SECOND EDITION

Fetal Therapy

Scientific Basis and Critical Appraisal of Clinical Benefits

EDITED BY Mark D. Kilby, Anthony Johnson and Dick Oepkes

CAMBRIDGE Medicine

Fetal Therapy

Second Edition

Fetal Therapy

Scientific Basis and Critical Appraisal of Clinical Benefits

Second Edition

Edited by

Mark D. Kilby
University of Birmingham

Anthony Johnson
University of Texas Health Science Center, Houston

Dick Oepkes
Leiden University Medical Center

CAMBRIDGE
UNIVERSITY PRESS

CAMBRIDGE
UNIVERSITY PRESS

University Printing House, Cambridge CB2 8BS, United Kingdom

One Liberty Plaza, 20th Floor, New York, NY 10006, USA

477 Williamstown Road, Port Melbourne, VIC 3207, Australia

314–321, 3rd Floor, Plot 3, Splendor Forum, Jasola District Centre, New Delhi – 110025, India

79 Anson Road, #06–04/06, Singapore 079906

Cambridge University Press is part of the University of Cambridge.

It furthers the University's mission by disseminating knowledge in the pursuit of education, learning, and research at the highest international levels of excellence.

www.cambridge.org
Information on this title: www.cambridge.org/9781108474061
DOI: 10.1017/9781108564434

First published 2020

Printed in Singapore by Markono Print Media Pte Ltd

A catalogue record for this publication is available from the British Library.

Library of Congress Cataloging-in-Publication Data
Names: Kilby, Mark, editor. | Johnson, Anthony, 1954– editor. | Oepkes, Dick, editor.
Title: Fetal therapy : scientific basis and critical appraisal of clinical benefits / edited by Mark D. Kilby, Anthony Johnson, Dick Oepkes.
Other titles: Fetal therapy (Kilby)
Description: Second edition. | Cambridge, United Kingdom ; New York, NY : Cambridge University Press, 2020. | Includes bibliographical references and index.
Identifiers: LCCN 2019008058 | ISBN 9781108474061 (hardback : alk. paper)
Subjects: | MESH: Fetal Diseases–therapy | Fetal Therapies–methods
Classification: LCC RG626 | NLM WQ 211 | DDC 618.3/2–dc23
LC record available at https://lccn.loc.gov/2019008058

ISBN 978-1-108-47406-1 Hardback

Additional resources for this publication at www.cambridge.org/9781108474061.

To the patients and families who entrust us with their most precious possession, their developing child, and those who have been our teachers and mentors over the years. A special thank you to each of our families for their support, tolerance, and understanding.

Contents

Contributors

Nimrah Abbasi MD
Fetal Medicine Unit, Ontario Fetal Centre, Mount Sinai Hospital, and Division of Maternal-Fetal Medicine, Department of Obstetrics and Gynaecology, University of Toronto, Toronto, Canada

N. Scott Adzick MD
The Center for Fetal Diagnosis and Treatment, The Children's Hospital of Philadelphia, Philadelphia, PA, USA

Karel Allegaert MD, PhD
Unit Woman and Child, Department of Development and Regeneration, Group Biomedical Sciences, Katholieke Universiteit Leuven, Leuven, Belgium

David W. Barrett PhD
Institute of Bioengineering, School of Engineering and Materials Science, Queen Mary University of London, London, UK

Margot M. Bartelings MD, PhD
Department of Anatomy and Embryology, Leiden University Medical Center, Leiden, the Netherlands

Ahmet A. Baschat MD
Johns Hopkins Center for Fetal Therapy, Department of Gynecology and Obstetrics, Johns Hopkins University School of Medicine, Baltimore, MD, USA

David Basurto MD
Department of Development and Regeneration, Cluster Woman and Child, and University Hospitals Leuven, KU Leuven, Leuven, Belgium

David Baud MD, PhD
Materno-Fetal and Obstetrics Research Unit, Department Woman – Mother – Child, Lausanne University Hospital, Lausanne, Switzerland

Marie H. Beall MD
Department of Obstetrics and Gynecology, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA

Michael A. Belfort MBBCH, MD, PhD
Department of Obstetrics and Gynecology (courtesy appointments in the Departments of Neurosurgery and Surgery) Baylor College of Medicine and Texas Children's Hospital Fetal Center Houston, Texas

Mar Bennasar PhD
Fetal Medicine Reseach Center, BCNatal, Hospital Clinic and Hospital Sant Joan de Déu, University of Barcelona, and Institut d'Investigacions Biomèdiques August Pi I Sunyer (IDIBAPS), Barcelona, Spain

Phillip Bennett BSc, PhD, MD, FRCOG, FMedSci
Institute for Reproductive and Developmental Biology and Department of Obstetrics and Gynaecology, Imperial College London and Imperial College Healthcare NHS Trust, London, UK

Guillaume Benoist MD
Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Nico A. Blom MD, PhD
Department of Pediatric Cardiology, Amsterdam University Medical Center, Amsterdam, and Leiden University Medical Center, Leiden, the Netherlands

Janet E. Brennand MD, FRCOG
The Ian Donald Fetal Medicine Centre, The Queen Elizabeth University Hospital, Glasgow, UK

David W. Britt PhD
Fetal Medicine Foundation of America, New York, NY, USA

Suzanne M. K. Buckley BSc (Hons), PhD
Elizabeth Garrett Anderson Institute for Women's Health, University College London, London, UK

Julene S. Carvalho MD, PhD, FRCPCH
Brompton Centre for Fetal Cardiology, Royal Brompton Hospital; Fetal Medicine Unit, St. George's University Hospital and Molecular and Clinical Sciences Research Institute, St. George's, University of London, London, UK

Gihad E. Chalouhi MD
National Reference Centre for the Management of Complicated Monochorionic Pregnancies, and Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Min Chen PhD
Department of Fetal Medicine and Prenatal Diagnosis, The Third Affiliated Hospital of Guangzhou Medical University, Obstetrics and Gynecology Institute of Guangzhou, The Medical Centre for Critical Pregnant Women in Guangzhou, Key Laboratory for Major Obstetric Diseases of Guangdong Province, and Key Laboratory for Reproduction and Genetics of Guangdong Higher Education Institutes, Guangzhou, China

K. W. Cheung MBBS, MRCOG
Birmingham Women's and Children's Hospital, Birmingham, UK; and Department of Obstetrics and Gynaecology, Queen Mary Hospital, University of Hong Kong, Hong Kong SAR, China

Hsu Phern Chong PhD
Fetal Medicine Centre, Birmingham Women's & Children's NHS Foundation Trust, Birmingham, UK

Tina T. Chowdhury PhD, SFHEA
Institute of Bioengineering, School of Engineering and Materials Science, Queen Mary University of London, London, UK

Claire L. Colmant MD
National Reference Centre for the Management of Complicated Monochorionic Pregnancies, and Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Isabel Couck MD
Department of Obstetrics and Gynecology, University Hospitals Leuven, and Department of Development and Regeneration, Biomedical Sciences, Katholieke Universiteit Leuven, Leuven, Belgium

Timothy M. Crombleholme MD
Fetal Care Center Dallas, Medical City Children's Hospital, Dallas, TX, USA

Jenifer Curtis ARDMS
Fetal Medicine Foundation of America, New York, NY, USA

Nicolas Dauby MD, PhD
Department of Infectious Diseases, CHU Saint-Pierre, and Institute for Medical Immunology, Université Libre de Bruxelles, Brussels, Belgium

Anna L. David MBChB, PhD, FRCOG
Elizabeth Garrett Anderson Institute for Women's Health, University College London, and National Institute for Health Research University College London Hospitals Biomedical Research Centre, London, UK

Joseph Davidson MBBS, MRCS
Stem Cell and Regenerative Medicine Section, Great Ormond Street Institute of Child Health, University College London, London, UK

Luc De Catte MD, PhD
Fetal Diagnosis and Therapy Unit, Division of Woman and Child, Department of Obstetrics and Gynecology, University Hospitals Leuven, Leuven, Belgium

Paolo De Coppi MD, PhD
Stem Cell and Regenerative Medicine Section, Great Ormond Street Institute of Child Health, University College London, London, UK

Guido de Wert MD
Department of Health, Ethics and Society, Faculty of Health, Medicine and Life Sciences, Research Schools of CAPHRI and GROW, Maastricht University, Maastricht, the Netherlands

Anne Debeer MD, PhD
Division of Woman and Child, Department of Neonatology, University Hospitals Leuven, Leuven, Belgium

Jan Deprest MD, PhD, FRCOG
Fetal Diagnosis and Therapy Unit, Division of Woman and Child, Department of Obstetrics and Gynecology, University Hospitals Leuven, Leuven, Belgium; and Department of Maternal Fetal Medicine, Institute for Women's Health, University College London, London, UK

Roland Devlieger MD, PhD
Department of Development and Regeneration, Cluster Woman and Child, and University Hospitals Leuven, KU Leuven, Leuven, Belgium

Koen Devriendt MD, PhD
Department of Human Genetics, University Hospitals Leuven, KU Leuven, Leuven, Belgium

Jodie Dodd MB BS, PhD, FRANZCOG, CMFM
Discipline of Obstetrics and Gynaecology, Women's and Children's Hospital, North Adelaide, SA, Australia

Wybo J. Dondorp MD
Department of Health, Ethics and Society, Faculty of Health, Medicine and Life Sciences, Research Schools of CAPHRI and GROW, Maastricht University, Maastricht, the Netherlands

Sascha Drewlo PhD
Department of Obstetrics and Gynecology, Michigan State University, Grand Rapids, MI, USA

Alex J. Eggink MD, PhD
Department of Obstetrics and Gynecology, Division of Obstetrics and Fetal Medicine, Erasmus MC, University Medical Center Rotterdam, Rotterdam, the Netherlands

Elisenda Eixarch PhD
Fetal Medicine Reseach Center, BCNatal, Hospital Clinic and Hospital Sant Joan de Déu, University of Barcelona and Institut d'Investigacions Biomèdiques August Pi I Sunyer (IDIBAPS), Barcelona; and Centre for Biomedical Research on Rare Diseases (CIBER-ER), Madrid, Spain

Åsa Ekblad PhD
Division of Obstetrics and Gynecology, Department of Clinical Science, Intervention and Technology, Karolinska Institutet, Stockholm, Sweden

Mark I. Evans MD
Fetal Medicine Foundation of America; and Comprehensive Genetics, Mount Sinai School of Medicine, New York, NY, USA

Shara M. Evans MSc, MPH
Department of Maternal and Child Health, Gillings School of Public Health, University of North Carolina, Chapel Hill, NC, USA

Alan W. Flake MD
Division of General, Thoracic and Fetal Surgery, Children's Hospital of Philadelphia, Philadelphia, PA, USA

Vicki Flenady RM, PhD
Centre of Research Excellence in Stillbirth, Mater Research Institute, University of Queensland, Brisbane, Australia

Philippa Francis-West BA, PhD
Cell and Developmental Biology, Centre for Craniofacial and Regenerative Biology, King's College London, London, UK

Helena M. Gardiner MD, PhD, FRCP, FRCPCH, DCH (retired)
Department of Obstetrics and Gynecology, McGovern Medical School, University of Texas Health Sciences Center, Houston, TX, USA

Janice L. Gibson MD, MRCOG
The Ian Donald Fetal Medicine Centre, The Queen Elizabeth University Hospital, Glasgow, UK

Adriana C. Gittenberger-de Groot PhD
Department of Cardiology, Leiden University Medical Center, Leiden, the Netherlands

Cecilia Götherström PhD
Division of Obstetrics and Gynecology, Department of Clinical Science, Intervention and Technology, Karolinska Institutet, Stockholm, Sweden

Eduard Gratacós PhD
Fetal Medicine Reseach Center, BCNatal, Hospital Clinic and Hospital Sant Joan de Déu, University of Barcelona and Institut d'Investigacions Biomèdiques August Pi I Sunyer (IDIBAPS), Barcelona; Institut de Recerca Sant Joan de Déu, Esplugues de Llobregat and Centre for Biomedical Research on Rare Diseases (CIBER-ER), Madrid, Spain

Lucy R. Green BSc, PhD
Assistant Director, Institute of Developmental Sciences, University of Southampton, University Hospital Southampton, Southampton, UK

Mark A. Hanson MA, DPhil, CertEd, FRCOG
British Heart Foundation Professor, Director, Institute of Developmental Sciences, University of Southampton, University Hospital Southampton, Southampton, UK

Alexander Heazell PhD, MRCOG
Maternal and Fetal Health Research Centre, School of Medical Sciences, Faculty of Biology, Medicine and Health, University of Manchester, and St. Mary's Hospital, Manchester University NHS Foundation Trust, Manchester Academic Health Science Centre, Manchester, UK

Gregory G. Heuer MD
The Center for Fetal Diagnosis and Treatment, The Children's Hospital of Philadelphia, Philadelphia, PA, USA

Alice E. Hughes BSc (Hons), BMBS, MSc
Department of Obstetrics and Gynaecology, University of Cambridge, Cambridge, UK

Edgar Jaeggi MD, FRCP(C)
Fetal Cardiac Program, Labatt Family Heart Center, Hospital for Sick Children, University of Toronto, Toronto, Canada

Monique R.M. Jongbloed MD, PhD
Departments of Anatomy and Embryology and Cardiology, Leiden University Medical Center, Leiden, the Netherlands

Brenda M. Kazemier MD, PhD
Department of Obstetrics and Gynecology, Amsterdam UMC, University of Amsterdam, Amsterdam, the Netherlands

Sarah Keating MD
Department of Laboratory Medicine and Pathobiology, University of Toronto, and Mount Sinai Hospital, Toronto, Canada

