without prebiotics

Nepro with Carb Steady (Abbott)	1.8	70	14	Calcium, magnesium, and sodium caseinates, milk protein isolate	96	43	High-oleic safflower oil, canola oil, soy ecithin	333	43	Corn syrup, sucrose, FOS	37	1370	27	695	665	Gluten and lactose free Kosher
Nutren Junior (Nestle)	1.0	30	12	Whey protein concentrate, milk protein concentrate	49.6	44	Soybean, canola and MCT oil, soy lecithin	110	44	Maltodextrin, sugar	20	1000	33.8	800	350	Fiber-containing version available 25% MCT oil Gluten and lactose free Low residue and kosher
Nutren 1.0 (Nestle)	1.0	40	16	Calcium, potassium caseinates	38	33	Canola, MCT, corn oil and soy lecithin	127	51	Maltodextrin, sucrose	38	1200	32	840	370	Fiber-containing version available 25% MCT oil Kosher, lactose and gluten free, low residue Higher calorie versions available
Nutren 1.5 (Nestle)	1.5	60	16	Calcium, potassium caseinates	68	39	MCT, canola, corn oil, soy lecithin	169	45	Maltodextrin	51	1000	48	1000	430–520	50 % MCT oil
Nutren 2.0 (Nestle)	2.0	80	16	Calcium, potassium caseinates	104	45	MCT and canola oil, soy lecithin, corn oil	196	39	Corn syrup solids, maltodextrin, sucrose	57	1340	49	1340	720	75% of fat as MCT oil Contains taurine, carnitine Kosher, lactose and gluten free, low residue
Nutrihep (Nestle)	1.5	40	11	L-amino acids, whey protein concentrate	21	12	MCT and canola oil, soy lecithin, corn oil	290	77	Maltodextrin	7	956	34	1000	790	66% fat as MCT oil High in BCAA Unflavored Kosher, lactose and gluten free, low residue
Osmolite (Abbott)	1.06	44	17	Sodium and calcium caseinates, soy protein isolate	35	29	Canola oil, MCT oil, soy lecithin, corn oil	144	54	Maltodextrin, corn syrup solids	40	760	40	760	300	84% free water 20% fat as MCT oil Unflavored High-calorie version available (1.2, 1.5 kcal/mL) Kosher, lactose and gluten free, low residue
PediaSure Enteral formula (Abbott)	1.0	30	12	Milk protein concentrate	40	35	High-oleic safflower oil, soy oil, MCT oil	133	53	Corn maltodextrin, sucrose	17	1055	34	845	335	84% free water 20% fat as MCT oil Kosher, lactose and gluten free, low residue
PediaSure with Fiber Enteral formula (Abbott)	1.0	30	12	Milk protein concentrate	38	34	High oleic safflower oil, soy oil, MCT oil	143	54	Corn, maltodextrin, sucrose, dextrose, soy fiber, scFOS	17	1055	34	844	345	85% free water 20% fat as MCT oil 13g fiber/L Kosher, lactose and gluten free
Pediasure 1.5 (Abbott)	1.5	58	16	Milk protein concentrate	67	41	High oleic safflower oil, soy oil, MCT oil	158	43	Corn maltodextrin	16	1458	42	1042	370	Lactose and gluten free Fiber-containing version
Pediasure Peptide (Abbott)	1.0	30	12	Whey protein hydrolysate, hydrolyzed caseinate	40	35	Canola and MCT oils	134	53	Corn maltodextrin	31	1055	34	844	250	
Pepdite Junior (Nutricia)	1.0	31	12	Free amino acids, hydrolyzed protein (pork, soy)	50	46	MCT, coconut oil, canola oil, safflower oil	106	42	Corn syrup solids	17	1130	35	940	430–440	44% Free amino acids 35% Fat as MCT oil Flavored version available
Peptamen (Nestle)	1.0	40	16	Enzymatically hydrolyzed whey protein	39	33	MCT oil, soybean oil, soy lecithin	127	51	Maltodextrin, corn starch	24	800	38	700	270–380	85% Free water Flavored version available 70% of fat from MCT Also in 1.5 kcal/mL
Peptamen Junior (Nestle)	1.0	30	12	Enzymatically hydrolyzed whey protein	38	33	MCT oil, canola oil, soybean oil	138	55	Maltodextrin, corn starch	20	1120	34	840	260–400	85% Free water Flavored version available
Peptamen Junior 1.5 (Nestle)	1.5	45	12	Enzymatically hydrolyzed whey protein	68	33	MCT oil, canola oil, soybean oil	180	55	Maltodextrin, corn starch	30	1652	51	1352	450	77% Free water 60% MCT 5.4 g/L PREBIO
Peptamen 1.5 (Nestle)	1.5	67.6	18	Enzymatically hydrolyzed whey protein	56	33	MCT oil, soybean oil	188	49	Maltodextrin, corn starch	44	1000	48	1000	550	77% Free water Flavored version available
Promote (Abbott)	1.0	62.5	25	Sodium caseinate, soy protein isolate	26	23	Safflower oil, MCT oil, soy oil and soy lecithin	130	52	Corn maltodextrin, sucrose	43	1200	51	1200	340	84% Free water 19% Fat as MCT oil Fiber-containing version available Kosher, lactose and gluten free, low residue
Pulmocare (Abbott)	1.5	63	17	Sodium and calcium caseinates	93	55	Canola oil, MCT oil, corn oil, high-oleic safflower oil, soy lecithin	106	28	Sucrose, corn maltodextrin	57	1060	50	1060	475	79% Free water 20% fat as MCT oil Low-carbohydrate formula Kosher, lactose and gluten free, low residue

Suplena (Abbott)	1.8	30	6	Milk protein isolates, sodium caseinate	96	43	High-oleic safflower oil, canola oil, soy lecithin	196	51	Maltodextrin, sucrose	35	1055	29	717	780	Low protein formula Kosher, lactose and gluten free, low residue Low in phosphorous, K, Ca, Na
Tolerex (Nestle)	1.0	21	8	Free amino acids	1.5	1.0	Safflower oil	230	91	Maltodextrin, modified food starch	22	550	30	551	550	84% Free water Free amino acid, high carbohydrate, low-fat formula Flavor packets available Lactose and gluten free, low residue
TwoCal HN (Abbott)	2.0	84	17	Sodium and calcium caseinates	90	40	High-oleic safflower oil corn oil, MCT oil, canola oil, soy lecithin	218	43	Corn syrup solids, maltodextrin, sucrose, FOS	63	1050	62	1050	725	70% Free water 19% of fat as MCT Multiple flavors available Kosher, lactose and gluten free, low residue, 5 g/L scFOS
Vital HN (Abbott)	1.0	42	17	Partially hydrolyzed protein blend (soy, collagen), whey protein concentrate, whey protein hydrolysate	11	9	Safflower oil, MCT oil	185	74	Corn maltodextrin, sucrose	25	667	36	667	500	Hydrolyzed protein, peptide based, elemental, high carbohydrate, low-fat formula 45% fat as MCT oil Gluten free, low residue
Vivonex TEN (Nestle)	1.0	38	15	Free amino acids	3	3	Safflower oil	210	82	Maltodextrin, modified corn starch	26	500	24	500	630	83% Free water Free amino acids High carbohydrate, low fat formula High protein version available Flavor packets available Lactose free
Vivonex Pediatric (Nestle)	0.8	24	12	Free amino acids	24	25	MCT oil, soybean oil	130	63	Maltodextrin, modified cornstarch	17	1120	31	840	360	89% Free water 68% fat as MCT oil Flavor packets available

Abbreviations: BCAA, branched-chain amino acid; FOS, fructooligosaccharides; MCT, medium-chain triglycerides; scFOS, short-chain fructooligosaccharides.

At the time of publication, every effort was made to ensure that all product information was accurate. The most current information can always be obtained by referring to the package label.

Websites and phone numbers:

Abbott Nutrition: http://abbottnutrition.com or 1-800-227-5767

Mead Johnson: http://www.meadjohnson.com or 1-847-832-2420

Nestle (Gerber): http://www.gerber.com or 1-800-284-9488

Nestle HealthCare Nutrition http://www.nestle-nutrition.com or 1-800-422-2752

Nutricia: http://www.nutricia-na.com or 1-800-365-7354

Source: Adapted with permission from Grunko A, Keenan E, Kinne J. Enteral nutrition. In: Sonneville K, Duggan C, editors. *Manual of Pediatric Nutrition*, 5th edition. Shelton, CT: People's Medical Publishing House USA; 2013.

TABLE III-3 Milk-Based Oral Supplements

Product	Manufacturer	Form	Serving Size (oz.)	kcal/ serving	Protein (g/serving)	Carbohydrate (g/serving)	Fat (g/ serving)
Boost High Protein powder[a]	Mead Johnson	Powder	8	300	14	43	8.5
Carnation Breakfast Essentials[a]	Nestle	Powder	9	280	13	39	8.5
Scandi-Shake[a]	Scandi-Pharm	Powder	9	600	13	69	29
Scandi-Shake Sugar Free	Scandi-Pharm	Powder	9	600	15	67	29
Scandi-Shake Lactose Free[b]	Scandi-Pharm	Powder	9	520	17	59	25

[a] Prepared with 8 oz. whole milk.

[b] Prepared with 8 oz. soymilk.

Source: Adapted with permission from Grunko A, Keenan E, Kinne J. Enteral nutrition. In: Sonneville K, Duggan C, editors. *Manual of Pediatric Nutrition*, 5th edition. Shelton, CT: People's Medical Publishing House USA, 2013.

TABLE III-4 Selected Modular Additives Used in Pediatrics

Product (Manufacturer)	kcal/g or kcal/mL	g/tbsp or mL/tbsp	Energy Source	Comments
Benecalorie (Nestle)	7.3	330 kcal/1.5 oz. serving	Calcium caseinate, high oleic sunflower oil, mono- and diglycerides	High calorie liquid 9% protein (7g/1.5 oz. serving), 91% fat (33g/1.5 oz. serving)
Beneprotein (Nestle)	3.5	1 scoop = 1 ½ tbsp = 7g, 6 g pro/scoop	Whey protein isolate, soy lecithin	Protein powder used to increase protein in formulas/foods 25 kcal/scoop
Canola oil	8.0	15	Canola	
Corn Oil	8.4	15	Corn	Nonemulsified
Cornstarch	3.8	8		30 kcal/tbsp
Corn Syrup (Karo)	3.8	15		60 kcal/tbsp
Duocal (Nutricia)	4.92	8.5	Hydrolyzed cornstarch, blend of refined vegetable oils (corn, coconut), MCT, mono- and diglycerides	59% carbohydrate, 41% fat (35% MCT, 65% LCT) 310 mOsm/kg water Carbohydrate and fat source used to increase caloric density of formula/foods 42 kcal/tbsp
MCT oil	7.7	15	Lipid fraction of coconut oil	Bile salts and lipase not necessary for digestion and absorption Does not contain essential fatty acids Nonemulsified 115 kcal/tbsp
Microlipid (Novartis)	4.5	15	Safflower oil	High in linoleic acid 50% fat emulsion that mixes easily and stays mixed with enteral formulas, beverages, and table food 68 kcal/tbsp
NutriSOURCE Fiber (Nestle)	4	3g fiber per 2 teaspoons, 2 tsp = 3.8g	Partially hydrolyzed guar gum	100% carbohydrate 15 kcal/tbsp
Promod liquid (Abbot)	100kcal/1 oz. 3.3 kcal/mL	10 g protein/ 1 oz.	Glycerine, hydrolyzed beef collagen	Fruit punch-flavored protein liquid that increased protein in formula/foods/beverages 60% carbohydrate, 40% protein
Safflower Oil	8.4	15	Safflower	Nonemulsified
Scandical (Aptalis Pharma)	5.4	6.5	Nonfat milk, corn syrup solids, maltodextrin, vegetable oil, MCT, coconut oil, soy oil	Tasteless powder used to increase calories in hot or cold foods 35 kcal/tbsp

Abbreviations: LCT, long-chain triglycerides; MCT, medium-chain triglycerides.
At the time of publication, every effort was made to ensure that all product information was accurate. The most current information can always be obtained by referring to the package label.

TABLE III-5 Enhancing the Caloric Density of Pumped Breast milk

	Quantity of Additive Needed	Total Volume

kcal/oz. desired by additive	of Breast milk[a] 25 mL	50 mL	100 mL
2 HMF	¼ tsp[b]	1 pkg	2 pkgs
4 HMF	1 pkg	2 pkgs	4 pkgs
2 Infant formula powder	1/8 tsp	¼ tsp	½ tsp
4 Infant formula powder	¼ tsp	½ tsp	1 tsp
6 Infant formula powder	½ tsp	¾ tsp	1 ½ tsp
2 MCT oil	0.2 mL	0.4 mL	0.9 mL
4 MCT oil	0.4 mL	0.9 mL	1.7 mL
2 Corn oil	0.2 mL	0.4 mL	0.8 mL
4 Corn oil	0.4 mL	0.8 mL	1.6 mL
1 Beneprotein powder	1/8 tsp	¼ tsp	½ tsp
2 Beneprotein powder	¼ tsp	½ tsp	1 tsp

Abbreviations: HMF, human milk fortifier; MCT, medium-chain triglycerides.

[a] For calories other than HMF, ingredients should be placed in container and breast milk added to make final volume (to account for displacement).

[b] Level measurements, not heaping, for all teaspoon measurements.

TABLE III-6 Selected Metabolic Disorders for Which Nutritional Therapy is Used

Disorder	Nutritional Therapy	Enteral Products Used
β-Ketothiolase deficiency	Low protein High carbohydrate	Ketonex-1[a]
Biotinidase deficiency	Biotin supplementation	
Galactosemia	Galactose restriction ± calcium supplementation	Soy based (Enfamil ProSobee,[b] Similac Soy Isomil[a])
Glutaricaciduria type 1	Lysine and tryptophan restriction Riboflavin supplementation Carnitine supplementation	XLys XTrp Analog[c] XLys XTrp Maxamaid[c] Glutarex-1,[a] Glutarex-2,[a] GA[b]
Glycogen storage disease types I and III	High carbohydrate (50%–60% of kcal) Low fat (15%–30%) Protein (5%–10% of kcal) Regular feeding regimen	Cornstarch

	Low cholesterol	
Hereditary fructose intolerance	Fructose, sorbitol restriction Vitamin C supplementation	Fructose, sorbitol, sucrose free
Homocystinuria (cystathionine β-synthase deficiency)	Methionine restriction Cystine supplementation ± vitamin B6 supplementation ± betaine supplementation ± folate supplementation	Milupa HOM 2[d], XMET Analog[c] XMET Maxamaid,[c] HCY 1,[b] HCY 2[b] Hominex-1,[a] Hominex-2[a]
Isovaleric acidemia	Leucine restriction Glycine supplementation ± carnitine supplementation	XLeu Analog,[c] XLeu Maxamaid[c] I-Valex-1,[a] I-Valex-2[a]
Long-chain acyl-CoA dehydrogenase deficiency	Low fat MCT acceptable Avoid prolonged fasting ± carnitine supplementation	Portagen[b] ProViMin[a] (add fat and carbohydrate) Tolerex, Vivonex[e] as overnight glucose source
Long-chain hydroxy acyl-CoA dehydrogenase deficiency	Low fat MCT acceptable Avoid prolonged fasting ± carnitine supplementation	ProViMin[a] (add fat and carbohydrate) Tolerex[e], Vivonex[e] as overnight glucose source
Maple syrup urine disease	Valine, isoleucine, and leucine restriction Thiamine supplementation	MSUD Analog[c], MSUD Maxamaid[c] Milupa MSUD 1,[d] MSUD 2[d] BCAD 1[b], BCAD 2[b], Ketonex-1[a]
Medium-chain acyl-CoA dehydrogenase deficiency	Low fat Avoid MCT ± carnitine supplementation Avoid prolonged fasting	
Methylmalonicacidemia	Methionine, valine, threonine, and isoleucine restriction ± vitamin B12 supplementation	XMTVI Analog,[c] XMTVI Maxamaid[c] OA 1,[b] OA 2[b], Milupa OS 2[d] Propimex-1,[a] Propimex-2[a]
Multiple acyl-CoA dehydrogenase deficiency (glutaricaciduria type II)	Fat restriction ± protein restriction Riboflavin supplementation Carnitine supplementation	
Phenylketonuria	Phenylalanine restriction	

	Tyrosine supplementation PKU 3,[d] Phenex-1,[a] Phenex-2[a] ±tyrosine supplementation	XPhe Analog,[c] XPhe Maxamaid[c] Phenyl-Free,[b] Milupa PKU-1[d], PKU 2,[d]
Propionic acidemia	Valine, isoleucine, methionine, and threonine restriction Carnitine supplementation ± biotin supplementation	XMTVI Analog,[c] XMTVI Maxamaid[c] OA 1,[b] OA 2[b], Milupa OS 2[d] Propimex-1,[a] Propimex-2[a]
Pyruvate dehydrogenase deficiency	High fat (>50% of kcal) Low carbohydrate (20%–30% of kcal) Thiamin supplementation	3232A[b] RCF[a]
Tyrosinemia	Phenylalanine and tyrosine restriction Regular feeding regimen	XPhe XTyr Analog,[c] XPTM Analog[c] XPhe XTyr Analog[c] XPhe XTyr Maxamaid[c], tyr 2[d] Tyros 1,[b] Tyros 2[b] Tyrex-1,[a] Tyrex-2[a]
Urea cycle disorders	Protein restriction Essential amino acid supplementation Arginine or citrulline supplementation Sodium benzoate and sodium phenylacetate (or sodium phenylbutyrate)	Cyclinex-1,[a] Cyclinex-2[a], Milupa UCD 2[d]

Abbreviation: MCT, medium-chain triglycerides.

[a] Abbott Nutrition, Columbus, Ohio.

