Seminars in Fetal & Neonatal Medicine 15 (2010) 113–118

Contents lists available at ScienceDirect

Seminars in Fetal & Neonatal Medicine

journal homepage: www.elsevier.com/locate/siny

Effects of maternal obesity on fetal growth and body composition: implications for programming and future health

Dilys J. Freeman*

Developmental Medicine, Faculty of Medicine, University of Glasgow, 2nd Floor McGregor Building, Western Infirmary, Dumbarton Road, Glasgow G11 6NT, UK

keywords:
Insulin
Metabolism
Obesity
Programming

SUMMARY

Since the hypothesis linking low birth weight and poor fetal growth with future risk of cardiovascular disease was first proposed, there has been much interest in the early origins of disease. As rates of obesity increase and as maternal obesity has become common, interest has been directed towards the early origins of obesity. It is likely that a complex interaction of inherited gene effects and in-utero environment may interact in the developing fetus to programme pathways leading to future obesity. It is clear that maternal metabolism is disturbed in pregnancy in obese women, and that offspring of obese mothers have a higher percentage of body fat and are insulin resistant. This review discusses the ideas contributing to the current working concept of obesity programming, and discusses several potential mechanisms that may underlie obesity programming and susceptibility to future metabolic and vascular disease.

1. Introduction

The concept of early origins of adult disease is popular but lacks definitive proof, especially in humans. Reports linking disturbances in offspring cardiovascular health (hypertension) to maternal ill-health and offspring low birth weight emerged in the late 1980s. Barker brought this information together and developed 'Barker's hypothesis'.[1] This stimulated a burgeoning area of research into the early (both fetal and childhood) origins of disease. Recently with the increasing prevalence of obesity in western societies,[2] interest in the early origins of obesity has grown.

Maternal obesity is increasing in prevalence[3] and is associated with adverse perinatal outcomes. It has been suggested that pregnancy in obese women has an in-utero influence on offspring leading to a cycling of risk factors through the generations.[4] This review considers a number of hypotheses and the evidence for in-utero origins of obesity, and discusses some of the potential mechanisms for such effects. Much of the evidence in humans to date has been, necessarily, based on epidemiological data and it is only recently that real attempts have been being made to identify molecular mechanisms for programming effects. It is clear that for the in-utero environment to have any real impact on offspring obesity, it must have long-lasting, permanent effects on biological processes such that the consequences are observed later in life. The appearance of obesity in offspring may be due to mechanisms with small, chronic cumulative effects or that have the potential to interact with future environmental factors (e.g. ageing, dietary intake, menopause). Obese offspring are more likely to suffer from metabolic diseases such as type 2 diabetes and cardiovascular disease later in life, but the extent to which the aetiology of obesity starts in utero is presently unknown.

2. Programming hypotheses

2.1. Barker's hypothesis

A succinct review of the evidence of the developmental origins theory according to Barker is provided by Barker himself.[5] Barker's hypothesis asserts that undernutrition in utero leads to permanent changes in tissue structure, function and metabolism that predispose to coronary heart disease (CHD) later in life. The hypothesis was built upon early epidemiological observations that geographical variation in CHD rates across England and Wales correlated with past variation in neonatal death rates. At the time of recording of these neonatal death rates, death was mostly attributable to low birth weight. Further longitudinal epidemiological studies from very diverse populations have corroborated the association between low birth weight and CHD incidence.[6–9] Associations with stroke,[8,10] glucose intolerance and type 2 diabetes[11,12] have also been observed. The associations are independent of gestation length and so are inferred to be the result of reduced fetal growth.

* Tel.: +44 0141 211 2785; fax: +44 0141 211 2012.
E-mail address: d.freeman@clinmed.gla.ac.uk

1744-165X/$ – see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.siny.2009.09.001

Glucose intolerance, metabolic syndrome and type 2 diabetes are arguably the most relevant pathologies when considering the programming of future obesity. The explanation for the link between these pathological states and low birth weight has been hypothesised by Hales and Barker to be due to a 'thrifty phenotype'.[13] This hypothesis suggests that poor nutrition in fetal life can lead to permanent changes in the glucose–insulin axis. This would occur by compromised development of the pancreas, including reduced β-cell number and function, resulting from the preservation of brain growth at the expense of other organs when in-utero nutrition is limited. Reduced ability to secrete insulin would not be detrimental to individuals who continue to be poorly nourished and are thin, but would be problematic in conditions of nutritional excess leading to insulin resistance, obesity, metabolic syndrome and type 2 diabetes later in life.[13]

