Role of the placenta in fetal programming: underlying mechanisms and potential interventional approaches

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ABSTRACT
Adverse influences during fetal life alter the structure and function of distinct cells, organ systems or homoeostatic pathways, thereby 'programming' the individual for an increased risk of developing cardiovascular disease and diabetes in adult life. Fetal programming can be caused by a number of different perturbations in the maternal compartment, such as altered maternal nutrition and reduced utero–placental blood flow; however, the underlying mechanisms remain to be fully established. Perturbations in the maternal environment must be transmitted across the placenta in order to affect the fetus. Here, we review recent insights into how the placenta responds to changes in the maternal environment and discuss possible mechanisms by which the placenta mediates fetal programming. In IUGR (intrauterine growth restriction) pregnancies, the increased placental vascular resistance subjects the fetal heart to increased work load, representing a possible direct link between altered placental structure and fetal programming of cardiovascular disease. A decreased activity of placental 11β-HSD-2 (type 2 isoform of 11β-hydroxysteroid dehydrogenase) activity can increase fetal exposure to maternal cortisol, which programmes the fetus for later hypertension and metabolic disease. The placenta appears to function as a nutrient sensor regulating nutrient transport according to the ability of the maternal supply line to deliver nutrients. By directly regulating fetal nutrient supply and fetal growth, the placenta plays a central role in fetal programming. Furthermore, perturbations in the maternal compartment may affect the methylation status of placental genes and increase placental oxidative/nitrative stress, resulting in changes in placental function. Intervention strategies targeting the placenta in order to prevent or alleviate altered fetal growth and/or fetal programming include altering placental growth and nutrient transport by maternally administered IGFs (insulin-like growth factors) and altering maternal levels of methyl donors.

INTRODUCTION
Genes and adult lifestyle factors have traditionally been regarded as the primary determinants for the risk of developing diseases like Type 2 diabetes and cardiovascular disease. More recently, a large body of epidemiological data has accumulated to suggest that adverse influences during early development, in particular...
during fetal life, increase the risk of developing disease in adult life [1–3]. This paradigm, referred to as ‘fetal programming’ or ‘developmental origins of health and disease’, may have a profound impact on public health strategies for the prevention of major illnesses. A link between intrauterine environment and adult disease was pioneered by Barker and co-workers, who reported associations between low birth weight and the risk of developing Type 2 diabetes [4,5] and cardiovascular disease [6,7]. A large number of experimental studies have subsequently provided clear proofs of principle for fetal programming [2,8,9]. The most commonly used approach to perturb intrauterine environment has been to alter maternal nutrition during pregnancy, typically subjecting the pregnant animal to protein malnutrition, which has been shown to result in various degrees of disturbance in glucose metabolism [10] and cardiovascular function [11] in the offspring. In addition to altered maternal nutrition, a number of other perturbations of maternal physiology, such as administration of corticosteroids [12], cytokines [13] or experimental reduction of uterine blood flow [14,15], have been shown to lead to fetal programming of diabetes and/or hypertension.

An adverse intrauterine environment can cause developmental disruption, exemplified by teratogenesis. In contrast, fetal programming involves developmental plasticity, where disturbances during critical periods of fetal development alter the structure and/or function of distinct cells, organ systems or homeostatic pathways [16]. This plasticity allows the conceptus/fetus to respond to environmental influences by following a developmental trajectory that may be associated with an adaptive advantage in utero. Furthermore, it has been suggested that fetal metabolism and growth are adapted to the predicted postnatal metabolic environment, represented by the nutrient supply during fetal life as the primary cue [2]. In the case of intrauterine nutrient restriction, the fetal predictive adaptive response may be inappropriate if postnatal life provides an abundance of nutrients. As a consequence of this mismatch, adult disease will develop [2]. The mechanisms linking an adverse intrauterine environment to adult disease are under intense investigation. The possibility that epigenetic regulation of key metabolic pathways represents a common pathway for fetal programming has attracted a great deal of attention [17,18]. Furthermore, there is strong support in the literature for involvement of alterations in the HPA (hypothalamic–pituitary–adrenal) axis [19] and leptin signalling [20] in the programming of fetal physiology in response to environmental cues.