[b] Mead Johnson Nutrition, Evansville, Indiana.

[c] Nutricia North America, Gaithersburg, Maryland.

[d] Milupa North America, Clarksburg, Maryland.

[e] Nestle HealthCare Nutrition, Florham Park, New Jersey.

Source: Adapted from Yannicelli S, Camp K. Appendix C: Metabolic medical foods for nutrition therapy of inherited metabolic diseases. In: Baker SS, Baker RD, Davis AM, editors. *Pediatric Nutrition Support*. Sudbury, MA: Jones and Bartlett; 2007. 570–579.

INDEX

Note: Page numbers followed by i or t indicate material in images or tables, respectively.

A

Absorptiometry, dual energy X-ray 12–13, 42
Absorption (*see also* Bioavailability; Digestion)
  development of function in gut 389–381
  gastrointestinal, in HIV 675–676
  laxatives affecting 308
  medications affecting 301–303
  of drugs, in malnutrition 298
  of fats 83–84
  of nutrients, secondary factors in 307
  of trace elements 101, 105, 109, 112, 120, 124
Abuse and neglect
  growth failure secondary to 713, 718t
  starvation as 713
Acanthosis nigrans in axillary skin 620t
Acceptable macronutrient intake level 163, 164
Acetylation, histone, and bioactive food components 355–356
Acidosis, lactic (*see* Lactic acidosis)
Acquired Immune Deficiency Syndrome (*see* HIV *entries*)
Acrodermatitis enteropathica 98
Adaptation, intestinal (*see* Intestinal adaptation)
Adaptive immunity
  breast milk effects on 512
  linked to innate immunity in neonatal gut 505
Adenoma in glycogen storage disease 762
Adenosine triphosphate (ATP)
  chromatin remodeling dependent on 356–357
Adequate intake (AI)
  in dietary reference intakes 164
  of carbohydrates in term and preterm infants 65
Adipose tissue in insulin resistance and MetS 621–622
Adiposity rebound 44 and rapid catch-up growth 691–692
Adolescence

and bone health 1007–22
as critical time for breast carcinogenesis 871–872
as time of bone accretion 1008
changes in bone growth during 47
Adolescent athletes, nutrition in 1091–98
dietary counseling for 1096–97
energy balance in 1095–96
fluids and electrolyte needs in 1094–95
macro- and micronutrients in 1092–94
Adolescent growth, and breast cancer 872–873
Adolescent nutrition
disturbances in, and bone health 1012–1013
U.S. nutritional recommendations for 166, 170, 251–253
Adolescents
diet of (*see* Diet, adolescent)
eating disorders in 1001–06
metabolic syndrome in 613–624
need for iron in 118, 127
with hyperlipidemia and cardiovascular disease 971–976
Adult disease, biological basis of development of 436
Age
and changes in body composition 43i, 46–47, 49i
as factor in measuring RMR and FFM 185i
microbial composition relative to 58i
muscle and organic metabolic rates relative to BMR 186t
Agenesis, pancreatic 839
Aging, impact of DNA methylation on 352–353
Agricultural transformation and child nutrition 649–660
Aids, mechanical, in supplementing lactation 531–532
AIDS (*see* HIV *entries*)
Air displacement plethysmography 13
in infants 44
Albumin 28
Allergens, food 825
labeling regulations 1100
Allergic disease(s)
eosinophilic gastroenteritis as 827
food 825–828
management of 830
novel therapies for 830–831
special diets for 1099–46
potential for probiotics in 175
proctocolitis as 827–828
Allergy

and duration of exclusive breastfeeding 577–578
and failure to grow 718
food (*see also* Food allergies) 823–833
support groups and resources for 1140
to fish 1107
Alpha-tocopherol (*see also* Vitamin E)
laboratory assessment of 32
Amino acid(s)
classified by FAO/WHO/UNU 73t
energy substrates and 196
enzyme disorders of 745t
in physiologic and metabolic function 74t
neonatal implications of supply of 477
nutritional classification of 73t
overview of 71–81
RDA for, relative to age and sex 77–78t
structural formula of 71i, 72–73t
systemic availability in neonatal pigs 80i
umbilical uptake of 472
Amino acid composition of parenteral formulas 1043t
Amino acid metabolism
fetoplacental 471–472
of fetal hindlimb 475
hepatic circulation and, fetal 474–475
Amino acid nutrition, placental function and 465–482
Amino acid profiles 28
Amino acid pools exchanged with body protein 75i
Amino acid requirements
current 80
methods for determining 79–80
Amino acid scoring pattern by age 79t
Amino acid transport
abnormal pregnancy outcome and 475–477
during pregnancy 470–474
placental 466–472
protein chemistry in 467–470
Ammonia metabolism in hepatocytes, enzymes in 751i
Anabolic steroids (*see* Steroids, anabolic)
Anaphylaxis, food-induced 825–826
Anatomy and physiology
in developing fetal and infant gut 387–389
in lactation 517–520
Anemia (*see also* Iron deficiency) 127–128
in glycogen storage disease 762

in pregnancy 425–426
in severe acute malnutrition 211, 221
pernicious, vitamin B_{12} in 157–158
Angioedema resulting from food hypersensitivity 828
Anionic transport system, light-chain placental proteins and 470
Anorexia and cancer treatment 881–882
Anorexia, nervosa (*see also* Eating disorders) 1001–06
and bone status 1014
Anthropometric data on child growth and economic development 654–657
Anthropometric techniques in field studies 40
Anthropometric *z*-scores for age and growth 635i
Anthropometrics
and body composition 7–11
body-mass index in 10–11
correlated with total body densitometry 276–277t
genetic growth potential in 10
ideal body weight percent 11
in children undergoing cancer therapy 882–883
in developmentally disabled children 731–737
in liver disease 854t
percent weight loss in 11
percentiles for age and sex in 10
triceps skinfold thickness and 12
WHO recommended measures for 707
Antibiotics, use of, for diarrhea 918–919
Antidepressant medication during lactation 498
Antihelmintic drugs in preventing morbidity 669
Antimalarial drugs in preventing morbidity 668–669
Antimicrobial activities of oligosaccharides and glycoconjugates 507–508
Antimicrobial proteins and peptides 508
Antioxidants
in adolescent athletes 1093
in HIV 680
Appetite
drugs affecting 316–319
enhancing and suppressing 319
drugs stimulating 683–684
Arm
as proxy for body composition 883
length of, by age 735i
measurement techniques and reference standards for 733–734
mid-upper, circumference of 11, 735
Arterial hypertension in MetS 616
Ascorbic acid (*see also* Vitamin C) 151–152

Aspiration, signs of, in failure to grow 716t
Assessment of nutritional status and body composition
  bioelectric impedance assessing lean body mass 27
  of iron status 134–137
Asthma, comorbid with food allergies 826–827
Atherosclerosis during childhood 971–973
  and lipoprotein level 973
Athletes, young, nutrition in 1091–98
Athletics, strenuous, in female adolescents 1004–05, 1014
Atopic dermatitis 828
Avoidant Restrictive Food Intake Disorder (ARFID) 1001

B

Bacteremia, catheter-related, in IF/SBS 938
Bacteria
  in presence of breast milk
  cellular reactivity to 510–511
  interacting with PPRs 509–510
  intestinal
  nutrient effects on 343
  relative to age 58i
  barrier function, in GI tract 391
Bacterial colonization of the human gut 503–515
Barrier functions, intestinal, development of 391, 505–506
Basal metabolism rate (BMR) (*see also* Metabolism)
  difficulties in measuring 183–187
  related to age and muscle and organic metabolic rates 186t
BCG vaccination rates and real income 659
Behavior, feeding, details of 719t
Behavioral assessment and therapy for child obesity 605–608
Behavioral disorders and feeding problems 702–703
Behavioral feeding problems in liver disease 858
Benign breast disease and adolescent diet 874
Beri-beri 152–155
Beverages, effect of, on drug bioavailability 306
Bifidobacteria, importance of 58
Biliary complications of parenteral nutrition 1065, 1068–69
Binding interactions between feedings and drugs 297
Binge eating disorder (*see also* Eating disorders) 1001–06
Bioactive food components
  DNA methylation and 354
  histone acetylation and 355–356
  possible role of microRNAs in 357–358
Bioactivity, IGH, in malnutrition 373–375

Bioavailability
of drugs, beverage types affecting 306
of iron 119–121
of trace elements 96–97
Biochemical abnormalities in HIV-infected children 678t
Biochemical assessment, variations in factors affecting 26
Biochemical composition of human milk 486–498
Biochemical correlates of trace element deficiency 99, 103, 109
Biochemical indices for measuring iron status 136–137t
Biochemical assessment of iron status 134–137
Biochemical markers of dietary intake 279–281
Bioelectric impedance analysis (BIA) in measuring body water 41
Biofortification in food crops 649
Biotin 158–159
in human milk 496
Biotinylation, histone 356
Birth weight
and later adult cancer 868–870
and risk of breast cancer in premenopausal women 869t
breastfeeding and, in developing economies 690–692
relative to later-life disease 435–443 (*passim*)
pediatric cancer and 866–867
Bisphosphonates, potential benefits and risks of 1917–18
Blood, GH and IGF in 369–370
Blood coagulation cascade, vitamin K in 150
Blood pressure in boys and girls, standard values of 617–619t
Blood stream infections, catheter-related 1069–71
Body cell mass (BCM), estimation of 38–39
Body composition
assessment of 37–54, 275–278
air displacement plethysmography assessment 13
chemical stability in 48–50
developmental changes in, and nutrition 294
effect of celiac disease on 813–815
ethnic and racial differences in 45
in cancer patients 883
in IBD 790, 801–802
in developmentally disabled children 736
metabolic reserves and 963
of HIV-infected children 675–676
of reference infant 567i
Body composition changes
by sex 44t
during growth 43–51, 44t

during malnutrition and disease 50
with age 43i, 44t
Body composition differences at NICU discharge 554–555
Body composition measurement 11, 37, 275–278
air displacement plethysmography in 13
and growth 37–34
characteristics of 278–279
dual-energy X-ray absorptiometry 12
mid-upper arm circumference 11
multicompartment models in 37–40
neutron activation analysis in 38–39
sexual and skeletal maturation in 13
terms used in study of 37t
triceps skinfold thickness in 12
Body composition multi-compartment models 37–40, 38i
atomic 38
cellular 39
equations and methods for determining 38–39
functional 39
molecular 38–39
whole-body 38–40
Body fat
and sports activities 45
as percentage of body composition 44t
changes with growth 43–45
distribution, ethnic and racial differences in 45
Body mass, lean, estimation of 27
Body mass index (BMI) 11
and cardiovascular risk in obesity 622i
correlated with skinfold thickness 278t
in obese children 603
in rural-migrant Indian population 694i
Body temperature
and energy expenditure 189, 194
Body water measurement, methods for 41–43
Body weight 38–39
during first days of life 537–538
percent, ideal, cautions concerning use in assessment 11
in HIV-infected children 677–678
Bolivia, prevention of malnutrition in 237–238
Bone complications in children receiving PN 1065, 1069
Bone density assessment in developmentally disabled children 736–737
Bone diseases 1013–15
in patients with intestinal failure 938

management of 1017–18
novel therapies for 1017–18
prevention of 1018
related to cyctic fibrosis 844–845
Bone health
assessment of 12, 13, 1009–22
exercise and 1009
maximizing 1008–12
nutritional influences on 1009–13
physiology and development of 1007–08
Bone mineral density (BMD), femoral 738i
Bone minerals, equations and methods for measuring 38–40t
Bones in severe acute malnutrition 212
Bowel disease, inflammatory (*See* Inflammatory bowel disease)
Brain development in SAM 212
Brain tumors and *N*-nitroso compounds 868
Breast(s), female
from infancy to lactation 518i
preparation of, for lactation 522–523
Breast cancer
diet and 866–874
carcinogenesis in adolescence and 871–872
risk of, and red meat and soy consumption 873–874
Breast disease, benign 874
Breast-fed infants
allergens affecting 1112
assessment of dietary intake in 274–275
compared to formula-fed infants 568–569
daycare for 529
growth in 484–485
nutritional adequacy in 577
revised estimate of protein intake in 74t
Breastfeeding (*see also* Lactation)
after NICU discharge 561–562
and complementary feeding, preventing malnutrition 232, 644–646
approach to 517–536
avoiding transmission of HIV in 498
discontinuance of 534
education about, desirability of 545
enhancing microbiota in infants 174, 176
exclusive
and real income 658i, 659
duration of 575–576
in low- and mid-income countries 642

strategies to promote 642
WHO recommendations for 666
failure to thrive despite 529–534
in severe acute malnutrition 220–221
initiation of, in low income countries 641
policy recommendations for 484
prenatal considerations in 521
return of interest in 565–566
return to work during 528
stages of 527–529
supplementation during 525
trends in 484
Breast-feeding women
DHA supplied to 89–90
Breast milk (*see also* Human milk)
as innate protection for neonates 391
and gene expression 338–339
in low- and mid-income countries 642–643
Breast pumps and storage of milk 528–529
Brown adipose tissue (BAT), stimulation of 189
Bulimia nervosa (*see also* Eating disorders) 1001–06

C

Cachexia
in cancer patients 878
in HIV-infected children 877–878
Calcium
and bone health 1011–12
dietary sources of 1018t, 1130, 1132–33
during peak bone growth 47
in human milk 497
in vegetarian diets 1137
in young athletes 1093
lactose-free sources of 1111t
RDA for 1012t
requirements during pregnancy 427–428
Calculations of REE/BMR to measure against critically ill children 1039t
Caloric deficiency in IBD 790
Calorie counts in assessing dietary intake 6, 7
Calorimetry, indirect, and REE 15
Canada
community nutrition in 244, 247, 248t (*see also* Community nutrition)
existing programs in 262, 263t, 264t
current nutrition guidelines in 265t

nutritional needs of special-needs children in 255, 256t
nutritional status of infants and youth in 255–256, 254t
resources for 265t, 266t
Cancer (*see also* specific type)
adolescent diet in 872–873
body composition measures in 883
connected to growth 865–866
in-utero diet/exposure and 866–868
laboratory evaluation in patients with 883–884
later-adult, and previous breastfeeding 870–871
malnutrition in 877–878
metabolic effects of 878–879
in treatment 879–881
nutritional assessment in 882–884
nutritional intervention in 884–886
pediatric, treatment of 877–889
Cancer prevention in childhood 865–876
Cancer survivors, nutritional concerns of
Cancer treatment in childhood 877–889, 879–881
food aversion in 881–882
side effects of 879–881
Carbohydrate balance in obesity 591–593
Carbohydrate component of human milk 486–488
Carbohydrate counting in diabetes 904–905
Carbohydrate disorders 66–68
Carbohydrate intake, recommendations for 64–66
Carbohydrate intolerance 1117–19
Carbohydrate metabolism
enzyme disorders of 745t
in critically ill children 961
in the fetus 445–456
in liver disease 851–852
Carbohydrate requirements in parenteral nutrition 149–151
Carbohydrate substrates, alternative, for fetal oxidation 451
Carbohydrates (CHO) 55–69
and sources, classification of 57t
as sources of glucose for placenta and fetus 446–456
composition of, during childhood 64
dietary, in diabetes mellitus 897–898
digestible, recommendations based on 64–65, 66t
digestion and absorption of 390
impact of, on drug kinetics 303
in cancer 878
in formula 570

in GI tract, overview of 55–69
in intestinal microbiota 56–57
in young athetes 1092
local and systemic effects of 57i
malabsorption of 66–67
recommended dietary intake of 64–66
required in preterm infants 543
Carbonated beverages and adolescent bone health 1013
Cardiac system in SAM 210
Cardiometabolic health, in post-discharge preterm infants 557–558
Cardiovascular disease
cholesterol-lowering diet in 1128–1130
in children and adolescents 971–986
Cardiovascular risk factors in obesity 622i
Carnitine
deficiency in 30
in formula 571
supplementation in lipid-based parenteral formulas 1048
Carotenoids (*see also* Vitamin A)
in human milk 493
Case-control studies of vitamin supplements in NTDs 286–287
Casein in human milk 491–492
Catabolic diseases
anabolic steroids in 379
of liver 751–752
Catabolic responses in critically ill children 957
Catch-up growth 364–366
nutritional needs for 720
rapid, and adiposity rebound 691–692
Catheter(s) in parenteral nutrition 1062–64
Catheter line infection in IF/SBS 938
Catheter-related blood stream infections (CRBSIs) 1069–71
prophylaxis of 1070–71
Catheter-related occlusion and thrombosis 1071–72
prophylaxis of 1071–1072
Caveats in use of nutraceuticals 178–180
Celiac disease 809–822
and bone disorders 1015
and duration of exclusive breastfeeding 577–578
and type 1 diabetes mellitus 905, 1117
catch-up growth in 365
classic presentation of 815i, 816i
clinical presentation of 811–812
diagnosis of 812–813

diet and 271–272
diets for 1115–17
effects on growth and body composition of 813–815
extra-intestinal manifestations of 816
natural history of 816–817
nutritional deficiencies in 815–816
prevalence and pathogenesis of 809–811
probiotics in 175
reaction to gluten protein in 828
treatment of 817
web information on 1117
Cell growth and nutrition 362
Cell signaling from intestinal lumen to mucosal immune system 339–345
Cellular reactivity to bacteria in presence of breast milk 510–511
Cellular regulation of growth, hormones in 362t
Cereal products as complementary foods 648
Cerebral palsy
and feeding difficulties 701–702
nutrition and feeding in 729–731
Chamomile 179
Chelation in sickle cell disease 134
Chemical analyses (*see also specific element*)
in vivo, for measuring body composition 42
Chemical composition
of body 38
of carbohydrates 48–50
Chemical stability of body weight 50–52
Chemokines and intestinal immune system 340–341
Chemotherapy, side effects of 879–881
Child abuse and neglect (*see also* Abuse)
Child development
conceptual, relative to malnutrition 230i
early, affecting later-life diseases 435–437
linked with nutrition and economics 653–661
Child health, global
and the nutrition transition 631–640
interventions for 663–672
Child Health Days, aiding at-risk children 670
Child nutrition (*see also* Nutrition, child)
and cancer prevention 865–876
and economic development 653–661
structural changes affecting 659–660
Childhood illnesses, integrated management of 669–670
Childhood obesity

behavioral assessment and therapy for 605–608
causes of 692–694
co-morbidities in 604
epidemiology and pathophysiology of 599–602
evaluation and management of 599–612
in developing economies 689–697
nutritional assessment of 604–605
prevention of 625–630
gaps in evidence on 628–629
recent trials of 628t
risk factors for 625–626
treatment of 606–610
weight-loss drugs and surgery for 608–609
Children
at risk for cardiovascular disease 972
at-risk, early interventions for 261–262
critically ill 957–970
injured, protein requirements for 959t
school-age, in U.S., nutritional recommendations for 250–252, 252t
with special needs (*see* Special-needs children)
Chile
interventions to prevent obesity in 694–695
prevention of malnutrition in 237–238
Chinese herbal medicine during pregnancy 430–431
Chloride in human milk 497
Cholera, treatment of 920
Cholestasis
and hepatobiliary disease 749
in intestinal failure 934–938
Cholesterol, and nutrition 977
Cholesterol-lowering diet in cardiovascular disease 1128–30
Cholesterol metabolism and transfer in placenta 467–458
Cholesterol screening, universal vs. selective 976–977
Choline requirements in parenteral nutrition 1057
Chromatin remodeling, ATP-dependent 356–357
Chromium 108–110
possible toxicity in parenteral solutions 1061
Cirrhosis
North American Indian 751
Clostridium difficile infections in diarrheal diseases 918
Cobalamin (*see also* Vitamin B_{12}) 157–158
Coenzyme A, conversion from pantothenic acid 159i
Coenzyme Q10 in human milk 494
Colitis