The generalisability and relevance of Barker's hypothesis has been questioned. There are technical concerns with the statistical analysis and suggestions that the data may be subject to inadequate accounting for confounding, selective use and publication bias.[14] Meta-analysis shows that the effect, at least on hypertension, is small.[14] It is unclear how applicable the hypothesis would be to the early origins of obesity as a prospective cohort study in the UK was unable to relate maternal nutritional intake to fetal growth in a well-nourished population.[15] Furthermore data from animal models are rather inconsistent and incorporate a variety of nutritional programming stimuli; thus mechanistic data to support the hypothesis are lacking.

2.2. Genetic origins

Hattersley and Tooke[16] proposed an alternative view to the intrauterine programming of insulin resistance, type 2 diabetes and CHD. They proposed that the link between low birth weight and future insulin resistance could be genetically determined. If an individual inherits genes that confer a propensity to insulin resistance, then that individual will have slower growth in utero, as insulin is the main fetal growth hormone, and hence will have a low birth weight. Later in life those same genes will interact with environment to predispose to obesity, insulin resistance and subsequent type 2 diabetes and CHD. Hence the low birth weight and propensity to metabolic disease are not directly linked but are co-associates of a particular genetic make-up.

The principle of this argument is neatly demonstrated in a study of a mutation in the glucokinase gene. Glucokinase is important in pancreatic β-cell sensing of blood glucose levels[17] and the mutation leads to the individual failing to detect blood glucose concentration properly. It was found that the presence or absence of a mutation in the mother and/or the baby could have marked effects on offspring birth weight.[18] When neither mother nor baby has the mutation, then birth weights are as expected on the 50th centile. When the mother has the mutation, but the baby does not, this leads to high maternal blood glucose levels which are transmitted across the placenta to the fetus via facilitated diffusion. This stimulates fetal insulin production and promotes fetal growth, leading to babies being born with a high birth weight around the 90th centile. When the baby has the mutation but the mother does not, then maternal glucose levels are normal but the fetus fails to sense these and fetal insulin production is decreased, leading to lower birth weights around the 25th centile. Interestingly, when both mother and baby have the mutation these two effects cancel out and babies are born with birth weights around the 50th centile. In the long term, offspring of mothers with a glucokinase mutation, who had been exposed to high intrauterine concentrations of glucose, did not grow up to have poorer β-cell function or glucose tolerance compared to offspring of fathers with the glucokinase mutation. Hence intrauterine exposure did not have any effect additional to that of the gene mutation per se.[19]

2.3. Intrauterine metabolic environment

Observations in Pima Indians indicated that type 2 diabetes in offspring was more common when the mother was diabetic at the time of pregnancy (45%) rather then when mothers were diabetic subsequent to pregnancy (i.e. pre-diabetic at pregnancy) (8.6%) or were not diabetic (1.4%).[20] This influence persisted after accounting for paternal diabetes, age of onset of diabetes and offspring weight. These data suggest that in addition to genetic factors, expression of maternal diabetic risk factors during pregnancy may influence the long-term health of the offspring. A study of 19 Pima Indian nuclear families demonstrated that in 15 of these families, diabetes was more common in siblings born after the mother was diagnosed with diabetes rather than before diagnosis (odds ratio: 3.7; 95% confidence interval: 1.3–11.3; $P = 0.02$).[21] This result did not hold for fathers (1.1; 0.3–2.5; $P = 0.80$). Furthermore offspring exposed to a diabetic intrauterine environment were more obese than their siblings born before the mother was diagnosed with diabetes ($P = 0.003$).