The placenta constitutes the active interface between the maternal and fetal blood circulations, regulating maternal physiological changes in pregnancy and fetal growth and is thought to play an important role in the development of many pregnancy complications. Considering that perturbations in the maternal compartment, such as altered nutrition, cytokine and cortisol levels and reduced utero–placental blood flow must be transmitted across the placenta in order to affect the fetus, it is surprising that the role of the placenta in fetal programming has attracted only limited attention. However, there is an increasing awareness that the placenta responds to and modulates perturbations in the maternal environment, thereby playing a key role in transmitting the programming stimuli to the fetus [21,22]. This review will focus on recent new insights into the nature of these responses. This information is essential for a better understanding of the mechanisms underlying fetal programming and will be crucial in the efforts to design strategies for intervention.

THE PLACENTA

The placenta maintains fetal homeostasis by performing a wide range of physiological functions, which after birth are carried out by the kidney, gastrointestinal tract, lungs and endocrine glands of the neonate. The primary functions of the placenta are to provide an immunological barrier between fetus and mother, mediate the transfer of respiratory gases, water, ions and nutrients, and produce and secrete a vast array of hormones, cytokines and signalling molecules. Maternal blood supply is established at the end of the first trimester and maternal blood enters the placenta via the spiral arteries, which delivers blood directly to the intervillus space (Figure 1). In order for maternal placental blood flow to increase normally, it is critical that invasive trophoblast cells migrate into the spiral arteries during the first half of pregnancy, resulting in the degeneration of elastic and smooth muscle tissue in spiral artery walls and the replacement of endothelial cells by trophoblasts. This process is normally completed by gestational weeks 16–18 and transforms the spiral arteries into dilated vessels, which are unresponsive to vasoconstrictors. On the fetal side, blood with low oxygen saturation and low nutrient concentrations enters the placenta through two umbilical arteries, which branch and ultimately form a capillary network in the terminal villi of the villous tree that is freely floating in the maternal blood of the intervillus space (Figure 1).

Perturbations in the maternal compartment that have been associated with fetal programming could affect one or several aspects of placental structure and function, which may be the direct link to an altered intrauterine environment triggering fetal adaptive responses. In this review, we will discuss changes in placental structure, 11β-HSD-2 [type 2 isoform of 11-β HSD (hydroxysteroid dehydrogenase)] activity, nitrification of placental proteins, gene methylation and the expression and activity of key placental nutrient transporters in response to an altered maternal physiology, and address the effects these alterations may have on the intrauterine environment.
PLACENTAL STRUCTURE

Placental size

Epidemiological data indicate that placental weight, albeit a crude proxy for placental structure, appears to provide information on the long-term outcome for the baby (reviewed in [21]). Indeed, it is likely that changes in placental growth represent an important link between perturbations in the maternal compartment (such as reduced placental blood flow, altered maternal nutrition and diabetes) and alterations in fetal growth. A lack of a normal increase in maternal placental blood flow, leading to ’placental insufficiency’, probably represents the most common underlying cause of IUGR (intrauterine growth restriction) in Western societies. Interestingly, both in animal studies in which uterine blood flow was reduced experimentally [23] and in clinical studies [24], a decreased placental weight/volume was evident before any decline in fetal growth rate was observed. At the other end of the growth spectrum, both Type 1 diabetes and GDM (gestational diabetes) represent pregnancy complications associated with increased placental and fetal weight, likewise implicating placental size in determining fetal growth. The relationship between maternal nutrition and placental size in humans is complicated. For example, low dietary intake of carbohydrates in early pregnancy has been reported to increase placental weight, in particular if combined with high protein intake in late pregnancy [25]. This observation is reminiscent of reports from the Dutch famine winter during World War II, showing that exposure to severe calorie restriction early in pregnancy and a high calorie intake in the second half of pregnancy results in increased placental growth [26]. In experimental models of severe maternal undernutrition, both in protein malnutrition and restriction of total calorie intake, placental and fetal growth is impaired [27–29]. The interdependence between placental and fetal growth is illustrated by the well-known positive correlation between placental and fetal weight [30]. Since fetal growth is related to adult outcomes [1], placental weight is likewise predictive of adult diseases, such as Type 2 diabetes and hypertension [21].