Crohn's, enteral nutrition in 796
ulcerative, nutritional therapy for 792–793
Colonization, neonatal intestinal 503–515
immune education in 509–512
Colostrum
energy composition of 487t
fatty acids in 489–490
Community nutrition
defined 245
effectiveness of 670
for malnutrition 213, 235–236
in Canada 244, 247, 248t, 253–254. 255, 256t, 263t, 264t, 266t
in industrialized countries 243–269
U.S. governmental programs for 243–262
web-based resources for 262–265
Complementary feeding
and content of diet 578–579
and health outcomes 575–582
during weaning 571–572
in low-income countries 644–646
interventions in practice of 649–650, 666–667
timing of introduction to 644i
Complementary foods
energy and density requirements of 644–645
nutrient requirements of 646–647
processed 648–649
safety in preparation of 649
Congenital disorders of glycosylation 747t
Congenital enteropeptidase deficiency 839
Congenital heart disease
and feeding difficulties 702
and hyperlipidemia, dietary restrictions in 1127–28
Congenital lipase/colipase deficiency 839
Contaminants, environmental, in human milk 498
Cooking methods influencing dietary metabolism 305
Copper
in human milk 497
in parenteral solutions, cautions about 1061
Copper deficiency 102–105
Copper metabolism, diseases involving 751
Cori disease 764–765
Coronary heart disease (CHD),
later-life mortality from related to birth weight 435i, 437–438
Cortisol and sex steroids 378–379

Costs of food and nutritional assistance in U.S. (FY2010) 257t
Counseling, dietary, for young athletes 1097–98
Cow’s milk
  and immune response in diabetes 894
  as percentage of infants fed 567
  as source of iron 579
  substitutions for 1112
  undesirability of, in early infancy 572
Cow’s milk allergens in breast milk 827–828, 1112
Cow’s milk allergy, challenges to 1115
Cow’s milk–free infant formulas 1112t
Cow’s milk protein-free diet 1110–15
Creatinine, urinary, assessing muscle mass 27
Critically ill children 957–970
  enteral nutrition in 1029
Crohn’s disease
  and protein metabolism 193
  dietary treatment in 1119
  elemental diet in 794
  enteral nutrition in 793, 796–797, 1029
  mechanisms of 799–801
  nutrient-gene interactions in 344–345
  nutritional compromise in, key factors in 789t
  nutritional therapy in 787–788, 794–796
  polymeric diet in 795
  responding to inaction in malnutrition 207–208
Cutaneous food hypersensitivity reactions 826
Cystic fibrosis 835–837, 841–846
  and bone disorders 1015–16
  and feeding difficulties 702
  bone disease in 844
  clinical manifestations of 837t
  enteral tube feeding in 847
  malnutrition in 842–844
Cytokines
  affecting energy expenditure 184
  and intestinal immunity 340–341
  in cancer 878–879
  in HIV-infected children 676

D

Dairy intake and bone health 1013
Daycare for breastfeeding infant 529
Daycare centers and treatment for malnutrition 235–236

Debranching enzyme deficiency 764–765
Decosahexaenoic acid (DHA), benefits of 177–178
Dehydration
  degree of, and gastroenteritis 916
  dietary therapy for 917–918
  hypernatremic 918–919
  in SAM 216–217
  severe
  hospital and outpatient therapy for 917
  signs of 915t
  treatment plans for 916t
Dehydroepiandosterone (DHEA), caveats concerning 1014
Demographic transition and structural changes affecting nutrition 659–660
Densitometry
  correlated with anthropomorphic measurements 276–277t
  in estimating body water 42
Dental benefits of fluorides 994–997
Dental caries
  etiology of 989–982
  fluoride preventing 111
  polyols reducing 991–992
  treatment of lesions in 997–998
Dental disease
  and diet 987–1000
  and sugar-containing food 990–991
Dentin, structure of 988
Depression, maternal, as factor in growth failure 711
Dermatitis, atopic, as food reaction 826
Developing countries and economies
  and the nutrition transition 637–639
  obesity and overweight in 689–697
  targeting interventions to achieve health goals 665–669
Development, economic (*see* Economic development)
Developmental disabilities, monitoring growth and nutrition in 729–741
Developmental origins of later-life disease 435–437
  hypothesis for 436–437
DHA-free formula feeding 177
Diabetes 891–910
  after kidney transplant 781
  and dyslipidemia 972
  as carbohydrate disorder 67
  carbohydrates in 897–898
  celiac disease and 1117
  dietary fiber in 898

dietary sweeteners in 900–901
energy, dietary intake in 896–897
fat, dietary, in 899–900
goals of medical nutrition therapy for 895
hypoglycemia in 906–907
lab assessment of 605t
macronutrients in 897
meal planning in 902–905, 906–908
nutritional management of 896
outpatient 905
prevalence, definition, and clinical description of 891–893
protein in 898
type 1
and eating disorders 702
celiac disease and 905
dietary factors in pathogenesis of 893–894
meal plan for 902–903
type 2
in later life, birth weight and 438–439
meal plan for 903–904
metabolic syndrome and 972
Dialysis, maintenance, energy requirements for 776–777
Diarrhea
acute bloody 920
home management of 913–914
chronic, hepatic and GI dysfunction in 648t
functional foods in 919
in SAM 216–217
persistent 920–921
sanitary measures to alleviate 657–659
treatment of, relative to income 657–659
with severe malnutrition 921–922
Diarrheal diseases 911–924
clinical assessment of 914–915
historical background to 911–912
pharmacologic therapy for 918–919
Diet(s)
adolescent
and benign breast disease 874
and breast cancer 873
red meat and soy consumption, risk of breast cancer and 873–875
affecting microbial composition of gut 57
and genetics in analysis of 284–286
balanced, plate method of 983i

childhood and age at menarche 874
cholesterol-lowering 1128–30
complementary feeding and 578–579
modification and diversity in 646
cow’s milk-protein-free 1110–1115
dental disease and 987–1000
egg-free 1100–02
elemental, in IBD 794
fluid-restricted 1127–28
for childhood obesity 607
for hereditary fructose intolerance 1122, 1123–24t
for GI disease 1115
for long-chain fatty acid disorders 1122
gluten-free diet in celiac disease 817–81, 115–178
gut microbiota in metagenomics era 399–409
high-calcium 1130–1133
high-energy, in growth failure 1126–27
high-fiber 1133, 1135–37
GI absorption in 305
immunity enhancing, for critically ill children 965–966
in etiology of IBD 787–788
interacting with microbiota and intestinal immune system 174i
iron-rich 1133, 1134–35t
lactose-free 1107–10
low-fat, to prevent CHD 978–979
low-phenylalanine 1119–21
maternal, and essential fatty acid supply to fetus 459
oral, in cancer paatients, 885
peanut-free 1103, 1105–06
polymeric, in Crohn’s disease 795
quality of, and food availability 657
rice-milk 219t
shellfish-free 1106–07
soy-free 1102–03
special (*see also* Special diets) 1095, 1099–46
tree-nut-free 1106
variations in, affecting response to drugs 293–322
vegan, as result of low food availability 657
vegetarian
health benefits of 1138
in infants 579
very-low-calorie, and drug metabolism 306
very-low-fat 1125t
very-low-fructose 1132, 1123–24t

Dietary adherence in diabetes 907–908
Dietary allowance, recommended (*see also* RDA)
  for amino acids by age and gender 77–78t
Dietary analysis
  compensating for missing data in 282–283
  genetics in 284–286
Dietary changes affecting drug kinetics 301–307
Dietary counseling for young athletes 1096–97
Dietary factors
  altering class II MHC 342
  in pathogenesis of diabetes 893–894
Dietary fats, and drug kinetics 304
Dietary fiber(s) (DFs)
  food sources for 1136t
  in IBD 788
  in vegetarian diets 1137
  intake recommendations based on 65, 66t
Dietary guidelines for Americans 167–169, 899
Dietary intake(s) (*see also* Dietary reference intakes and Appendix II on pp. 1201–15 for detailed information)
  assessment methods in 272–275
  in breast-fed babies 274–275
  biochemical markers of 277–282
  driving obesity in developing countries 692–694
  history of 5–6, 274
  in breast-fed infants
  adding solid foods in 527
  assessment of 6, 275
  in children with CP 730
  in hypercholesterolemia 1128
  of energy, and food availability 657
  of fats 337
  in infants and children 85–86
  tracking 281
Dietary management
  in chronic kidney disease 774
  in SAM 218–220
Dietary nutrients
  absorption in the gut 173–175
  affecting gene expression 335i
Dietary patterns, effects of urbanization on 637–638
Dietary recommendations to reduce triglyceride and HDL levels 977–980
Dietary records for assessing nutrient intake 274t
Dietary reference intakes (DRIs)

accepted macronutrient distribution range 164
adequate intake in 164
data used to formulate 166
establishing values for 171–172
for macronutrients 167–168
in the U.S. 6, 14, 163–171
life-stage groups in pediatric population in 165–166
macronutrient recommendations 166–168
practical aspects of 168
RDAs in 164
tolerable upper limit in 164–165
Dietary regulation and enterocyte gene expression 334i, 334t
Dietary requirements
for bone health 1009–12
for iron 117–119, 128–129
for trace elements 97, 101–102, 105, 108, 110, 113
Dietary restrictions affecting drug metabolism 306
Dietary sources (*see also* Food sources)
of calcium 1018t
of folate 156
of iodine 113
of niacin 154
of riboflavin 153
of vitamin B_{12} 158
Dietary supplements (*see also specific supplement*)
fish oil, benefits of 177–178
in SAM 221
potential risk of 177–180
toxic side effects of 179–180
widespread use of 177
Dietary therapy for dehydration 917–918
Digestion (*see also* Absorption)
fatty acid
comparison of chain length in 390t
in premature infants 389–390
of fats 83–84
in exocrine pancreatic disorders 840, 846
Digestive absorptive function, development of 389–391
Disabilities, developmental (*see* Developmental disabilities)
Disaccharides 55–56
Disease(s)
adult, and early pre- and postnatal growth 635–636
and food association 271–272
changes in body composition during 50

communicable, and undernutrition 636
effects on energy expenditure during 192–198
hepatobiliary 749
involving inborn errors of metabolism 743–756
later-life
developmental hypothesis for,
origins in early nutrition and growth 435–443
role of genes and environment in 441–442
role of placenta in 440
metabolic, featuring hepatic and GI dysfunction 745–748t
occurring during economic development, treatment of 657–659
of iron metabolism 133t
protein metabolism in 192–194
renal, acute and chronic 769–786
Disease burden, global trends in 631–632
Disease prevention
by food fortification 287–288
interventions for 667–668
Disease-specific growth charts 1192–93
Disease states, GH in 372–373
DNA methylation 351–354
bioactive food components and 354
during lifetime 352–353
embryonic development and 352
folate deficiency and 353
nutritional influence on 353–354
Docasahexaenoic acid (DHA)
supplied to pregnant and breastfeeding women 89–90
Doping agents in young athletes 1094–95
DPT vaccination rates and real income 659i
Drivers of overweight and obesity in developing economies 692–694
Drug absorption in malnutrition 298
Drug bioavailability, affected by beverages 306
Drug clearance, hepatic, in malnutrition 299
Drug distribution in malnutrition 298–299
Drug kinetics
affected by nutritional status 293–327
effects of malnutrition on 298–301
effects of diet on 301–307
factors having an impact on 293–299
Drug metabolism, factors affecting 305–306
Drug-nutrient interactions 5t, 293–327
examples of 309–311t
Drug therapy and the role of nutrition 293–327

Drugs
affecting absorption 301–303
affecting nutrient transport 309–316
altering glucose metabolism 312–314
and appetite 316–319
antihelmintic and antimalarial, in preventing morbidity 668–669
causing weight gain in obese children 604t
chemotherapeutic, in cancer 880–881t
effects of, on nutrient status 309
in human milk 11, 13, 498
in hyperlipidemia 981t
in kwashiorkor 299
in marasmus 299–300
inducing fluid and electrolyte imbalance 309–312
interacting with grapefruit juice 305
interacting with vitamins 320–321t
lipid-lowering 980
rectal administration of 295–296
weight-loss 608–609
Dual energy X-ray absorptiometry (DXA) 12
in estimating body water 42
in evaluating bone mineral density 1016
Dust Bowl, nutritional problems in 2
Dysentery, treatment of 920
Dyslipidemia
after renal transplant 781
drugs associated with 315–316t
in metabolic syndrome 616
major, diagnosis of 974–976
pediatric, and CVD 971–972
treatment for 978–979

E

Eating disorders
adolescent 1001–06
clinical assessment of 1002
feeding problems with 702–704
long-term consequences of 1004–05
precursors to 721–722
refeeding syndrome in 1003
treatment of 1004
Eating patterns, healthy 169–170
Echinacea 178
Economic development and child nutrition 653–661

Ecosystem, intestinal, and microbiota 393–394
Education
  nutritional, in fighting malnutrition 234
  of mucosal immune system by factors in breast milk 509–512
  women's, improving health environment via 232
Egg-free diet 1100–02
  substitutions for egg in 1102
Eicosapentaenoic acid, benefits of 177–178
Electrolyte imbalance
  drug-induced 309–312
  in SAM 216–217
Electrolyte homeostasis in acute kidney injury 772
Electrolyte metabolism in critically ill children 962–963
Electrolyte requirements in parenteral nutrition 1051–54
  in VLBW infants 1054–55
Electrolytes
  changing during malnutrition 206
  disturbances in, in chronic kidney disease 779–780
  needed in young athletes 1094
  required in preterm and VLBW infants 544, 1051–55
Elemental diet in IBD 794
Embryonic development, DNA methylation in 352
Emergency treatment for SAM 213–222
Emotional deprivation and growth retardation 376–377
Emotional stimulation in SAM 222
Enamel
  dental, structure of 988
  mottled 992–994
Endocrine system
  and growth 361–385
  changes in, in severe acute malnutrition 208
Energy
  and substrate regulation in obesity 583–597
  dietary fiber and 1137
  resting (*see* Resting Energy expenditure)
Energy balance
  in cancer 879
  in young athletes 1095–96
Energy content
  enriching formula after NICU discharge 558
  of human milk 486
Energy deficits, after time in NICU 554–555
Energy expenditure 189
  and disease states 192–198

and malnutrition 191–192
and utilization of energy substrate 194
cost of, in protein synthesis 188
in critically ill children 960–961
in Crohn's disease 801–802
in cystic fibrosis 843–844
in disabled children 730
in growth, estimation of 187
in liver disease 853
in obesity 589–593
imbalance of 625
in parenteral nutrition 1037–41
physiologic determinants of 183–190
twenty-four-hour 190
Energy intakes
and food availability 657
in breast-fed vs. formula-fed infants 569
in cystic fibrosis 842–843
in diabetes mellitus 896–897
in disabled children 737–739
in GH-treated patients 373i
in HIV-infected children 674–675
in obesity 589, 593
in severe acute malnutrition 295–206
statistical models for assessing 283–284
total, in epidemiologic studies 283–284
energy metabolism in critically ill children 960–961
Energy regulation in obesity 588–589
Energy requirements 28–31
complementary feeding and 578–579, 644–645
estimated average 592i
in acute kidney injury 779–771
in developmentally delayed children 711t
in chronic kidney disease 776–777
in health and disease 183–200
in parenteral nutrition 1037–41
in preterm and VLBW infants 1040–41
in term infants 568
normal 738–739i
Energy-protein requirements in preterm infants 541
Energy substrate
and amino acids 196
balance of, in obesity 591–593
effects of disease on 194–195