Sundeep G. Keswani MD
Fetal Center, Division of Pediatric General, Thoracic and Fetal Surgery, Texas Children's Hospital and Baylor University School of Medicine, Houston, TX, USA

Asma Khalil MBBCh, MD, MRCOG, MSc (Epi), DFSRH, Dip (GUM)
Fetal Medicine Unit, St George's Hospital NHS Foundation Trust, London, UK

Mark D. Kilby DSc, MD, FRCOG, FRCPI
Institute of Metabolism and Systems Research, University of Birmingham, and Birmingham Women's Hospital NHS Foundation Trust, Birmingham, UK

John Kingdom MD
Maternal-Fetal Medicine Division, Mount Sinai Hospital, and Department of Obstetrics and Gynaecology, University of Toronto, Toronto, Canada

Marianne Leruez-Ville MD
Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Tak Yeung Leung MD FRCOG
Department of Obstetrics and Gynaecology, Faculty of Medicine, The Chinese University of Hong Kong, Hong Kong

Liesbeth Lewi MD, PhD
Fetal Diagnosis and Therapy Unit, Division of Woman and Child, Department of Obstetrics and Gynecology, University Hospitals Leuven, and Department of Development and Regeneration, Biomedical Sciences, Katholieke Universiteit Leuven, Leuven, Belgium

David Lissauer PhD, MBChB
Malawi-Liverpool-Wellcome Research Institute, Blantyre, Malawi; and Institute of Translational Medicine, University of Liverpool, Liverpool, UK

Enrico Lopriore MD, PhD
Division of Neonatology, Department of Pediatrics, Leiden University Medical Center, Leiden, the Netherlands

Fiona L. Mackie MBChB, MRes, PhD
Obstetrics and Gynaecology Academic Department, Birmingham Women's Hospital NHS Foundation Trust, Birmingham, UK

Eamonn R. Maher MD
Academic Department of Medical Genetics, Addenbrooke's Treatment Centre, Addenbrooke's Hospital, Cambridge, UK

Katarzyna M. Maksym MD, MRCOG
Institute for Women's Health, University College London, London, UK

Arnaud Marchant MD, PhD
Institute for Medical Immunology, Université Libre de Bruxelles, Brussels, Belgium

Josep Maria Martinez PhD
Fetal Medicine Reseach Center, BCNatal, Hospital Clinic and Hospital Sant Joan de Déu, University of Barcelona and Institut d'Investigacions Biomèdiques August Pi I Sunyer (IDIBAPS), Barcelona; and Centre for Biomedical Research on Rare Diseases (CIBER-ER), Madrid, Spain

Fergus P. McCarthy MB ChB, PhD, MRCOG
Anu Research Centre, Department of Obstetrics and Gynaecology, University College Cork, Cork, Ireland

Dominic McMullan PhD
West Midlands Regional Genetics Laboratory, Birmingham Women's and Children's NHS Foundation Trust, Birmingham, UK

Catherine L. Mercer BA, BM, PhD, MRCPCH
Centre for Human Development, Stem Cells and Regeneration, Faculty of Medicine, University of Southampton, Southampton, UK

Isabelle Miletich DDS, BSc, MSc, PhD
Centre for Craniofacial and Regenerative Biology, King's College London, London, UK

Tim J. Mohun PhD
The Francis Crick Institute, London, UK

Ben W. Mol MD, PhD
Department of Obstetrics and Gynaecology, School of Medicine, Monash University, Clayton, Australia

Julie S. Moldenhauer MD
The Center for Fetal Diagnosis and Treatment, The Children's Hospital of Philadelphia, Philadelphia, PA, USA

Fionnuala Mone PhD
Fetal Medicine Centre, Birmingham Women's and Children's NHS Foundation Trust, Birmingham, UK

Rachel Katie Morris MBChB, PhD, MRCOG
Birmingham Women's and Children's Hospital, and The Institute of Metabolism and Systems Research, University of Birmingham, Birmingham, UK

Sarah Murray MBChB, MSc, PhD, MRCOG
University of Edinburgh MRC Centre for Reproductive Health, Edinburgh, UK

Jane E. Norman MD, MBChB, FRCOG, FRCPE, FMedSci, FRSE
Faculty of Health Sciences, University of Bristol, Bristol, UK

Dick Oepkes MD, PhD, FRCOG
Division of Fetal Medicine, Department of Obstetrics, Leiden University Medical Center, Leiden, the Netherlands

Oluyinka O. Olutoye MD, PhD
Department of Surgery, Nationwide Children's Hospital, Ohio State University, Columbus, OH, USA

Emily A. Partridge MD, PhD
Division of General, Thoracic and Fetal Surgery, Children's Hospital of Philadelphia, Philadelphia, PA, USA

Jonna Petzold PhD
Centre for Craniofacial and Regenerative Biology, King's College London, London, UK

Robert E. Poelmann PhD
Department of Animal Science and Health, Leiden University, Leiden, the Netherlands

Léo Pomar MSc
Materno-foetal and Obstetrics Research Unit, Obstetric Service, Department "Femme-Mère-Enfant," University Hospital, Lausanne, Switzerland; and Department of Obstetrics and Gynecology, Centre Hospitalier de l'Ouest Guyanais Franck Joly, Saint-Laurent-du-Maroni, France

Judith Rankin BSc (Hons), PhD, FFPH
Maternal and Child Health, Institute of Health and Society, Newcastle University, Newcastle-upon-Tyne, UK

Michael G. Ross MD, MPH
Obstetrics and Gynecology and Public Health, David Geffen School of Medicine at UCLA, Los Angeles, and Department of Obstetrics and Gynecology, Harbor-UCLA Medical Center, Torrance, CA, USA

Francesca Russo MD, PhD
Department of Development and Regeneration, Cluster Woman and Child, University Hospitals Leuven, KU Leuven, Leuven, Belgium

Greg Ryan MD
Fetal Medicine Unit, Ontario Fetal Centre, Mount Sinai Hospital, and Division of Maternal-Fetal Medicine, Department of Obstetrics and Gynaecology, University of Toronto, Toronto, Canada

Mike Seed MBBS
Department of Pediatrics, University of Toronto, and Division of Cardiology, The Hospital for Sick Children, Toronto, Canada

Alireza A. Shamshirsaz MD
Department of Obstetrics and Gynecology (courtesy appointment in the Department of Surgery) Baylor College of Medicine and Texas Children's Hospital Fetal Center Houston, Texas

Femke Slaghekke MD, PhD
Department of Obstetrics, Division of Fetal Medicine, Leiden University Medical Center, Leiden, the Netherlands

Gordon C. S. Smith MD, PhD, DSc, FRCOG, FMedSci
Department of Obstetrics and Gynaecology, University of Cambridge, Cambridge, UK

Marjolijn S. Spruijt MD
Division of Neonatology, Department of Pediatrics, Leiden University Medical Center, Leiden, the Netherlands

Regine P. M. Steegers-Theunissen MD, PhD
Department of Obstetrics and Gynecology, Erasmus MC, University Medical Center Rotterdam, Rotterdam, the Netherlands

Julien Stirnemann MD
National Reference Centre for the Management of Complicated Monochorionic Pregnancies, and Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Dorota Szumska PhD
Department of Cardiovascular Medicine, BHF Centre of Research Excellence, and Wellcome Trust Centre for Human Genetics, University of Oxford, Oxford, UK

Danielle R. M. Timmermans PhD
Department of Public and Occupational Health, Amsterdam Public Health Research Institute, Amsterdam UMC, Vrije Universiteit Amsterdam, Amsterdam, the Netherlands

Lisanne S. A. Tollenaar BSc
Division of Fetal Medicine, Department of Obstetrics, Leiden University Medical Center, Leiden, the Netherlands

Rosemary Townsend MBChB, MRCOG
Fetal Medicine Unit, St George's University of London, London, UK

Sanne van der Hout PhD
Department of Health, Ethics and Society, Faculty of Health, Medicine and Life Sciences, Research Schools of CAPHRI and GROW, Maastricht University, Maastricht, the Netherlands

Lennart Van der Veeken MD
Fetal Diagnosis and Therapy Unit, Department of Obstetrics and Gynecology, Division of Woman and Child, University Hospitals Leuven, Leuven, Belgium

Inge L. van Kamp MD, PhD
Division of Fetal Medicine, Department of Obstetrics, Leiden University Medical Center, Leiden, the Netherlands

Jeanine M. M. van Klink PhD
Division of Child and Adolescent Psychology, Department of Pediatrics, Leiden University Medical Center, Leiden, the Netherlands

Tim Van Mieghem MD, PhD
Fetal Medicine Unit, Department of Obstetrics and Gynaecology, Mount Sinai Hospital and University of Toronto, Toronto, Canada

Janneke van 't Hooft MD, PhD
Department of Obstetrics and Gynecology, Amsterdam UMC (Academic Medical Center), Amsterdam, the Netherlands

Maud D. van Zijl MD
Department of Obstetrics and Gynecology, Amsterdam UMC, University of Amsterdam, Amsterdam, the Netherlands

Guillermo Villagomez Olea PhD
Centre for Craniofacial and Regenerative Biology, King's College London, London, UK

Yves Ville MD
National Reference Centre for the Management of Complicated Monochorionic Pregnancies, and Department of Obstetrics and Fetal Medicine, Paris Descartes University, Assistance Publique-Hôpitaux de Paris, Hôpital Necker-Enfants-Malades, Paris, France

Melissa Walker MD, MSc
Department of Obstetrics and Gynaecology, University of Toronto, Toronto, Canada

Wolfgang Weninger, PhD
Center for Anatomy and Cell Biology, Medical University of Vienna, Vienna, Austria

Eleanor Whitaker BA, BM BCh
University of Edinburgh MRC Centre for Reproductive Health, Edinburgh, UK

Clare L. Whitehead MB ChB, PhD, FRANZCOG
Department of Obstetrics and Gynaecology, University of Adelaide, Adelaide, Australia

David I. Wilson BA, MBBS, PhD, FRCP
Centre for Human Development, Stem Cells and Regeneration, Faculty of Medicine, University of Southampton, UK

William E. Whitehead MD
Department of Neurosurgery (courtesy appointment in the Department of Obstetrics and Gynecology) Baylor College of Medicine and Texas Children's Hospital Fetal Center
Houston, Texas

Robert Wilson PhD
The Francis Crick Institute, London, UK

Dian Winkelhorst MD
Department of Obstetrics, Leiden University Medical Center, Leiden, the Netherlands

Carolien Zwiers MD, PhD
Division of Fetal Medicine, Department of Obstetrics, Leiden University Medical Center, Leiden,
the Netherlands

Foreword

The dawn of fetal therapy occurred over five decades ago. Sir William Liley pioneered the first successful fetal therapy when he transfused donor red blood cells into the peritoneal cavity of an anemic fetus in a pregnant woman with Rh(D) alloimmunization. What is most remarkable is that this procedure was accomplished before the introduction of obstetrical ultrasound. Liley used radiopaque dye injected into the amniotic cavity to outline the fetus as an *amniogram* in order to target the fetal peritoneal cavity. Since these early days, remarkable progress has been achieved in the areas of fetal diagnosis and therapy. With ultrasound, using increasingly sophisticated technology, becoming part of routine obstetrical practice most fetal structural anomalies are easily diagnosed. Rapid acquisition, high resolution magnetic resonance imaging has further refined these diagnoses. Chromosomal microarray and whole exome sequencing have led to new diagnostic capabilities. Invasive procedures to acquire chorionic villi or amniotic fluid are rapidly being replaced by analyzing free fetal DNA in the maternal circulation.

These tools have led to a rapid evolution in fetal therapy. Early attempts to correct major congenital anomalies such as lower urinary tract obstruction, diaphragm hernia and sacrococcygeal teratoma were attempted by the pediatric surgical community through open hysterotomy. Premature delivery or fetal demise was often the result, leading many to question the future of fetal therapy for structural anomalies. A renewed interest in fetoscopy, once used primarily as a diagnostic tool, occurred when laser photocoagulation of placental anastomoses proved successful in the treatment of severe twin-twin transfusion syndrome (TTTS). Open hysterotomy returned to the spotlight with interest in correcting fetal myelomeningocele (MMC) – the first non-lethal congenital condition where fetal therapy attempted to improve lifelong morbidity instead of perinatal mortality.

A notable shift in the mindset of fetal therapy has occurred in the last decade. New therapies are no longer accepted as the standard of care after a period of simple innovation. Randomized clinical trials for laser therapy for TTTS and fetal MMC repair have proven these therapies to be scientifically sound. Tracheal occlusion for the treatment of fetal diaphragm hernia is currently being evaluated in such a trial. Multicenter alliances such as the EUROFOETUS group and the North American Fetal Treatment Network have been established to further research collaboration.

This second edition of *Fetal Therapy: Scientific Basis and Critical Appraisal of Clinical Benefits* builds on this new paradigm of an evidence-based approach to therapeutic maneuvers to aid the unborn patient. The editors have assembled a renowned group of international experts in their respective fields. Many aspects of fetal therapy that have evolved since the publication of the first edition are now addressed in this updated version. Notably a new section on the pathophysiology and prevention of preterm birth has been added. Ongoing research and potential therapies to ameliorate neurologic sequelae in cases of severe growth restriction, congenital heart disease, TTTS and the premature infant in general are included in new chapters. Evolving therapies such as the artificial womb, fetoscopic repair of fetal MMC, and the use of stem cells to address the issue of premature rupture of the membranes after fetoscopy are included in this edition.

This text deserves a prominent place in the library of any provider of fetal medicine. Its owner will be well served with a contemporary and authoritative reference on the care of the unborn patient with complex issues.