Placental vasculature and exchange barrier

There are few studies reporting effects of specific maternal perturbations in human pregnancy on placental morphometry. However, anaemia increases fetal angiogenesis in the first trimester [31] and vascular adaptations at high altitude include dilatation of capillary sinusoids and thinning of the exchange barrier [32,33], findings that are in line with observations in animal models in which the dam is exposed to hypoxia [34,35] or anaemia [36]. In the IUGR placenta, the emerging picture is that, in cases of absent or reversed end-diastolic flow in the umbilical artery, as assessed by Doppler velocity waveform analysis, villi are poorly branched and capillarized, and the exchange barrier is thickened [37–39]. In contrast, less severe IUGR with positive end-diastolic umbilical artery flow appears to be associated with a normal pattern of stem artery development, increased capillary angiogenesis and terminal villous development [37].
The impact on specific maternal nutritional manipulations on the development of the placental vasculature and exchange barrier has been studied in some detail in animal experiments. For example, in the rat, maternal protein restriction (8% dietary protein compared with the normal 20%) increased the villous surface area, whereas the underlying vascular volume did not increase [40]. In contrast, maternal calorie restriction (dams fed 50–70% of normal) in the guinea pig decreased the villous surface area [28]. Whether the discrepancies between these studies are due to species differences or related to the differences in the dietary manipulations is not clear. Recently, Rutland et al. [41] reported a reduction in the length of the labyrinthine vessels and decreased expression of vascular endothelial adhesion molecules in the murine placenta in response to maternal protein malnutrition, compatible with the possibility that alterations in maternal nutrition changes placental vascular function.

The possible link between changes in placental structure and the risk of adult disease is particularly evident for the fetal programming of cardiovascular disease. It is well established that IUGR is characterized by an increased resistance to blood flow in the umbilical artery [42]. As proposed by Thornburg and Louey [43], this subjects the IUGR fetal heart to an increased pressure work load, which is suggested to result in an adaptation that may be advantageous in the short-term perspective, but could contribute to cardiovascular disease postnatally. Furthermore, a subgroup of IUGR fetuses are hypoxic in utero [44], which may decrease the number of cardiomyocytes and make the heart more sensitive to hypoxic insults later in life [45]. Thus the thicker placental exchange barrier (a primary cause of fetal hypoxia in IUGR) and the increased placental vascular resistance in IUGR may represent alterations in placental structure that are directly involved in the fetal programming of cardiovascular disease [43].

**PLACENTAL 11β-HSD-2 AND PROGRAMMING**

It is well-established that maternal administration of glucocorticoids to pregnant animals and humans causes IUGR [46]. Furthermore, offspring of glucocorticoid-exposed dams have altered activity in the HPA axis and are susceptible to developing hypertension and hyperglycaemia in adult life [12,47,48]. This may not be a direct effect on the fetus, since studies in the sheep have shown that giving corticosteroids directly to the fetus does not result in IUGR [49–51], compatible with the possibility that the placenta mediates the effects of corticosteroids on fetal growth.

Despite the lipophilic nature of glucocorticoids and, therefore, having the ability to cross the placental barrier quite readily, circulating levels of cortisol are markedly higher in the mother than in the fetus [52]. Indeed, placental 11β-HSD-2 forms a functional barrier restricting the free transfer of cortisol between the maternal and fetal compartments by converting cortisol into its much less active 11-keto form, cortisone [53–55]. In contrast, synthetic corticosteroids, such as dexamethasone, are poor substrates for 11β-HSD-2 [56] and, therefore, readily cross the placenta. It has been proposed that attenuation of placental 11β-HSD-2 activity may expose the placenta and fetus to inappropriately high levels of corticosteroids and result in IUGR and fetal programming of adult disease [57]. This hypothesis is supported by associations observed between human placental 11β-HSD-2 expression and/or activity and birth weight in several studies [58,59], albeit not in all [60]. Inhibition of 11β-HSD activity using carbenoxolone in pregnant rats induces IUGR and programmes hypertension in the offspring [47,61]; however, placental-specific inhibition of 11β-HSD is needed in order to unequivocally determine a cause and effect relationship between low placental 11β-HSD activity and reduced fetal growth. Nevertheless, decreased 11β-HSD activity, resulting in dysfunction in the placental glucocorticoid barrier and exposure of the placenta and fetus to excess corticosteroids, constitutes a direct link between altered placental function and fetal programming.