Enrichment of formula and human milk in preterm infants 558–559
Enteral feeding
and drug interactions 297
devices used for 941t
drug-nutrient interactions in 1031
in cancer patients 885
in cystic fibrosis 847
interventional approaches to 394–395
in LBW and VLBW infants 389–396, 542–543t, 546t
methods for advancement in 942
minimal 394–395
optimization of 546t
Enteral formulas 885, 1023–34
composition of 1025–27
disease-specific 1026–27
for infants 1025–26
Enteral intake as proportion of human milk in preterm infants 556i
Enteral nutrition (*see also* Tube feeding) 1023–1034
access to
complications in 1030–1031
management of 1029
as maintenance therapy in IBD 796–797
at home 1031
contraindications to 1024–25
gastrointestinal complications of 1030
in critically ill children 964–965
in Crohn’s disease 792–794, 796–797
in growth failure 798–799
in HIV-infected children 683
in intestinal failure and SBS 929–931, 940–941
in IBD 794
mechanism of 799–801
in preterm infants 394–398, 544–547
in specific disease states 1029
in VLBW infants, challenges for 387–398
infectious complications of 1030
initiation and advancement of 545–547
mechanical complications of 1030–31
modes of delivery of 1028
role in growth and development 798–799
pulmonary complications of 1030
routes of administration for 1027–28
steroids vs., 795–798
Enteral products, infant and noninfant [see Appendix III on pp. 1217–1231 for

detailed tables]
Enterocolitis, food protein-induced 828
Enterocytes (*see also* Intestinal epithelial cells)
and diet 57
adapting to nutritional cycle 336–339
genetic expression in 336i
in fatty acid uptake 338
in hormonal regulation of iron 123i
Enterokinase deficiency affecting the pancreas 839, 841
Enteropeptidase deficiency, congenital 839
Enteroplasty, serial transverse, technique for 943i
Environment
role of, in later-life diseases 441–442
shaping gut microbiota 402–403
Environmental contaminants in human milk 498
Environmental vs. genetic factors in obesity 586
Enzyme deficiency disease, debranching 764
Enzyme disorders of liver and GI tract 745–748t, 751–752
Enzymes
in human milk 492–493
in HMO metabolism 59t
Eosinophilic esophagitis (EoE) in food reaction 827
Ephedra 178
Epidemic, HIV and pediatric AIDS 673
Epidemiologic studies (*see also* Anthropometrics; Field studies)
models accounting to total energy input in 283–284
types of 286–288
Epidemiologic variables, preservation of data in 285–286
Epidemiology
nutritional 271–291
of childhood obesity 599–600
of feeding disorders 699
of growth failure 798–799
of iron deficiency 126
of pediatric growth failure 707–708
Epidermal growth factor in intestinal adaptation 933
Epigenetic modification in nutrition 329
Epigenetics and nutrition 351–360
Epithelial cells (*see also* Enterocytes; Intestinal epithelial cells)
polarity of 334
role in absorption and in immune system 173, 329–349
Equations for measuring body composition 38–39t
Erythropoietic regulation of iron 124
Esophagitis, eosinophilic 827

Essential fatty acids (EFAs; *see also* Fatty acids) 30
  availability of, to fetus 458–460
  deficiency in 30
  intravenous lipid emulsions for 1046–48
  maternal diet and 459
Estimated average requirements (EAR) 163–164
  for amino acids by age and gender 77–78t
Ethnic differences
  in body composition 45
  in stature 50
Etiologic features of trace element deficiencies 99–100, 104, 106–107
Etiology
  of anorexia in cancer patients 881–882
  of dental caries 989–992
  of feeding disorders 701–703
  of growth failure 710t
  in IBD 798–799
  of IBD 787–788
Evidence-based results of health interventions 669
Exclusive Liquid Diet therapy in children with Crohn's disease 1119, 1120t
Exercise (*see also* Physical activity)
  affecting bone health 1009
  and body fat 45
  in diabetes mellitus 906
Exocrine pancreatic disorders 835–849
  absorption and digestion of fats in 840, 846
  nutrition in 841–847
  protein absorption and digestion in 841–844
Extracellular fluid (ECF), estimation of 39
Extrauterine growth restriction 538
  during NICU hospitalization 553i
Extremely-low-birth-weight (*see* Low-birth-weight; Very-low-birth-weight)
Eye care in SAM 222

F

Factorial approach in determining protein or amino acid requirements 79
Failure to let down milk 533–534
Failure to thrive (*see also* Growth failure)
  in outpatient setting 707–727
  maternal causes of 533–534
  risk factors for 708–713
  while breastfeeding 529–534
Failure to wean 700
FAO/WHO/UNU, assessment of energy needs by 190

Familial dyslipidemias 974–976
Famine
as political weapon 2
effects of, on human health 871–872
Fasting
and energy substrate 192
effects of
on GH release 371–372
on somatomedin and nitrogen balance 375i
growth hormone release in 372t
Fasting adaptation, inborn errors of 757–768
Fat(s),
absorption in, biochemistry of 84
balance of, in obesity 591–593
contents of milk, considerations in reducing 578–579
dietary intake of 337
and drug kinetics 304
in diabetes 899–900
digestion of 83–84
in breast-fed infants 83
in cystic fibrosis 839–841
ectopic, stored in muscle and liver 622–623
in breast-fed vs. formula-fed infants 570
in human milk 488–489
malabsorption of in CF 846–847
needed in young athletes 1092
Fat distribution
ethnic and racial differences in 45
relative to gender during adolescence 45
Fat-free mass (FFM)
and resting metabolic rate 184i, 185i
changes during lifespan 49i
changes relative to growth 48
fat metabolism in liver disease 852
Fat-soluble vitamins 31–32
deficient, in liver disease 853, 854
Fatty acid(s) (*see also* Essential fatty acids)
biochemistry of 86–87
chain length of 389–390
conversion in premature infants 389–390
essential 87–89
in human milk and colostrum 489–490
long-chain polyunsaturated, in liver disease 852–853
macronutrient assessment of 30–31, 83–93

mitochondrial, oxidation of 754i
omega-3, in VLBW infants 396
oxidation defects in 746–747t
relative to maternal plasma 89i
required in preterm infants 544t
structural formulas of 87i
uptake, metabolism, and transport in placenta 456–457
Fatty acid deficiency 88t
Fatty acid metabolism
genetic variation in 88–89
Fatty acid transfer, placental 89
Fatty acid uptake
adaptation of enterocytes in 338
comparison of chain length in 390t
trophoblast 90i
Feeding
complementary (*see also* Complementary feeding)
health outcomes and 575–582
in countering malnutrition 232
in low-income countries 641–652
difficulties in 699–704
enteral, in LBW and VLBW infants 389–396, 543–547
infant
gastrodevelopment and 387–398
trends in 565–566
methods of, during complementary food period 580
oral, development of mechanical function for 388–389
parenteral, in preterm infants 547–549
physiology of 699–700
school programs for, effectiveness of 670
thermic effect of 187–188
tube, administration and tolerance for 547
Feeding behavior, observation of 719t
Feeding devices
mechanical difficulties in 1030–1031
postpyloric 1028
Feeding difficulties in disabled children 729–730
Feeding disorders 699–705, 711–712, 719t
assessment and management of 703–704
classification of 712–713t
post-traumatic 702–703
Feeding problems, behavioral, in liver disease 858
Feeding skills
interrupted by cancer treatment 882

normal development of 700
Feeding tubes, gastric access to 1027–1028
Female athlete triad
energy balance and 1096
of eating disorders 1004
Ferritin in assessing iron status 134–138
Fetal fat content 43 (*See also items under* Gestation *or* Gestational)
Fetal glucose and lipid metabolism 445–460
Fetal growth, and risk of adult disease 635–636
Fetal growth restriction (FGR), amino acid transport in 475–477
Fetal hepatic circulation, amino acid metabolism and 474–475
Fetal iron needs 117–118
Fetal nutrition
amino acids supplying 465–482
later-life disease and 439
Fetal oxidation, amino acids and 472–474
Fetal-placental interrelationship 445–460 (*passim*)
Fetus
carbohydrate and lipid metabolism in 445–463
essential fatty acid accumulation in 459–460
glucose in 446–456
hyper- and hypoglycemia in 453–456
lipid uptake, metabolism, and transfer to 456–460
Fever, effects of, on energy substrate 194
Fibers, dietary
in diabetes 898
in vegetarian diets 1137
GI absorption in 305
recommendations based on 65
sources of 1136–37t
to modify lipid profiles in dyslipidemia 979
Field studies, anthropometric techniques in 40
Fish allergy 1107
Fish oil supplements
benefits of 177–178
during pregnancy 431
Flavor of breast milk vs. formula 569
Fluid(s) and electrolytes
needed in young athletes 1094
required in preterm infants 544, 1055–1951
Fluid imbalance, drug-induced 309–312
Fluid-restricted diet 1127–28
Fluoridation of water supply, benefits of 995–996
Fluoride(s) 11–112

excess, toxicity of 992–994
human health and 992–998
in human milk 497
in preventing dental caries 111
in tablets, toothpaste, and oral rinses 996–997
intake by infants 111
mechanisms of action of 997
naturally available 994–995
Fluorosis 111
Folic acid 156–157
dietary sources of 156
DNA methylation in deficiency 353–354
in HIV-infected children 680
in human milk 495–496
metabolism of 156–157
status in pregnancy 426–427
Folic acid deficiency
and neural tube defects 286–288
DNA methylation and 353–354
laboratory assessment of 33
use of vitamin supplements in 287–288
Folic acid requirements in pregnancy 287–290, 426–427
Food (*see al*so Complementary food; Drug-nutrient interactions)
and nutritional assistance plans, in U.S. 253–262
as political weapon 2–3
availability of, and per-capita income 657
complementary (*see also* Complementary feeding; Complementary foods)
insecurity about availability of 670
interacting with drugs 296
functional, in diarrheal disease 919
reintroduction of, after exclusive enteral feeding 796
safe preparation of 649
solid, added in breast-fed infants 527, 571–572
Food allergies (*see also* Celiac disease *and* Allergy *entries*) 823–833
clinical disorders of 825–828
management of 830, 1099–11
novel therapies for 830
pathophysiology of 824
reaction mechanisms in 825
Food aversion 702–703
in cancer patients 881–882
Food crops, biofortification in 649
Food hypersensitivity reactions 826–828
Food information laws 1100

Food ingredients, functional, overview of 173–181
Food intake
  and drug-nutrient interactions 297
  questionnaire in assessing dietary intake 6, 273–274
  reduction in, and rate of metabolism 183
  24-hour recall of 272–273
Food record (*see also* Dietary records) 273
Food safety in patients receiving HSCs 884–885
Food science, beginnings of 3
Food sources (*see also* Dietary sources)
  of chromium 110
  of copper 195
  of iodine 113
  of riboflavin 153
  of selenium 108
  of vitamin D 146
  of zinc 102
Food supply, fortifying
  to prevent iron deficiency 129–130
  to prevent NTDs 287–288
Forbes disease 764–765
Formula(s)
  as risk factor for childhood obesity 626
  benefits in preterm infants 555
  composition of 566, 570–571
  cow’s milk-based, contraindications for 1108
  cow’s milk–free 1112t
  DHA-free, deficits of 177
  disease-specific 1026–27
  enteral (*see* Enteral formulas)
  in glycogen storage disease 760–761
  for children with liver disease 856t
  for parenteral nutrition, amino acid composition of 1043t
  for preterm infants 546
  after NICU discharge 560t
  isocaloric nutrient enrichment of 558–559
  in diarrheal diseases 917–918
  in intestinal failure/SBS 940
  kidney-specific 773t
  MCT-based 1126
  substances added to 571
Formula-fed vs. infants at term 566–571
Fortification of food supply
  to prevent NTDs 287–288

iron 129–130
of human milk after NICU discharge 559–560
Frankfort plane
in assessing length 7i, 732i
Free radicals in severe acute malnutrition 208–209
Frontal occipital circumference (*see* Head circumference)
Fructose
controversial opinions concerning 63
dietary, in diabetes 900
metabolic aspects of 62
sources to be avoided 1122t
Fructose intake, high, potential risks in 62
Fructose intolerance 67
hereditary 750, 1122
Fructose load, metabolic fate of 61i
Fruits, sucrose content of 1118t
Fungemia, catheter-related, in IF/SBS 938

G

Galactosemia 1107
acute manifestations of, in infant 750
as carbohydrate diease 67
low-lactose cheese allowed in 1108
Garlic 178–179
Gastric access feeding devices 1027–1028
Gastric acids and innate defense mechanisms 391
Gastric bypass surgery for obesity 300, 609
Gastric emptying and intestinal transit 294–295, 388–389
Gastric lipase (*see* Lipase, gastric)
Gastroenteritis
allergic eosinophilic 827
bacterial, and intestinal epithelium 335–336
therapy of, based on degree of dehydration 916
Gastrointestinal development
implications for infant feeding and 387–398
microbiota and 399–409
Gastrointestinal diseases
diets for 1115–1119
enteral nutrition in 1029
complications in 1030
Gastrointestinal food hypersensitivity disorders 827–828
Gastrointestinal sphincter tone, development of 388
Gastrointestinal tract
absorption, in HIV-infected children 675–676

adverse effect of medications on 307–312
development of defense and barrier function in 391–393
in SAM 210–211
inborn errors of metabolism in 744–749
influenced by milk oligosaccharides and prebiotics 63i
pediatric, role of carbohydrates in development of 55
side effects of medication for HIV in 675t
Gene expression
and breast milk 338–339
and nutritional regulation 329–349
Gene-diet interactions 284–286
signal transduction mediating 335i and growth hormones 367–368
therapeutic implications of 344–345
Genes
and growth hormones 367–368
role in later-life diseases 441–442
Genetic disorders
exocrine pancreatic 835–837
hepatic 749–755
of iron overload 132–134
Genetic growth potential
calculation of 714t
midparental length in 10
Genetic syndromes with obesity phenotype 602t
Genetic variation in fatty acid metabolism 88–89
Genetic vs. environmental factors in obesity 586
Genetics
and bone health 1008
of childhood obesity 601
Genetically modified foods, overview of 2
Genomics in assessing nutritional status 34
Gestation (*See also* Fetal *entries*)
body fat accumulation during 43
changes in body composition during 44i
fetal and placental glucose during 452
Ginger 179
Ginko biloba 178
Global child health
and the nutrition transition 631–640
interventions in 663–672
Global Database on Child Growth and Malnutrition (WHO) 854
Global distribution of underweight in children 633i
Global trends in child mortality and disease 631–632
Globalization and malnutrition 234, 632–634

Glomerular filtration rate, estimation of 769, 770t
Glucose
  effect of medications on 312–313
  in human milk 486–487
Glucose-galactose malabsorption 1118–19
Glucose metabolism
  medications altering 312–314
  normal 757
Glucose regulation, disordered 616
Glucose-6-phosphatase deficiency 759–763
Glucose supply to placenta and fetus 446–456
Glucose tolerance, impaired, birth weight and 435t
Glucose transport in MetS 621i
Glucose transporter expression 337
Glutamine supplements in VLBW infants 396
Glutamine transporter systems 330–332
Gluten
  and possible relationship to diabetes 894
  in immune system responses in celiac disease 810i
  in pathogenesis of celiac disease 809–810
Gluten challenge in uncertain diagnosis 1117
Gluten-free diet in celiac disease 817–818, 1115–17
Glycemic control in critical illness 961–962
Glycemic index 897–898
Glycemic outcomes and meal planning 905–906
Glycemic targets for type 1 diabetes 896t
Glycoconjugates, human milk, antimicrobial and prebiotic activities of 507–508
Glycogen phosphorylase deficiency 765–766
Glycogen phosphorylase kinase deficiency 765–766
Glycogen storage disease(s) (GSDs) 67, 757–768
  nutritional management of 758–759, 760–761, 763–764, 766
  type 0 757–759
  type I and Ia 759–763
  type Ib 763–764
  type III 764–765
  type VI and type IX 765–766
Glycogen synthase deficiency, hepatic 757–759
  nutritional management of 758–759, 760–761, 763–764, 766
Glycosade in treatment of GSD type I 761
Ginkgo biloba 178
Glycosylation
  congenital disorders of 747t
  hepatic disease due to 753
Goblet cells protecting developing gut lining 391–392

Governmental programs to prevent malnutrition 245–267
Governments, responsive, in preventing malnutrition 233
Graft-versus-host disease after HSC transplant 887–889
Grafts, type of, in intestinal transplantation 946–947
Grapefruit juice and drug interactions 305
Green Revolution 2
Growth
  after intestinal transplantation 948t
  after kidney transplant 780–781
  after NICU discharge 557–558
  interventions supporting 558–61
  and body composition 37–51
  assessment of 275–278
  in HIV-infected children 675–676
  and cancer, connection between 865–866
  and nutritional deficits at NICU discharge 553–556
  attenuated, linear and velocity curves for 715t
  catch-up 363–365, 437–438
  nutritional needs for 720
  rapid, and adiposity rebound 691–692
  compositional changes during 41–49
  effect of celiac disease on 813
  energy expenditure in 187
  estimation of 187
  humoral regulation of, and nutrition 361–385
  factors affecting 361–366
  fats and fatty acids in 83–93
  feeding disorders and 699
  Fenton’s growth charts for 539–541i
  fetal, and risk of adult disease 635–636
  in breast-fed vs. formula-fed infants 569
  in children and adolescents, breast cancer and 872–873
  in chronic kidney disease 773–779
  in disabled children, monitoring 729–741
  in LBW infants 537–541
  in infancy and childhood, later-life diseases and 437–439
  in young athletes 1091–92
  of breast-fed infants, assessing 484–485
  normal, insulin in 370–371
  rapid in infancy, as predictor of obesity 626
  retardation of, in utero 538
  thyroid hormones in 377–378
  tissue 363–364
  velocity in 364–365

