

**The transgenerational epigenetic transmission of traits allows future generations to be maximally competitive in their environment. Under this assumption, adaptive gene programs acquired during the parental life span persist in the subsequent generation.**

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## Developmental Programming and Transgenerational Transmission of Obesity

by Mark H. Vickers

### Key insights

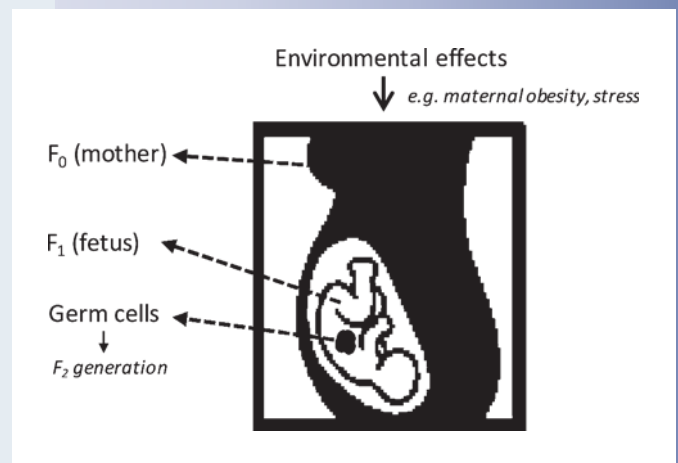
The effects of developmental programming may be transmitted not only from parent to offspring, but to generations beyond. Data from human and animal studies indicate that metabolic disorders and adverse environmental influences may be perpetuated to the F<sub>2</sub> and F<sub>3</sub> generations via both the maternal and paternal lineages.

### Current knowledge

Environmental factors can modulate the activity of the genome through several mechanisms. The most widely studied mechanism is epigenetics, with the classic example being the influence of the maternal phenotype and intrauterine environment on the regulation of fetal energy metabolism via altered DNA methylation of specific genes. However, environmental influences during key periods in development can also alter the germline of the fetus, thereby affecting the F<sub>2</sub> offspring and beyond.

### Practical implications

Exposure to environmental stressors such as poor early-life nutrition can result in maladaptive epigenetic traits that are passed on to the offspring. Over several generations, these have the potential to manifest as a population-wide phenotype. The rapid perpetuation of conditions such as obesity is especially relevant to populations undergoing the transition between traditional and Western lifestyles. There is an urgent need to de-



Developmental programming is a complex transgenerational process involving not only the F<sub>0</sub> and F<sub>1</sub> generations, but also the generations beyond. Thus, an adverse maternal environment (F<sub>0</sub>) affects both the development of the fetus (F<sub>1</sub>) and the fetal germ cells which will give rise to the F<sub>2</sub> generation.

fine the mechanisms that underpin the transmission of developmental programming in order to modulate the phenotype of future generations.

### Recommended reading

Aiken CE, Ozanne SE: Transgenerational developmental programming. *Hum Reprod Update* 2014;20:63–75.

# Developmental Programming and Transgenerational Transmission of Obesity

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## Key Messages

- Experimental and human evidence to date suggests that developmental programming is a transgenerational phenomenon. A number of potential mechanisms underlie the transmission of metabolic traits, including epigenetic effects via the germline, a suboptimal reproductive tract environment or altered maternal adaptations to pregnancy.
- Evidence from human cohorts is limited, although paternal line transmission of ill-health has been reported through to the F2 generation; data from animal studies describe transgenerational transmission of metabolic disorders to the F3 generation through both the paternal and maternal lineage following a range of altered maternal (F0) environments.
- Many studies reported to date are up to the F2 generation, whereas true transgenerational transmission is the F3 generation and beyond where there is no exposure to the initial environmental challenge; however, data in the F3 generation are limited and often variable depending on the model used.

