Timing of nutrient restriction and programming of fetal adipose tissue development

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It is apparent from epidemiological studies that the timing of maternal nutrient restriction has a major influence on outcome in terms of predisposing the resulting offspring to adult obesity. The present review will consider the extent to which maternal age, parity and nutritional restriction at defined stages of gestation can have important effects on fat deposition and endocrine sensitivity of adipose tissue in the offspring. For example, in 1-year-old sheep the offspring of juvenile mothers have substantially reduced fat deposition compared with those born to adult mothers. Offspring of primiparous adult mothers, however, show increased adiposity compared with those born to multiparous mothers. These offspring of multiparous ewes show retained abundance of the brown adipose tissue-specific uncoupling protein 1 at 1 month of age. A stimulated rate of metabolism in brown fat of these offspring may act to reduce adipose tissue deposition in later life. In terms of defined windows of development that can programme adipose tissue growth, maternal nutrient restriction targeted over the period of maximal placental growth results in increased adiposity at term in conjunction with enhanced abundance of mRNA for the insulin-like growth factor-I and -II receptors. In contrast, nutrient restriction in late gestation, coincident with the period of maximal fetal growth, has no major effect on adiposity but results in greater abundance of specific mitochondrial proteins, i.e. voltage-dependent anion channel and/or uncoupling protein 2. These adaptations may increase the predisposability of these offspring to adult obesity. Increasing maternal nutrition in late gestation, however, can result in proportionately less fetal adipose tissue deposition in conjunction with enhanced abundance of uncoupling protein 1.

Maternal nutrient restriction: Fetal adipose tissue development: Endocrine sensitivity: Adult obesity

Adipose tissue is laid down early in fetal life, and during normal development there appears to be distinct lineages of white and brown adipocytes (Moulin et al. 2001). Fetal fat exhibits brown and white characteristics, demonstrating an ontogenic increase in the brown adipose tissue-specific uncoupling protein (UCP) 1 (Clarke et al. 1997a) together with a more modest rise in leptin synthesis (Yuen et al. 1999), a product primarily of white adipocytes. Fetal adipose tissue is made up of a combination of multilocular and unilocular cells (Yuen et al. 2003), of which the latter possess few, if any, mitochondria. However, their proportional abundance increases after birth as white adipose tissue dominates (Gemmel et al. 1972). Following birth, which results in substantial endocrine stimulation of UCP1 synthesis (Symonds et al. 2003), there is normally a gradual loss of UCP1 to undetectable levels at 1 month of age in sheep (Mostyn et al. 2003). The decline in UCP1 is paralleled by a gradual rise in plasma leptin and mRNA for leptin up to 1 week after birth (Bispham et al. 2002). The increase in adipose tissue deposition after 1 week occurs independently of any change in leptin. These adaptations represent a major transition that has pronounced implications for future adipose tissue development. The magnitude of these and subsequent changes within fat are determined by the maternal, and therefore fetal, nutritional environment. As a consequence modest perturbations in fetal adipose tissue growth and endocrine sensitivity can have very-long-term outcomes. In the present review the extent to which maternal age, parity and nutritional restriction at defined stages of gestation can have important effects on fat deposition and endocrine sensitivity of adipose tissue in the resulting offspring will be considered.

Abbreviations: IGF, insulin-like growth factors; UCP, uncoupling protein.
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Ontogeny of fetal and postnatal adipose tissue development

From very early in fetal development adipose tissue is present, and in the sheep the most abundant site is the perirenal–abdominal region, which comprises approximately 80% of the total fat stores (Clarke et al. 1997a). At 90 d gestation (term 147 d), when only 1–2 g adipose tissue are laid down, the first expression of mRNA for both UCP1 and leptin occurs (Yuen et al. 1999; Budge et al. 2004). There is then a gradual increase in the mRNA and/or protein for UCP1 and leptin, plus endocrine growth factors, up to term. This process is accompanied by increased plasma concentrations of a range of endocrine stimulatory factors, including triiodothyronine, noradrenaline, cortisol, prolactin and insulin-like growth factors (IGF), in parallel with an up-regulation of respective receptor populations (Symonds et al. 2003). These factors are all important in promoting adipose tissue deposition and ensuring UCP1 abundance is maximal around the time of birth. The abundance of some of these receptors continues to rise during postnatal life (e.g. IGF) and may be important in ensuring sufficient fat depots are laid down after birth. For example, a large increase in adipose tissue growth occurs over the first week of life (Bispham et al. 2002). The nutritional signal for this adaptation is likely to be abundant milk supply from the mother, which has a high fat content (Clarke et al. 1997b). Between 2 and 4 d after birth an approximately tenfold increase in mRNA for both IGF-I and -II receptors (Bispham et al. 2003a) is coincident with a doubling of adipose tissue mass, indicating the potential importance that IGF may play in promoting fat growth (Fig. 1).