Growth charts and tables
CDC (2000) 9
Disease-specific (Appendix I, pp. 1192–93)
for screening and diagnosis 713–714
WHO (2006) 9
Growth cycle of bone 1007–08
Growth factors in human milk 492
Growth failure
after liver transplantation 860
and enteral feeding 932
consequences and prognosis of 722–723
etiology of 710t
high-energy diet in 1126–27
in IBD 790
epidemiology and etiology of 798–799
in utero 43
psychosocial 376–377
in outpatient setting 707–727
management of 720–722
screening and evaluation of 713–720
secondary to abuse or neglect 718t
Growth hormone(s) (GHs) (*see also* Insulin; Insulin-like growth factors) 366–373
axis with IGF
effects of undernutrition on 374t
physiological regulation 368–369
binding proteins (GH-BP) 367
concentrations in blood 369–370
decreased, in emotional deprivation 376
direct vs. indirect actions of 369
in disease states 372–373
in intestinal adaptation 931–933
in malnutrition 371–372
Growth patterns after NICU discharge 556–557
Growth potential, genetic 10
Growth velocity, incremental 9–10
Gut (*see also* Intestine *and* Intestinal *entries*)
homeostasis in, restoration of 175–176
immunologically active in 340–342
“leaky” 393i
mature, innate mucosal immune response in 503–506
microbiome, interacting with diet and immune system 174i
neonatal
bacterial colonization of 503–515
immunoglobulin in 510–511

nutrient transport in 337
Gut microbiota
in IBD 788
in obesity and metabolic disease 406–407
probiotics modulating 407
shaped by diet and environment 399–409

H

Head circumference
in assessing nutritional status 8
in disabled children 733
reduced at discharge from NICU 553–554
Health outcomes and complementary feeding 575–582
Height
anthropometric data on, in developing countries 654–657
weight relative to 10
Height potential, genetic, calculation of 714t
Helsinki Birth Cohort and later-life disease 435–437, 441
Hematologic tests as indicators of nutritional status 33–34
Hematopoietic stem cell transplant (HSCT) in cancer 881
food safety recommendations for patients receiving 884–885
Hemochromatosis, perinatal and juvenile 750
Hemoglobin in assessing iron status 134
Hepatic circulation, fetal, amino acid metabolism and 474–475
Hepatic complications in parenteral nutrition 1065
Hepatic diseases and inborn errors of metabolism 743–756
Hepatic glycogen synthase deficiency 757–759
Hepatic injury and pathophysiology 749
Hepatic involvement in celiac disease 816
Hepatic metabolism and nutrient-drug interactions 295
Hepatic pathology from peroxisomal disease 752–753
Hepatocellular necrosis 749
Hepatomegaly 752–753
Hepcidin in iron regulation 123i
Herbal supplements (*see also* Chinese herbal medicine *and specific supplement*) 178–179
in pregnancy 430–431
Herbal teas during lactation 526–527
Hereditary exocrine pancreatic disorders 835–839
Hereditary fructose intolerance 750
very-low-fructose diet for 1122, 1123–24t
Heritability
of childhood obesity 600–601
of individual components of MetS 621t

Hers disease 765–766
High-energy diet in growth failure 1126–27
High-fiber diet 1133, 1135–37
Histones
  acetylation of 355–356
  biotinylation of 356
  DNA methylation modifying 354–356
History, medical, in assessing growth failure 714–716
HIV in human milk, precluding breastfeeding 498
HIV-associated metabolic disturbances 678–679
HIV-infected children
  diagnostic evaluation in 680–681
  wasting in 674–677
HIV infection 772
  avoiding transmission of, by breastfeeding 498
  in severe acute malnutrition 212, 218
  pediatric 673–687
Home-based devices for nutritional feeding 1073–75
  complications and infections in 1073
  enteral, 1031
  long-term monitoring of 1074
  parenteral 1073–75
Home management of acute diarrhea 913–914
Homeostasis
  electrolyte, after acute kidney injury,
  intestinal, restored by probiotics 175–176
  iron 121–124
Hormonal preparation post partum for lactation 519i
Hormonal regulation of iron efflux 123
Hormonal status affecting bone health 1009
Hormone secretion, nutrient stimulation of 332–334
Hormones (*see also* Growth hormones; Insulin-like growth factors *and specific hormones*)
  acting on body compartments 363t
  affecting intestinal adaptation after resection 935t
  and growth factor and nutrients 361–362
  in human milk 492
  thyroid 377–378
Hospital treatment
  monitoring 1064–65, 1074
  of dehydration 917
  of malnutrition 213–216, 234–235
Hospitalized child, assessing nutritional needs of 5–17, 1147–51
Human capital, investing in, by lessening malnutrition 233

Human immunodeficiency virus (*see* HIV *entries*)
Human milk (*see also* Breast milk)
  adequacy after 6 months 578
  affording immune protection 176
  biochemical composition of 486–498
  and maternal nutrition 642–643
  cellular reactivity to bacteria in presence of 510–511
  compared with formula 560t
  composition in preterm and term infants 456t
  deficits in, in preterm infants 555
  donor's vs. mother's 545
  education mucosal immune system 509–510
  enriched after NICU discharge 559–560
  factors modulating response to bacteria 510t
  in at-term infants 565–573
  breast-feeding vs. formula–feeding 566–572
  in preterm infants 545
  initiation of secretion of 520–251
  macro- and microminerals in 496–498
  macronutrients in 58t
  microbiome in 511
  nutritional properties of 483–502
  production of 485–486
  protective properties of, in neonatal gut 503–515
  protein and micronutrient content of 643t
  PPR-bacteria interactions in presence of 509–510
  pumping and storage of 528–529
  shaping intestinal microbiota 507
  supplementation of, during breastfeeding 525
  synthesis and secretion of 519–521
  TGF-β in immune education 511–512
Human milk oligosaccharides (HMOs) 487–488
  antimicrobial and prebiotic activities of 507–508
  metabolized by bifidobacteria 59t, 59–61
  monosaccharide composition of 61t
Human population, microbiome variation, in 403–405
Humoral regulation of growth, and nutrition 361–385
Hydration in young athletes 1094
Hydrolysis of fatty acids 84i
Hypercholesterolemia
  dietary therapy for 1128–30
  severe, other therapies for 980
Hyperglycemia
  affected by medications 313–314t

maternal, and placenta and fetus 455–456
in critical illness 961–962
Hyperinsulinism 67
Hyperlactatemia in GSD type Ia 761
Hyperlipidemia
and cardiovascular disease 971–986
diet for 1131t
dietary restrictions in 1127–28
medications treating 981t
secondary, causes of 975
Hypernatremic dehydration 918–919
Hypersensitivity to food, diagnostic methods for 829–830
Hypertension
arterial, in MetS 616
classification of, in children and adolescents 629t
later-life, relative to birth weight 438–439
potential link of fructose to 62i
Hypervitaminosis 319–322
Hypoalbuminemia 28
Hypoglycemia
affected by medications 313–314t
in diabetes 906–907
placenta and fetus and 453–454
in SAM 213–214, 216
Hypothermia in SAM 216
Hypothyroidism, catch-up potential in 365i

I

Identification of children at risk of obesity 627
Illness, chronic, affecting bone health 1009
Imaging techniques in measuring body water 42
Immune defense mechanisms, micronutrients involved in 34
Immune responses
adaptive and innate, in celiac disease 810i
and bovine insulin 894
in SAM 208–209
innate mucosal, in mature gut 503–506
Immune system
influenced by milk oligosaccharides and prebiotics 63i
interacting with microbiota and diet 174i
intestinal 173–175
prenatal 504i
Immune system, mucosal
educated by factors in breast milk 509, 511

TGF-β in 511–512
signaling from intestinal lumen to 339–345
Immunity
adaptive, breast milk effects on 512
cell-mediated, in SAM 209–210
diets to enhance, in critical illness 965–966
humoral, in SAM 210
Immunization, campaign for, in developing countries 659
Immunoglobulin E (IgE)
diagnostic methods for 828–830
mediating reactions to food 825
Immunoglobulins in neonatal gut 510–511
Immunological tests for nutritional status 34
Inborn errors
of fasting adaptation 757–768
of metabolism
and diseases of the liver and GI tract 743–756, 859
dietary treatment for 1119
of trace element metabolism (*see also specific disease*) 96
Income
food and water availability and sanitation, relative to 657–659
household, related to CHD 437t
low to middle, in nonwestern countries, and childhood obesity 628–629, 689–697
real, and child nutrition 653–661
India, BMI related to rural/urban status 694t
Industrialized countries, community nutrition in 243–269
Inequity of food availability in undernutrition 670
Infancy, nutrition during, later-life disease and 435–443 (*passim*)
Infant feeding
breastfeeding vs. formula-feeding 566–573
in low- and mid-income countries 641–652
initiation of solid food in 527
GI development and 387–389
preterm and neonatal 387–398
Infants
air-displacement plethysmography in 42
breast-fed
assessing growth of 484
development of gut microbiota in 406
dietary assessment in 274–275
iron levels in 130
role in initiating lactation 523–524
vs. formula-fed 568–572
enteral formulas for 1025–26

failure to thrive in, interventions for 720–721
fluoride intake by 111
growth in, later-life development of disease and 437–438
health benefits of breastfeeding in 483–485
in malaria-endemic areas 131
in U.S.
community nutrition concerns for 249–250
nutritional recommendations for 166, 250t, 251t
LC-PUFA supply to 90–91, 389–390
need for iron in 117–118
nutrient requirements of 566–568
in cystic fibrosis 846–847
preterm or LBW (*see also* Preterm infants)
energy requirements for 1040–41
enteral nutrition in 387–398
fat absorption in 84
iron deficiency in 126–127
parenteral nutrition in 1036–37
trace element deficiency in 96
reference, body composition and weight gain in 567i
suckling disorders in 530–531
term 565–573
very-low-birth-weight (VLBW)
enteral and parenteral nutrition in 387–398
vitamin K deficiency bleeding in 151
Infection(s)
and iron, interaction between 131
and nutrition, relation of 206–207
as complication of enteral feeding 1030
breastfeeding and 576–577
catheter-related 1069–72
effects of, on energy expenditure 194–195
in SAM 218
pediatric HIV 673–687
with *Clostridium difficile,* in diarrhea 918
Infectious diseases
and undernutrition 636
sanitary interventions to prevent 657–659
Inflammation, systemic, affecting bone health 1009
Inflammatory bowel disease (IFB) 787–808
and gut microbiota 175, 788
bone disorders and 1015
in glycogen storage disease type Ib 763
role of pre- and probiotics in 173–175

Inflammatory syndrome, pharmacologic approach to 196–197
Injury, acute kidney 769–772
Innate immunity linked to adaptive immunity in neonatal gut 505
Innate mucosal immune response in mature gut 503–506
Insulin 370–371
  bovine and immune response 894
  in malnutrition 375–376
  nutrient stimulation of 332
  parenteral, and intestinal adaptation 933–934
  pharmacodynamics, profiles of 892t
Insulin level
  fetal, 451
  in elderly, relative to birth weight and genes 441t
Insulin-like growth factors (IGFs) 366–375
  and intestinal adaptation 932
  and somatomedin hypothesis 366–367
  in malnutrition 373–375
  nutrient stimulation of 332–334
  regulation by 375
Insulin-like growth factor-binding proteins (IGFBPs) 367–368, 367–368
  and genes 367–368
  and immune system 342
  in malnutrition 373–375
Insulin-mediated signal transmission in glycogen synthesis 621i
Insulin resistance
  and glucose regulation disorder 616
  as pathomechanism of MetS 619
  molecular basis of 619–621
  potential link of fructose to 62t
  role of adipose tissue in 621–622
Insulin secretion, fetal 452–453
Interleukins, effect of, on energy expenditure 184
Interventions
  global health, evidence-based results of 669
  nutritional, for growth failure 720–722
  in HIV-infected children 682–684
  targeted towards women and children 665–669
  to prevent malnutrition in Canada and U.S. 247–267
  to support growth after NICU discharge 558–561
Intestinal adaptation 927–929
  after resection 935t
  nutritive and non-nutritive factors in 929–934
Intestinal barrier, neonatal, components of 505–506
Intestinal damage in celiac disease 813i

Intestinal environment
in infants 387–398
microbiota and 393–394
Intestinal epithelial cells (IECs) 329–349 *passim*
nutritional factors and gene expression in 333–334
Intestinal epithelium, genes of immunologic importance in 340–342
Intestinal failure (IF) 925–956
and short bowel syndrome (SBS) 925–956
cholestasis in 934–938
distribution of causes of 927t
enteral feeding in 941–943
devices used for 941t
methods for advancement of 942
epidemiology of 926
management of and outcomes in 939–945
medical support in 940–943
physiology and pathophysiology of 934–939
small intestinal bacterial overgrowth in 938–939
surgical support for 943–944
underlying diseases of 925–926
Intestinal failure-associated liver disease (IFALD) 934–938
as metabolic complication of parenteral nutrition 1065–1067
unbalanced PN solution related to 1067–1068
Intestinal flora in SAM 218
Intestinal fluid output in diarrheal diseases 912
Intestinal homeostasis, probiotics restoring 175–176
Intestinal immune system 173–175
development of innate defenses in 391
genes expressed in 340–342
Intestinal maturation, cortisol in 378
Intestinal microbiota (*see also* Gut microbiota)
as essential component of intestinal tract 173–175
breast milk factors modulating 507
carbohydrates and 56–57
development of, in infants 406, 506–507
nutrient effects on 343–344
Intestinal motility, development of 388–389
Intestinal mucosal barrier 173t, 391–392
Intestinal pattern recognition receptors 174t
Intestinal transit and gastric emptying 294–295
Intestinal transplantation for IF/SBS 945–949
(*see also* Transplanation, intestinal)
Intestinal villi, immunofluorescent 393i
Intestine(s)

anatomic development in 387–389
mature, innate mature response in 503–506
Intolerance (*see also* Food allergies)
carbohydrate 1117–19
food, management of 1099–1100
Intrauterine growth charts 8
Intrauterine growth restriction and fetal glucose metabolism 454–455
Intravenous lipid emulsions (ILEs) in PN support 1044–49
in VLBW infants 1048–49
MCT/LCT-based 1045
olive oil-based 1045–46
fish oil-based 1046
for parenteral supply 1047t
soybean oil-based 1044–45
stability and structure of 1049
Intravenous therapy in SAM 217
Iodine 112–113
deficiency in 112–113
food sources of 113
in human milk 497
Iron 117–141
biochemical assessment of 134–137, 135t, 136–137t
dietary requirements for 128–129
distribution and exchange pools in 121i
during pregnancy 425–426
heme 119–120
homeostasis of 121–124
in breast-fed vs. formula-fed infants 569
in human milk 497
in young athletes 1094–95
inborn errors of metabolism, and 750
inorganic 120–121
metabolic diseases of 133t
need for in pregnancy 130
storage or trafficking of 122–124
Iron absorption, factors affecting 120i, 124
Iron balance, factors negatively influencing 118t
Iron content of various foods 119–121
Iron deficiency 125–131
and obesity 121
ferritin as biomarker of 134–138
food fortification and supplementation for 129–131
in malaria-endemic areas 131
in pregnancy 425–426

transferrin receptor in screening for 137–138
Iron efflux, hormonal regulation of 123
Iron overload 131–134
genetic disorders of 132–134
Iron poisoning 131–132
Iron requirements during complementary food period 579
Iron-rich diet 1133, 1134–35t
Iron status, assessment of 134–138
and cutoff points by age 135t
biochemical indices for 136–137t
Iron supplements in pregnancy to prevent undernutrition 665
Iron toxicity, acute, stages of 132t
Iron transport, cellular 125i
Irritable bowel syndrome, probiotics in 175
Isocaloric nutrient enrichment of formula 558–559
Isotope dilution methods in estimating body water 41
Isotope tracer methods to measure amino acid oxidation 79–80

J

Johanson-Blizzard syndrome 838

K

Kidney (*see also* Renal *entries*)
Kidney disease 769–786
acute 769–772
chronic 769–786
nutrition in 769–786
Kidney injury 769–772
Kidney transplantation, management and growth after 780–782
Kinetics
drug (*see* Drug kinetics)
transport, of glucose in placenta and fetus 448, 449
Kwashiorkor 204–205, 208
drugs in 299

L

Laboratory testing
confounders of interpretation of 19, 25–28
in assessing nutritional status 13, 19–36
in critically ill children 964
in cancer patients 883–884
standardization of 26
to ascertain growth failure 719