## Key Words

Developmental programming · Transgenerational transmission · Obesity · Epigenetic inheritance · Germline · Methylation · Maternal nutrition

## Abstract

The global obesity pandemic is often causally linked to marked changes in diet and lifestyle, namely marked increases in dietary intakes of high-energy diets and concomitant reductions in physical activity levels. However, far less attention has been paid to the role of developmental plasticity and alterations in phenotypic outcomes resulting from environmental perturbations during the early-life period. Human and animal studies have highlighted the link between alterations in the early-life environment and increased susceptibility to obesity and related metabolic disorders in later life. In particular, altered maternal nutrition, including both undernutrition and maternal obesity, has been shown to lead to transgenerational transmission of metabolic disorders. This association has been conceptualised as the developmental programming hypothesis whereby the impact of environmental influences during critical periods of developmental plasticity can elicit lifelong effects on the physiology of the offspring. Further, evidence to date suggests that this developmental programming is a transgenerational phenomenon, with a number of studies showing transmission of programming effects to subsequent generations, even

in the absence of continued environmental stressors, thus perpetuating a cycle of obesity and metabolic disorders. The mechanisms responsible for these transgenerational effects remain poorly understood; evidence to date suggests a number of potential mechanisms underpinning the transgenerational transmission of the developmentally programmed phenotype through both the maternal and paternal lineage. Transgenerational phenotype transmission is often seen as a form of epigenetic inheritance with evidence showing both germline and somatic inheritance of epigenetic modifications leading to phenotype changes across generations. However, there is also evidence for non-genomic components as well as an interaction between the developing fetus with the in utero environment in the perpetuation of programmed phenotypes. A better understanding of how developmental programming effects are transmitted is essential for the implementation of initiatives aimed at curbing the current obesity crisis.

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## Background

The developmental programming hypothesis suggests that the early-life environment influences offspring characteristics in later life, including susceptibility to the development of obesity and related metabolic disorders. Growing evidence now indicates that the effects of developmental programming may also be transmitted to future generations in the absence of further environmental stressors. The developmental programming hypothesis has opened up a new research paradigm for understanding chronic disease risk that moved beyond the simplistic explanations based on genetic and lifestyle influences. A more integrated approach has developed examining the interaction between genetic inheritance and lifestyle factors (including diet) but also incorporating the role of developmental plasticity – the ability of changes in gene function to generate a range of phenotypic outcomes based on environmental exposures [1].

The transmission of developmental programming effects is often viewed as a form of epigenetic inheritance, either via the maternal or the paternal line. Evidence exists for both germline and somatic inheritance of epigenetic modifications which may be responsible for phenotypic changes in further generations [2]. It must be noted,

however, that the definition of ‘transgenerational’ is not straightforward in the setting of developmental programming [2, 3]. As per figure 1, when a perturbation is applied to the mother (F0, e.g. altered maternal diet), it directly effects the developing fetus in utero (F1) and also the germ cells that will form the F2 generation. Thus, strictly speaking, only later generations (F3 and beyond) can truly be deemed ‘transgenerational’ and not a consequence of the initial early-life exposure [2, 4]. However, except for a few rodent studies (with varying and, in some cases, conflicting data), very limited data are available on the F3 generation and beyond.

The initial epidemiological studies by Barker [5] and other researchers linked fetal growth restriction to later disease, implying that fetal nutritional deprivation may be a strong programming stimulus. This led to a range of experimental animal models that primarily utilised maternal dietary manipulations to induce fetal growth restriction, e.g. maternal calorie, protein or macronutrient deficiency during critical periods of development, to examine transgenerational phenotype transmission. However, in many societies, maternal and postnatal nutrition are now either sufficient or excessive. As a result, excessive weight gain and/or maternal obesity are the more common nutritional problems complicating pregnancy in developed countries. Thus, in view of the rising prevalence of obesity in pregnancy and its association with gestational diabetes, there is now

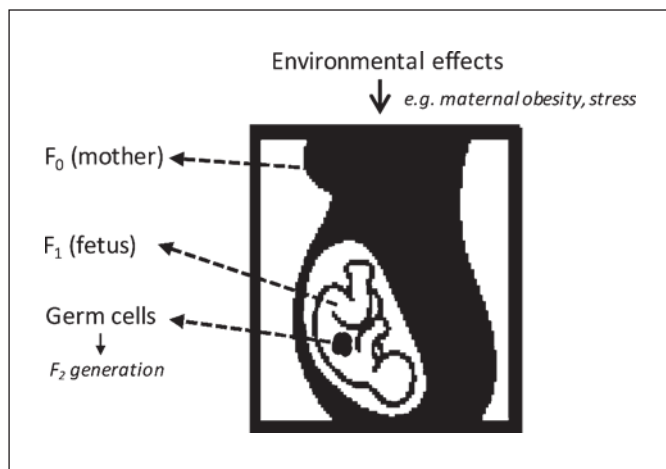
also an increasing interest in the detrimental influence of maternal obesity and excess maternal nutrition on the risk of disease in the F1 generation and beyond. Of note, both ends of the maternal nutrition spectrum can elicit similar phenotypic outcomes in the offspring with both maternal