**PLACENTAL OXIDATIVE AND NITRATIVE STRESS**

Oxidative stress, as defined as an imbalance between the production of ROS (reactive oxygen species), such as superoxide, and the ability to rapidly scavenge them, can be detected in the placenta in normal pregnancy. When NO (nitric oxide) and superoxide radicals are produced simultaneously in the same subcellular compartment, NO can react with superoxide resulting in the formation of peroxynitrate, which is a powerful oxidant with a multitude of cellular effects. In particular, peroxynitrate nitrates aromatic amino acids, such as tyrosine, thereby producing nitrative stress and potentially resulting in altered activity in signal transduction pathways [62]. The functional consequence of protein nitration is often inhibition, although occasionally activation is observed [62]. It has been reported recently that pre-eclampsia is associated with an increased nitrination of placental phosphorylated p38 MAPK (mitogen-activated protein kinase), resulting in reduced catalytic activity [63]. The p38 MAPK signalling pathway is activated in cellular responses to environmental stressors and cytokines, and modifications in p38 MAPK activity may therefore have marked effects on cellular function. In IUGR, diabetes and pre-eclampsia, which are pregnancy complications associated with fetal programming, placental oxidative stress is markedly increased [64–66] and eNOS
(endothelial NO synthase) is up-regulated in placental villous tissue [67]. It has been proposed that hypoxia and oxidative and nitrite stress alter the function of placental proteins in key signalling and metabolic pathways and may be a general mechanism underlying fetal programming [22]. However, the link between, for example, nitration of specific placental proteins and a permanent alteration of fetal physiological regulatory circuits remains to be established.

**EPIGENETIC REGULATION OF KEY METABOLIC PATHWAYS IN THE PLACENTA**

Epigenetic modification refers to heritable changes in gene expression that are not mediated by alterations in DNA sequence [68]. Epigenetic mechanisms include modifications at the N-terminal region, such as methylation, acetylation and phosphorylation of histones. A particularly important mechanism for epigenetic regulation of genes is methylation of cytosine residues in specific regions of DNA called CpG islands, present in low abundance throughout the genome and particularly associated with promoter regions of genes. DNA methylation regulates gene expression in that hypermethylation of promoter regions is commonly associated with transcriptional repression, whereas hypomethylation often increases transcription [69]. Maternal nutritional status, possibly by altering the availability of methyl donors, such as folate, has been shown to influence the methylation status of the fetal genome [70]. Imprinted genes are genes whose expression depends on their parental origin. In a paternally imprinted gene, for example, the maternal allele is silenced by DNA methylation [71]. Approx. 70 imprinted genes have been discovered so far in mice, and for most of these genes the imprinting status is conserved in humans [72]. A substantial proportion of imprinted genes are involved in the control of fetal growth and, in general, paternally expressed imprinted genes (e.g. *IGF-2*, a gene encoding IGF (insulin-like growth factor) 2, and *Slc38a4*, a gene encoding SNAT4 (sodium-coupled neutral amino acid transporter 4)) enhance fetal growth, whereas maternally expressed ones suppress it (e.g. *IGF-2r*, a gene encoding IGF-2 receptor). Most of the imprinted genes are expressed in the placenta, suggesting that imprinted genes have central roles in controlling both the fetal demand for, and the placental supply of, maternal nutrients [73,74].