Laboratory values, nutritional, reference ranges for 20–25(t)
LactAid nursing training system 532i
Lactating women, maternal nutrition in 525–527
Lactation
  anatomy and physiology of 485–486, 517–520
  caveats about herbal teas during 526–527
  initiating 523–525
  oxytocin and prolactin release in 521
  preparation of the breasts for 522–523
  supplemental devices for 531–532
Lactation failure, let-down reflex in 533–534
Lactic acidosis in short bowel syndrome 939
Lactic acidosis syndromes, enzyme defect in 747t
LactMed database of the NLM 534
Lactoferrin, adaptation of enterocytes to 339
Lactose
  in human milk 486–487
Lactose-derived oligosaccharides, monosaccharide linkages in 61t
Lactose-free diet 1107–10
Lactose-free sources of calcium 1111t
Lactose intolerance 1, 1118
La Leche League 528
Latin America, undernutrition in 230i
Laxatives affecting drug absorption 308
Lead exposure, effects on children of 420–421
Leg measurement, techniques in 733–735
Length
  after discharge from NICU, deficits in 553–554, 559t
  in measuring nutritional status 7–8
  of leg in disabled children 732–734
  weight relative to 10
Let-down reflex
  failure of, prolactin’s role in 527
  in lactation 521
Leukemias, initiation of, in utero 866
Life-stage groups, pediatric
  and DRIs 165–166
  factors affecting obesity in 690–692
Lifestyle factors
  changed by nutrition transition 637
  in childhood obesity 606–607
  in developing countries, obesity prevention programs in 694–698
Linear growth during development 50
Linoleic acid

deficiency in 30, 87–88
in premature infants 389–390
Lipase/colipase deficiency, congenital 839
Lipase, gastric
adaptation of enterocytes in 338
in pancreatic insufficiency 83
lipid digestion in infants 389
Lipid biochemistry and CVD in children 973–974
Lipid-lowering drugs in children 980
Lipid metabolism
fetal 456–460
in cancer 878
in critically ill or injured children 962
in SAM 208
lipid requirements, and intravenous lipid emulsions 1044–1049
Lipid values in childhood 924t
Lipids
changes during growth 48
effects of medications on 314–316
in metabolic syndrome 616
in treating pancreatic insufficiency 846
intravenous, in cholestasis 937
macronutrient assessment of 30–31
metabolism and transfer to fetus 456–460
long-chain *n*-3 polyunsaturated fatty acids (PUFAs) 87–88, 177–178
parenteral requirements and 1048
required in preterm infants 543
transport of 85
Lipodystrophy, HIV-associated 678–679
Lipoprotein levels and atherosclerosis 973
Lipoproteins, assembly of 85
Liver (*see also* Hepatic *entries*)
changes in, in SAM 210
disorders caused by inborn errors of metabolism 743–756, 859
fat storage in, in MetS 622–623
Liver disease
acute 858
acute and chronic 851–863
chronic 859
clinical manifestations of 854t
formulas for children with 856t
Liver transplantation affecting nutritional status 859–860
Long-chain fatty acid disorders, diet for 1122, 1124–26
Long-chain polyunsaturated fatty acids (LC-PUFAs) 88–89

in formula 571
in liver disease 852–853
in premature infants 389–390
perinatal supply of 88
Longitudinal intestinal lengthening and tailoring, technique for 943i
Low birth weight and later-life diseases 436–437
Low-birth-weight infants (*see also* Preterm infants)
growth charts for 8–9, 539–541t
inpatient care of 537–551
Low-income countries
complementary feeding in 644–646
infant and child feeding in 641–652
Low phenylalanine diet in PKU 1119–1121
Lutein in formula 571

M

Ma huang (ephedra) 178
Macrominerals in human milk 496–498
Macronutrient assessment
of energy 28–31
estimates of requirements for 29
overfeeding syndrome and the RQ 29
production from nutrients 29
respiratory quotient in 29
of lipids and fatty acids 30–31
and carnitine 30–31
deficiency of EFAs 30
of protein 27–28
acute-phase response and plasma proteins 28t
albumin 28
amino acid profiles as indicator 28
nitrogen balance 27
plasma 28
prealbumin 28
somatic 27
retinol-binding 28
transferrin 28
Macronutrient distribution 897
ranges in, acceptable (AMDRs) 163, 164
Macronutrients
and bone health 1009
dietary reference intakes for 167, 168
in mature human and cow’s milk 59t
needed in young athletes 1092

Macrophages in regulation of iron 123i
Magnesium in human breast milk 497
Major histocompatibility complex (MHC)
  class II, and invariant chain expression 341–342
Malabsorption
  consequences of, in CF 841–842
  glucose–galactose 1118–19
  in cystic fibrosis 844–846
  in exocrine pancreatic disorders 839–841
  in SAM 211
  of fats 846–847
Malabsorption disorders, vitamin 748t
Malaria, interventions to prevent morbidity 668
Malaria-endemic areas, iron deficiency in 131
Malnutrition
  affecting body composition 50–51
  and effects on energy expenditure 191–192
  and energy substrate 19
  and micronutrients 636–637
  as global health problem 230–240
  best treatment strategies for 238–239
  causes of, in HIV-infected children 674t
  cortisol in 378–379
  drug absorption and disposition in 298
  governmental programs to prevent 245–262
  growth hormone in 371–372
  hepatic drug clearance in 299
  in cystic fibrosis 842–843
  in diarrheal diseases, oral rehydration for 921t
  in disabled children 729
  in HIV-infected children, algorithm to monitor 681i
  in hospital, risk assessment of 1148–49
  in liver disease 851–854
  in pediatric cancer 877–878
  insulin, IGFs, and IGF-BPs in 373–375
  indicators of 230–232
  Latin American experience treating 230i, 237–240
  moderate to severe, prevention strategies for 234–237
  prevention and control at national level 229–242
  RUTF to combat 236
  severe acute (*see* Severe acute malnutrition)
  severe
  cost-effectiveness in therapy of 237i
  diarrhea with 921–922

thyroid hormones and 378
Waterlow criteria for categorizing 708t
women's education alleviating 232
Mammary alveolus, synthesis and secretion of milk in 518i, 520i
Mammary gland, fat and ductal system in 518i
Manganese 110–111
caveats against, in liver disease 857–858
in human milk 497
Marasmus 204–205
drugs in 299–300
Markers
biochemical 459
of dietary intake 279–281
Maternal causes of failure to thrive 533–534
Maternal diet
and essential fatty acid supply to fetus 459
childhood cancer and 867–868
Maternal education preventing malnutrition 233
Maternal nutrition
and later-life diseases 439
and volume and composition of breast milk 642–643
during pregnancy 425–433
in lactating women 525–527
micronutrient and protein energy supplementation of 665–666
MCT oil, to replace medium-chain fats 1128–29
Meal(s)
composition of, and TEF 188
plan for
in diabetes 902–906
in young child 1114t
timing of, in young athletes 1095
Measles, vaccination against, in developing economies 660i
Measurement of body composition 38–42
anthropometric techniques in 40
body water assessment in 41–43
equations and methods for 38–39t
in vivo chemical analysis in 40
Meat (red) consumed in adolescence and risk of breast cancer 873
Medical history
in evaluating growth failure 714–716
in nutritional screening 5–6
Medical nutrition therapy
for diabetes 891, 893, 901–902
goals of 895

Medical risk factors for growth failure 709–710
Medical support in IF/SBS 940–945
Medications (*see also* Drugs *and* Drug *entries*)
  affecting nutrient absorption 307–312
  food affecting absorption of 301–307
Medium-chain triglycerides (MCTs)
  and fat absorption 84–85
  energy content of 85i
  to replace long-chain fats 1128–29
Menarche, age at, and childhood diet 874
Menkes' syndrome 96, 102–104
Metabolic abnormalities after HIV treatment 678–679
Metabolic aspects of fructose 62
Metabolic cost of protein synthesis 188
Metabolic complications
  of enteral nutrition 1031
  of parenteral nutrition 1065–1069
  IFAD and 1066–1068
  of biliary tract, liver, bone, and kidney 1068–69
  short-term 1066t
Metabolic disorders
  enteral products for 1230–1231
  featuring liver and GI dysfunction 745–748t
Metabolic disease, gut microbiota in 406–407
Metabolic effects of cancer and its treatment 878–881
Metabolic fate of oral fructose load 61i
Metabolic function, amino acids in 74t
Metabolic programming in obesity 586–588, 600–601
Metabolic reserves, and body composition 963
Metabolic response
  mechanisms and pathways of 958
  to critical illness 957
Metabolic stress and plasma concentrations of micronutrients 31t
Metabolic syndrome (MetS) 613–624
  and risk of CVD 972
  etiology and pathogenesis in 616–622
  heritability of individual components of 621i
  prevalence, prevention, and therapy for 623–624
  role of adipose tissue in 621–622
Metabolism
  and amino acid nutrition in utero 465–482
  carbohydrate
  in liver disease 851–852
  and lipid, in fetus and placenta 445–460

cooking methods influencing 305
effect of drugs on 308–309
glucose
drugs altering 312–314
in fetus and placenta 445–456
normal 757
hepatic, and nutrient-drug interactions 295
histone methylation, one-carbon metabolism of 356
in critically ill children 958– 963
in HIV-infected children 676
in malnutrition 299
inborn errors of, in liver disease 859
lipid
and fatty acid 456–460
and transfer to placenta and fetus 456–460
of fatty acids 88–89
of fluorides 996
of HMOs by bifidobacteria 59–61
of iron 133t
of protein 74–75
disease states affecting 192–194
of trace elements 101, 105, 107, 109, 112
one-carbon, and histone methylation 356
Metabolomics
in assessing nutritional status 35
in studying gut microbiota 401
Metagenomics describing human microbiota 399–409
Metal metabolism disorders 748t
Methylation
DNA (*see* DNA methylation)
histone, one-carbon metabolism and 356
Microbes (*see also* Microbiome; Microbiota)
in human milk, educating neonatal immune system 511
modulated by breast milk 507
Microbial composition of gut, and age 58i
Microbiome, core, in studying gut organisms 402
Microbiota
characterization of 57
gut (*see* Gut microbiota)
interacting with diet and immune system 174i
intestinal 173–175, 334–335, 393–394, 399–409
and carbohydrates 56–57
development of 506–507
Microminerals in human milk 497–498

Micronutrient composition of human milk 643t
Micronutrient deficiency
  in IBD 791–792
  underlying causes of 717–718t
Micronutrient supplements
  maternal 665, 667
  to complementary foods 649
Micronutrients
  and immune defense mechanisms 34 t
  and major minerals, assessment of 31–33
  and malnutrition 636–637
  in acute kidney injury 771–772
  in HIV-infected children 679–680
  needed in young athletes 1092–1094
  plasma concentrations of, and metabolic stress 31t
Microorganisms contributing to dental caries 989–992
MicroRNAs, DNA methylation and 357–358
Middle-income countries, early feeding in 641–642
Migrants, rural to urban, and obesity 694
Migration and shift of disease risk 872
Milk (*see also* Cow's milk; Human milk)
  macronutrients in 58t
  supplementing breast milk 525
  undesirability of, in early infancy 572
Milk oligosaccharides (*See also* Human milk oligosaccharides)
  and gut microbiota 58–59
  influencing the intestinal immune system 63i
  monosaccharide composition of 61t
  milk proteins, functional role of 492–493
Millennium Development Goals (MDGs) to promote child health 663
Mineral mix solution in SAM 219t
Mineral supplementation 319–322
  in children on special diets 1130
  in children with allergies to milk 1108
  in pregnancy 429–430
Mineralized tissues 988–989
Minerals
  deficits in, after discharge from NICU 555
  drug kinetics and 304
  equations measuring amounts in body 38–40t
  reference intervals for 20–25t
  required in preterm infants 543
Mitochondrial dysfunction in hepatic disease 753–755
Mitochondrial energy production dysfunction 747t

Models
multicompartmental, of growth 37–40
of child development, conceptual 230i
Molecular basis of insulin resistance in MetS 619–621
Molybdenum in parenteral nutrition, cautions about 1061–62
Monitoring food intake, necessity for 167
Monitoring PN at home or hospital 1064–1065, 1074
Monogenic causes of obesity 601–602
Monosaccharide linkages in prebiotics 62t
Monosaccharides 55–56
in HMOs and PBOs 61–62
Morbidity, malarial, interventions to prevent 668–669
Mortality, child, global trends in 632
Mother's vs. donor's milk 546
Motor impairment, oral, and feeding difficulties 701
Mottled tooth enamel, classification system for 994
Mucosal barrier, intestinal 173t
Mucosal healing in IBD 801
Mucosal immune system (*see also* Immune system, mucosal)
education of by factors in breast milk 509
innate response in 503–506
Multivitamin formulations for parenteral nutrition 1059t
Multivitamin supplements during pregnancy 429–430
and risk of childhood cancer 868
Muscle, storage of ectopic fat in 622
Muscle mass
creatinine levels in 27
skeletal, changes during growth 47
Muscle metabolic rates relative to age and BMR 186t
Muscle protein, status of 27

N

Nasogastric tube feeding in cancer patients 885
Necrosis, hepatocellular 749
Necrotizing enterocolitis, probiotics in 175
Neglect, child (*see* Abuse and neglect)
Neonatal gut
bacterial colonization of 509–512
immunoglobulins in 510–511
innate and adaptive immunity linked in 507
Neonatal immune system, educated by microbes in human milk 511
Neonatal Intensive Care Unit (NICU)
growth and nutritional deficits after discharge 553–556
nutritional management after discharge 553–563

Neonatal intestinal barrier, components of 505–506
Neonates
  drug clearance rates in 293i
  implications of fetal amino acids in 477–478
  recommendations for breastfeeding of 168
Neural tube defects (NTDs)
  and folate deficiency 286–288
  DNA methylation and 353–354
  epigenetic role of DNA methylation and 353–354
Neurodevelopmental disorders and feeding difficulties 702, 729–731
Neurological development after discharge from NICU 557
Neurological impairment, nutrition in children with 256t
Neurological involvement in celiac disease 816
Neutron activation analysis in measuring body composition 38–39
Niacin 154–155
  in human milk 495
  laboratory assessment of 33
NICU (*see* Neonatal Intensive Care Unit)
Nipple, normal 521i, 522i, 523i 524i, 532i, 534i
Nitrogen, non-protein, in human milk 491, 493
Nitrogen balance
  effects of fasting on 375i
  laboratory assessment of 27
  to determine protein or amino acid requirements 79
Nitrogen loss, calculating 27
N-nitroso compounds and brain tumors 868
Non-cystic fibrosis pancreatic diseases 837–839
Nongovernmental organizations (NGOs) in preventing malnutrition 233
Non-nutritive factors in intestinal adaptation 931–934
Nonprotein nitrogen (NPN) in human milk 491, 493
North American Indian cirrhosis 751
Nucleotides in breast milk vs. formula 571
Nursing (*see* Breastfeeding *and* Lactation)
Nutraceuticals, overview of 173–181
  cautions about 178–180
  nonphytochemical 177–178
  pediatric considerations in 177–180
  phytochemical 178–181
Nutrient balance in obesity 591–593
Nutrient densities in complementary foods, recommended 648t
Nutrient-drug interactions (*see also* Drug-nutrient interactions) 5t, 293–327, 309–311t
Nutrient effects on intestinal bacteria 343–344
Nutrient-enriched formula after NICU discharge 558–559

Nutrient functions, dietary references intakes for 167–168
Nutrient-gene interactions
signal transduction mediating 335i
therapeutic implications of 344–345
Nutrient-nutrient interactions 320t
Nutrient recognition and signal transduction 342–343
Nutrient requirements
by age, based on UNICEF report and DRI 647t
from complementary foods 646
in pancreatic insufficiency 845t
of infants and young children 646
Nutrient transition
and global child health 631–640
in developing countries 637–639
Nutrient transport
and drug absorption 296–297t, 309–316
in the gut 337
Nutrients
absorption of, secondary mechanisms in 307
and cell and tissue growth 362–363
and drug kinetics 309–305
and hormones and growth 361–371
changing view of 3
IGF regulation by 375
metabolism of 308–309
problem, in complementary foods 646
required in term infants 566–568
stimulating hormone secretion 332–334
storage of 188
genes regulating 331–332
Nutrition (*see also* Child nutrition)
amino acid in utero 465–482
and anthropomorphic data in developing countries 654–657
and cholesterol in children 977
child, linked with economic development, 653–661
structural changes affecting 659–660
community 243–269 (*see also* Community *entries*)
drug therapy interacting with 293–327
enteral (*see* Enteral nutrition) 1023–1034
epigenetics and 351–360
feeding difficulties and 699
fetal, carbohydrate and lipid metabolism in 445–463
gene expression and 329–349
historic overview of 1–4

humoral regulation of growth and 361–385
in cancer survivors 886–887
in cystic fibrosis 841–847
in disabled children 729–741
in exocrine pancreatic disorders 841–844
in low-birth-weight infants 394–398
in renal disease 769–786
in young athletes 1091–1098
inadequate, in growth retardation 538–539
individual, meal plans for 904–905
influencing DNA methylation 353–354
interacting with infection 206–207
maternal
and pregnancy outcome 425–433
and volume and composition of breast milk 642–643
in lactating women 525–527
later-life disease and 439–441
parenteral (*see* Parenteral nutrition) 3, 547–549, 1035–40
Nutrition programs, community, in Canada 262, 263t, 264t, 265t, 266t
Nutrition Support Teams (NSTs) 1147–1151
in assessing risk of malnutrition in hospitalized children 1148
Nutritional adequacy of breast milk 577
Nutritional assessment 5–17 (*see also* Appendix I on pp. 1153–1199 for pertinent charts and tables)
in childhood obesity 604–605
in children at risk for HIV infection 680–681
in disabled children 731
in exocrine pancreatic disorders 844–847
in liver disease 854–855
in pediatric cancer 882–884
in severe acute malnutrition 205–207
in weight-loss surgery 610
Nutritional complications of IBD 789–792
Nutritional deficiencies (*see specific nutrient or condition*)
Nutritional disease states and impaired bone health 1014–15
Nutritional disturbances, and adolescent bone health 1012–13
Nutritional epidemiology 271–291
Nutritional influences on bone health 1009–12
Nutritional interventions
for growth failure 720–722
in HIV-infected children 682–684
to mitigate dyslipidemia 929–930
Nutritional management
after discharge from NICU 553–563

after renal transplant 780, 781–782
in diabetes 896–901, 905
in glycogen storage diseases 758–759, 760–761, 763–764, 766
Nutritional needs of a hospitalized child 5–17
Nutritional properties of human milk 483–502
Nutritional provision in critically ill children 964–965
Nutritional recommendations
in infants, children, and adolescents in U.S. 250–253
Nutritional regulation of gene expression 329–343
Nutritional requirements (see Appendix II on pp. 1201–1214 for pertinent charts and tables)
for trace elements 96–97
in preterm infants 541–544
Nutritional screening
brief
dietary history in 5–6
medical history in 5–6
physical examination in 5–6
in community nutrition 247t, 247–248
Nutritional status (*See also* Macronutrient assessment; Micronutrient and Major mineral assessment)
clinical assessment of 5–17, 19t
during economic development 657–659
effect of drugs on 307
effect of liver transplantation on 859–860
emerging technologies in assessment of 34–35
hematological tests of 33–34
immunological tests of 34
laboratory assessment of 19–36
confounders in 19, 25–26
in critically ill children 964
maternal, and volume and composition of breast milk 642–643
serum IGF as index of 374
specific tests of 26–35
Nutritional studies of oral tissues and structures 988–989
Nutritional support
in intestinal failure/SBS 940–943
in liver disease 855–856
perioperative, in IBD 793
Nutritional treatment
for diabetes 896–907
for IBD 792–793, 794–796
for ulcerative colitis 792–793
Nutritive factors in intestinal adaptation 929–931