undernutrition and maternal obesity leading to increased adiposity and related metabolic disorders in the offspring; nevertheless, whether the mechanisms are similar remains poorly defined.

## Human Populations

A systematic search of the literature for both human population and animal studies was recently undertaken by Aiken and Ozanne [2]. Data to date are primarily derived from rodent models due to the short timeframe required to generate offspring. Human evidence is under-

*The developmental programming hypothesis suggests that the early-life environment influences offspring characteristics in later life, including susceptibility to the development of obesity and related metabolic disorders.*



**Fig. 1.** The effects of a single environmental exposure can be transmitted transgenerationally. An adverse maternal environment (F<sub>0</sub>) affects not only the development of the fetus (F<sub>1</sub>) but can also affect the germ cells which form the F<sub>2</sub> generation.

standably limited due to long generation times for prospective studies and the quality of data records for retrospective studies. Two well-cited human population studies are those of the Dutch Famine cohort [6–8] and the Avon Longitudinal Study of Parents and Children (ALSPAC) and Overkalix cohorts, where both cohorts have been linked to transmission of ill-health into the F<sub>2</sub> generation [9].

The study by Painter et al. [6] on the Dutch Famine cohort did not find transgenerational effects of prenatal exposure to famine on birth weight nor on cardiovascular and metabolic disease rates. However, F<sub>1</sub> famine exposure in utero was associated with increased F<sub>2</sub> neonatal adiposity and poor health in later life. Offspring of prenatally undernourished fathers, but not mothers, were heavier and more obese than offspring of fathers and mothers who had not been undernourished prenatally [7]. In a recent study on this cohort, no evidence of transgenerational effects of grandmaternal undernutrition during gestation on the health of the F<sub>2</sub> offspring were reported, but it was suggested that the observed increases in adiposity in the F<sub>2</sub> offspring of prenatally undernourished fathers may lead to increased chronic disease rates in the future [7]. Of note, reports from the Dutch Famine cohort have been varied. One study observed that F<sub>1</sub> offspring were born smaller and that these effects persisted to the F<sub>2</sub> generation [8]; however, a subsequent report by the same authors failed to reproduce some of the initial findings [10].

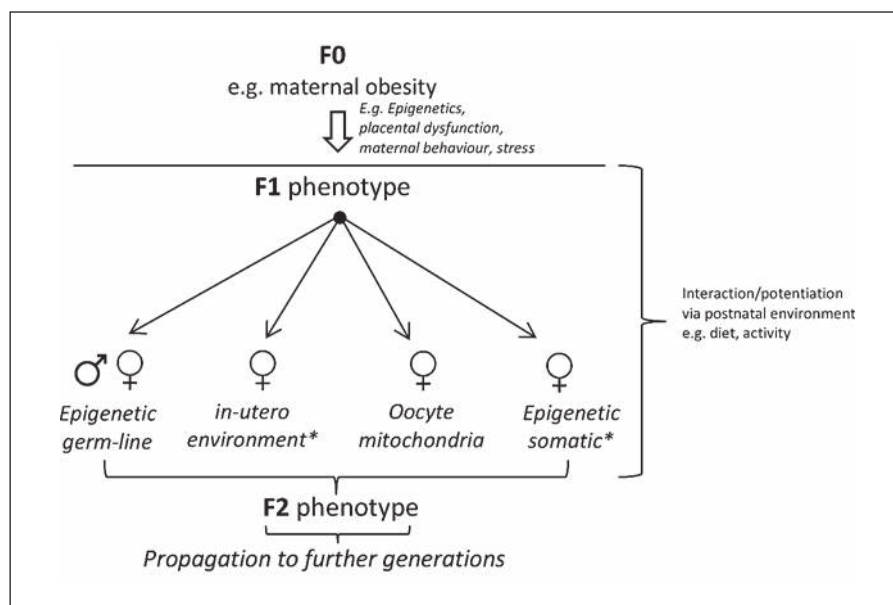
In the ALSPAC/Overkalix cohorts from 1890, 1905 and 1920, male-line transgenerational responses were reported; the paternal grandfather's food supply was only linked to the mortality risk ratio of grandsons, while the paternal grandmother's food supply was only associated with the granddaughters' mortality risk ratio. These transgenerational effects were observed with exposure during the slow-growth period (both grandparents) or fetal/infant life (grandmothers) but not during either grandparent's puberty [9]. Although these studies suggest transgenerational effects induced by environmental factors, molecular evidence to date does not support a direct transfer of epigenetic information via the gametes, and it is possible that transgenerational effects of this type could be explained by societal factors [11].