Kenneth J. Moise, Jr., MD
Professor of Obstetrics, Gynecology and Reproductive Sciences and Pediatric Surgery
McGovern School of Medicine – UTHealth
Co-Director, The Fetal Center
Children's Memorial Hermann Hospital
Houston, TX, USA

Chapter 1

The Rationale for Fetal Therapy

Ahmet Baschat

Introduction

In 1982, a group of subspecialists in fetal medicine, pediatric surgery, pediatrics, radiology, genetics, and bioethics reported on a meeting that discussed the emerging field of 'fetal therapy' [1]. Their summary statement laid down the foundation and principles for the treatment of prenatally diagnosed congenital anomalies where the natural history of the disease can potentially be influenced by intervention before birth (Table 1.1). In principle, this document defines the criteria of candidate conditions for fetal therapy, the goals of fetal treatment, and the appropriate setting for where fetal therapy should be performed. Since this original publication there have been significant advances in prenatal diagnostic and prognostic assessments of the fetus, the scope of treatments, and the care settings where fetal therapy is offered that require consideration [2].

Prenatal Diagnosis and Prognostic Assessment – Defining Candidate Conditions for Fetal Treatment

Fetal therapy targets specific conditions that carry significant risk for the fetus where prenatal intervention can be anticipated to significantly improve outcome. In order to be certain that a disease meets these fundamental criteria, a precise prenatal diagnosis and prognostic assessment is required. The principle diagnostic tools include a combination of ultrasound modalities, magnetic resonance imaging (MRI), or specialized computerized tomography (CT) imaging [3]. Following the formulation of a primary and differential diagnosis a major determining factor for eligibility for fetal treatment is the presence of any underlying untreatable conditions that affect outcome. Major advances have been made in genetic testing since the inception of fetal therapy. The range of prenatal genetic studies now ranges from traditional karyotyping to microarray analysis, targeted single gene testing, and exome sequencing [4, 5]. Another significant advance since the inception of fetal therapy is the transition of infection testing to polymerase chain reaction (PCR) for viral particles or viral culture from amniotic fluid [6, 7]. This contemporary approach to prenatal genetic testing and infection testing increases the diagnostic yield for significant underlying genetic or other abnormalities, and can now more deliberately identify fetuses that may benefit from prenatal

Table 1.1 Criteria for the advancement of fetal therapy: 1982

Topic	Viewpoint
Nature of the disorder	The disorder must be of a significant nature and should be a simple structural defect that interferes with organ development, whose alleviation might allow fetal development to proceed normally
Appropriateness criteria	The fetus should be a singleton without concomitant anomalies according to advanced ultrasonographic examination and amniocentesis for karyotype, α-feto protein, and cultures
Candidate diseases	Selection for treatment must be based on careful clinical evaluation and sound knowledge of the natural history of the fetal disease; intervention can be ethically justified only if there is reasonable probability of benefit
Goals of treatment	The family should be fully counseled about risks and benefits and should agree to treatment, including long-term follow-up to determine efficacy
Maternal safety and autonomy	*Implied but not stated:* maternal risks should be minor and acceptable to mother and family
Center infrastructure	There should be access to a level III high-risk obstetric unit and bioethical and psychosocial counseling
Checks and balances	A multidisciplinary team, including a perinatal obstetrician experienced in fetal diagnosis and intrauterine transfusion, an ultrasonographer experienced in the diagnosis of fetal anomalies, and a pediatric surgeon and neonatologist who will manage the infant after birth, should concur on the plan for innovative treatment and obtain approval of an institutional review board
Reporting requirements	All case material should be reported, regardless of outcome, to a fetal treatment registry or in the medical literature (or both)

interventions and exclude those who do not. The importance of this approach is illustrated by the outcomes of shunting for fetal hydrocephaly, which was abandoned in an era where the exclusion of diseases with no anticipated benefit was not uniformly applied. Now that a group of fetuses with isolated aqueductal stenosis is more likely to be identified, fetal therapy for this specific subset of patients may need to be re-explored [8].

Concurrently with rendering a precise diagnosis of the fetal condition, assessing the severity of the fetal condition is part of identifying suitable candidates for fetal therapy. It is important to recognize that despite the prominent role of ultrasound in evaluating physical abnormalities of the fetus, MRI is complementary in many conditions, including spina bifida and congenital diaphragmatic hernia, in delineating the abnormality as well as its prognosis [9, 10]. Since most fetal conditions that are currently offered fetal therapy are considered severe, most prognostic assessments measure the mortality or irreversible damage that is associated with a particular condition rather than morbidity. To render a prognosis, several specific parameters have been described that offer disease-specific quantification of severity. These include the traditional [11] and observed to expected lung-to-head ratio [12] for congenital diaphragmatic hernia and the cyst-volume ratio [13] for cystic adenomatoid malformations of the lung. In addition to individual measurements, combinations of several parameters in scoring or staging systems have been described to grade the severity of fetal cardiovascular disease [14, 15], hydrops [16], or twin-twin transfusion syndrome (TTTS) [17, 18, 19]. The utilization of standardized prenatal prognostic markers is of critical importance from several perspectives. The relationship with outcome forms the basis of the risk-benefit assessment and the selection of appropriate candidates for fetal therapy. Uniform assessment of conditions allows the study of natural disease evolution and a more consistent case selection facilitates a more robust evaluation of the impact of fetal therapies. The ability to re-evaluate defining key prognostic indicators also allows appropriately targeted monitoring for resolution following fetal treatment.

The evaluation of any prenatal abnormality should ideally reach the highest level of certainty about the condition, any underlying contributors to lifelong health impacts, and the severity of the condition in terms of its anticipated prenatal and postnatal outcome if left untreated. Only when this level of information is available can the risks of the natural disease be weighed against the risks of the therapy and parents be provided with the opportunity to select the appropriate scope of treatment. At any point in these decisions it is the obligation of the fetal medicine provider to put safeguards in place to protect the pregnant women from undue risk. For conditions not meeting intervention criteria longitudinal observations at the appropriate surveillance intervals are often required in order to ensure that deterioration to a degree that meets treatment criteria is detected. This is often required for complicated monochorionic multiple gestation [20], or fetal anemia due to red cell alloimmunization [21].

The Scope and Goals of Fetal Therapy

Fetal therapy may involve medical and surgical treatments that are performed before separation of the fetus from the placenta during birth. Within this scope, fetal therapy can be divided into medical or surgical approaches that aim to achieve either a complete prenatal resolution, alleviate severe pediatric developmental or functional deficiencies, or optimize the fetal transition to extrauterine life. In the latter two instances, treatment requires completion after birth and therefore relies on an appropriate pediatric subspecialty setting (**Figure 1.1**).

Fetal interventions carry different levels of complexity both in terms of required operator training and experience and the systems requirement to safely administer the treatment. At the most basic level, ultrasound-guided needle procedures have been adapted from the sampling of amniotic fluid or chorion villus tissue. Fetal therapy techniques based on this approach include fetal blood sampling, intrauterine transfusions [22], shunt placements for renal or thoracic abnormalities [23], balloon valvuloplasty for cardiac lesions [24], and interstitial coagulation techniques utilizing laser, radiofrequency ablation or microwave technology [25, 26]. A greater level of complexity exists for diagnostic or operative fetoscopic procedures. While the insertion of the instrumentation relies on ultrasound guidance, the instrumentation required is more complex and most optimally used in an operative room setting. Fetoscopic techniques now encompass laser ablation of communicating vessels in TTTS [27], umbilical cord occlusion [28], tracheal balloon occlusion and reversal [29], amniotic band release [30], laser ablation for lower urinary tract obstruction [31], and more complex surgical procedures such as myelomeningocele (MMC) repair [32].

The highest level of complexity involves open fetal surgery that is performed through a hysterotomy through the muscular portion of the myometrium or the *ex utero* intrapartum treatment (EXIT), which is a specialized delivery technique that enables securing of the fetal airway on a placental bypass. These types of procedures require a specific approach guided by the anatomy of the fetus and have high system requirements for monitoring of maternal and fetal well-being at the time of the procedure and afterwards, as well as the ability to immediately respond to complications such as obstetric hemorrhage or maternal cardiopulmonary collapse [33]. Open fetal surgeries are most frequently performed for MMC repair [34] and less often for resection of lung masses or teratomas [35]. The EXIT delivery technique is specifically intended for the management of anomalies that compromise the newborn's airway at birth [36, 37, 38].

These treatment techniques evolved following the consideration of the fetal, neonatal and lifelong risks of the untreated condition as well as the potential fetal benefits of treatment and the risks to the mother and the fetus. With regards to the fetal benefits, treatments may achieve prenatal cure or alleviation of damage. Examples for approaches that aim to achieve a prenatal cure include fetal blood transfusions for anemia [22] and

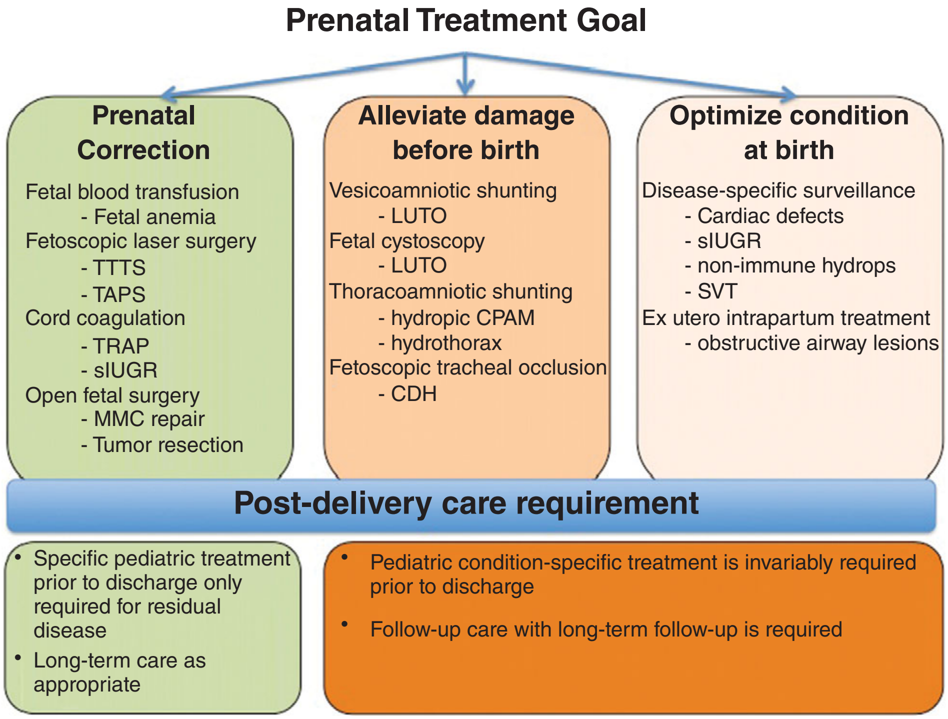

Figure 1.1 Treatment goals in fetal therapy. The schematic represents the prenatal treatment goals for various fetal interventions and their associated postnatal care needs. TTTS, twin-twin transfusion syndrome; TAPS, twin anemia polycythemia sequence; TRAP, twin reversed arterial perfusion; sIUGR, selective intrauterine growth restriction; MMC, myelomeningocele; LUTO, lower urinary tract obstruction; CPAM, cystic pulmonary airway malformation; CDH, congenital diaphragmatic hernia; SVT, supraventricular tachycardia.

fetoscopic laser dichorionization of the placenta in TTTS [39]. In addition to the appropriate care setting, the ability to achieve the intended outcome with these low- to medium-complexity therapies relies on operator experience and ongoing caseload [40–44]. Because the mortality of the underlying conditions in the absence of treatment is high, thresholds for the establishment of treatment centers are lower than for more complex treatments. A treatment that is also aimed at prenatal correction is fetal MMC repair. However, irrespective of whether an open or fetoscopic approach is chosen the multidisciplinary nature of the treatment team requires an appropriate resource setting to achieve the desired outcomee [45]. Since fetal MMC is not a lethal condition and treatment is also complex because of the maternal care requirements, prenatal repair can only be offered in an appropriately resourced setting. In fact it is the significant maternal risk with open fetal MMC repair that is one of the driving forces to transition to a viable fetoscopic technique that maintains the fetal benefits [46].

An example for a treatment that does not achieve prenatal cure, but rather alleviates prenatal damage until definitive postnatal repair can occur, is fetoscopic tracheal occlusion (FETO) for severe congenital diaphragmatic hernia (CDH) [12]. After successful FETO, delivery of the neonate at a center with expertise in the management of CDH is required to complete the treatment. It is important to recognize that caseload and operator experience improve outcomes in both the prenatal and postnatal components of FETO followed by post-delivery surgical CDH management [47, 48]. The ideal setting for a FETO program with an experienced fetal team would therefore be at a facility with a coexisting high-volume experienced pediatric CDH program [49]. The importance of the appropriate pediatric care setting at delivery is particularly evident for anomalies such as cardiac defects, where the primary contribution of the fetal medicine specialist is to optimize delivery circumstances to facilitate post-delivery surgical repair [50]. Accordingly, as the management goal of fetal therapies shifts from prenatal cure to alleviation of damage the emphasis on delivery in an appropriate pediatric care setting increases. With the exception of fetal therapies carried out prior to viability the need for a high-level neonatal intensive care unit (NICU) is universal for all fetal therapy centers [1, 2, 45].