Recently, it was reported that maternal dietary protein restriction in the rat induces alterations in the methylation of specific genes, such as the glucocorticoid receptor, in the liver of the offspring and that these changes could be prevented by folate supplementation [18]. In a similar experimental paradigm, it was shown that folate supplementation during pregnancy prevented much of the adverse effect of maternal protein restriction on cardiovascular function in the offspring [75]. IUGR due to placental insufficiency in the rat is associated with epigenetic modifications of hepatic DUSP-5 (dual specificity phosphatase-5) [76] and renal p53 in the offspring [17]. These studies indicate that epigenetic regulation of fetal genes represents an important mechanism mediating fetal programming and raises the possibility that genes expressed in the placenta, an organ directly exposed to maternal blood, are modified epigenetically by perturbations in the maternal compartment. With the exception of a recent report that the methylation pattern of placental *H19* and *IGF-2*, two imprinted genes, was unaltered in response to maternal protein restriction in the rat [77], no data are currently available. Also, maternal environmental influences before or at the time of conception may alter the methylation status of trophoblast genes, which could result in a permanent change in placental function and structure [78]. Furthermore, it is conceivable that changes in placental transport of folate, vitamin B12 and choline could alter the availability of these methyl-donors in the fetus, providing a direct link between placental function, gene methylation and fetal programming. It is anticipated that these areas of research will attract a great deal of interest in the near future.

**PLACENTAL NUTRIENT TRANSPORT AND FETAL PROGRAMMING**

Notwithstanding that the associations between fetal growth and adult disease often extend across the normal birth weight range, it is IUGR and LGA (large-for-gestational age) babies that are at particular risk of developing diabetes and cardiovascular disease later in life. The primary determinant of fetal growth is nutrient supply, which is directly dependent on placental transport functions. Thus elucidating the factors regulating placental nutrient transport will provide critical information on mechanisms underlying altered fetal growth and fetal programming.

**Placental nutrient transport in altered fetal growth**

The growth-restricted human fetus has reduced plasma concentrations of certain key amino acids [79] and some are hypoglycaemic and hypoxic *in utero* [44]. Although generally accepted that IUGR is associated with limitations in nutrient and oxygen supply, the mechanisms underlying IUGR are largely unknown. Similarly, the accelerated fetal growth often observed in pregnancies complicated by diabetes, resulting in the delivery of a LGA baby, has been attributed to an excess glucose delivery to the fetus due to maternal hyperglycaemia. However, in modern clinical management of the pregnant woman with diabetes, maternal glucose levels are rigorously controlled throughout the second and third trimester making it...
Changes in the activity of placental transporters in IUGR

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Table 2 Changes in the activity of placental transporters in fetal overgrowth in pregnancies complicated by diabetes

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* Only GDM.
† Only Type 1 diabetes.
‡ Different results have been reported in a previous study [135].

difficult to explain the high incidence of LGA infants in pregnancies complicated by diabetes. Recent advances in the study of placental transport functions have provided important information suggesting that alterations in the expression and activity of placental nutrient and ion transporters may play a key role in regulating fetal growth in normal and complicated pregnancies.

Net transport of nutrients across the placenta is dependent on several factors, including the concentration gradient between maternal and fetal blood, placental blood flow and metabolism, and the expression and activity of specific membrane-bound transporters. Measurements of maternal placental blood flow and volume blood flow in the umbilical circulation clearly suggest that blood flows are reduced on both sides of the placental exchange barrier in association with IUGR [80,81]. However, it is unlikely that the blood flow reduction is a sufficient explanation for the decreased levels of various nutrients in the fetal circulation, as the transplacental transport of nutrients, such as glucose and amino acids, is limited primarily by transfer across the placental barrier rather than blood flow [82]. There are only two cell layers constituting the barrier between maternal and fetal blood in the human placenta: the syncytiotrophoblast, the transporting epithelium, and the fetal capillary endothelium (Figure 1). Of these two cell layers it is the syncytiotrophoblast and, in particular, its two polarized plasma membranes that constitute the primary barrier to the transport of molecules such as glucose, ions and amino acids. This is the rationale for studying the transporter activity and expression in MVM and BM (microvillus and basal plasma membranes respectively) isolated from pregnancies with altered fetal growth in order to model the materno–fetal transport of amino acids, glucose and ions in IUGR (Table 1) and LGA pregnancies (Table 2). IUGR is characterized by a reduced activity of certain placental amino acid transporters (transporters for taurine, leucine and cationic amino acids as well as System A) [83–88], whereas the activity and expression of glucose transporters in the placental barrier remain unaltered in IUGR [84,89]. With regard to essential amino acids, these in vitro findings are compatible with a study in pregnant women in which Paolini et al. [90] demonstrated, using stable isotope techniques, that placental transfer of the essential amino acids leucine and phenylalanine is reduced in IUGR. Accelerated fetal growth in pregnancies complicated by Type 1 diabetes [91], but not GDM [92], is associated with increased glucose transporter activity and protein expression in BMs. These alterations might explain the occurrence of large babies in pregnancies complicated by Type 1 diabetes despite ‘normal’ maternal blood glucose levels [93]. Moreover, the activity of the transporter for the essential amino acid leucine is increased in GDM with accelerated fetal growth, whereas we found an increased activity of placental system A in both Type 1 diabetes and GDM [94].