### Animal Models

Evidence for the transgenerational programming of adverse metabolic outcomes has been shown in a number of experimental paradigms, primarily in the rodent. These include challenges such as nutrient restriction or over-feeding during pregnancy and lactation, restriction of uterine blood flow, intrauterine exposure to high levels of glucocorticoids and experimental gestational diabetes. Although a number of primary research papers have used small animal models of developmental programming to examine at least through to the F<sub>2</sub> offspring, only a few have examined obesity as an endpoint, with many focusing on insulin sensitivity and glucose tolerance, cardiovascular outcomes or changes in DNA methylation [2].

While a large number of animal studies have shown the effects of undernutrition during fetal/perinatal development on glucose metabolism in exposed animals (F<sub>1</sub>) in adulthood [12], several studies have reported that glucose metabolism is also altered in the offspring (F<sub>2</sub>) of F<sub>1</sub> females undernourished in utero, even when the F<sub>1</sub> females have been well-nourished after weaning [13, 14]. Maternal protein restriction in the rat adversely affects the glucose metabolism of male and female F<sub>2</sub> offspring in a gender- and developmental time window-specific manner (i.e. gestation and/or lactation) [15]. The offspring and grand-offspring of female rats fed low-protein diets during pregnancy and lactation, but fed nutritionally adequate diets thereafter, exhibit altered insulin sensitivity in adulthood [16]. However, Benyshek et al. [17] showed that maternal energy restriction did not consistently program reduced insulin sensitivity in offspring over three consecutive generations. The reasons for this remain unclear, although it is possible that the transgenerational transmission of de-

**Fig. 2.** Proposed mechanisms by which developmental programming in the F0 generation can be transmitted to the F3 generation and beyond via either the maternal or paternal lineage. The asterisks denote pathways of de novo phenotype propagation. Adapted in part from Aiken and Ozanne [2].



velopmentally programmed insulin resistance is determined in part by the relative insulin sensitivity of the mother during pregnancy/lactation.

Furthermore, it has been reported that the glucose metabolism of the grandoffspring (F3) of female rats malnourished during development is also adversely affected, but these effects are diminished as compared to those observed for the F2 generation [17]. These data may suggest a normalisation in the F3 generation when the maternal diets of F2 dams and post-weaning diets of F3 animals were adequate, and may provide further evidence of an eventual intergenerational ‘resolution’ of the altered glucose-insulin metabolism [17–19]. Whether such an intergenerational normalisation can be accelerated by manipulating the diet of insulin-resistant F2 dams remains to be seen. A meta-analysis examining transgenerational effects of maternal caloric restriction on appetite revealed a weak and statistically non-significant overall effect on offspring’s appetite [20]. However, it also showed that a lower protein content of restricted diets was associated with higher food intake in female offspring. Importantly, these data suggest that a main source of variation among studies arises from whether, and how, food intake was adjusted for body mass.