Risk-Benefit Assessment for Fetal Therapy

Fundamental to the endeavor of fetal treatment is the construction of a risk-benefit assessment that considers potential benefits to the fetus, newborn and mother balanced against the risks to these parties. Since all fetal therapy, medical and surgical, must pass through the mother it cannot be performed without her informed consent, given with the necessary safeguards in place and full consideration of maternal and fetal risks. Assuming an accurate prenatal diagnosis, this assessment rests on a reasonable degree of certainty about the natural history of the condition, the likelihood of treatment success, and the preparedness for the potential of unintended consequences. Fetal therapy is unique in that the potential complications for a given procedure may include the mother or fetus. For surgical interventions the potential for unintended consequences depends on the complexity of the procedures as well as operator experience and caseload. The correct risk-benefit estimate therefore relies on all of these factors.

The neonatal risks relate to the likelihood of premature delivery and the additional outcome impact of the underlying condition, and are partly mitigated when delivery occurs at a facility with the appropriate level of neonatal care [1]. For conditions that require surgical correction after birth risks may arise from the combination of residual morbidity after fetal therapy and superimposed neonatal complications. As prematurity is a risk factor associated with many fetal treatments, accurate representation of institution-specific, disease-specific outcomes of centers that perform fetal therapy is most pertinent to gauge the overall impact on outcome [1, 2, 45]. Over time advances in any of the subspecialties involved in the care of the fetal patient potentially alter outcome, and therefore ongoing reappraisal of the risk-benefit ratio is required whenever such developments occur. Examples include the transition from open fetal surgery to maternal steroid use as a primary treatment of congenital pulmonary airway malformations [51], or reappraisal of the relative safety of CO_2 insufflation for operative fetoscopy [52]. Once a risk-benefit assessment for a fetal treatment has been completed administration in the appropriate care setting is essential to mitigate some of the adverse effects.

Care Settings for Fetal Therapy

Since all fetal therapies pass through the mother the need to establish the most appropriate maternal care setting is universal for all fetal therapies. The resources required to ensure maternal safety may range from obstetric care facilities, including obstetric anesthesia, all the way to medical and intensive care facilities [38]. These requirements depend on the complexity of the fetal treatments performed. Ultrasound-guided procedures such as amniocentesis and chorion villous sampling have a negligible miscarriage rate and overall procedure-related risks ranging from 0.4% to 1% in high-risk populations [53, 54]. Fetal blood sampling and transfusion require a higher level of operator skill and carry a 5–10% risk of fetal bradycardia and a pregnancy loss rate of up to 25% in complex fetal conditions [55, 56, 57]. Fetal shunt procedures and fetoscopic laser ablation for TTTS involve larger diameter uterine instrumentation and accordingly can carry an up to 40% risk of obstetric complications, including preterm premature rupture of membranes (PPROM), preterm labor, and preterm birth [58, 59]. If intervention for fetal status is appropriate as part of the management plan or significant obstetric risks are recognized complications such fetal treatments should be performed in the vicinity of a Labor and Delivery unit to ensure that obstetric management, including delivery if appropriate, can be achieved in a timely fashion. Procedures that are performed at viability, carry significant obstetric risks, or require multidisciplinary effort may benefit from a dedicated intervention suite near Labor and Delivery. Fetal cardiac interventions have fetal mortality rates of 10–30% and may require additional treatment of complications such as bradycardia and hemopericardium in 27–52% of procedures [60, 61]. FETO with subsequent balloon removal is associated with a 47% rate of PPROM and the need for emergency balloon removal in over 50% of cases. Inability to remove the balloon prior to birth in the latter setting may lead to neonatal death in almost 5% of patients [29]. Hybrid or open fetal surgeries, including fetoscopic spina bifida repair [62] and EXIT, naturally require an appropriately staffed operation room setting [6, 63]. The significant risk for healing complications of the uterotomy with partial or complete dehiscence in 2.3% of patients, and the need for blood transfusion at delivery in 8%, emphasize the importance of follow-up dedicated obstetric care [34]. As integration of subspecialties is one of the core achievements that drives a fetal treatment center, the complete integration of the required level of maternal care is necessary. For the highest risk procedures this requires the in-house availability of an appropriate level of maternal care services, including intensive care and adult medical specialty availability.

As all neonates that are delivered after fetal therapy require post-delivery assessment, stabilization and potentially further management, a high-level NICU is recommended for all fetal therapy centers offering treatment after viability [1, 2, 45]. This level of care is recommended since most conditions targeted by fetal therapy have neonatal care requirements that reach beyond prematurity-related complications, and management of anomalies and associated problems is required [64, 65]. Specifically for neonates with congenital abnormalities such as CDH, MMC or cardiac defects, the in-house presence of the appropriate pediatric surgical specialties is highly desirable. In the US the 'Task Force for Children's Surgical Care' defines the highest level of center by its ability to manage congenital anomalies in an in- and outpatient setting [66]. The improved outcome for neonates cared for in such centers has been documented for several conditions, including CDH and MMC, and is in part attributable to infrastructure, higher surgical volumes and an enhanced ability to triage, recognize and manage complications compared with lower volume centers [67–69].

Requirements for a Fetal Therapy Center

A Fetal Therapy Center is ethically obliged to consider both maternal and fetal well-being and complications of *any* fetal intervention that may be offered. In order to provide safe care the appropriate infrastructure, dedicated institutional support and oversight are required. The level of infrastructure and support are dictated by the level of the maternal, fetal and neonatal care needs that arise as a consequence of the fetal intervention. Once the appropriate multidisciplinary care context has been established the monitoring and reporting of outcomes allows for effective oversight monitoring at the institutional level. It has been considered essential for centers that perform invasive fetal procedures to report their maternal, fetal, and newborn outcomes as transparently as possible to allow for ongoing scientific scrutiny [1, 2, 45]. This can be in the form of institutional, national, regional, or international registries or trials. Examples include treatment registries for FETO for severe CDH [29] or fetal cardiac interventions [24] as well as randomized trials for laser therapy for TTTS [27, 70] and open fetal MMC repair [34]. Particularly for procedures that are still considered as innovative or under research a multidisciplinary institutional oversight committee is important, and ideally includes individuals not directly involved in the clinical care of patients. Such committees may sometimes also serve as reviewing bodies for the purposes of institutional or ethical review board submissions.

An important obligation of a Fetal Therapy Center is also to provide education for physicians and other healthcare personnel to train the next generation of fetal therapists. While there are currently no formal training programs for fetal therapy it is only a matter of time before a curriculum will be formalized and an appropriate training model developed in which junior faculty are gradually allowed to develop the necessary skill set to operate independently.

Conclusion

As both diagnostic and surgical techniques continue to evolve so does the role of fetal therapy in conditions that can be prenatally diagnosed. With advances in fetal treatment techniques and the management of maternal risks the focus is likely to shift from just enabling survival to improving quality of life (e.g. fetal MMC repair). The formalization of appropriate care settings and potentially levels of care is likely to not only ensure the safety of the mother and fetus but also expand the rationale for fetal therapy in the future.

References

[1] Harrison MR, Filly RA, Golbus MS, Berkowitz RL, Callen PW, Canty TG, et al. Fetal treatment 1982. *N Engl J Med*. 1982; 307: 1651–2.

[2] Moon-Grady A, Baschat A, Cass D, et al. Fetal treatment 2017: the evolution of fetal therapy centers – a joint opinion from the International Fetal Medicine and Surgical Society (IFMSS) and the North American Fetal Therapy Network (NAFTNet). *Fetal Diagn Ther*. 2017; 42: 241–8.

[3] Snyder E, Baschat A, Huisman TAGM, Tekes A. Value of fetal MRI in the era of fetal therapy for management of abnormalities involving the chest, abdomen, or pelvis. *Am J Roentgenol*. 2018; 210: 998–1009.

[4] Wapner RJ, Martin CL, Levy B, Ballif BC, Eng CM, Zachary JM, et al. Chromosomal microarray versus karyotyping for prenatal diagnosis. *N Engl J Med*. 2012; 367: 2175–84.

[5] Drury S, Williams H, Trump N, Boustred C, GOSGene, Lench N, Scott RH, Chitty LS. Exome sequencing for prenatal diagnosis of fetuses with sonographic abnormalities. *Prenat Diagn*. 2015; 35: 1010–7.

[6] Reddy UM, Baschat AA, Zlatnik MG, Towbin JA, Harman CR, Weiner CP. Detection of viral deoxyribonucleic acid in amniotic fluid: association with fetal malformation and pregnancy abnormalities. *Fetal Diagn Ther*. 2005; 20: 203–7.

[7] Adams LL, Gungor S, Turan S, Kopelman JN, Harman CR, Baschat AA. When are amniotic fluid viral PCR studies indicated in prenatal diagnosis? *Prenat Diagn*. 2012; 32: 88–93.

[8] Emery SP, Greene S, Hogge WA. Fetal Therapy for Isolated Aqueductal Stenosis. *Fetal Diagn Ther*. 2015; 38: 81–5.

[9] Micu R, Chicea AL, Bratu DG, Nita P, Nemeti G, Chicea R. Ultrasound and magnetic resonance imaging in the prenatal diagnosis of open spina bifida. *Med Ultrason*. 2018; 20: 221–7.

[10] Madenci AL, Sjogren AR, Treadwell MC, Ladino-Torres MF, Drongowski RA, Kreutzman J, Bruch SW, Mychaliska GB. Another dimension to survival: predicting outcomes with fetal MRI versus prenatal ultrasound in patients with congenital diaphragmatic hernia. *J Pediatr Surg*. 2013; 48: 1190–7.

[11] Metkus AP, Filly RA, Stringer MD, Harrison MR, Adzick NS. Sonographic predictors of survival in fetal diaphragmatic hernia. *J Pediatr Surg*. 1996; 31: 148–51.

[12] Jani J, Nicolaides KH, Keller RL, Benachi A, Peralta CF, Favre R, et al. Observed to expected lung area to head circumference ratio in the prediction of survival in fetuses with isolated diaphragmatic hernia. *Ultrasound Obstet Gynecol*. 2007; 30: 67–71.

[13] Crombleholme TM, Coleman B, Hedrick H, Liechty K, Howell L, Flake AW, Johnson M, Adzick NS. Cystic adenomatoid malformation ratio predicts outcome in prenatally diagnosed cystic adenomatoid malformation of the lung. *J Pediatr Surg*. 2002; 37: 331–8.

[14] Wieczorek A, Hernandez-Robles J, Ewing L, Leshko J, Luther S, Huhta J. Prediction of outcome of fetal congenital heart disease using a cardiovascular profile score. *Ultrasound Obstet Gynecol*. 2008; 31: 284–8.

[15] Huhta JC, Paul JJ. Doppler in fetal heart failure. *Clin Obstet Gynecol*. 2010; 53: 915–29.

[16] Kim SA, Lee SM, Hong JS, Lee J, Park CW, Kim BJ, et al. Ultrasonographic severity scoring of non-immune hydrops: a predictor of perinatal mortality. *J Perinat Med*. 2015; 43: 53–9.

[17] Quintero RA, Morales WJ, Allen MH, Bornick PW, Johnson PK, Kruger M. Staging of twin-twin transfusion syndrome. *J Perinatol*. 1999; 19: 550–5.

[18] Rychik J, Tian Z, Bebbington M, Xu F, McCann M, Mann S, Wilson RD, Johnson MP. The twin-twin transfusion

syndrome: spectrum of cardiovascular abnormality and development of a cardiovascular score to assess severity of disease. *Am J Obstet Gynecol*. 2007; 197: 392. e1–8.

[19] Shah AD, Border WL, Crombleholme TM, Michelfelder EC. Initial fetal cardiovascular profile score predicts recipient twin outcome in twin-twin transfusion syndrome. *J Am Soc Echocardiogr*. 2008; 21: 1105–8.

[20] O'Donoghue K, Cartwright E, Galea P, Fisk NM. Stage I twin-twin transfusion syndrome: rates of progression and regression in relation to outcome. *Ultrasound Obstet Gynecol*. 2007; 30: 958–64.

[21] Oepkes D, Seaward PG, Vandenbussche FP, Windrim R, Kingdom J, Beyene J, Kanhai HH, Ohlsson A, Ryan G, DIAMOND Study Group. Doppler ultrasonography versus amniocentesis to predict fetal anemia. *N Engl J Med*. 2006; 355: 156–64.

[22] Zwiers C, Lindenburg ITM, Klumper FJ, de Haas M, Oepkes D, Van Kamp IL. Complications of intrauterine intravascular blood transfusion: lessons learned after 1678 procedures. *Ultrasound Obstet Gynecol*. 2017; 50: 180–6.

[23] Manning FA, Harrison MR, Rodeck C. Catheter shunts for fetal hydronephrosis and hydrocephalus. Report of the International Fetal Surgery Registry. *N Engl J Med*. 1986; 315: 336–40.

[24] Moon-Grady AJ, Morris SA, Belfort M, Chmait R, Dangel J, Devlieger R, et al. International Fetal Cardiac Intervention Registry: a worldwide collaborative description and preliminary outcomes. *J Am Coll Cardiol*. 2015; 66: 388–99.

[25] Bebbington MW, Danzer E, Moldenhauer J, Khalek N, Johnson MP. Radiofrequency ablation vs bipolar umbilical cord coagulation in the management of complicated monochorionic pregnancies. *Ultrasound Obstet Gynecol*. 2012; 40: 319–24.

[26] Stephenson CD, Temming LA, Pollack R, Iannitti DA. Microwave ablation for twin-reversed arterial perfusion sequence: a novel application of technology. *Fetal Diagn Ther*. 2015; 38: 35–40.

[27] Senat MV, Deprest J, Boulvain M, Paupe A, Winer N, Ville Y. Endoscopic laser surgery versus serial amnioreduction for severe twin-to-twin transfusion syndrome. *N Engl J Med*. 2004; 351: 136–44.