Regulation of placental nutrient transporters

The finding of altered activity and expression of placental nutrient transporters in pregnancy complications has stimulated interest in regulation studies. Glucocorticoids have been shown to decrease the expression of placental glucose transporters [95]. Most previous [96], but not all [97], studies indicate that insulin does not affect placental glucose transporters at term. In contrast, glucose transport activity was increased after treatment of a first trimester trophoblast cell line with insulin, IGF-1 or IGF-2. © 2007 The Biochemical Society
Figure 2  Placenta as a nutrient sensor: a hypothesis

It is suggested that placental nutrient and ion transporters are regulated in response to a primary event, such as a lack of increase in placental blood flow, maternal malnutrition or hyperglycaemia. Alterations in placental transport activity result in changes in nutrient delivery to the fetus which, in turn, affects fetal growth. Hormones produced by the placenta or the mother, hypoxia and nutrient-sensing mechanisms 'intrinsic' to the placenta (such as mTOR) may be involved.

[98,99]. Recent studies in primary villous fragments support the conclusion that placental glucose transporters are regulated early in pregnancy, but not at term. Glucose transporter activity was not affected by hormones, such as leptin, GH (growth hormone), IGF-1, insulin and cortisol, at term [100]. In contrast, insulin stimulated glucose uptake in primary villous fragments obtained at 6–8 weeks of gestation [101], which may be related to the presence of the insulin-sensitive glucose transporter GLUT4 in the cytosol and MVMs of the syncytiotrophoblast in the first trimester [101].

With regard to the regulation of placental amino acid transporters, IGF-1 stimulates System A activity in cultured trophoblast cells [99,102,103], and insulin increases transport of neutral amino acids in the perfused human lobule [104] and in cultured trophoblast cells [99,105]. In addition, System A transporter activity and expression are decreased by hypoxia [106]. Leptin and insulin stimulated System A activity uptake by 50–60% in primary villous fragments at term [107]. Furthermore, NO and oxygen radicals have been shown to reduce the activity of several placental amino acid transporters [108,109].

Does the placenta function as a 'nutrient sensor'?

In a situation such as IUGR, where fetal plasma concentrations of amino acids are decreased [79,110,111], it might be expected that placental transporters would be up-regulated in an attempt to increase transport. Similarly, in situations with maternal (and fetal) hyperglycaemia (diabetes), a down-regulation of placental glucose transporters may seem as an appropriate biological response. However, available results (summarized above and in Tables 1 and 2) indicates the opposite. We have developed a working hypothesis, 'the placenta as a nutrient sensor', that we believe takes into account the results that we, and others, have obtained and provides a testable model for further study. According to this hypothesis, the placenta acts as a nutrient sensor, coordinating nutrient transport functions with maternal nutrient availability [93]. Thus the ability of the maternal supply line to deliver nutrients (i.e. placental blood flow, maternal nutrition, substrate and oxygen levels in maternal blood etc.) regulates key placental nutrient transporters (Figure 2). With this perspective, placental transport alterations represent a mechanism to match...
A simplified schematic representation of the mTOR signalling pathway. mTOR is a protein kinase shown to be regulated by growth factors and nutrients. mTOR exists in the cell in two complexes: mTORC1 (mTOR complex 1) and mTORC2 (mTOR complex 2). When activated, mTORC1 signalling promotes translation, mediated via phosphorylation of 4E-BP1 and S6K1, and transcription (via unknown effectors) and as a result a number of proteins associated with cell growth are produced. In contrast, mTORC2 signalling affects actin organization. The function of the mTOR signalling pathway in the placenta is currently unknown. We propose that mTOR represents a molecular mechanism for placental nutrient sensing, integrating information on nutrient availability and growth factor signalling, and regulating placental nutrient transporters and, as a consequence, fetal growth. Rapamycin inhibits mTORC1. eIF-4E, eukaryotic initiation factor.