O

Obesity (*see also* Overweight) 3–4
affecting body composition 50, 51
alternative therapies for, information about 611
and drug availability 300–301
and iron deficiency 121
and risk of CVD 972
and therapy for malnutrition 239
as carbohydrate disorder 67–68
childhood (*see* Childhood obesity)
dietary prevention of 1138–40
drivers of, in developing economies 692–694
energy and substrate regulation in 583–597
evaluation of 602–606
genetic vs. environmental factors in 586
in children and adolescents (*see also* Childhood obesity) 588–612, 625–630
in developing economies 689–697
in glycogen storage diseases 762–763
in young athletes 1095–96
metabolic programming in 586–588, 600–601
monogenic causes of 601t
offset by nonexercise physical activity 191
pathophysiology of 600–601
potential link of fructose to 62t
risks of 584–586
in children 625–626
role of gut microbiota in 406–407
rural-to-urban migration in 694
substrate balance in 591–593
treatment of 593, 606–610
visceral, in MetS 615–616
oil-based emulsions in parenteral nutrition 1044–1046
Occlusion, catheter, in parenteral nutrition 1071–1072
25-OH-cholecalciferol, lab assessment of 32
Oligosaccharides 56, 59–61
in breast milk 569
lactose-derived, monosaccharide linkages in 61t
milk (*see also* Milk oligosaccharides) 58–59, 487–488
prebiotic
affecting the gut 59–61
linkage specificity of 61t
monosaccharide composition of 61t
Omega-3 fatty acids (*see also* Fatty acids, omega–3)

in critically ill children 966
in vegetarian diets 1138
to modify lipid profiles 979–980
Oral cavity
diseases of 989–992
soft tissues in 988
Oral feeding, mechanical functional development in 388–389
Oral-motor impairment in feeding difficulties 701, 729–730
Oral rehydration solutions (ORS)
choice of 920–921
composition of commercial preparations 913t
in severe malnutrition 921t
physiologic basis for 912–913
Oral rehydration therapy (ORT)
limitations of 918
obstacles to good practice of 922
Oral rinses, fluoride, dental benefits of 997
Oral tolerance in food allergies 824
Organs
genes controlling nutritional requirements of 330–331
metabolic rate in, relative to age and BMR 186t
protein synthesis rate in 75i
osteomalacia, factors contributing to 1013
outliers, statistical, in dietary analysis 282–283
relative contribution of, to body weight 49t
Osteoporosis 1013
in glycogen storage diseases 762
in IBD 790–791
predisposing conditions for 1010t
Overfeeding syndrome and RQ 29
Overnutrition 3–4
Overweight
dietary prevention of 1138–40
energy and substrate regulation in 583–597
in developing economies 689–697
prognostication for 45
Oxidation, fetal and placental 472–474
Oxygen consumption, fetal, glucose and carbon substrates in 451
Oxytocin release in lactation 521

P

Pancreatic agenesis 839
Pancreatic disease (*see also* Exopancreatic disorders) 835–839
Pancreaticobiliary secretion as defense mechanism in neonates 391

Pancreatitis, hereditary 838
Paneth cells in developing intestine 392
Pantothenic acid 159–160
  in human milk 496
Parasites in SAM 218
Parental behavior in feeding disorders 703
Parental factors in early feeding of infant 580
Parental stress as risk factor for failure to thrive 711
Parenteral nutrition 3, 547–549, 1035–1090
  as proxy for prevalence of IF/SBS 926
  catheter-related complications of 1069–72
  choline requirements for 1057
  chronic, and quality of life 944
  cyclic vs. continuous infusion in 1072–73
  drug metabolism and 306–307
  energy requirements in 1037–41, 1040t
  in preterm and VLBW infants 1040–41
  essential fatty acids needed in 1046–1048
  home-based 1073–75
  long-term monitoring of 1074, 1075t
  survival and mortality in 1074
  immediate establishment of 547
  in active Crohn’s disease 794
  in critically injured children 965
  in essential fatty acid deficiency 1046–1048
  in HIV-infected children 683
  in hospital setting, initiation and monitoring of 1064–65
  in liver disease 857–858
  in pediatric intensive care unit 1039–40
  in preterm infants 547–549, 1036–37
  initiating and monitoring in hospital and home 1064–65, 1073–75
  intravenous lipid emulsions in 1044–1949
  maintenance of trace elements during 97t, 394
  metabolic complications of 1065–1069
  biliary 1065, 1068–69
  bone 1065, 1069
  renal 1065, 1069
  peripheral 1075–76
  potentially detrimental in VLBW infants 394
  protein requirements in 1041–44
  -related side effects of chemotherapy 880–881t, 882i
  relative to MDGs 663
  risk of IFALD in 1973
  role of education in 233–234, 267

role of placenta in 440–441
routes of, in acute kidney injury 772
social development and 229–230
tracking 281
weaning from 1074
Parenteral nutrition solutions
carbohydrate requirements in 1049–51
delivery bags for 1078–79
fluid, electrolyte, and mineral requirements in 1051, 1052t
in VLBW infants 1054–1055
individually tailored vs. standard 1078–80
lipid requirements in 1044–1049
molybdenum in, cautions concerning 1061–62
multivitamin formulations in 1059t
safety of 1078
trace elements in 1058–62
cautions concerning 1059–62
unbalanced, and risk of IFALD 1067–68
vitamin requirements for 1055–58
Pathophysiology, hepatic 749
Pattern recognition receptors (PRRs) 174
and their ligands 174t
interacting with bacteria in presence of breast milk 509–510
Peanut-free diet 1103, 1105–06
Pearson's marrow pancreas syndrome 837, 838t
Pediatric Intensive Care Unit (PICU)
nutritional support in 1039–40
Pellagra 157
Peptides, antimicrobial
in gut 341
in human milk 508
Performance-enhancing substances in young athletes 1094–95
Perinatal growth failure
consequences of 538
risk factors for 708–709
Peroxysomal biosynthesis disorder 748t, 752–753
Pharmaceuticals (*see also* Drugs)
in human milk 498
Pharmacodynamic profiles of insulin preparations 892t
Pharmacokinetics (*see* Drug kinetics)
Pharmacologic approach to inflammatory syndrome 196–197
Pharmacologic interventions to lower lipids 980, 981t
Phenotype, obesity, genetic syndromes with 602t
Phenylalanine, tolerance to and exchange foods for 1121

Phenylketonuria
avoidance of high protein/phenylalanine in 1120–21
contraindications for sugarless gum in 998
dietary treatment of 1119–1121
permissible foods in 1121t
phenylalanine-free L-amino acid supplements in 1121
Phospholipids in human milk 491
Phosphorus in human milk 497
Physical activity
and body fat 45
assessment of 281–282
energy needs of 190,191–966
in diabetes 906
in obesity in developing economies 693–694
in program for weight maintenance 1140
linked to child nutrition 267
low, as risk factor for obesity 626
nonexercise, offsetting obesity 191
Physical examination
in assessing growth failure 716–719
in nutritional assessment 5–6
Physical stimulation in SAM 222
Physiologic function, amino acids in 74t
Physiology
of feeding 699–700
of lactation 485–486, 517–520
Phytochemicals, overview of 173–181
Phytoestrogens, potential bad effects of 179
Phytosterols and cholesterol levels 17
Pituitary-thyroid axis 377i
Placenta
fatty acid transfer to 89, 456–460
fetal-placental interrelationships 456i
glucose in formation of 446–456
role of, in birth weight and nutrition 440–441
Placental oxidation, amino acids and 472–474
Placental transfer of amino acids 466–472
Plaque related to dental caries 991–992
Plasma proteins
laboratory assessment of 28
reference range for 20–25t
Plasticity, developmental, biological basis for 436
Plethysmography, air displacement, in body composition assessment 13
Poisoning, iron 131–132

Policy recommendations and breastfeeding trends 484
Pollen food allergy syndrome 827–828
Polycomb repressive complex (PRC) 357
Polymeric diet in Crohn’s disease 795
Polyols in reducing dental caries 991–992
Polysaccharides 56
Polyunsaturated fatty acids
  in formula 571
  in severe acute malnutrition 208–209
  long-chain 88–89
  in liver disease 852–853
  supply in infants 90
Ponderal index at birth relative to CHD 437t
Population studies, anthropometric techniques in 40
Porphyria, hepatic disease due to 755
Postnatal growth relative to later disease risk 634–636
Postpyloric feeding devices 1028
Post-traumatic feeding disorder 702–703
Potassium
  in human milk 497
  total body 41
  in severe acute malnutrition 206
Potato Famine, Irish 1–2
Poverty
  and relationship of stunting and obesity in developing countries 692
  as risk factor for failure to thrive 710
  child health and 638
  reduction of, in preventing malnutrition 233
Prealbumin, laboratory assessment of 28
Prebiotic activity of oligosaccharides and glycoconjugates 507–508
Prebiotics
  affecting the gut 59–61, 60i, 63i
  definition of 176
  in diarrheal diseases 919
  monosaccharide composition of 61t, 62t
  overview of 173–181
Pregnancy
  calcium requirements in 427–428
  DHA supplied in 89–90
  FGR during 475–477
  fish oil supplements in 431
  folate supplementation in 287–288, 426–427
  food intake during 868
  herbal supplements in 430–431

multivitamin and mineral supplements in 429–430
and childhood cancer 868
need for iron in 118–119
placental amino acid transport during 470–477
protein gain in last trimester of 538i
Pregnancy outcomes and maternal nutrition 425–433
Pregnant women in U.S., community nutrition for 249–250
Pregnant youth, programs for 262
Premature infants
growth charts and tables 8–9
Prenatal considerations in breastfeeding 521
Prenatal growth and risk of adult disease 635–636
Prenatal gut, elements of immune system in 504i
Prenatal intake of fruits and vegetables, and childhood cancer 868
Prenatal risk exposure and leukemia 866
Preparation, safe, of processed foods 649
Presystemic clearance 295
Preterm infants (*see also* LBW and VLBW infants)
energy requirements in 1040–1041
enteral nutrition in 543t, 544–547
formula for 546
inpatient care of 537–551
mothers' and donors' milk in 545
nutritional management of 553–563
optimizing feeding practices in 546
patterns of growth after NICU discharge 556–557
tube feeding in 547
Prevalence estimates of obesity and overweight based on income group 691i
Prevention
of childhood obesity 625–630
recent trials in 628t
of severe acute malnutrition 223–224
Probiotics
in diarrheal diseases 919
in restoring intestinal hemostasis 175–176
modulating gut microbiota 407
overview of 173–181
safety concerns in infants 396
Processed complementary foods 648–649
Prognostic significance of overweight 4
Progressive familial intrahepatic cholestasis syndromes 748t, 749
Prolactin levels in let-down failure 533–534
Prolactin release in lactation 521
Protein(s)

acute phase, in SAM 207–208
airborne, in food allergies 827
amino acids and 469–470
and drug kinetics 303–304
antimicrobial in breast milk 508
dietary recommendations for 75–80
digestion and absorption of 75
distribution of, in various organs 75
immunologically active, in gut 340–342
in developing gut 395
in formula 570
in human milk 491–493
in vegetarian diets 1137
macronutrient assessment of 27–28
metabolism and distribution in 74–75
overview of 71–81
requirements of, for deposition 76t
Protein deficits in preterm infants 554–555
Protein deposition rates by age and sex, mean protein requirements for 76t
Protein digestion and absorption in exocrine pancreatic disorders 841
Protein-energy malnutrition
and community and government networks 233i
in preterm infants, irreversible effects of 541
Protein-energy supplementation to counter poor maternal health 666
Protein exchange with amino acid pools 751
Protein gain in last trimester of pregnancy 538I
Protein intake
during complementary food period 579
high, and risk of IBD 788
in breast-fed infants 74t, 569
in children with chronic kidney disease 777t
in liver disease 856–857
in severe acute malnutrition 205–206
Protein maldigestion in exocrine pancreatic disorders 839–841
Protein metabolism
and disease states 192–194
Crohn’s disease and 193
in critically ill children 958–960
in inflammatory bowel disease 801–802
in cancer 878
in liver disease 852
in rheumatoid arthritis 193
Protein needs in children by age 76t
Protein requirements

current 80
for term infants 568
in acute kidney injury 771
in chronic kidney disease 777–779
in critically injured children 959t
in diabetes 898–899
in preterm infants 541–542
in VLBW infants 1044
in young athletes 1092
methods for determining 79–80
Protein synthesis rate in organs 75i
Proteomics in assessing nutritional status 34–35
Psychosocial factors
in growth failure 710–713
in HIV-infected children 676–677
Psychosocial growth failure 376–377
Puberty
changes in bone growth during 49
changes in stature during 52
delayed, effects of malnutrition on 379
Public health programs
interventions to prevent obesity in developing economies 694–695
malnutrition as global problem in 232–240, 245
programs to improve nutrition in industrialized countries 262–267
Pulmonary complications of enteral feeding 1030
Pumping and storing milk during breastfeeding period 528–529

Q

Quality of life in patients with chronic parenteral nutrition 944, 1075
Questionnaire to assess eating disorder 1002t

R

Race, changes in stature relative to 50
Racial differences in body composition 45
Rapid early growth in utero as risk factor for obesity 626
Reaction mechanisms in food allergies 825
Ready-to-use therapeutic food (RUTF)
in SAM 213, 218–220
Recommended dietary allowances (RDAs) (*see also* Appendix II, pp. 1201–14)
in neonates 168
of iron 129–130
reference intakes 163, 164
Recommended dietary intake (RDI) (*see also* Appendix II, pp. 1201–14)

of carbohydrates 64–66
Rectal administration of drugs 295–296
Refeeding syndrome
in anorexic or starved children 1003
in SAM 221–222
Reference nutrient intakes (*see* Appendix II for detailed tables)
daily RDIs (U.S.) 1142t
daily RNIs (U.K.) 1141t
daily reference intakes (E.U.) 1143
Reference standards in disabled children 732, 733, 734
Rehabilitation in severe acute malnutrition
hospital vs. community 213–223
national guidelines for 229–242
Rehydration solutions, oral (*see* Oral rehydration solutions)
Rehydration therapy, oral (*see* Oral rehydration therapy)
Renal calculi in short bowel syndrome 939
Renal complications in parenteral nutrition 1065, 1069
Renal disease, nutrition in 769–786
Respiratory quotient (RQ) 15, 28
and overfeeding syndrome 28
clinical utility of 29
Respiratory reactions in food hypersensitivity 828
Respiratory system in SAM 210
Resting energy expenditure (REE) 14
and indirect calorimetry 15
predicting from body weight 739t
Resting metabolic rate (RMR), relative to fat-free mass 183, 184i, 185i
Resveratrol 356
Retinoids (*see also* Vitamin A) 144
in human milk 493
Retinol-binding protein (RBP), laboratory assessment of 28
Rheumatoid arthritis (RA), and protein metabolism 193
Riboflavin (*see also* Vitamin B_2) 153–154
in human milk 495
interconversion of co-factors and 153i
laboratory assessment of 33
Risk factors
for childhood obesity 625–627
for growth failure 708–713
Risk(s)
of adult disease, and fetal and early postpartum growth 635–636
potential, of high fructose intake 62, 62i
RNAs (*see* MicroRNAs)

S

Safety in preparation of complementary foods 649
Saint John's wort 178
Sample collection, variable factors in 26
Salt intake during complementary food period 579
Sanitation
  changing during economic development 657–659
  in global initiative to prevent morbidity 667–668
Schofield prediction equations 14, 15
School-based programs to promote health 267
School-based strategies to counter obesity in developing world 695–696
School feeding programs, effectiveness of 670
Screen time as risk for obesity 626
Screening (*see also* Nutritional screening)
  and diagnosis in growth failure 713–714
  cholesterol, in pediatric patients at risk for CVD 976–977
Scurvey and diet 151, 271–272
Sedentary behavior as risk factor for obesity 626
Selenium 105–108
Serum markers of bone turnover 1017
Severe acute malnutrition (SAM) (*see also* Malnutrition) 203–228
  clinical management of 212–224
  complications of 213–218
  criteria for diagnosis 203–204
  kwashiorkor in 204–205, 208, 209, 211, 222
  marasmus in 204
  oral rehydration for 921t
  pathophysiology of 205–212
  WHO-supported guidelines for 213–233
Sex
  relative to body composition by age and 46t
Sex steroids and cortisol 378–379
Sexual maturation in assessing body composition 13
Shellfish-free diet 1106–07
Short bowel syndrome (*see also* Intestinal failure)
  and intestinal failure and transplantation 925–956
  enteral nutrition in 1029
  management of and outcomes in 939–935
  medical support in 940–943
  renal calculi in 939
  surgical support in 943–944
  transplantation for 945–949
Shwachman-Diamond syndrome 835, 837, 838t

Sickle cell disease (SCD)
  chelation in 134
Signal transduction in nutritional gene expression 330i, 342–343
Singapore, interventions to prevent child obesity in 694–695
Skeletal maturation in assessing body composition 13
Skeletal muscle mass, changes during growth 47
Skin, hair, and teeth in SAM 211–212, 222
Skinfold thickness measurement
  correlated with BMI 277–278t
  in disabled children 736
  in measuring subcutaneous fat 40
Sleep, poor, as risk factor for obesity 626
Snacking in dental disease 991–992
Social class relative to hypertension and birth weight 441t
Social development, nutrition as key in 229–230
Socioeconomic development and nutrition transition 637–638
Sodium
  in diabetic diets 900
  in human milk 497
Sodium-restricted diet 1128, 1129t
Solid food(s) (*see also* Complementary foods)
  added to diet in breast-fed infants 527
Solutions for oral rehydration therapy 913, 921–922
Somatic proteins 27
Somatomedin(s)
  axis with GH 368i
  effects of fasting on 375i, 901
Somatomedin hypothesis 366–377
Sorbitol in diabetic diets 901
Soy-free diet 1102–03
Soy intake, adolescent, and breast cancer 873–874
Special diets (*see also specific diets*)
  cow’s milk-protein free 1110–1115
  egg-free 1100–02
  in young athletes 1095, 1099–1046
  lactose-free 1107–10
  low-phenylalanine 1119–21
  peanut-free, 1103, 1105–06
  shellfish-free 1106–07
  soy-free 1102–03
  tree-nut-free 1106
Special-needs children
  Head Start programs for 261
  nutrition for 254–255