[28] Quintero RA, Reich H, Puder KS, Bardicef M, Evans MI, Cotton DB, Romero R: Brief re- port: umbilical-cord ligation of an acardiac twin by fetoscopy at 19 weeks of gestation. *N Engl J Med* 1994; 330: 469–471.

[29] Jani JC, Nicolaides KH, Gratacós E, Valencia CM, Doné E, Martinez JM, Gucciardo L, Cruz R, Deprest JA. Severe diaphragmatic hernia treated by fetal endoscopic tracheal occlusion. *Ultrasound Obstet Gynecol*. 2009; 34: 304–10.

[30] Richter J, Wergeland H, DeKoninck P, De Catte L, Deprest JA. Fetoscopic release of an amniotic band with risk of amputation: case report and review of the literature. *Fetal Diagn Ther*. 2012; 31: 134–7.

[31] Sananes N, Cruz-Martinez R, Favre R, Ordorica-Flores R, Moog R, Zaloszy A, Giron AM, Ruano R. Two-year outcomes after diagnostic and therapeutic fetal cystoscopy for lower urinary tract obstruction. *Prenat Diagn*. 2016; 36: 297–303.

[32] Kohl T, Hering R, Heep A, Schaller C, Meyer B, Greive C, et al. Percutaneous fetoscopic patch coverage of spina bifida aperta in the human – early clinical experience and potential. *Fetal Diagn Ther*. 2006; 21: 185–93.

[33] Abraham RJ, Sau A, Maxwell D. A review of the EXIT (Ex utero Intrapartum Treatment) procedure. *J Obstet Gynaecol*. 2010; 30: 1–5.

[34] Adzick NS, Thom EA, Spong CY, Brock JW 3rd, Burrows PK, Johnson MP, et al. A randomized trial of prenatal versus postnatal repair of myelomeningocele. *N Engl J Med*. 2011; 364: 993–1004.

[35] Cass DL, Olutoye OO, Ayres NA, Moise KJ Jr., Altman CA, Johnson A, Cassady CI, Lazar DA, Lee TC, Lantin MR. Defining hydrops and indications for open fetal surgery for fetuses with lung masses and vascular tumors. *J Pediatr Surg*. 2012; 47: 40–5.

[36] Liechty KW, Crombleholme TM, Flake AW, Morgan MA, Kurth CD, Hubbard AM, Adzick NS. Intrapartum airway management for giant fetal neck masses: the EXIT (ex utero intrapartum treatment) procedure. *Am J Obstet Gynecol*. 1997; 177: 870–4.

[37] Mychaliska GB, Bealer JF, Graf JL, Rosen MA, Adzick NS, Harrison MR. Operating on placental support: the ex utero intrapartum treatment procedure. *J Pediatr Surg*. 1997; 32: 227–30.

[38] Norris MC, Joseph J, Leighton BL. Anaesthesia for perinatal surgery. *Am J Perinatol*. 1989; 6: 39–40.

[39] Slaghekke F, Lopriore E, Lewi L, Middeldorp JM, van Zwet EW, Weingertner AS, et al. Fetoscopic laser coagulation of the vascular equator versus selective coagulation for twin-to-twin transfusion syndrome: an open-label randomised controlled trial. *Lancet*. 2014; 383: 2144–51.

[40] Peeters SH, Van Zwet EW, Oepkes D, Lopriore E, Klumper FJ, Middeldorp JM. Learning curve for fetoscopic laser surgery using cumulative sum analysis. *Acta Obstet Gynecol Scand*. 2014; 93: 705–11.

[41] Inglis SR, Lysikiewicz A, Sonnenblick AL, Streltzoff JL, Bussel JB, Chervenak FA. Advantages of larger volume, less frequent intrauterine red blood cell transfusions for maternal red cell alloimmunization. *Am J Perinatol*. 1996; 13: 27–33.

[42] Edwards AG, Teoh M, Hodges RJ, Palma-Dias R, Cole SA, Fung AM, Walker SP. Balancing Patient Access to fetoscopic laser photocoagulation for twin-to-twin transfusion syndrome with maintaining procedural competence: are collaborative services part of the solution? *Twin Res Hum Genet*. 2016; 19: 276–84.

[43] Perry KG Jr., Hess LW, Roberts WE, Allbert JR, Floyd RC, McCaul JF, Martin RW, Martin JN Jr., Morrison JC. Cordocentesis (funipuncture) by maternal-fetal fellows: the learning curve. *Fetal Diagn Ther*. 1991; 6: 87–92.

[44] Chang YL, Chao AS, Chang SD, Hsieh PC, Wang CN. Short-term outcomes of fetoscopic laser surgery for severe twin-twin transfusion syndrome from Taiwan single center experience: demonstration of learning curve effect on the fetal outcomes. *Taiwan J Obstet Gynecol*. 2012; 51: 350–3.

[45] Cohen AR, Couto J, Cummings JJ, Johnson A, Joseph G, Kaufman BA, et al. Position statement on fetal

myelomeningocele repair. *Am J Obstet Gynecol*. 2014; 210: 107–11.

[46] Belfort MA, Whitehead WE, Shamshirsaz AA, Ruano R, Cass DL, Olutoye OO. Fetoscopic repair of meningomyelocele. *Obstet Gynecol*. 2015; 126: 881–4.

[47] Araujo Júnior E, Tonni G, Martins WP, Ruano R. Procedure-related complications and survival following Fetoscopic Endotracheal Occlusion (FETO) for severe congenital diaphragmatic hernia: systematic review and meta-analysis in the FETO Era. *Eur J Pediatr Surg*. 2016; 27: 297–305.

[48] Grushka JR, Laberge JM, Puligandla P, Skarsgard ED, Canadian Pediatric Surgery Network: effect of hospital case volume on outcome in congenital diaphragmatic hernia: the experience of the Canadian Pediatric Surgery Network. *J Pediatr Surg*. 2009; 44: 873–6.

[49] Snoek KG, Greenough A, van Rosmalen J, Capolupo I, Schaible T, Ali K, Wijnen RM, Tibboel D. Congenital diaphragmatic hernia: 10-Year evaluation of survival, extracorporeal membrane oxygenation, and foetoscopic endotracheal occlusion in four high-volume centres. *Neonatology*. 2018; 113: 63–8.

[50] Sanapo L, Moon-Grady AJ, Donofrio MT. Perinatal and delivery management of infants with congenital heart disease. *Clin Perinatol*. 2016; 43: 55–71.

[51] Loh KC, Jelin E, Hirose S, Feldstein V, Goldstein R, Lee H. Microcystic congenital pulmonary airway malformation with hydrops fetalis: steroids vs open fetal resection. *J Pediatr Surg*. 2012; 47: 36–9.

[52] Baschat AA, Ahn ES, Murphy J, Miller JL. Fetal blood gas values during fetoscopic myelomeningocele repair performed under carbon dioxide insufflation. *Ultrasound Obstet Gynecol*. 2018; 52: 400–402.

[53] Wulff CB, Gerds TA, Rode L, Ekelund CK, Petersen OB, Tabor A, Danish Fetal Medicine Study Group: risk of fetal loss associated with invasive testing following combined first-trimester screening for Down syndrome: a national cohort of 147,987 singleton pregnancies. *Ultrasound Obstet Gynecol*. 2016; 47: 38–44.

[54] Enzensberger C, Pulvermacher C, Degenhardt J, Kawacki A, Germer U, Gembruch U, Krapp M, Weichert J, Axt-Fliedner R. Fetal loss rate and associated risk factors after amniocentesis, chorionic villus sampling and fetal blood sampling. *Ultraschall Med*. 2012; 33: E75–9.

[55] Society for Maternal-Fetal Medicine, Berry SM, Stone J, Norton ME, Johnson D, Berghella V. Fetal blood sampling. *Am J Obstet Gynecol*. 2013; 209: 170–80.

[56] Bigelow CA, Cinelli CM, Little SE, Benson CB, Frates MC, Wilkins-Haug LE. Percutaneous umbilical blood sampling: current trends and outcomes. *Eur J Obstet Gynecol Reprod Biol*. 2016; 200: 98–101.

[57] Wilson RD, Gagnon A, Audibert F, Campagnolo C, Carroll J, Genetics committee: prenatal diagnosis procedures and techniques to obtain a diagnostic fetal specimen or tissue: maternal and fetal risks and benefits. *J Obstet Gynaecol Can*. 2015; 37: 656–68.

[58] Papanna R, Block-Abraham D, Mann LK, Buhimschi IA, Bebbington M, Garcia E, Kahlek N, Harman C, Johnson A, Baschat A, Moise KJ Jr. Risk factors associated with preterm delivery after fetoscopic laser surgery for twin-twin transfusion syndrome. *Ultrasound Obstet Gynecol*. 2014; 43: 48–53.

[59] Ruano R, Sananes N, Sangi-Haghpeykar H, Hernandez-Ruano S, Moog R, Becmeur F, Zaloszyc A, Giron A, Morin B, Favre R. Fetal intervention for severe lower urinary tract obstruction: a multicenter case-control study comparing fetal cystoscopy with vesicoamniotic shunting. *Ultrasound Obstet Gynecol*. 2015; 45: 452–8.

[60] Moon-Grady AJ, Morris SA, Belfort M, Chmait R, Dangel J, Devlieger R, et al. International fetal cardiac intervention registry: a worldwide collaborative description and preliminary outcomes. *J Am Coll Cardiol*. 2015; 66: 388–99.

[61] Araujo Júnior E, Tonni G, Chung M, Ruano R, Martins WP. Perinatal outcomes and intrauterine complications following fetal intervention for congenital heart disease: systematic review and meta-analysis of observational studies. *Ultrasound Obstet Gynecol*. 2016; 48: 426–33.

[62] Belfort MA, Whitehead WE, Shamshirsaz AA, Ruano R, Cass DL, Olutoye OO. Fetoscopic repair of meningomyelocele. *Obstet Gynecol*. 2015; 126: 881–4.

[63] Johnson MP, Bennett KA, Rand L, Burrows PK, Thom EA, Howell LJ, et al. The Management of Myelomeningocele Study: obstetrical outcomes and risk factors for obstetrical complications following prenatal surgery. *Am J Obstet Gynecol*. 2016; 215: 778. e1–778. e9.

[64] Crenshaw C Jr., Payne P, Blackmon L, Bowen C, Gutberlet R. Prematurity and the obstetrician. A regional neonatal intensive care nursery is not enough. *Am J Obstet Gynecol*. 1983; 147: 125–32.

[65] American Academy of Pediatrics Committee on Fetus and Newborn. Policy statement: Levels of Neonatal Care. *Pediatrics*. 2012; 130: 587–97.

[66] Task Force for Children's Surgical Care. Optimal resources for children's surgical care in the United States. *J Am Coll Surg*. 2014; 218: 479–87.

[67] Birkmeyer JD, Stukel TA, Siewers AE, Goodney PP, Wennberg DE, Lucas FL. Surgeon volume and operative mortality in the United States. *New Engl J Med*. 2003; 349: 2117–27.

[68] Grayson AD, Moore RK, Jackson M, Rathore S, Sastry S, Gray TP, Schofield I, Chauhan A, Ordoubadi FF, Prendergast B, Stables RH. north west quality improvement programme in cardiac interventions: multivariate prediction of major adverse cardiac events after 9914 percutaneous coronary interventions in the north west of England. *Heart*. 2006; 92: 658–63.

[69] Wright JD, Herzog TJ, Siddiq Z, Arend R, Neugut AI, Burke WM, Lewin SN, Ananth CV, Hershman DL. Failure to rescue as a source of variation in hospital mortality for ovarian cancer. *J Clin Oncol*. 2012; 30: 3976–82.

[70] Slaghekke F, Lopriore E, Lewi L, Middledorp JM, van Zwet EW, Weingertner AS, et al. Fetoscopic laser coagulation of the vascular equator versus selective coagulation for twin-to-twin transfusion syndrome: an open-label randomised controlled trial. *Lancet*. 2014; 383: 2144–51.

Chapter 2

A Fetal Origin of Adult Disease

Mark Hanson and Lucy Green

Introduction

There is a worldwide epidemic of non-communicable diseases (NCDs), including cardiovascular disease (CVD), type 2 diabetes, chronic lung disease, and some forms of cancer; predisposition to these is linked to obesity. This is despite efforts by individuals to modify their diet and lifestyle, and government and global programs aimed at promoting healthy eating or increased physical activity. Some initiatives have begun to target childhood eating and activity. But a strong and international body of scientific and epidemiological data suggests that health interventions should be focused on a much earlier period of development: pregnancy. Expectant couples are often focused on the immediate result of their pregnancy – a viable baby. It may come as a surprise to many of them to hear that the finer details of building a baby are in fact the foundation of lifelong health.

A number of potentially serious clinical conditions originate during fetal life, and these include neurological handicap, premature birth, fetal growth restriction (FGR), and pulmonary hypoplasia. These are often viewed as 'pathophysiological' conditions where normal development (or physiology) has been disrupted by a challenge *in utero* that has had immediate and longer-term damaging effects. But, current concepts suggest that the developing organism may respond to cues (e.g. nutrient supply, maternal stress) from the environment and that its development is 'channelled' (rather than disrupted) to give a phenotype optimized for the subsequent postnatal environment. However there may be limits to the responses that the developing organism (e.g. fetus) can make or the postnatal environment may not turn out to be what was expected. Either or both of these circumstances may lead to an increased disease risk in adulthood [1]. In this chapter we will explore human and animal studies that investigate how cues from the environment (e.g. nutrient supply) before and during gestation invoke one or several fetal adaptive strategies involving the timing of birth, fetal growth, metabolism, and cardiovascular control. These strategies are not simply linked to immediate survival but may put the offspring at a disadvantage in terms of health in later life.