Overweight/obese women with normal glucose tolerance have neonates who are born with an increased percentage body fat.[22,23] Both GDM and obese pregnancy are associated with abnormalities in maternal lipid and carbohydrate metabolism and inflammatory status.[24] Catalano proposed that the metabolic abnormalities associated with obese pregnancy could be communicated to the offspring via the intrauterine environment[4] resulting in development of obese offspring hence cycling obesity through the generations. Offspring of obese pregnancies have a metabolic profile consistent with insulin resistance.[22] Offspring obesity could result from fetal overnutrition (high maternal circulating glucose and lipid concentrations), from upregulation of placental transporters, or could be due to the influence of the metabolic environment on placental and offspring tissue gene expression or metabolism.[25]

2.4. Maternal metabolites: quantity or quality?

Data already exist supporting a role for maternal metabolism in influencing offspring future cardiovascular health. The earliest form of the atherosclerotic plaque is the fatty streak and this can be observed in children and young adults. Using postmortem material, Napoli *et al.* found that maternal hypercholesterolaemia is associated with enhanced fatty streak formation in fetal arteries[26] and suggested that this may be related to the strong correlation between maternal and fetal plasma cholesterol levels in fetuses of less than 6 months of age. Animal data indicate that increased maternal concentrations of plasma cholesterol can increase placental transport to the fetus.[27] These early atherosclerotic lesions appear to regress as the children grow older but the rate of regression is less in those offspring of hypercholesterolaemic mothers.[28] Since maternal third trimester plasma cholesterol levels do not differ between lean and obese pregnant women[24] the relevance of this as a potential mechanism for increased disease in the offspring of obese mothers might be questioned. However, studies of lipoprotein metabolism have shown that once plasma triglyceride levels increase above a threshold of 1.5 mmol/L then plasma low density lipoprotein (LDL), the main carrier of cholesterol in blood, is converted from larger, less dense particles to smaller, denser LDL particles.[29] These small dense LDL (or LDLIII) are easily oxidised and are more able to infiltrate subendothelial spaces and are regarded as being highly atherogenic.[30] During pregnancy, plasma triglyceride levels clearly exceed the threshold of 1.5 mmol/L particularly in obese women.[24] The LDL subfraction

profile is demonstrably shifted to a significantly higher proportion of LDLIII in the third trimester in obese pregnant women compared to lean women (D.J. Freeman *et al.*, unpublished data). In support of this, in GDM women, obesity attenuated the decreased susceptibility of LDL to oxidation associated with late gestation.[31] Placenta has both LDL[32] and oxidised LDL (OLR-1)[33] receptors at the maternal side. Hence, either direct delivery of oxidised LDL to the fetus or indirect formation of oxidised LDL in the fetus due to increased placental cholesterol transfer from the mother[27] may cause increased fatty streak formation in the offspring. Interestingly, placental OLR-1 expression is increased in obese pregnancy.[33] Napoli *et al.* observed that the fatty streaks in the fetuses of hypercholesterolaemic mothers did contain oxidised LDL.[26] These data suggest that it is not only the quantity of metabolites and nutrients that may influence the offspring, but also the quality. It has been proposed that such early exposure to abnormal metabolic, and also inflammatory, environment may start offspring on a steeper trajectory towards disease and hence programme a propensity to atherosclerotic disease later in life.[34]

3. Potential mechanisms for programming effects

3.1. Effects of maternal metabolism on offspring tissue function

Clearly suboptimal functioning of some key tissues may have a large potential impact on whole-body insulin sensitivity and appetite regulation which may lead to an increased propensity for obesity. It has been shown in pancreatic β-cells that high intracellular cholesterol concentration promotes dimerisation of neuronal nitric oxide synthase, leading to downregulation of glucokinase and hence impaired glucose sensing.[35] Furthermore, exposure of pancreatic β-cells to oxidised LDL results in β-cell apoptosis and a reduction in insulin secretion.[36] Thus if the fetus is exposed to high cholesterol or high triglycerides and a degree of oxidative stress, this might lead to a reduced β-cell capacity in the pancreas which could result in premature pancreatic failure in later life and development of insulin resistance, obesity and high cardiovascular risk.

Recently there has been substantial interest in programming effects on the brain, particularly in the potential for influencing appetite regulation pathways. The hypothalamus has receptors for both insulin and leptin[37] that regulate the expression of neural peptides in distinct neuronal populations in the hypothalamic arcuate nucleus. This includes downregulation of the orexigenic neuropeptide Y which in turn leads to upregulation of the anorexigenic hormone corticotrophin-releasing hormone in the paraventricular nucleus. Insulin also activates the melanocortin system, leading to production of the anorexigenic α-melanocyte stimulating hormone. Thus both leptin and insulin signalling in the brain may regulate complex pathways involved in appetite regulation. In addition, insulin has been shown to affect neuronal survival and plasticity.[37] Since both maternal[24] and cord blood[22] insulin and leptin concentrations are increased in offspring of obese mothers, it is possible that in-utero exposure to higher concentrations of these hormones may influence the development of appetite regulation pathways in the offspring, leading to a propensity to become obese later in life. Poston and Taylor have developed a rat model of obesity programming and have shown that offspring of obese rats have an amplified and prolonged neonatal leptin surge.[38] Postnatal molecular changes in the hypothalamus are evident and leptin-induced appetite suppression is reduced. These authors propose that the excess exposure to postnatal leptin in the offspring of obese rats leads to leptin resistance and may programme for obesity.