Fetal growth rate to a level which is compatible with the amount of nutrients that can be provided by the maternal supply line, making the placenta a key player in the regulation of fetal growth and, as a consequence, fetal programming. In the case of IUGR, a lack of normal increase in placental blood flow or maternal malnutrition may be sensed by the placenta and, as a consequence, some key placental transporters are down-regulated in order to decrease fetal growth. Similarly, hyperglycaemia early in pregnancy (which is common even in the well-regulated patient with Type 1 diabetes) may convey a ‘good nutrition’ signal to the placenta, resulting in up-regulation of glucose and amino acid transporters (Figure 2).

The mechanisms conveying information about the ability of the maternal supply line to deliver nutrients and regulating placental nutrient transporters remain speculative. However, it is likely that the activity of key placental nutrient transporters in a particular situation represents an integrated response dependent on information from a number of signalling pathways. For example, we have provided evidence that maternal nutrition influences placental transporters and fetal growth by altering the levels of metabolic hormones, such as insulin, IGF-1 and leptin [29], which have all been shown to regulate placental nutrient transporters [99,101–103,105,107]. In IUGR, a pregnancy complication associated with reduced placental blood flow, hypoxia may also down-regulate placental amino acid transporters [106]. In addition, we have recently pursued the possibility that the mTOR (mammalian target of rapamycin) signalling system represents an ‘intrinsic’ placental nutrient-sensing mechanism (Figure 3). mTOR is a serine/threonine kinase and represents an important nutrient-sensing pathway in mammalian cells by controlling cell growth through regulation of translation and transcription in response to nutrient availability, in particular branched chain amino acids [112], hypoxia [113] and cellular energy status [114]. The downstream effects of mTOR are mediated by phosphorylation of 4E-BP1 (eukaryotic initiation factor 4E binding protein 1) and S6K1 (p70 ribosomal S6 kinase 1) [115]. Our results indicate that mTOR protein is highly expressed in the cytosol of...
the syncytiotrophoblast and that the activity of the placental mTOR signalling pathway, as measured by the expression of S6K1 phosphorylated at Thr^{389}, is markedly down-regulated in IUGR [116]. When primary villous fragments from human placenta were incubated for 4 h in rapamycin, a specific inhibitor of mTOR, leucine uptake mediated by the L amino acid transporter was almost completely inhibited, suggesting that mTOR signalling regulates placental amino acid transport [116]. Thus these initial observations are compatible with the hypothesis that placental mTOR is involved in nutrient sensing, regulating placental transport according to resources available in the maternal supply line, thereby regulating fetal growth (Figure 3).

**PLACENTAL PHENOTYPES OF INTRAUTERINE GROWTH**

It has been argued that birth weight, albeit convenient, is a poor indicator of an adverse intrauterine environment since not all fetuses subjected to an abnormal intrauterine environment have altered growth and altered fetal growth is not always a result of a response to an environmental perturbation [2,21,117]. We have proposed that ‘placental phenotype’ is a better representation of the intrauterine environment than birth weight [118]. In particular, specific changes in placental nutrient transporter activity/expression characteristic of an intrauterine environment with decreased or increased delivery of nutrients [82], together with results on placental morphology [119] and blood flows [42], constitute the ‘placental phenotype’. For example, an altered placental phenotype will provide a marker of compromised intrauterine environment even when the birth weight lies within the range considered normal. It has been proposed that placental phenotyping will provide much better information concerning the risk of developing diseases later in life than the crude proxies of intrauterine exposure that are currently used [118].