An Early Origin for the Epidemic of Adult Non-communicable Disease

The Problem

CVD affects more than 40% of adults in the UK. Despite the fall in death from coronary heart disease (CHD) in the second half of the twentieth century, the combination of unhealthy lifestyle in the young and an ageing population is expected to increase the number of people suffering CVD such as heart failure. Globally, it is estimated that 17.9 million people died from CVD in 2016 and, without intervention, this number is projected to rise [2]. The number of people with diabetes rose from 108 million in 1980 to 422 million in 2014 [3]. Obesity is a component of metabolic syndrome and is considered to be an intermediate risk factor for CVD, even in the young. The speed with which the incidence of these diseases has escalated has been attributed to changing lifestyles, especially the consumption of high glycemic index foods with a high fat and salt content and a sedentary lifestyle. However, not all individuals have the same risk of developing pathological conditions, even in the same environment. In the last 30 years it has emerged that the developmental environment (periconceptionally through early postnatal life) influences an individual's response to their adult environment and lifestyle, hence determining in part their risk of disease.

The DOHaD Concept

Epidemiological studies show that small size at birth and during infancy is associated with a greater risk of CHD, hypertension and stroke in later life [4]. Importantly, the degree of these changes, and hence disease risk, is graded across the normal range of size at birth, i.e. it is not just a consequence of FGR. The Developmental Origins of Health and Disease (DOHaD) concept suggested that the low birth weight-disease risk association may underestimate the true influence of the early environment effect. Birth size is one measure of fetal environment and DOHaD might be better viewed as a later consequence of a normal developmental response to environmental cues. The risk of adult CHD is particularly increased if small size at birth and during infancy is followed by rapid childhood weight gain [5]. Recent proposals suggest that the developing organism responds to its environment to develop a phenotype optimal for survival to reproduce in the postnatal environment in which it predicts that it will live, and that a mismatch between the *in utero* and childhood nutritional environments increases risk of CVD [1, 6]. The degree of mismatch will be increased by an unhealthy lifestyle (unbalanced diet, reduced physical activity, smoking, and excessive alcohol consumption) and this is of particular concern in light

https://doi.org/10.1017/9781108564434.003

of the rising incidence of childhood obesity and the links between obesity and CVD.

Human and Animal Evidence

There is potential for the embryo and fetus to be exposed to a range of such cues, including environmental toxins, 'maternal constraint' (from e.g. body composition, stature, nutrition, age, and parity), maternal stress, umbilical-placental complications (including resultant hypoxia/asphyxia), and maternal diseases. Environmental factors such as maternal nutrition can channel development (i.e. influence developmental plasticity). The adaptations that are made might be of immediate adaptive value and help survival, or could confer little or no immediate benefit but nonetheless be predictive of the postnatal environment. If the postnatal environment is not as predicted this may increase the risk of disease [1]. But studies attempting to investigate DOHaD should distinguish these sort of adaptive responses from pathophysiological effects of the environment (disrupting development, e.g. toxins or umbilical-placental complications) with no obvious adaptive value at any point in the life course. This is important since these simply disrupt the normal pattern of development and do not necessarily lead to an increased risk of disease.

There are a few key human cohorts in which the DOHaD concept has been investigated [7]. In addition a number of experimental animal models have been developed in a range of species. Ascertaining the risk of disease is usually not possible in animal models, but making sure that the challenges are of physiological rather than pathophysiological magnitude and of a type relevant to DOHaD remains crucial to progress in this field. In this chapter, we focus primarily on maternal constraint-type cues for which a cohesive and persuasive body of evidence exists.

Many women in the UK consume unbalanced or 'imprudent' diets, including during pregnancy. The influence of a poor intrauterine environment on later CVD was highlighted in the Dutch winter famine cohort [8]. Maternal body composition and metabolism provide the backdrop against which more acute changes in diet act and influence the compartmentalization of nutrients between the mother, placenta, and fetus. In England 15.6% of women are obese (body mass index (BMI) ≥ 30 kg/m^2) at the start of pregnancy and a smaller proportion (2.88%) are underweight (BMI < 18.5 kg/m^2) [9]. In the Southampton Women's Survey by the Institute of Medicine Standards (2009), excessive (49%) and inadequate (21%) weight gain in pregnancy are prevalent [10]. Both extremes of maternal weight profile are thought to pose a significant threat to maternal and fetal/neonatal well-being and may have substantial ramifications for cardiovascular health in later life. Excessive weight gain is linked to offspring obesity [10, 11] and to higher systolic blood pressure into early adulthood (21 years [12]). Human data suggest that whilst gestational weight gain is associated with adverse cardiovascular risk factors at 9 years, pre-pregnancy weight has a greater overall impact [11]. Slimness in mothers is linked to CHD and raised blood pressure, while high maternal weight/adiposity is linked to CHD [13, 14]. In this regard, new guidelines on pregnancy weight management were issued in 2010 [9] and their implementation may serve to break the cycle of obesity and reduce the incidence of CVD.

Numerous studies in animal models (rodents, guinea pigs, sheep, and non-human primates) corroborate the idea that maternal diet during gestation and breastfeeding are very important in determining adult propensity to obesity, cardiovascular and renal dysfunction [15–23], and left ventricular hypertrophy [23, 24], in ways that mimic predisposition to CVD with increasing age. The phenotypic effects of an altered early environment include altered adult growth, glucose intolerance and insulin resistance [15, 25, 26] and changes in sympathoadrenal function and hypothalamic-pituitary-adrenal (HPA) axis responses [20, 24, 27, 28], and these may constitute part of the mechanism by which cardiovascular control is affected. There is emerging evidence that the nature of the response is sex dependent [15, 23]. It is striking that, without further dietary challenges in the F1 pregnancy, features of the cardiovascular dysfunction in adult guinea pig offspring following F0 maternal diet challenge can persist into the F2 generation [24, 29]. In sheep, maternal obesity abolishes the normal leptin spike in their neonatal offspring (important for development of hypothalamic appetite circuitry) and this effect is also observed in their granddaughters [30]. Importantly, maternal dietary restriction even before conception can induce effects on vascular function in adult offspring [31], which emphasizes the importance of life-long good nutrition. Maternal body composition can be reliably manipulated through diet in sheep and it can induce long-term adverse metabolic effects and skeletal muscle structural alterations, along with cardiovascular and renal effects, in offspring [32, 33]. This underlines the concept that these effects are part of a coordinated strategy affecting development of a range of tissues, as opposed to a pathological effect.

A comparatively small number of animal studies have tested directly the concept that a mismatch between the *in utero* and childhood nutritional environments increases risk of CVD. In sheep, cardiovascular dysfunction in offspring exposed to either prenatal or postnatal undernutrition alone was not seen when pre- and post-weaning environments were similar (**Figure 2.1**) [23]. Also, undernutrition in early-mid gestation was associated with more renal lipid deposition in young adult obese sheep [34]. Late gestation undernutrition in sheep increased the neonatal appetite for fat, changed the pattern of fat deposition [35], and predisposed adult sheep offspring to hypercholesterolemia in an obesogenic environment [36]. In rats, dietary manipulation to minimize the mismatch between pre- and post-weaning nutrition minimizes endothelial dysfunction and the disruption of mechanisms regulating appetite and energy expenditure in offspring [17, 37]. In rats, a greater pre- and postnatal dietary mismatch worsened liver function [38] and decreased life span [39]. In pigs, the coronary atherosclerotic effects of a high-fat diet were prevented by prior feeding of a similar diet to the pregnant mother [40].

https://doi.org/10.1017/9781108564434.003

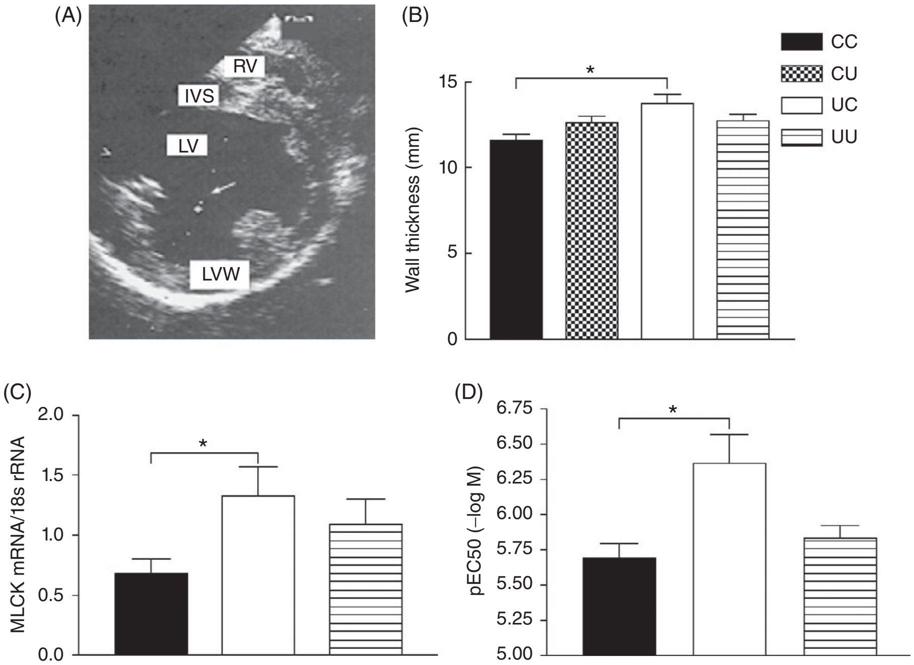

Figure 2.1 Altered cardiac morphology and coronary function in male adult sheep are absent when the mismatch of pre- and postnatal nutrition is minimized. Sheep were fed a control diet throughout pre- and postnatal life (CC), or they were exposed to moderate undernutrition either during early gestation (1–31 days' gestation, where term is 147 days; UC), during early postnatal life (12–25 weeks; CU), or both (UU). (A) An echocardiograph showing the right ventricle (RV), interventricular septum (IVS), left ventricle (LV), and left ventricular wall (LVW) of the ovine heart. (B) Thickness of the intraventricular septum; CC (n = 14), CU (n = 10), UC (n = 14), and UU (n = 14). Also shown are myosin light chain kinase (MLCK) relative mRNA expression in the coronary artery of male sheep as measured by real-time PCR [CC (n = 7), UC (n = 7), and UU (n = 4)] (C) and vascular response to acetylcholine in the coronary artery [CC (n = 10), UC (n = 9), and UU (n = 7)] (D). *, P <0.05, significantly different from CC (by one-way ANOVA). Values are mean ± SEM. Insufficient PCR and myography data were obtained from CU animals. Reproduced, with permission, from Cleal et al. [23]

The Fetus Responds to Its Environment

Adaptive or Disruptive?

Early detection of individuals who are at risk of disease is a cornerstone of predictive and personalized medicine. Children can show early signs of CVD, including atherosclerosis, and lower birth weight is associated with impaired endothelial function [41] and altered cardiac structure [42] in 8–9 year olds. In sheep, elevated blood pressure and HPA axis responsiveness were observed in 3-month-old offspring of ewes fed 85% total requirements for the first half of gestation [20]. However the fetus offers the potential for detection of an individual's risk of disease even earlier in life, and may provide future early routes for intervention. But, rather than being viewed as the start of a pathological process, current thinking suggests that some of these fetal changes might be of immediate adaptive value (prioritize and conserve energy use) and optimize a phenotype for better chance of survival over the life course [1]. Such prenatal physiological adaptive responses may operate over a broader range of normal development (Figure 2.2).

The cardiovascular system is a key part of a coordinated adaptive response, designed to get nutrients where they are really needed. Any redistribution of the cardiovascular resources might preserve the growth of some organs at the expense of others. There are also likely to be limits to the extent that the fetus can cope through cardiovascular or growth adaptations (stretched to the limit by duration or severity of challenge), at which point the cue from the maternal environment becomes 'disruptive' [1], or if the adaptations that it has made do not suit the postnatal environment this may lead to impaired function after birth and longer-term health problems. An alternative strategy would be for the fetus to be born early, and this may be a good course of action when the *in utero* environment is so hostile that life outside the womb confers a greater chance of survival. The effect of maternal adiposity and gestational weight gain on fetal blood pressure, blood flow and tissue perfusion is less well investigated.

These sorts of environmental challenges from modern western diets are relatively recent problems and ones that humans are unlikely to have evolved protective mechanisms against [1]. Unlike undernutrition, they are thought to disrupt development (nonadaptive fetal responses) in a way that might prove to be of some immediate benefit to the fetus but could lead to profound defects or perinatal death. In this section we summarize some of the evidence of fetal phenotypic changes (fetal CV homeostasis, organ perfusion, organ growth and function) in response to a suboptimal intrauterine environment (with occasional reference to overfeeding or obesity studies) and it will be obvious that with ethical restrictions on human fetal investigations, animal models have been crucial in advancing this area of research.