3.2. Gene expression and epigenetic effects

The study of the effects of maternal hypercholesterolaemia on offspring arterial gene expression has been carried out in LDL receptor-deficient mice on normal or high-fat diets.[39] Maternal hypercholesterolaemia doubled atherosclerotic lesion size in male offspring. A microarray analysis of the expression of 11 000 murine genes in the descending aorta indicated that 139 genes were significantly regulated in offspring of hypercholesterolaemic mothers. These results were validated for a subset of genes using semiquantitative reverse transcription–polymerase chain reaction and comparative immunostaining. The functions of most of the identified genes were unknown, but some were target genes of nuclear receptors such as RXRα. The difference in gene expression was apparent in offspring at three months of age; thus, if it had been induced by in-utero environment then it had been retained for some considerable time. The differences in degree of expression were modest, implying that programming resulted in subtle changes in gene expression lasting over a long period of time rather than in acute regulation. Offspring had identical lipid levels.

In rats, folate supplementation during pregnancy improves offspring cardiovascular function induced by protein restriction.[40] Folate is an important vitamin involved in the generation of S-adenosyl methionine (SAM), a one-carbon unit donor that is involved in DNA methylation. DNA methylation is a characteristic of eukaryotic genomes and involves the addition of a methyl group to the 5' carbon of cytosine within CpG-rich regions of the genome. These regions are common within gene promoters and the initial exons of genes. Methylation patterns can be copied into daughter DNA after mitosis and have been suggested to form a 'cellular memory'. Patterns of global gene methylation may reflect overall gene expression levels. Hypermethylation at a specific gene leads to decreased transcription (probably by physically restricting access by transcription factors) whereas hypomethylation leads to increased transcription. One of the first experiments showing direct evidence for programming was published by Waterland and Jirtle in 2003.[41] Mouse coat colour is coded for by the agouti gene (A^{vy}PS1A) and can vary between yellow and dark brown. When the A^{vy}PS1A gene is hypomethylated the yellow colour phenotype is observed; when the gene is maximally methylated the dark brown colour is observed. Pregnant A^{vy} mice had their diet either unsupplemented or supplemented with dietary methyl donors (folic acid, vitamin B_{12}, choline and betaine) during pregnancy and the number of offspring of each coat colour was counted. Offspring from mothers with the dietary supplement were more likely to have darker coat colour and had a higher degree of A^{vy}PS1A methylation. Thus a short-term dietary intervention, admittedly rather severe and unphysiological, during pregnancy was able to have a long-lasting influence on offspring phenotype.

Further work from this group has shown that maternal dietary genistein supplementation during pregnancy (at a level comparable to a human high soy protein diet) also shifted coat colour from yellow towards dark brown.[42] CpG methylation at the A^{vy} gene is inversely correlated with ectopic *Agouti* expression and obesity incidence. Genistein increased tissue methylation during early embryonic development and the effect continued into adulthood. The supplemented animals were also protected from obesity. Thus this is the first evidence, in a mouse model, that dietary in-utero genistein supplementation switches off ectopic *agouti* gene expression by altering the epigenome (via enhanced histone acetylation) and hence influences susceptibility to obesity in adult life. Others have found that pregnant rats fed a 'programming diet' (protein restriction) had a persistent reduction in methylation of the *PPARα* promoter in offspring.[43] This could be retrieved by

maternal folate supplementation during pregnancy. These animal studies show that epigenetic changes in offspring may be induced by maternal dietary manipulation and lead to long-term changes in expression of genes involved in energy metabolism and obesity, suggesting that this is a potential mechanism for the programming of obesity. The relevance of these studies to the human situation remains to be explored. It is interesting to note that obese pregnant women tend to have a poorer diet and have been shown to be folate deficient.[44]