Maternal undernutrition during pregnancy in the mouse programs reduced birth weight, impaired glucose tolerance and obesity in both F1 and F2 offspring. The sex-specific transmission of these phenotypes suggests complex mechanisms including alterations in the mater-

nal metabolic environment (transmaternal inheritance of obesity), gene expression mediated by developmental and epigenetic pathways (transpaternal inheritance of low birth weight), or both (impaired glucose tolerance) [21]. Using a model of intrauterine hyperglycaemia, Ding et al. [22] reported transgenerational glucose intolerance with *Igf2/H19* epigenetic alterations in mouse islets. In this model, a high risk of impaired glucose tolerance appeared as early as 3 weeks in F2 offspring and progressed through both parental lineages, particularly the paternal line. In a model of placental insufficiency, fetal exposure to maternal hypertension and hypoleptinaemia was associated with altered leptin and growth patterns in mature female offspring in the F1 generation but was not perpetuated to the F2 generation [23]. First-generation female diabetic offspring of F0 rats treated with streptozotocin during pregnancy had F2 offspring with altered glucose and carbohydrate metabolisms. These studies suggest that the mechanisms involved in developmental programming are likely epigenetic rather than due to DNA sequence mutations [24].

Other studies have reported a transgenerational passage of effects resulting from treatment of pregnant rats with dexamethasone (DEX) by either the maternal or paternal lineage. Male offspring of female rats that had been prenatally exposed to DEX, but not manipulated in their own pregnancy, had reduced birth weight, glucose intolerance and elevated hepatic PEPCK activity. Similar transgenerational programming was observed in off-

spring of male rats prenatally exposed to DEX mated with control females. However, these effects were resolved in the F3 generation [18]. Experimental models in the guinea pig (maternal DEX and low-protein diet) have shown DNA methylation changes, altered hypothalamic-pituitary axis function and cardiovascular impairment as well as delayed neurodevelopment in the F2 offspring [25, 26]. Maternal diets deficient in micronutrients have also been shown to result in F2 phenotypes including vitamin D and zinc deficiencies [27–29].

As reported in the rodent undernutrition models, the work by Pentinat et al. [30] also suggests a partial resolution of the phenotype in subsequent generations in models of early-life overnutrition. A model of neonatal overfeeding (ON) in the F0 generation was used to examine the development of the metabolic syndrome in male offspring (ON-F1) and grandoffspring (ON-F2) of ON-F0 male mice, which were not overfed during lactation. ON-F1 mice developed fed and fasting hyperinsulinaemia, hypertriglyceridaemia, insulin resistance and glucose intolerance, but not obesity, by the age of 4 months. In contrast, ON-F2 male mice showed a more moderate phenotype and only developed fasting hyperglycaemia and glucose intolerance at 4 months of age [30].

The impact of paternal obesity in transgenerational inheritance has also been reported in recent work by Fullston et al. [31]. Paternal obesity was shown to initiate metabolic disturbances in two generations of mice albeit with incomplete penetrance to the F2 generation. Diet-induced paternal obesity modulated the sperm microRNA content and germ methylation status, which are potential signals that program offspring health and initiate the transmission of obesity to future generations. Dunn and Bale [32] have previously reported that maternal high-fat diet exposure in mice results in an increase in body size and reduced insulin sensitivity that persisted to the F2 generation via both maternal and paternal lineages. However, as described above, as the first generation's primordial germ cells may be affected by gestational exposure, analysis of phenotype transmission into the F3 generation is necessary to determine whether stable epigenetic programming has occurred. Further work looking at the F3 generation in this model revealed that only females displayed an increased body size at F3, and this effect was only passed on via the paternal lineage. The finding of a paternally transmitted phenotype to F3 fe-

male offspring further supports a stable germline-based transgenerational mode of inheritance, thus suggesting that imprinted genes may be involved in such epigenetic programming [33].

In addition to studies in the rat and mouse, studies in large animal models have also reported transgenerational programming effects including those in the sheep, pig and primate [34–36]. In the sheep, maternal DEX administration to F0 mothers abolished the neonatal leptin peak in female offspring possibly by inhibition resulting from elevated cortisol levels in the DEX F2 offspring. The DEX F2 offspring displayed hyperphagia, increased weight gain and adiposity during an ad libitum feeding challenge concomitant with a decreased insulin responsiveness following a glucose tolerance test [36]. Using methylation micronutrient-supplemented diets in the pig, gene ex-

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***The mechanisms underlying transgenerational effects in the setting of developmental programming remain poorly understood.***  
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pression, DNA methylation and carcass composition differences were observed between F2 offspring of supplemented and non-supplemented groups, with the F2 control offspring tending towards increased adiposity

compared to the F2 offspring of supplemented pigs. These effects were transmitted through the male lineage [34]. In the primate, also utilising a maternal DEX model, F2 and F3 offspring presented higher cholesterol levels, with significantly more low-density lipoprotein cholesterol, although body weights were not affected [35].