Bench to Bedside, and Back Again

Imaging of the fetus by ultrasound is now commonplace in developed countries as an obstetric tool to assess fetal growth and identify structural abnormalities of the fetus and blood flow anomalies through the major fetal organs, the umbilical cord, and uteroplacental circulation. Major technological advances have extended the application of this tool to the assessment of fetal movement and blood flow. Power Doppler is most commonly used to evaluate blood flow through vessels within solid organs, while color and spectral Doppler both reveal the direction of blood flow. This information, combined

https://doi.org/10.1017/9781108564434.003

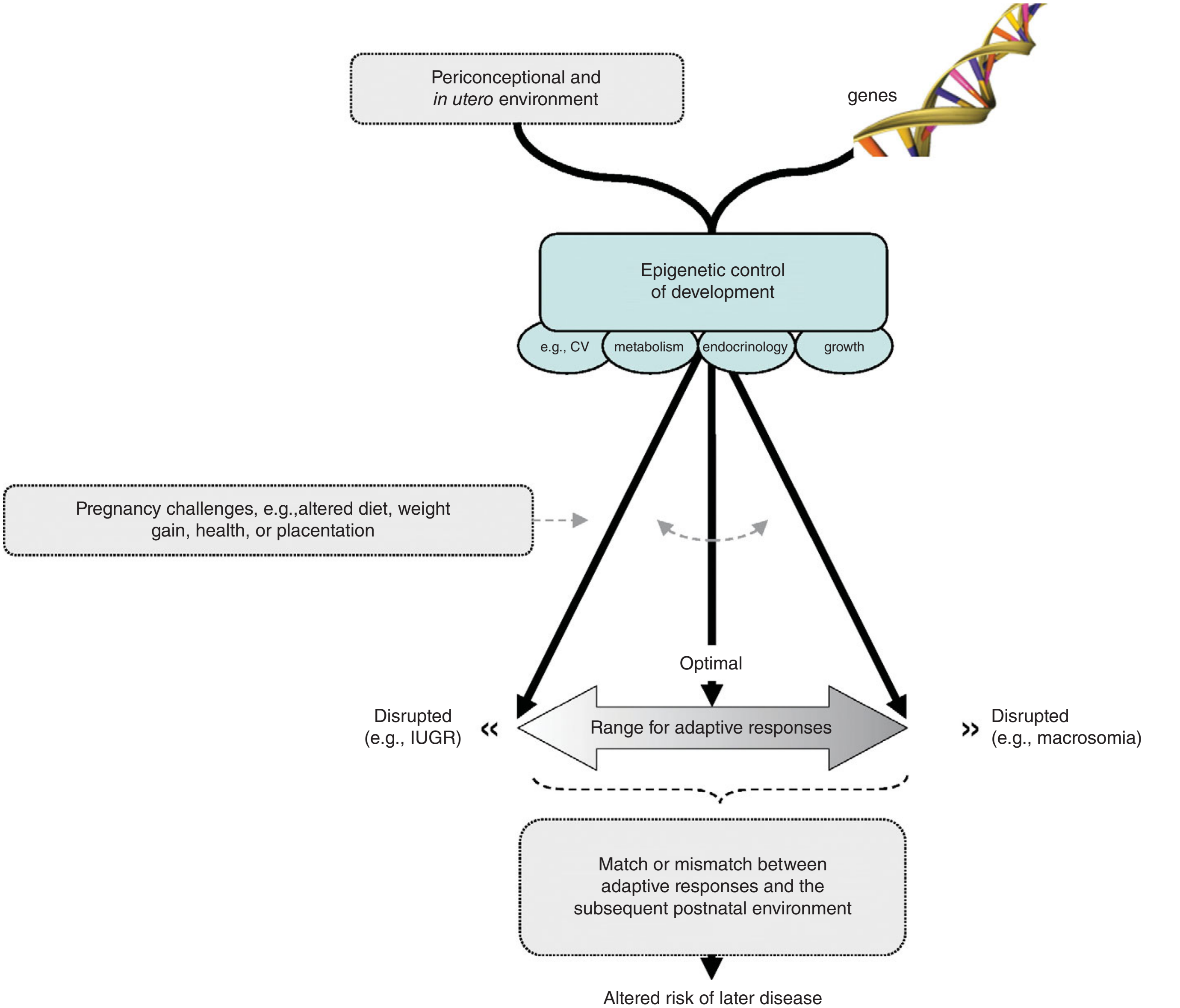

Figure 2.2 Fetal adaptive responses to challenges in prenatal life. An individual's development (cardiovascular [CV] etc.) and hence their risk of later disease is controlled by an interaction between genes and the pre-pregnancy, pregnancy, and postnatal environment. It is against this backdrop that additional pregnancy challenges may alter the individual's trajectory by instigating fetal adaptive responses. Such adaptations may be matched or mismatched to the subsequent postnatal environment and cause a progressive increased risk of later disease (e.g. hypertension, diabetes, fat deposition). At the extreme (e.g. intrauterine growth restriction [IUGR] or macrosomia), development is disrupted with risk for example of early delivery or dystocia in labor.

with fetal heart rate monitoring and calculation of amniotic fluid volume and fetal gestational age-related weight, is crucial to obstetric management aimed at identifying growth-restricted fetuses at risk of severe intrauterine hypoxia, monitoring their health, and delivering them if an adverse outcome is imminent. There is additional value of such measurements in predicting neonatal health and survival [43].

Combining such techniques with sampling of umbilical cord tissue and blood at term, and measurement of genes and products linked to growth, cardiovascular function and metabolism, might provide mechanistic insight, inform predictive markers of later dysfunction and disease risk, and lead to therapeutic interventions. However, ethical constraints on investigations in humans are a significant impediment to such mechanistic studies. Thus in parallel with clinical research activity, animal models have been developed to investigate clinical concerns that originate during pregnancy and have pathophysiological (e.g. FGR, neurological handicap, respiratory distress syndrome) or adaptive (with or without long-term consequences – see later) effects on the fetus. Observations in human cohorts frequently drive the development and direction of animal experiments.

Small rodent animal models have distinct advantages in terms of relatively low cost of purchase and housing, easy handling, a short gestation length and life span that facilitates transgenerational studies [29], and with the genome being fully sequenced, gene targeting and transgenic work is well established. Disadvantages are that they are polytocous species and intrauterine growth rates differ from monotocous species, and rats and mice give birth to young that are relatively immature compared with humans. Moreover, the scope for *in vivo* investigations of the fetal cardiovascular system in a small animal model is limited. The most widely used model of fetal physiology is the sheep. They are readily available in many countries and, cost aside, their benefits include similarities to humans in the proportion of singleton offspring (an important influence on fetal growth pattern) and in the timing of organ development (they are a precocious species in terms of brain development and have a full complement of cardiomyocytes and nephrons at term). Sheep fetuses are big enough to instrument surgically from approximately 70% gestation using aseptic technique under general anesthesia. This allows the implantation of vascular catheters, electrodes and other devices via which normal late gestation fetal physiology (cardiovascular, metabolic, respiratory, growth, brain activity) can be studied over a period of weeks, without the complicating effects of anesthesia [44]. This longitudinal developmental approach can be extended into postnatal life and is proving to be important in investigating the long-term implications of changes in the prenatal environment on later physiology and health [23]. Parallel approaches in sheep and humans, e.g. using Doppler ultrasound measurements of blood flow, are important for translation between basic and clinical science. The closer relation of non-human primates to man may be of use in extrapolating animal model data to humans by combining controlled manipulations of the maternal environment with human obstetrical monitoring tools such as Doppler ultrasound [45].

Cardiovascular Control

Several studies implicate the early developmental nutrient environment in fetal cardiovascular homeostasis (including endothelial function) and the perfusion of fetal organs. During hypoglycemia the sheep fetus redistributes blood flow away from organs such as the liver and skeletal muscle, and towards the adrenal gland. This pattern is similar to the fetal response to hypoxia [44]. Our work in sheep links lower maternal gestational weight gain with lower fetal liver weight and higher adrenal and brain-to-liver weight ratio, consistent with a redistribution of nutrient provision in favor of the adrenal gland [46]. Maternal hypoglycemia in sheep alters the fetal cardiovascular responses to a subsequent challenge such as umbilical cord occlusion [47]. In the Southampton Women's Survey, fetuses of slimmer mothers with lower body fat stores and those eating an unbalanced diet had greater liver blood flow and shunted less blood through the ductus venosus at 36 weeks' gestation [48]. Indeed, low pregnancy weight gain (rather than pre-pregnancy maternal BMI) has a strong influence on blood flow distribution between the right and left human fetal liver lobes, sparing the left lobe [49]. This evidence that the maternal environment, even prior to conception, can influence late gestation fetal cardiovascular control is corroborated by sheep studies in which early gestation undernutrition altered later gestation fetal blood pressure and flow [20, 21], resistance vessel function from the hind limb circulation [50, 51] (**Figure 2.3**), and cardiovascular responses to a further period of nutrient restriction [44]. In some studies maternal undernutrition during the last 20% of gestation in sheep increased fetal blood pressure [52]. Candidate mechanisms underlying the effects of altered maternal undernutrition on fetal cardiovascular control include the redistribution of resources through fetal carotid body sensing of glucose (known to occur in adults) [46], the hypothalamo-pituitary axis, and the renal-renin angiotensin system (RAS). The impact on fetuses differs between the sexes and is greater in twins.

Growth and Function of the Fetus – at What Level?

The early epidemiological literature links size at birth, across the normal birth weight range, and adult health (the DOHaD concept, see above). However size at birth is not an accurate measure of the prenatal environment, because different patterns of fetal growth can result in similar birth weights, and the processes affecting fetal growth are complex. In clinical practice, size at birth at the 50th centile is often assumed to assure the best birth outcome. But a growing body of evidence suggests that birth weight between the 80 and 90th centiles, indicative of some maternal constraint in operation, is better for perinatal survival [53]. A World Health Organization initiative is leading the development of revamped fetal growth charts to reflect multiple populations. This could improve the prediction and diagnosis of obstetrical complications, perinatal mortality, child morbidity, and adult health risk [54].

In human and animal studies, size continues to be reported both as an outcome variable and an additional marker of the challenge imposed to which other outcome variables can be related. However, the way in which fetal growth is reported varies considerably between studies from body weight/ dimensions (e.g. crown-rump length), to individual organ weights, to cell number (e.g. skeletal myofibers or nephron number), to the expression of growth-regulating genes such as *igf1*. Gaining an overall picture of the growth strategy taken by fetuses in the face of altered maternal nutrition is therefore complex. However in terms of fetal weight and length, sheep fetuses reduce growth following severe [55] but not moderate maternal undernutrition [21, 44]. The slowing of growth in response to severe late gestation maternal undernutrition is influenced by growth rate and maternal nutrition in early gestation [55]. Importantly, moderate undernutrition, with

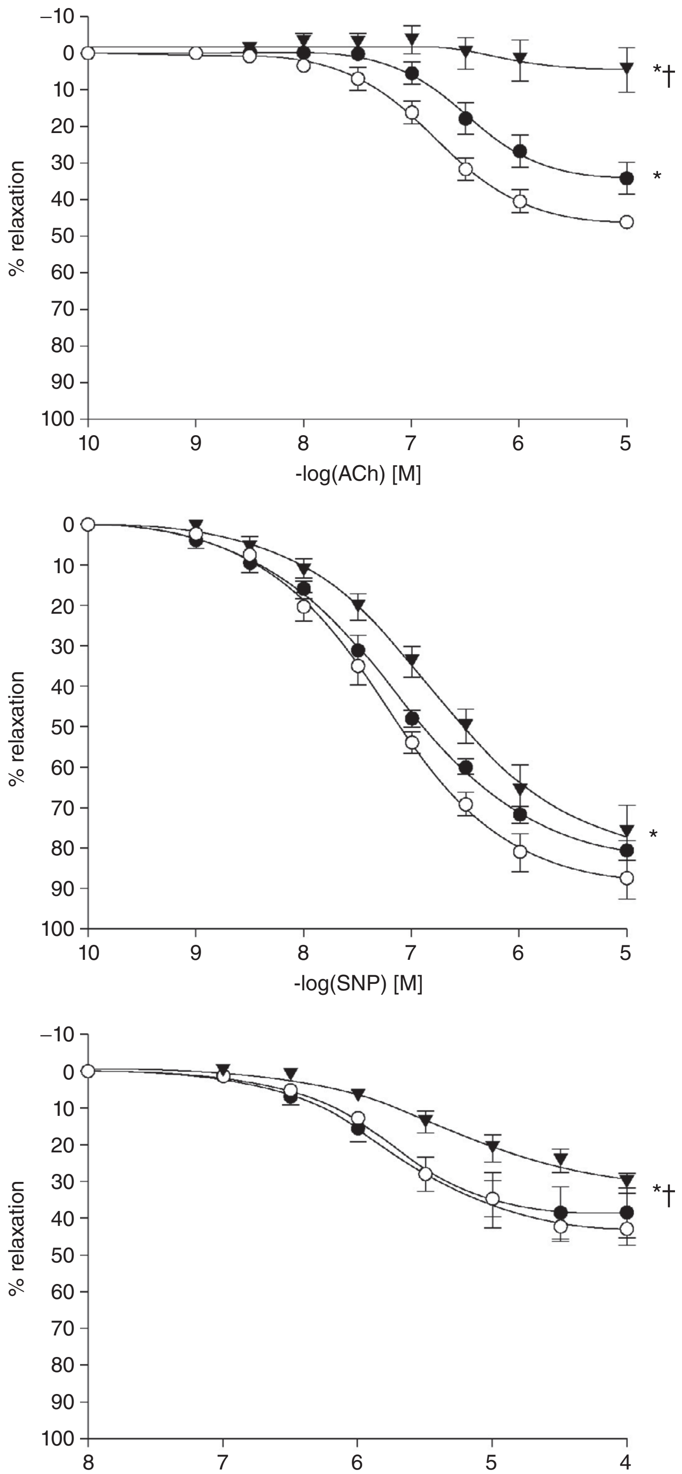

Figure 2.3 Vasorelaxation to acetylcholine (ACh), sodium nitroprusside (SNP) and UK14304 (α_2 adrenoceptor agonist) in femoral arteries of fetuses from control and nutrient-restricted ewes. Data are expressed as a percentage of initial preconstriction. Values are mean ± SEM in control (○), 70% global (●), and 70% protein (▲) fetuses. * $P < 0.05$ (ANOVA), significantly different from control; † $P < 0.05$ (ANOVA), significantly different from global group.
Reproduced, with permission, from Nishina et al. [50]

effects on cardiovascular control, has been linked to altered growth at the organ and cellular level [46, 56] and therefore cellular markers of growth rather than basic fetal biometry are important.