3.3. Vascular endothelium

Data from mouse experiments have suggested that the endothelium is another organ whose development may be influenced by maternal environment. Rodents do not naturally develop cardiovascular disease and have a plasma lipid profile completely unlike humans. However, laboratory animal models have been developed that do show development of atherosclerotic lesions similar to, but not exactly identical to, humans especially when the animals are fed high fat diets. One of the most common models is the apolipoprotein E knockout mouse (*apoE*$^{-/-}$) which has a lipid profile more like humans and is also pro-inflammatory and has increased oxidative stress. The *apoE* knockout mouse develops atherosclerotic lesions in response to a high fat diet. Interestingly when offspring from *apoE*$^{-/-}$ and *apoE*$^{+/+}$ mothers were compared there was a significantly lower carotid artery endothelial cell volume in offspring from *apoE*$^{-/-}$ mothers.[45] The *apoE*$^{-/-}$ mothers had an exaggerated cardiovascular risk profile during pregnancy; the authors suggested that intrauterine exposure to maternal cardiovascular risk factors may have been responsible for this effect and proposed that a reduced endothelial cell volume would lead to a greater susceptibility to atherosclerosis later in life.

Endothelial progenitor cells (EPCs) arise from a population of circulating mononuclear cells and have the capacity to form new blood vessels and contribute to vascular repair.[46] There is interest in EPCs in the field of cardiovascular medicine as it has been observed in middle-aged men, with no history of cardiovascular disease, that numbers of circulating EPCs are inversely associated with the number of cardiovascular risk factors (Framingham risk score) and positively associated with endothelial function.[47] EPCs are also detectable in cord blood and it has been noted that levels of EPCs are significantly reduced in offspring of pregnancies complicated by pre-eclampsia[48] and type 1 diabetes.[49] Thus it is possible that offspring who have been exposed to an adverse intrauterine metabolic environment, that might affect EPC viability, start life with fewer EPCs and therefore have a compromised ability to effect vascular repair later in life, leading to premature expression of vascular disease.

4. Potential for intervention

If exposure to an adverse metabolic environment does exert a malign influence on offspring, then the opportunities for intervention are worth considering. Fortunately metabolic disturbances, especially those associated with obesity, can be easily remedied by changes in lifestyle and small amounts of weight loss.[50] However, lifestyle changes are notoriously difficult to implement, especially in pregnancy. Clearly other interventions may be more successful. In the mouse studies of atheroma in offspring of hypercholesterolaemic mothers, treatment of the mothers with cholestyramine (which reduced maternal plasma cholesterol levels) or vitamin E (an antioxidant that does not influence plasma cholesterol levels) resulted in offspring with fewer atherosclerotic lesions, similar in number to offspring of normocholesterolaemic