The timing of maternal nutrient restriction and subsequent effects on fetal adiposity

Nutrient restriction between early and mid gestation

One of the most striking epidemiological findings relating to the timing of maternal nutrient restriction and subsequent predisposals to adult obesity comes from the Dutch famine of 1944–5. During the 5-month period of the famine mean energy intake was 3-2 MJ/d compared with 6.3 MJ/d immediately afterwards (Roseboom, 2000). Dietary restriction during early gestation was shown to have the greatest effect on placental size:birth weight and to result in a much greater risk of adult CHD and obesity (Roseboom et al. 2000a,b). It is interesting to note that when using sheep models of undernutrition that broadly adopt the same magnitude of nutrient restriction as that imposed under the Dutch famine there is seldom any effect on birth weight but a pronounced effect on placental mass (Dandrea et al. 2001). The consistent finding that a 50% variation in maternal food intake can determine placental mass may have further relevance to contemporary human populations, for which a similar range between the upper and lower quartiles in energy intake is found in both early and late gestation (Godfrey et al. 1996).

It has now been established that maternal, and therefore fetal, nutrition are primary regulators of fetal adipose tissue deposition and hormone receptor mRNA abundance, particularly when nutrient restriction is targeted over the period of maximal placental growth (Bispham et al. 2003b). In terms of the impact of maternal nutritional manipulation on the fetus, the consequences for adipose tissue deposition are strongly dependent on the time of the nutritional intervention. For sheep, nutrient restriction up to 110 d gestation promotes adipose tissue deposition (Gopakumar et al. 2001) whereas nutrient restriction after this time decreases it (Fig. 2). Then, following the restoration of the maternal diet to the same level as controls, after at least 80 d gestation, a further stimulus for fat deposition occurs (Bispham et al. 2003b). This adaptation is accompanied by an increased abundance of mRNA for IGF-I and -II receptors, which is likely to increase adipose tissue sensitivity to the potential anabolic effects of IGF (Teruel et al. 1996). A combination of enhanced responsiveness to IGF in conjunction with increased abundance of GLUT1 in fetuses that have been previously nutrient restricted (Dandrea et al. 2001) may act to promote the anabolic effect of glucose on fetal fat growth (Stevens et al. 1990).
In summary, prolonged (i.e. >30 d) maternal nutrient restriction up to 80–110 d gestation results in enhanced fetal fat deposition. This increase occurs in combination with enhanced IGF receptor abundance and glucose supply, which may exacerbate the deposition of fat following the restoration of the maternal diet (Bispham et al. 2003b).

Maternal nutrient restriction, insulin-like growth factors, glucocorticoids and adiposity

The enhanced adipose tissue deposition following reduced maternal nutrition between early and mid gestation provides further evidence that fetal adiposity can be programmed in utero. The findings suggest that the increased incidence of obesity in adults born to mothers exposed to the Dutch famine during early pregnancy (Roseboom et al. 1998) and all animals were fed to 100% of requirements for the remainder of gestation. Values are means with their standard errors represented by vertical bars. Mean values were significantly different from control values: *P<0.05. (Adapted from Gopalakrishnan et al. 2001 and Symonds et al. 1998.)

In both sheep and man the majority of fetal adipose tissue mass in the ovine fetus. The magnitude of maternal nutrient restriction was 50% of total metabolisable energy requirements for that stage of gestation (see Symonds et al. 1998) and all animals were fed to 100% of requirements for the remainder of gestation. Values are means with their standard errors represented by vertical bars. Mean values were significantly different from control values: *P<0.05. (Adapted from Gopalakrishnan et al. 2001 and Symonds et al. 1998.)