**STRATEGIES FOR INTERVENTION**

Efforts promoting the health of the pregnant woman and her fetus are likely to not only decrease perinatal morbidity and mortality, but also have a significant impact on the incidence of major diseases, such as diabetes and cardiovascular disease, in future generations. Therefore the development of intervention strategies in order to prevent or alleviate the adverse consequences of fetal programming are of critical importance for public health. However, since fetal programming involves plasticity, allowing the fetus to respond to environmental influences by following a developmental trajectory that may be associated with an adaptive advantage, any intervention aimed at minimizing the long-term adverse effects of a suboptimal environment may jeopardize any short-term advantages of fetal intrauterine adaptations.

The lack of detailed information on the mechanisms underlying fetal programming precludes firm statements on treatment and intervention options. This is particularly true with regard to interventions specifically targeting placental structure and function. Restricted placental transport of nutrients results in IUGR, which is associated with fetal programming. Therefore alleviating IUGR by increasing placental nutrient transport could represent an effective way of preventing fetal programming of adult disease. Maternal IGF-1 administration stimulates placental and fetal growth and has been forwarded as a possible therapy in cases of IUGR [120,121]. The mechanisms involved have been proposed to be effects on placental function and possibly the transfer of nutrients across the placenta [120,121], compatible with in vitro studies demonstrating that IGF-1 stimulates trophoblast glucose and amino acid uptake [99,103]. Recently, it was reported that maternal IGF-1 administration in the pregnant guinea pig increased fetal growth [122], possibly by increasing placental glucose transport capacity [123]. Thus maternal IGF-1 administration is an example of an intervention targeting placental function that may be of value in order to prevent fetal programming associated with restricted fetal growth. Similarly, maternal administration of IGF-2 appears to stimulate fetal growth by affecting placental structure [122,123]. However, it should be emphasized that there are no studies available investigating the effect of maternal administration of IGFs on placental function in cases of placental insufficiency. Of particular significance is recently published compelling results suggesting a down-regulation of placental IGF-1 receptors and some of the components of the signal transduction pathway of the IGF-1 receptor in IUGR [124], alterations that may make maternal IGF administration ineffective in these cases. Another, more speculative, alternative to manipulate placental nutrient transporters is targeting the placental mTOR signalling system, which has recently been shown to regulate the placental system L amino acid transporter [116], a key transporter across the placental barrier for a number of essential amino acids. Placental mTOR inhibition may be particularly relevant in situations of fetal overnutrition and overgrowth, such as maternal diabetes, which is associated with up-regulation of placental nutrient transporters [91,94].

In our model of the placenta as a nutrient sensor (Figure 2), we are suggesting that in IUGR down-regulation of placental nutrient transporters is a response to a primary perturbation, typically a lack of normal increase in placental blood flow. Thus interventions that increase maternal placental blood flow could potentially alleviate IUGR and fetal programming. However, the uteroplacental circulation is less sensitive than the systemic circulation to most established vasodilators, resulting in a
decreased placental blood flow due to the lowered mean arterial blood pressure. In contrast, low doses of ANP (atrial natriuretic peptide) appear to selectively dilate the placental circulation in experimental animals [125] and pregnant women [126]. More speculative treatment options include altering the methylation status of placental genes by, for example, maternal folate supplementation, up-regulating the activity of placental 11β-HSD-2 and decreasing placental oxidative/nitrative stress. However, in order to design more specific interventions with these placental targets, more research is needed.

CONCLUSIONS

Perturbations in the maternal compartment, such as altered nutrition, cytokine and cortisol levels, and reduced utero-placental blood flow, programme the fetus to develop disease in adult life. The programming effects on the fetus may be mediated by placental responses to these perturbations, which includes altered thickness of the placental barrier, abnormal villous branching and capillarization, changes in 11β-HSD-2 activity, altered expression and activity of key placental nutrient transporters, epigenetic modification of placental genes and increased oxidative/nitrative stress. Potential intervention strategies targeting the placenta in order to prevent or alleviate altered fetal growth and/or fetal programming include influencing placental growth and nutrient transport by maternally administered IGFs and changing maternal levels of methyl donors. Further elucidation of the role of the placenta in fetal programming will not only increase our understanding of the origins of adult disease, but may also provide novel approaches to intervention.

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