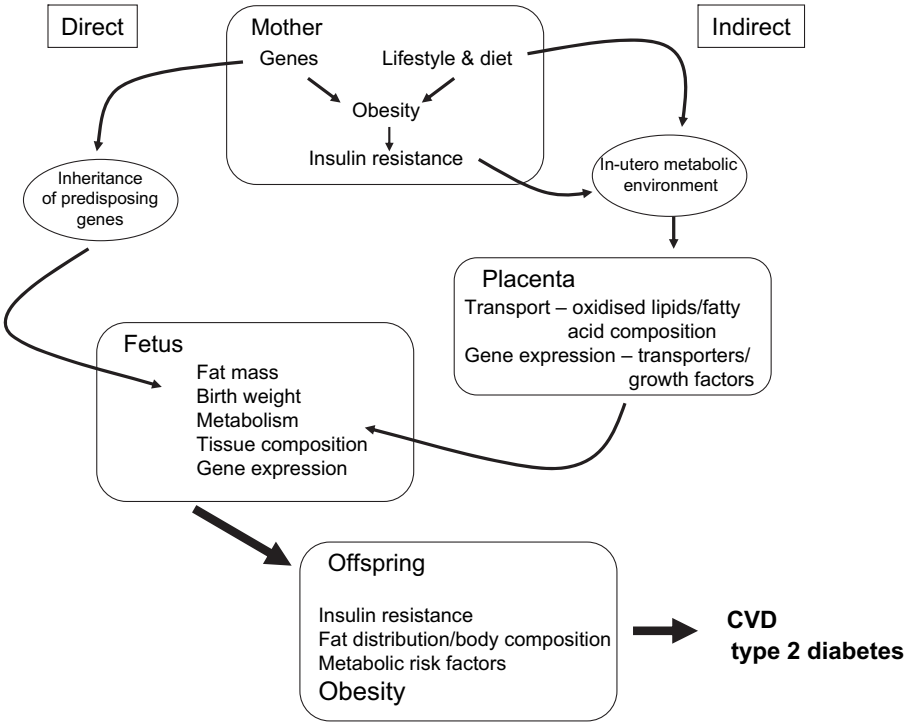

Fig. 1. Programming of offspring obesity. Fetal body composition, metabolism and gene expression may be influenced by the mother to develop patterns resulting in future obesity. Direct influences include inheritance of predisposing genes. Indirect influences include the in-utero metabolic environment, which is determined by maternal lifestyle, diet and metabolism. The placenta may alter transport processes, gene expression and metabolism in response to the in-utero environment, transmitting effects to the fetus. Patterns of gene expression and metabolism may lead to the offspring developing insulin resistance, adverse metabolic risk factor profile and obesity, hence increasing their risk for premature cardiovascular disease and type 2 diabetes. CVD, cardiovascular disease.

mothers.[34] Therefore although pharmacological therapies are undesirable in pregnancy, nutritional therapy or supplementation might be effective.

5. Summary

The concept of the developmental origins of disease has broadened to incorporate elements of all current hypotheses discussed as well as postnatal effects (Fig. 1). In terms of programming of human obesity, it is clear that both maternal genes and environment (particularly lifestyle and diet) have an influence on maternal obesity which is commonly associated with maternal insulin resistance. There may be direct effects on fetal metabolism and tissue development via inheritance of maternal obesity susceptibility genes, but there are also likely to be indirect effects via the supply of nutrients/metabolites to the fetus (both in terms of quantity and quality). These direct and indirect effect may combine to influence neonatal body composition and metabolism such that the impact of environmental stimuli throughout life lead to fat accumulation, expression of cardiovascular risk markers and earlier development of CVD and type 2 diabetes. The aim now is to collect sufficient mechanistic data to attempt to support this hypothesis.

Research directions

- The impact of maternal obesity on maternal metabolites, both in terms of quantity and quality in humans.
- The impact of maternal obesity on offspring metabolism and gene expression.
- The impact of maternal obesity on placental gene expression especially with respect to transporters and nuclear receptors.
- Development of good animal models for obesity programming.

Conflict of interest statement

None declared.

Funding source

Supported by a British Medical Association Obesity Research Award.