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The enhanced adipose tissue deposition following reduced maternal nutrition between early and mid gestation provides further evidence that fetal adiposity can be programmed in utero. The findings suggest that the increased incidence of obesity in adults born to mothers exposed to the Dutch famine during early pregnancy (Roseboom et al. 2000a) may be a direct consequence of adaptations in the endocrine sensitivity of fetal adipose tissue.

In both sheep and man the majority of fetal adipose tissue is deposited during the final third of gestation (Symonds & Lomax, 1992). Over this period there is an increased abundance within the fetal circulation of hormones that are important in regulating fetal adipose tissue growth (Symonds & Stephenson, 1999), including IGF-I and leptin (Lorenzo et al. 1993; Yuen et al. 1999). The extent to which mRNA abundance within adipose tissue is increased or decreased at term during or following nutritional manipulation is determined by the level of maternal nutrition throughout gestation. In vitro, IGF-I has been shown to have an anabolic effect on fetal adipose tissue growth (Lorenzo et al. 1993). The fetal plasma concentration of IGF-I is normally positively related to fetal glucose supply (Owens et al. 1994). An increase in nutrient supply to the fetus may also contribute to greater mRNA abundance for the receptor, thereby increasing tissue sensitivity to IGF-I. As a consequence, IGF-I may have an anabolic effect on the growth of specific tissues in the absence of a marked change in plasma concentration. This effect may also explain the loss of the relationship normally seen between fetal dimensions and organ weights at term and plasma IGF-I, which has previously been described in nutrient-restricted offspring (Heasman et al. 2000). Fetal overgrowth following in vitro fertilisation is associated with a reduction in IGF-II receptor mRNA abundance in a range of fetal tissues, including the liver, kidney, heart and muscle (Young et al. 2001), although fat has not been investigated. It has been suggested, therefore, that reduced abundance of the IGF-II receptor acts to remove some of the fetal constraints on growth; however, it is not known whether this action extends to adipose tissue growth. These findings, therefore, indicate that although IGF-I and II mRNA are both abundant within adipose tissue, an up-regulation of their receptor mRNA can promote adipose tissue deposition (Bispham et al. 2003b). It is likely that fat growth may be further enhanced after birth, particularly following a period of excess nutrient availability.

Stimulatory effects of maternal undernutrition in early gestation on mRNA abundance in adipose tissue during late gestation are not confined to IGF receptors, but extend to the glucocorticoid receptor and the enzyme 11β-hydroxysteroid dehydrogenase type 1 (Whorwood et al. 2001). This enzyme acts predominantly as an 11-oxygenase catalysing the conversion of cortisone to bio-active cortisol (Bamberger et al. 1996; Stewart & Krozowski, 1999). Transgenic mice in which 11β-hydroxysteroid dehydrogenase type I is over expressed show substantially increased visceral adipose tissue deposition at 18 weeks of age (Masuzaki et al. 2001). The extent to which altered abundance of mRNA for IGF-I or -II mRNA may act as a signal of unilocular fat mass when visceral adipose tissue deposition remains to be confirmed.

Maternal nutrient intake in late gestation and the programming of fetal leptin sensitivity

Plasma leptin is normally low in the late-gestation ovine fetus and does not respond to decreased maternal nutrition over the final month of gestation (Yuen et al. 2002). It has been shown to be positively correlated with both body weight and leptin mRNA abundance in perirenal adipose tissue in some (Yuen et al. 1999; Bispham et al. 2002), but not all (Devasker et al. 2002), studies to date. In the fetus leptin may act as a signal of unilocular fat mass when maternal nutrient intake is fixed at or above maintenance (Muhlhauser et al. 2002). A primary factor that could determine this relationship may be the prevailing plasma leptin concentration in conjunction with fat mass, as in the latter study plasma leptin was found to be much higher than in all other studies to date, i.e. 2–9 ng/ml (Muhlhauser et al. 2002) compared with 1–1·5 ng/ml (Bispham et al. 2002; Yuen et al. 2002). Moreover, in growth-restricted fetuses that are hypoglycaemic with very low adipose tissue stores fetal plasma leptin is similarly reduced (Forhead et al. 2002; Fig. 3).