The fetal response to undernutrition involves the redistribution of resources that may favor vital organs such as the heart, and give rise to organ-specific effects on growth and function [44]. In sheep, maternal undernutrition from mid pregnancy onwards alters the mechanisms by which isolated fetal coronary vessels relax [57]. Maternal undernutrition around conception [58] or during early to mid gestation was associated with compensatory growth, and increased expression of associated genes, of both heart ventricles by mid gestation [59, 60], and even with re-alimentation this altered ventricular development will persist into late gestation [61] and can be accompanied by increased pericardial adiposity. Overfeeding-induced maternal obesity in sheep from before conception and during pregnancy was associated with thicker heart walls and enlarged ventricles, with fat deposition, inflammation, and impaired isolated-heart function in mid-late gestation fetal sheep [62].

Many studies suggest that perfusion of the fetal periphery, i.e. limbs, is decreased during maternal undernutrition. There was no effect on body or organ weights, but muscle-specific structural changes have been observed. Moderate (40–50% restriction) early- and late-gestation undernutrition reduced skeletal myofiber and capillary density in *tricep brachii* but not *soleus* muscles from late-gestation sheep fetuses, and a rise in mRNA levels of the insulin receptor and type 1 insulin-like growth factor (IGF) receptor were observed in fetuses from the late-gestation group, possibly as a compensatory response to maintain growth [56] (Figure 2.4). Indeed in cows, 85% of metabolizable energy requirements (for the second half of pregnancy) did not appear to impact overall fetal growth (~0.9 gestation), but an increased mRNA abundance of genes related to the IGF axis and insulin sensing was observed in the fetal *longissimus dorsi* but not *semitendinosus* muscles [63]. In sheep, even periconceptional and preimplantation undernutrition have been observed to alter the expression in late-gestation fetal *quadriceps* muscle of genes regulating myogenesis [64] and insulin signaling [65], and are important in setting growth trajectories and the growth response to later intrauterine periods of nutrient restriction. A prenatal origin of diseases of the liver (e.g. non-alcoholic fatty liver) has been implicated probably due to pathophysiological-type responses to maternal obesity [66]. Also, maternal undernutrition in periconception, early or mid-late pregnancy in sheep can alter the fetal metabolic profile [67] and is associated with more adiposity and changes in the thermogenic capacity and the insulin and fatty acid oxidation signaling pathway in visceral fat [68], altered adrenal growth and HPA function [20], and altered liver growth, gluconeogenesis and lipid metabolism pathways in fetal liver [69, 70]. Overall, these fetal adaptive-like responses to maternal nutrition in muscle, liver and fat are also likely to have metabolic ramifications in later life.

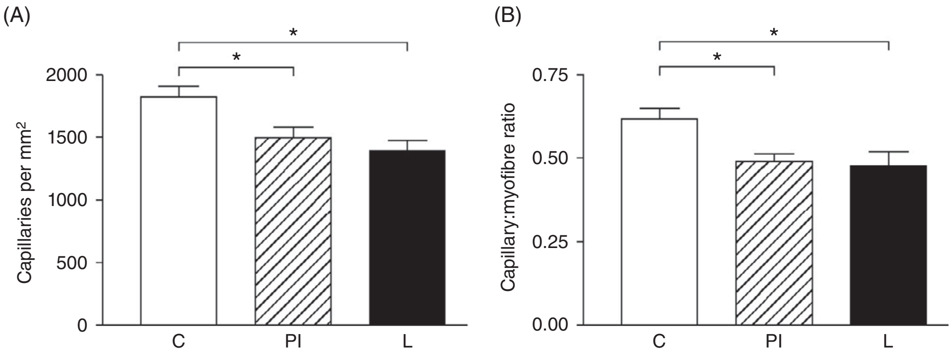

Figure 2.4 Capillary density and capillary:myofiber ratio in the *triceps brachii* muscle of late-gestation fetuses. Capillary density (A) and capillary : fiber ratio (B) in control (C, $n = 6$), peri-implantation restricted (PI, $n = 9$) and late restricted (L, $n = 6$) groups. Values are mean ± SEM. * $P < 0.05$. Reproduced, with permission, from Costello et al. [56]

Timing of Birth

Spontaneous preterm labor is of serious obstetrical concern but discussion of its etiology is beyond the scope of this chapter. Nonetheless, adjusting the timing of labor is one potential strategy that the fetomaternal unit could adopt in the face of a change in the maternal nutrient environment. In sheep, poor maternal weight gain in the first 20% of gestation (resulting from undernutrition) increased birth weight and gestation length in male twin pregnancies [25]. It seems likely that the underlying mechanisms involve the HPA axis through changes in the preparturient fetal cortisol surge (essential for the initiation of normal labor) since the early nutrient restriction also reduced maternal plasma cortisol levels at 30 days of gestational age (dGA), which might have influenced the development of the fetal HPA axis. Indeed mild undernutrition (a 15% reduction in maternal nutrient intake) during early gestation reduced pituitary and adrenal responsiveness in late-gestation ovine fetuses [27]. In contrast, accelerated maturation of the fetal HPA axis is seen with a more severe undernutrition challenge [71], which appears to trigger early maturation and delivery of the fetus. Taken together, these findings suggest that the effect of the maternal environment on the timing of labor depends greatly on the timing, duration, and severity of the insult. Early delivery could make sense if the environmental challenge exceeds the limits of fetal adaptation (e.g. growth and cardiovascular control), while increased gestation length, however modest, could thus be part of a strategy for allowing more time *in utero* for maturation and growth in the face of a milder challenge.

The Pathway to Fetal Adaptive Responses

Candidate mechanisms for alterations in fetal cardiovascular control and growth in the face of an altered developmental nutrient environment include glucocorticoids (i.e. HPA axis), the renal-RAS, and the IGF axis. Additional work has been conducted on the mechanisms by which dietary imbalance in pregnancy may be transmitted to the fetus. These include the carotid chemoreceptors, which mediate rapid fetal cardiovascular adaptations to hypoxia and are responsive to reductions in circulating plasma glucose [46, 72]. In this section the role of maternal adaptations, placental adaptations, and epigenetic mechanisms in the pathway to fetal adaptations will be considered.

Maternal Adaptations to Pregnancy

Nutritional restriction markedly reduced concentrations of total α-amino acids (particularly serine, arginine-family amino acids, and branched-chain amino acids) and polyamines in maternal uterine artery and fetal plasma, and in fetal allantoic and amniotic fluids, at both mid and late gestation. During normal pregnancy, there is a redistribution of cardiac output in favor of the reproductive tract. In humans this is contributed to by vasodilatation in the uterine bed via vascular endothelial growth factor (VEGF)/nitric oxide mechanisms. In the rat, dietary restriction of the mother (50%) resulted in maintained maternal liver blood flow at the expense of the pregnant uterus [73] and maternal low-protein diet impaired vasodilator response to VEGF in late pregnancy uterine arteries *in vitro* [74]. In sheep, 40% restriction of maternal nutrition decreased uterine artery blood flow [75]. In hypertensive rat offspring of protein-restricted dams the mesenteric vasodilator responses to acetylcholine are blunted [18]. Subsequent studies suggested that the vascular dysfunction induced by a low protein diet in the maternal uterine/mesenteric arteries and in the offspring mesenteric vessels (and hypertension) could be ameliorated by dietary supplementation with folate, a key player in gene methylation, or the conditionally essential amino acid glycine [76]. Uterine vascular dysfunction of female offspring is likely to contribute to intergenerational effects observed in F2 generation offspring of raised blood pressure and mesenteric artery endothelial dysfunction in adult life [29].

The Role of the Placenta

The environment of the preimplantation embryo is well known to influence the allocation of cells to the inner cell mass (which becomes the fetus) and the trophectoderm (which becomes the placenta) [77]. Once the placenta is established, it can act as a simple conduit for nutrients from the maternal circulation to the fetus. But there is mounting evidence that the

https://doi.org/10.1017/9781108564434.003

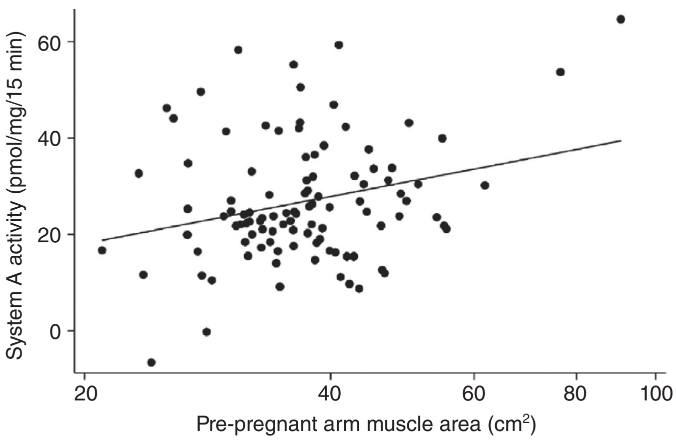

Figure 2.5 Placental system A activity at birth is associated with pre-pregnant maternal upper arm muscle mass. $r = 0.27$, $P = 0.007$, $n = 103$. Reproduced, with permission, from Lewis et al. [82]

placenta may take a more active role in mediating the fetal response to maternal constraints on development, and it is suggested that the way in which the placenta responds will be subject to evolutionary selective pressures to increase Darwinian fitness (i.e. reproductive success) [78]. The size and function of the placenta is known to be influenced by a wide range of hormones and nutrients from the mother and fetus. In humans, maternal upper-arm muscle mass before pregnancy, an index of lean mass, is related to activity of the amino acid transporter system A in the term placenta [78] (Figure 2.5). These data provide the first link between maternal body composition and placental function, which could influence the environmental signal that is transmitted to the fetus. In Rhesus macaques, placental perfusion (maternal spiral artery Doppler) was lower following a low protein diet from conception and throughout pregnancy, possibly due to altered spiral artery remodeling in early pregnancy involving nitric oxide and/or tissue inflammation [79]. In addition, the placenta adapts its function (e.g. glucose and system A transporters) to help maintain fetal growth when placental growth is compromised by general maternal undernutrition or even a specific deficiency in vitamin D [80]. In mice, elegant mechanistic studies using genetic models of overgrowth and fetal-placental mismatch suggest that the placenta can fine-tune the supply of maternal resources to the fetus via p110α (PIK3CA) in accordance with both the fetal drive for growth and the maternal ability to supply the required nutrients [81].

Epigenetic Modifications

The persistent effects of the early nutrient environment on later cardiovascular phenotype of the offspring may be mediated by epigenetic processes that alter gene expression without changing DNA sequence. These could set up later responses to transcription factors and their impact may not become apparent until later life, perhaps when an additional challenge is received. Depletion of vitamin B12, folate and methionine in the periconceptional diet of ewes led to widespread altered DNA methylation in the liver of adult offspring, which was associated with elevated blood pressure [83]. In rats, maternal protein restriction in pregnancy caused increased methylation and decreased expression of genes (e.g. peroxisome proliferator-activated receptor [PPAR] gamma and glucocorticoid receptor [GR]) that regulate cardiovascular and metabolic function. These effects can be prevented by maternal dietary folate supplementation and can be transmitted to the F2 generation [84]. In sheep fetuses, periconceptional undernutrition altered epigenetic regulatory mechanisms (i.e. histone acetylation and methylation) in hypothalamic POMC (pro-opiomelanocortin) and GR genes that influence food intake, energy expenditure, and glucose homeostasis in later life [85]. In the baboon, hypomethylation of the *PCK1* gene promotor region in the fetal liver was observed following moderate maternal undernutrition between 0.16 and 0.9 gestation [86]. In sheep, prenatal and postnatal undernutrition altered methylation of imprint control regions of imprinted gene clusters *DLK1*/*MEG3* and *IGF2* in adult offspring liver in a sex-specific manner [87] and may underlie the metabolic and growth phenotype observed in these offspring. Overall, the evidence to date suggests that epigenetic modifications provide a mechanism by which environmental influences on cardiovascular control, metabolism and growth might be integrated into a life course response, starting from very early in development.

Adjusting Our Focus on Interventions for Adult Health

The substantial body of work on adaptation by the fetus to the early environment, from animal physiology and human epidemiology and trials, has fundamentally refocused how we view what makes us healthy. The emergent role of early development in setting an individual's risk of disease in later life substantially undermines hopes that health interventions in adult (sometimes in childhood) lifestyle and nutrition will succeed in combating the epidemic of NCDs. One hope is that the animal model data combined with current clinical diagnostic tools (e.g. umbilical cord sampling, non-invasive fetal heart rate monitoring, and ultrasound assessments of the fetal circulation and growth) will help derive early life markers of cardiovascular risk and inform future interventions in early (even fetal) life. Methylation of several genes in human umbilical cord tissue (including the promoter of the long non-coding RNA ANRIL, CDKN2A) have now been associated with newborn/childhood body composition, bone mass and cardiovascular risk, and in some cases additionally linked to markers of the prenatal environment [87, 88]. Indeed, the safety of more targeted interventions continues to be debated, including pharmacological approaches (e.g. statins or metformin) and gene therapy to improve uteroplacental perfusion [89]. However the biggest change may come from adjusting the focus of the diet and lifestyle advice from adult life to much earlier in the life course. The outcomes from randomized controlled trials of complex interventions targeting diet and physical activity from 15–18 weeks' gestation in pregnant women who

https://doi.org/10.1017/9781108564434.003