References

*1. Barker DJ. The fetal and infant origins of adult disease. *Br Med J* 1990;**301**:1111.
2. Haslam D, Sattar N, Lean M. ABC of obesity. Obesity – time to wake up. *Br Med J* 2006;**333**:640–2.
3. Kanagalingam MG, Forouhi NG, Greer IA, et al. Changes in booking body mass index over a decade: retrospective analysis from a Glasgow Maternity Hospital. *Br J Obstet Gynecol* 2005;**112**:1431–3.
*4. Catalano PM. Obesity and pregnancy – the propagation of a viscous cycle? *J Clin Endocrinol Metab* 2003;**88**:3505–6.
5. Barker DJ. The origins of the developmental origins theory. *J Intern Med* 2007;**261**:412–7.
6. Forsen T, Eriksson JG, Tuomilehto J, et al. Mother's weight in pregnancy and coronary heart disease in a cohort of Finnish men: follow up study. *Br Med J* 1997;**315**:837–40.
7. Leon DA, Lithell HO, Vagero D, et al. Reduced fetal growth rate and increased risk of death from ischaemic heart disease: cohort study of 15 000 Swedish men and women born 1915–29. *Br Med J* 1998;**317**:241–5.
8. Rich-Edwards JW, Stampfer MJ, Manson JE, et al. Birth weight and risk of cardiovascular disease in a cohort of women followed up since 1976. *Br Med J* 1997;**315**:396–400.
9. Stein CE, Fall CH, Kumaran K, et al. Fetal growth and coronary heart disease in south India. *Lancet* 1996;**348**:1269–73.
10. Lawlor DA, Ronalds G, Clark H, et al. Birth weight is inversely associated with incident coronary heart disease and stroke among individuals born in the 1950s: findings from the Aberdeen Children of the 1950s prospective cohort study. *Circulation* 2005;**112**:1414–8.
11. Lithell HO, McKeigue PM, Berglund L, et al. Relation of size at birth to non-insulin dependent diabetes and insulin concentrations in men aged 50–60 years. *Br Med J* 1996;**312**:406–10.
12. Phipps K, Barker DJ, Hales CN, et al. Fetal growth and impaired glucose tolerance in men and women. *Diabetologia* 1993;**36**:225–8.
13. Hales CN, Barker DJ. The thrifty phenotype hypothesis. *Br Med Bull* 2001;**60**:5–20.
14. Huxley R, Neil A, Collins R. Unravelling the fetal origins hypothesis: is there really an inverse association between birthweight and subsequent blood pressure? *Lancet* 2002;**360**:659–65.
15. Langley-Evans AJ, Langley-Evans SC. Relationship between maternal nutrient intakes in early and late pregnancy and infants weight and proportions at birth: prospective cohort study. *J R Soc Promot Health* 2003;**123**:210–6.
*16. Hattersley AT, Tooke JE. The fetal insulin hypothesis: an alternative explanation of the association of low birthweight with diabetes and vascular disease. *Lancet* 1999;**353**:1789–92.
17. Matschinsky FM. Banting Lecture 1995. A lesson in metabolic regulation inspired by the glucokinase glucose sensor paradigm. *Diabetes* 1996;**45**:223–41.
18. Hattersley AT, Beards F, Ballantyne E, et al. Mutations in the glucokinase gene of the fetus result in reduced birth weight. *Nat Genet* 1998;**19**:268–70.
19. Singh R, Pearson ER, Clark PM, et al. The long-term impact on offspring of exposure to hyperglycaemia in utero due to maternal glucokinase gene mutations. *Diabetologia* 2007;**50**:620–4.
20. Pettitt DJ, Aleck KA, Baird HR, et al. Congenital susceptibility to NIDDM. Role of intrauterine environment. *Diabetes* 1988;**37**:622–8.
*21. Dabelea D, Hanson RL, Lindsay RS, et al. Intrauterine exposure to diabetes conveys risks for type 2 diabetes and obesity: a study of discordant sibships. *Diabetes* 2000;**49**:2208–11.
*22. Catalano PM, Presley L, Minium J, et al. Fetuses of obese mothers develop insulin resistance in utero. *Diabetes Care* 2009;**32**:1076–80.
23. Sewell MF, Huston-Presley L, Super DM, et al. Increased neonatal fat mass, not lean body mass, is associated with maternal obesity. *Am J Obstet Gynecol* 2006;**195**:1100–3.
*24. Ramsay JE, Ferrell WR, Crawford L, et al. Maternal obesity is associated with dysregulation of metabolic, vascular, and inflammatory pathways. *J Clin Endocrinol Metab* 2002;**87**:4231–7.
25. Radaelli T, Lepercq J, Varastehpour A, et al. Differential regulation of genes for fetoplacental lipid pathways in pregnancy with gestational and type 1 diabetes mellitus. *Am J Obstet Gynecol*; 2009.
*26. Napoli C, D'Armiento FP, Mancini FP, et al. Fatty streak formation occurs in human fetal aortas and is greatly enhanced by maternal hypercholesterolemia. Intimal accumulation of low density lipoprotein and its oxidation precede monocyte recruitment into early atherosclerotic lesions. *J Clin Invest* 1997;**100**:2680–90.
27. Burke KT, Colvin PL, Myatt L, et al. Transport of maternal cholesterol to the fetus is affected by maternal plasma cholesterol concentrations in the Golden Syrian hamster. *J Lipid Res* 2009;**50**:1146–55.
28. Napoli C, Glass CK, Witztum JL, et al. Influence of maternal hypercholesterolaemia during pregnancy on progression of early atherosclerotic lesions in childhood: Fate of Early Lesions in Children (FELIC) study. *Lancet* 1999;**354**:1234–41.
29. Griffin BA, Freeman DJ, Tait GW, et al. Role of plasma triglyceride in the regulation of plasma low density lipoprotein (LDL) subfractions: relative contribution of small, dense LDL to coronary heart disease risk. *Atherosclerosis* 1994;**106**:241–53.
30. Griffin BA. Lipoprotein atherogenicity: an overview of current mechanisms. *Proc Nutr Soc* 1999;**58**:163–9.
31. Sanchez-Vera I, Bonet B, Viana M, et al. Changes in plasma lipids and increased low-density lipoprotein susceptibility to oxidation in pregnancies complicated by gestational diabetes: consequences of obesity. *Metabolism* 2007;**56**:1527–33.
32. Alsat E, Bouali Y, Goldstein S, et al. Characterization of specific low-density lipoprotein binding sites in human term placental microvillous membranes. *Mol Cell Endocrinol* 1982;**28**:439–53.
33. Ethier-Chiasson M, Forest JC, Giguere Y, et al. Modulation of placental protein expression of OLR1: implication in pregnancy-related disorders or pathologies. *Reproduction* 2008;**136**:491–502.
34. Palinski W, Napoli C. The fetal origins of atherosclerosis: maternal hypercholesterolemia, and cholesterol-lowering or antioxidant treatment during pregnancy influence in utero programming and postnatal susceptibility to atherogenesis. *FASEB J* 2002;**16**:1348–60.
*35. Brunham LR, Kruit JK, Verchere CB, et al. Cholesterol in islet dysfunction and type 2 diabetes. *J Clin Invest* 2008;**118**:403–8.
36. Abderrahmani A, Niederhauser G, Favre D, et al. Human high-density lipoprotein particles prevent activation of the JNK pathway induced by human oxidised low-density lipoprotein particles in pancreatic beta cells. *Diabetologia* 2007;**50**:1304–14.
37. van der Heide LP, Ramakers GM, Smidt MP. Insulin signaling in the central nervous system: learning to survive. *Prog Neurobiol* 2006;**79**:205–21.