The abundance of leptin mRNA in fetal adipose tissue can be markedly suppressed by increased nutrition, when the mother is fed to appetite in late gestation (Bispham et al. 2003b). Any extra available energy may be used to promote...
the growth of fetal tissues other than adipose tissue, which has a much greater energetic requirement. Other potential roles for leptin in the fetus remain to be established, although it is interesting to note that intracerebroventricular infusion of leptin into hypoleptinaemic fetuses dampens the increase in fetal corticotropin and cortisol pulses near term (Howe et al. 2002). The fetus, however, has no direct regulation of food intake and its growth is limited by nutrient availability, with increased nutrition promoting somatotrophic growth rather than fat deposition. It has yet to be investigated whether a reduction in leptin secretion from fetal adipose tissue would potentially benefit the fetus at a time when nutrient supply is not limited.

Maternal nutritional manipulation in late gestation
Manipulation of both the maternal metabolic and hormonal environment as a consequence of increasing or decreasing food intake in late gestation can act to reduce adipose tissue deposition in the resulting offspring (Symonds et al. 2003). Maternal energy restriction in late gestation, coincident with the period of maximal fetal growth and a parallel rise in fetal fat depots, results in reduced adiposity if this energy restriction is accompanied by a reduction in fetal glucose supply (Symonds et al. 1998). This adaptation in nutrient-restricted mothers can be overcome by chronic maternal adaptations that accompany cold exposure (Symonds et al. 1992), thereby increasing fetal plasma glucose concentration (Thompson et al. 1982) and promoting fat deposition. As a consequence these offspring are better adapted to meet the cold challenge of the extraterine environment as a result of an increased total thermogenic capacity within brown adipose tissue (Symonds et al. 1992). A similar overall adaptation can occur by increasing maternal food intake in late gestation (Budge et al. 2000). However, although this adaptation promotes fetal growth and total UCP1 abundance, the fetus possesses less fat when expressed in either absolute terms or relative to body weight. These divergent responses emphasise the complex control of nutrient partitioning within fetal adipose tissue.

Fetal number and adipose tissue sensitivity
A further determinant of the outcome of any nutritional manipulation in late gestation is fetal number (Budge et al. 2003). Nutrient restriction of twin fetuses causes an up-regulation of specific mitochondrial proteins within fetal adipose tissue, an adaptation that is not found in singletons. The abundance of both voltage-dependent anion channel and UCP2 are enhanced (Budge et al. 2003; Mostyn et al. 2003). These adaptations within the mitochondria are predicted to substantially alter adipocyte function. Voltage-dependent anion channel and UCP2 are both implicated in the regulation of the rate of apoptosis and necrosis (Crompton, 1999; Voehringer et al. 2000) as well as in regulating the supply of mitochondrial ATP and ADP (Gottlieb, 2000). One potential mechanism related to the increased sensitivity of adipose tissue of twins compared with singletons following maternal undernutrition may relate to the relative abundance of the prolactin receptor (Budge et al. 2003), which is greater in the adipose tissue of twins than in that of singletons irrespective of nutritional status. Twins subsequently go on to deposit more fat over the first month of postnatal life (Pearce et al. 2002). In fetal fat there is a marked developmental ontogeny for the prolactin receptor, which may act to determine the abundance of specific mitochondrial proteins (Symonds et al. 1998). Support for this proposal comes from the finding that increased maternal nutrition not only stimulates UCP1 abundance but also promotes protein abundance of the long form of the prolactin receptor (Budge et al. 2000). It is likely, therefore, that an increase in fetal plasma prolactin in late gestation specifically promotes UCP1 mRNA expression and protein translation (Budge et al. 2002).