Sphingolipids in human milk 491
Sports activities and body fat 45
Sports drinks vs. energy drinks 1094
Standardization of laboratory tests 26
Standardized BMI curves 40
Stanols in interventions for CVD 979–980
Starch 56, 901
  resistance to, in diabetic diets 901
Starvation as child abuse 713
Statistical models for assessing energy intake 283–284
Stature
  in disabled children 732–733
  in linear growth 50
  in measuring nutritional status 7–8
Steatosis, 752
Steroids
  anabolic
  in catabolic disease 379
  in young athletes 1094–95
  vs. enteral nutrition in Crohn’s disease 795–796
Sterols
  defects in synthesis of 748t
  in human milk 491
Stimulants, appetite, in HIV-infected children 683–684
Storage
  intracellular, of iron 122–124
  of pumped milk 528–529
Storage disorders, hepatic disease due to 747t, 754–755
Strategies
  for delivering targeted interventions 669–670
  school-based, to prevent obesity in developing countries 695–696
  to promote exclusive breastfeeding 642
Streptococcus mutans in dental disease 989–992
Stress, parental, as risk factor for growth failure 711
Stress response, metabolic, in critically ill children 957
Structural changes affecting child nutrition 659–660
Stunting
  and obesity, economic relationship between 692
  anthropometric data on, and income 654–657
  as side effect of cancer therapy 887
  prevalence of 664i
Substrate, energy
  effects of fasting on 192
  regulation in obesity 583–597

Suck-swallow coordination, development of 383
Suckling disorders in infants 530–531
Sucrase isomaltase deficiency 1118
Sucrose diets
  in diabetes 900
  in sucrase isomaltase deficiency 1118
Sucrose intake, adaptation of enterocytes to 336
Sugar(s) (*see also specific sugar*) 56
  during complementary food period 577
Sugar alcohols in diabetic diets 900–901
Sugar-containing food and dental disease 990–991
Sugar-sweetened drinks as risk factor for obesity 626
Sulfur dioxide and sulfites as allergens, sources of 1107
Supplemental feedings in chronic kidney disease 775–776
Supplemental nutrients, amino acid, caveats concerning 80–81
Supplements
  DHEA, cautions against 1014
  fluoride, to prevent caries 996–997
  formula, kidney-specific 773t
  herbal, during pregnancy 430–431
  micronutrient 649, 665, 667
  multivitamin, and childhood cancer 868
  oral, enteral, and parenteral, in HIV-infected children 682–683
  protein-energy, to counter poor maternal health 666
  vitamin and mineral 319–322
  during breastfeeding 525
  during pregnancy 428–430
  in chronic kidney disease 778–779
  in liver disease 857
  in SAM 221
  with vaccination 659–660
  zinc, in diarrheal diseases 920
Suppressants, appetite 319
Surgery
  in children with IF/SBS 943–944
  weight-loss 609–610
Survivors of pediatric cancer, nutrition in 886–887
Sweeteners in diabetic diet 900–901
Sympathetic nervous system (SNS) and energy expenditure 188

T

Tanner staging
  in assessing body composition 13
Taurine in formula 571

Teeth
hypoplasia in 988–989
in SAM 211–212
Temperature, body (*see* Body temperature)
Temporal difference in interpreting laboratory values 25
Term infant 565–573
Thermic effect of feeding (TEF) 187–188
Thermogenesis 189–190
Thiamine (*see also* Vitamin B_1) 152–153
in human milk 495
laboratory assessment of 32
Thrombosis, catheter-related 1071–73
Thyroid hormones 377–378
and nutrients 362–363
critical periods in 363–364
Tissue(s)
major, contributing to body weight 48t
oral cavity
mineralized 988–989
soft 988
Tissue growth
and nutrients 362–363
critical periods in 363–364
Tobacco, and adolescent bone health 1013
Tolerable upper intake levels 163t, 167
Tolerance, oral, in food allergies 824
Tomography, quantitative computed, to assess BMD 1016–17
Toothpaste, fluoride, benefits/cautions in using 996–997
Total body water, estimation of 38–39
Total energy expenditure (TEE) 14
Toxic side effects of dietary supplements 179
Toxicity
amino acid 70–71
chromium 108
copper 104–105
iodine 113
iron 131–132
manganese 857–858
mineral, in liver disease 857–858
of excess fluoride 992–994
selenium 107
zinc 101
Trace elements (*see also individual element*) 95–116
biologic roles of 95

chromium 108–110
copper 102–105
deficiencies in 96–97
clinical features of 98–99, 103, 106–107, 108–109
lab analyses to detect 97–98
dose-response of 95–96
fluoride 111–112
iodine 112–113
in common foods 102t
in HIV-infected children 680
in human milk 497
in parenteral nutrition 1058–62
interacting with nutrients 312
manganese 110–111
metabolism of, inborn errors of 96
nutritional requirements for 97
selenium 105–108
supplemental 319–322
Trace element requirements for solutions 1060t
Trace mineral metabolism in critically ill children 963
Transferrin
laboratory assessment of 28
receptor, in screening for iron deficiency 137–138
Transforming growth factor-beta (TGF-β)
immune education by 511–512
Transplantation
for intestinal failure and short bowel syndrome 945–949
graft types used 946–947
indications for 946t
nutritional care afterward 947–948
outcomes in 948–949
hematopoietic stem cell, in cancer 881t
kidney, management after 780–782
liver, affecting nutritional status 859–860
Transport, iron 125
Transport mechanisms of carbohydrates and lipids in placenta and fetus 445–460
Transport systems, amino acid, and abnormal pregnancy outcomes 475–477
Tree-nut-free diet 1106
Triage for rehabilitation in SAM 213
Triceps skinfold thickness 12, 40
Triglycerides
medium-chain
and fat absorption 84–85
energy content of 85i

Tube feeding (*see also* Enteral nutrition)
in cancer patients 885
in cystic fibrosis 847
in preterm infants, administration and tolerance of 547
mechanical complications of 1030–31
nasogastric, in cancer 885
Tube placement in, bedside evaluation of 1030t
Tumors (*see also specific locus*)
Twenty-four-hour energy expenditure (24hEE), assessment of 190
Twenty-four-hour recall in dietary assessment 272–273
Tyrosinemia in early infancy 752

U

Ulcerative colitis, nutritional therapy for 792–793
Umbilical uptake of amino acids 472
Underfeeding affecting energy expenditure 191
Undernutrition (*see also* Malnutrition)
and economic development 654–657
and mortality risk 632–634
and susceptibility to communicable diseases 636
effects of, on GH-IGF axis 374t
food insecurity and 670
global interventions to prevent 665–669
in IBD 799
prevalence by age 709t
protein-energy, irreversible effects of 541
Underweight, prevalence of 664i
United States
community nutrition and programs in 243–267
dietary references intake for 163–171
Urbanization
affecting dietary patterns 637–638
as factor in child obesity 694
Urea cycle and related diseases 745–746t, 751
Urticaria as hypersensitivity reaction to food 826

V

Vaccination
against measles, relative to income 660i
in developing economies 659, 669
Valerian root 179
Variables, uncontrolled, in determining nutritional intake 25
Variation, intraindividual and analytical, in interpreting lab results 25–26

Vascular access to parenteral nutrition solutions 1062–1063
Vegan diet, default, as result of poverty 657
Vegetables
  and drug kinetics 304–305
  sucrose contents of 1118t
Vegetarian diets
  health benefits of 1138
  in infants 579
Vegetarianism 1135, 1137
  and drug metabolism 305
Very-low-birth weight (VLBW) infants (*see also* Low-birth-weight infants; Preterm infants)
  energy requirements in 1040–1041
  fluid, electrolytes, and minerals required for 1054–1055
  intravenous lipid emulsions in 1048–49
  protein requirements in 1044
Very-low-fat diet 1125
Very-low-fructose diet for hereditary fructose intolerance 1122–23
Virus, human immunodeficiency (*see* HIV entries)
Visceral mass, changes during growth 47–48
Visceral obesity in the MetS syndrome 615–616
Vitamin A (*see also* Retinoids) 143–146
  in HIV-infected children 679–680
  laboratory assessment of 31, 32
  supplementing measles vaccine 659–660
  visual cycle 145i
Vitamin B_1 (*see also* Thiamine) 152–153
  laboratory assessment of 33
Vitamin B_2 (*see also* Riboflavin) 153–154
  laboratory assessment of 33
Vitamin B_3 (*see also* Niacin)
  laboratory assessment of 33
Vitamin B_5 (*see* Pantothenic acid)
Vitamin B_6, 155–156
  -dependent enzymes in carbon metabolism 155i
  in human milk 495
  laboratory assessment of 33
Vitamin B_{12}
  fortifying food with 287–288
  in HIV-infected children 680
  in human milk 495
  laboratory assessment of 33
Vitamin C

in human milk 495
laboratory assessment of 33
Vitamin D 146–148
and adolescent bone health 1011–1012
DRI/RDA for 1012t
in human milk 493–494
in severe acute malnutrition 212
laboratory assessment of 32
metabolism of 147i
supplementation
during pregnancy 428
in chronic kidney disease 778–779
Vitamin deficiency 30
epigenetic role of SNA methylation in 354
in CF-related bone disease 844
Vitamin E
in human milk 494
laboratory assessment of 32
Vitamin K
in blood coagulation cascade 150i
in human milk 496–497
laboratory assessment of 32
Vitamin K deficiency bleeding (VKDB) 151
Vitamin malabsorption disorders 748t
Vitamin metabolism in critically ill children 963
Vitamin requirements in parenteral nutrition 1055–1059
Vitamins 143–162
drug interactions with 307–309, 320–321t
fat-soluble 31–32
food sources for 167–168
in HIV-infected children 679–680
in human milk 493–496
needed in young athletes 1093
reference ranges for 20–25t
supplementation with 319–322
during pregnancy 428–430
in SAM 221
to prevent NTDs 286–287
in vegetarian diets 1138
Vitamin and mineral supplementation
in children with allergies to milk 1108
in liver disease 857
von Gierke disease 759–763

W

Waist circumference relative to ethnicity and gender 614–615t
Wasting
  in childhood cancer 878
  in HIV-infected children 674–677
  related to income 654–657
Water
  as percentage of body composition by age and sex 44t
  fluoridation of, dental benefits of 995–996
  in preventing disease in at-risk children 667
Water loss, insensible, in preterm infants 541t
Water-soluble vitamins, laboratory assessment of 32–33
Water supply
  access to, relative to income 657–659
  sanitary, in preventing infectious diseases 657
Waterlow criteria for categorizing malnutrition 708t
Weaning
  from breastfeeding 527–528
  complementary foods in 571–572
  failure of 700
  premature 534
  from parenteral nutrition 1074
Web-based resources
  for community nutrition 262–265
  for treatment of obesity 610–611
Weight
  birth, affecting health in developing countries 690–692
  body (*see* Body weight)
  for height and length 10–11
  in disabled children 732–34
  in measuring nutritional status 7
  loss or gain, during development 52
Weight control to prevent obesity 1138–1140
Weight gain
  as risk factor for diabetes 895
  drugs causing, in obese children 604t
  in reference infant 567
  poor, in infants 530–531
Weight loss, therapy for 608–611
Weight-related behavior in young athletes 1094–96
Whey in human milk 492
Women, status of, and alleviation of malnutrition 232
World Health Organization (WHO)

criteria for assessing SAM 205–206, 222
guidelines for oral rehydration 912
guidelines undergirding protocols for treating SAM 213–233
manual for aids in physical and emotional stimulation 222
Worms, screening for, to target intervention 668

X

Xylitol
in dental disease 997–998
in diabetic diets 901

Z

Zellweger disease 752–753
Zinc (*see also* Trace elements)
in human milk 497
interacting with nutrients 312
requirements during complementary food period 579
supplementation
in at-risk children 667
in diarrheal diseases 920
Zinc deficiency 98–102
neuropsychological consequences of 420

目录

Cover 1
Title Page 2
Copyright Page 3
Sales and Distribution 5
Dedications 7
Contents 8
Contributors 11
Preface 26
Part I: General Concepts 28
Chapter 1: Nutrition: The Driving Force for Health, Disease Prevention, and Development 28
Chapter 2: Clinical Assessment of Nutritional Status 38
Chapter 3: Laboratory Assessment of Nutritional Status 71
Chapter 4: Body Composition and Growth 110
Chapter 5: Carbohydrates 153
Chapter 6: Proteins and Amino Acids 187
Chapter 7: Macronutrient Requirements for Growth: Fats and Fatty Acids 211
Chapter 8: Trace Elements 238
Chapter 9: Iron 294
Chapter 10: Vitamins 353
Chapter 11: Dietary Reference Intakes in the United States 403
Chapter 12: Functional Food Ingredients: Probiotics, Prebiotics, Nutraceuticals, and Phytochemicals 424
Part II: Physiology and Pathophysiology 447
Chapter 13: Energy Requirements in Health and Disease 447
Chapter 14: Severe Acute Malnutrition: Pathophysiology, Clinical Consequences, and Treatment 496
Chapter 15: Childhood Malnutrition: Prevention and Control at the National Level 558
Chapter 16: Community Nutrition and Its Impact on Children: 591

Highly Industrialized Countries 591

Chapter 17: Nutritional Epidemiology 653
Chapter 18: Drug Therapy and the Role of Nutrition 702
Part III: Perinatal Nutrition 777
Chapter 19: Nutrition and Gene Expression 777
Chapter 20: Nutrition and Epigenetics 830
Chapter 21: Nutrition and the Humoral Regulation of Growth 855
Chapter 22: Gastrointestinal Development: Implications for Infant Feeding 915
Chapter 23: Diet, Environment, and Gut Microbiota in the Metagenomic Era 945
Chapter 24: Nutrition and Neuropsychological Development 970
Chapter 25: Maternal Nutrition and Pregnancy Outcome 1003
Chapter 26: Human Growth and Disease in Later Life 1026
Chapter 27: Nutrition and Development of the Fetus: Carbohydrate and Lipid Metabolism 1047
Chapter 28: Amino Acid Nutrition in Utero: Placental Function and Metabolism 1092
Chapter 29: Human Milk: Nutritional Properties 1137
Chapter 30: Protective Properties of Human Milk and Bacterial Colonization of the Neonatal Gut 1184
Chapter 31: Approach to Breastfeeding 1218
Chapter 32: The Low-Birthweight Infant: Inpatient Care 1263
Chapter 33: Nutritional Management of Preterm Infants Post-Discharge 1293
Chapter 34: The Term Infant 1317
Chapter 35: Complementary Feeding and Health Outcomes 1337
Part IV: Obesity 1356
Chapter 36: Energy and Substrate Regulation in Obesity 1356
Chapter 37: Evaluation and Management of Childhood Obesity 1391
Chapter 38: The Metabolic Syndrome in Children and Adolescents 1423
Chapter 39: Childhood Obesity Prevention 1448
Part V: Nutrition in Low- and Middle-Income Countries 1461

Chapter 40: The Nutrition Transition and Global Child Health 1461
Chapter 41: Infant and Young Child Feeding in Low- and Middle-Income Countries 1483
Chapter 42: Child Nutrition and Economic Development 1511
Chapter 43: Global Interventions in Child Health 1530
Chapter 44: Pediatric HIV Infection 1552
Chapter 45: Obesity in Developing Economies 1588
Part VI: Nutritional Aspects of Specific Disease States 1608
Chapter 46: Feeding Difficulties 1608
Chapter 47: Failure-to-Thrive: Growth Failure in the Outpatient Setting 1626
Chapter 48: Developmental Disabilities 1672
Chapter 49: Inborn Errors of Metabolism and the Liver 1706
Chapter 50: Inborn Errors of Fasting Adaptation—Glycogen Storage Disease 1734
Chapter 51: Acute and Chronic Kidney Disease: Nutrition in Renal Disease 1762
Chapter 52: Inflammatory Bowel Disease 1807
Chapter 53: Celiac Disease 1861
Chapter 54: Food Allergies 1892
Chapter 55: Exocrine Pancreas 1919
Chapter 56 Acute and Chronic Liver Disease 1956
Chapter 57 Cancer Prevention 1987
Chapter 58 Cancer Treatment 2017
Chapter 59 Diabetes Mellitus 2049
Chapter 60 Diarrheal Diseases 2099
Chapter 61 Intestinal Failure, Short Bowel Syndrome, and Intestinal Transplantation 2132
Chapter 62 The Critically Ill Child 2208
Chapter 63 Hyperlipidemia and Cardiovascular Disease 2239
Chapter 64 Diet and Dental Disease 2275
Chapter 65 Adolescent Eating Disorders 2307
Chapter 66 Adolescence: Advancing Bone Health 2320
Part VII: Approach to Nutritional Support 2360
Chapter 67 Enteral Nutrition and Formulas 2360

Chapter 68 Parenteral Nutrition 2389
Chapter 69 Nutrition in the Athlete 2527
Chapter 70 Special Diets 2548
Chapter 71 Nutrition Support Teams 2642
Appendices 2654
Appendix i: Nutritional Assessment 2654
Appendix ii: Nutritional Requirements 2706
Appendix iii: Enteral Products 2720
Index 2730