*38. Kirk SL, Samuelsson AM, Argenton M, et al. Maternal obesity induced by diet in rats permanently influences central processes regulating food intake in offspring. *PLoS One* 2009;**4**:e5870.
39. Napoli C, de Nigris F, Welch JS, et al. Maternal hypercholesterolemia during pregnancy promotes early atherogenesis in LDL receptor-deficient mice and alters aortic gene expression determined by microarray. *Circulation* 2002;**105**:1360–7.
40. Torrens C, Brawley L, Anthony FW, et al. Folate supplementation during pregnancy improves offspring cardiovascular dysfunction induced by protein restriction. *Hypertension* 2006;**47**:982–7.
41. Waterland RA, Jirtle RL. Transposable elements: targets for early nutritional effects on epigenetic gene regulation. *Mol Cell Biol* 2003;**23**:5293–300.
42. Dolinoy DC, Weidman JR, Waterland RA, et al. Maternal genistein alters coat color and protects Avy mouse offspring from obesity by modifying the fetal epigenome. *Environ Health Perspect* 2006;**114**:567–72.
43. Lillycrop KA, Phillips ES, Torrens C, et al. Feeding pregnant rats a protein-restricted diet persistently alters the methylation of specific cytosines in the hepatic PPAR alpha promoter of the offspring. *Br J Nutr* 2008;**100**:278–82.
44. Laraia BA, Bodnar LM, Siega-Riz AM. Pregravid body mass index is negatively associated with diet quality during pregnancy. *Public Health Nutr* 2007;**10**:920–6.
45. Alkemade FE, Gittenberger-de Groot AC, Schiel AE, et al. Intrauterine exposure to maternal atherosclerotic risk factors increases the susceptibility to atherosclerosis in adult life. *Arterioscler Thromb Vasc Biol* 2007;**27**:2228–35.
46. Robb AO, Mills NL, Newby DE, et al. Endothelial progenitor cells in pregnancy. *Reproduction* 2007;**133**:1–9.
47. Hill JM, Zalos G, Halcox JP, et al. Circulating endothelial progenitor cells, vascular function, and cardiovascular risk. *N Engl J Med* 2003;**348**:593–600.
48. Xia L, Zhou XP, Zhu JH, et al. Decrease and dysfunction of endothelial progenitor cells in umbilical cord blood with maternal pre-eclampsia. *J Obstet Gynaecol Res* 2007;**33**:465–74.
49. Ingram DA, Lien IZ, Mead LE, et al. In vitro hyperglycemia or a diabetic intrauterine environment reduces neonatal endothelial colony-forming cell numbers and function. *Diabetes* 2008;**57**:724–31.
50. Tuomilehto J, Lindstrom J, Eriksson JG, et al. Prevention of type 2 diabetes mellitus by changes in lifestyle among subjects with impaired glucose tolerance. *N Engl J Med* 2001;**344**:1343–50.

*The most important references are indicated with an asterisks.