The positive effects of maternal nutritional manipulation on mitochondrial protein abundance are not restricted to the period immediately after birth but extend into postnatal life. Offspring of energy-restricted mothers possess adipose tissue (and lungs) in which the abundance of both voltage-dependent anion channel and UCP2 remain raised at 1 month of age (Mostyn et al. 2003). A higher abundance of UCP2 may have deleterious consequences in relation to later development, as a reduced abundance of UCP2 increases resistance to infection from, for example, toxoplasmosis (Arsenijevic et al. 2000). It has also been shown that in obese women UCP2 gene exon 8 may affect susceptibility to obesity through an interaction with leptin (Cassell et al. 1999). Enhanced abundance of both UCP2 and voltage-dependent anion channel could result in an accelerated rate of apoptosis (Voehringer et al. 2000) and greater predisposition to adult disease, as a consequence of chronic impairment in lung function together with excess fat deposition. Epidemiological studies have indicated that nutrient restriction of pregnant women can predispose the resulting offspring to obesity or chronic lung disease in adulthood (Barker et al. 1991; Roseboom et al. 2000a), although it remains to be established if these two adaptations are linked. The findings in sheep indicate that the development of adipose tissue mitochondria can be reprogrammed in late gestation and may be an important mechanism by which infants of nutrient-restricted mothers could subsequently become obese.
Maternal age as a determinant of adiposity in the resulting offspring

It is recognised that birth weight normally increases with parity, even when maternal body weight has changed very little between the first and second pregnancies. This adaptation is apparent in sheep, independent of whether the ewe has a singleton or twin pregnancy (Symonds et al. 2004). The increase in birth weight with parity can, however, be compromised by previous nutrient restriction during pregnancy. For example, females born to mothers exposed to the Dutch famine in early gestation have further shown a decline in birth weight by 250–350 g with increasing parity. This information may have substantial implications for contemporary populations even when adequate nutrition is available, given the substantial decline in birth rates within the UK and Europe. It is now the norm for many women to have only one child. It is possible, therefore, that in addition to the pronounced impact that contemporary lifestyles have on increased prevalence of childhood obesity and diabetes, being born from a first pregnancy may also be an additional risk factor. The percentage of the UK population for which this situation is relevant is now far greater than that used for the majority of epidemiological studies that have involved individuals born 50–60 years ago.

It has recently been found that the offspring of primiparous mothers deposit appreciable more adipose tissue than those born to multiparous mothers (Pearce et al. 2003; Fig. 4). Critically, these effects are observed despite similar growth rates, and are therefore apparent whether tissue weights are expressed in absolute terms or relative to current body weight. Similar effects are not found in the other major organs, including the brain and heart. Maternal parity is, therefore, a major factor determining subsequent adiposity, although the extent to which this factor may have deleterious longer-term consequences has yet to be established. The enhanced adiposity in offspring of primiparous mothers is accompanied by accelerated loss of the brown adipose tissue-specific UCP1. A suppressed rate of metabolism within brown fat of these offspring may act to promote adipose tissue deposition in later life. It has been established that transgenic mice lacking UCP1 subsequently go on to develop obesity (Lowell et al. 1993), but the extent to which obesity may be prevented by enhanced UCP1 abundance in early life has been overlooked. One explanation for this oversight has been that small mammals, unlike sheep and man (Lean, 1989; Mostyn et al. 2003), always retain brown adipose tissue through their life cycle, which is likely to have profound effects on fat metabolism, and hence its growth.

The critical importance that maternal age as well as first pregnancy may play in relation to adipose tissue growth in the resulting offspring is further emphasised by studies comparing fat deposition in sheep born to juvenile or adult mothers. These studies have demonstrated that one factor that may be protective in terms of subsequent adiposity is maternal age. In 1-year-old sheep the offspring of juvenile mothers have substantially reduced fat deposition compared with those born to adult mothers (Fig. 5). The weights of all other major organs (e.g. liver, kidney, heart) are unaffected by maternal age.

![Fig. 5. Effect of maternal age on adipose tissue deposition in the resulting offspring at 1 year of age. (□), Pericardial; (■), perirenal and abdominal; (▲), omental. Values are means with their standard errors represented by vertical bars.](https://example.com/fig5.png)
Conclusion

The amount, metabolic activity and endocrine sensitivity of adipose tissue are highly dependent on the maternal nutritional, metabolic and hormonal environment. Intervention strategies aimed at these critical periods of development are very likely to reduce an individual’s predisposition to obesity in adult life. Given the annual increase in the incidence of obesity within Europe and the USA, this approach would contribute to substantial improvements in health and economic performance.

References


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