### **Mechanisms**

The mechanisms underlying transgenerational effects in the setting of developmental programming remain poorly understood. Human studies of transgenerational programming are observational in nature and therefore have limited value in determining the mechanisms underlying these phenomena [37]. Data derived from animal models suggest a number of potential mechanisms which may underpin the transgenerational transmission of the programmed phenotype, including persistence of the abnormal environment across generations, programmed effects on maternal physiology and the transmission of epigenetic information through the germline (fig. 2) [2, 19, 38].

Although evidence to date indicates that multiple mechanisms are in play in the interaction between nutritional imbalance and the transgenerational transmission of obesity and related metabolic phenotypes, it is the role

of epigenetics that has gained increasing momentum in recent years [21]. Environmentally induced transgenerational epigenetic inheritance of adult-onset disease involves a variety of phenotypic changes, suggesting a general alteration in genome activity. Combined observations demonstrate that all tissues derived from the epigenetically altered germline develop transgenerational transcriptomes unique to the tissue, but common epigenetic control regions in the genome may coordinately regulate these tissue-specific transcriptomes [39]. Data from Burdge et al. [40] suggest that the regulation of energy metabolism during pregnancy and lactation within a generation is influenced by the maternal phenotype in the preceding generation and the environment during the current pregnancy. These transgenerational effects on phenotype are associated with altered DNA methylation of specific genes in a manner consistent with a de novo induction of epigenetic marks in each generation.

In environmentally mediated inheritance, it is suggested that the epigenetic marks in the parents modify their behaviour in a way that causes the same epigenetic marks in their offspring (e.g. altered hypothalamic DNA methylation), and that these behavioural changes recreate the epigenetic marks de novo at each subsequent generation [38]. An example of this are alterations in maternal care where there is evidence for the behavioural transmission of postpartum behaviour from mothers to female offspring [41, 42]. The mechanisms underpinning this transmission have been explored in rats and implicate oestrogen-oxytocin interactions and the differential methylation of hypothalamic oestrogen receptors [41]. However, a recent study has also shown that programmed obesity in offspring of obese mothers is independent of the level of maternal care, although only first-generation effects were investigated [43].

In germline epigenetic inheritance, environmental influences during the period of developmental plasticity leads to an epigenetic change within the first-generation offspring's germline that is then transmitted to F2 offspring and beyond [38]. In this setting, a number of environmental factors (e.g. toxicants) have now been shown to promote the transgenerational epigenetic inheritance of disease and phenotypic variation [44]. A well-cited example of this relates to exposures to endocrine disruptors such as bisphenol A. Perinatal exposure of male rats to bisphenol A leads to perturbations in the expression profile of testicular steroid receptor co-regulators through to the F3 generation [45]. Of note, maternal behaviour may also be affected by bisphenol A exposure [46], thus potentially leading to behaviour-mediated effects on future gen-

erations [41]. Ancestral exposure to the insecticide dichlorodiphenyltrichloroethane in the rat has been reported to promote transgenerational epigenetic inheritance of obesity through to the F3 generation where over 50% of males and females developed obesity [47]. The transgenerational transmission of disease was through both the female and male germlines, and the F3 generation displayed sperm epimutations and differential DNA methylation regions. Further, perinatal exposure of 4-nonylphenol, at environmentally relevant doses, can lead to obesity in both male and female F1 offspring. This effect progresses to the F2 offspring through the maternal line [48].

Mutations in folate metabolism can cause epigenetic instability and transgenerational effects on development. Embryo transfer experiments have revealed that methionine synthase reductase deficiency (necessary for the utilisation of methyl groups from the folate cycle) in mice leads to two distinct, separable phenotypes: adverse effects on their wild-type daughters' uterine environment, leading to growth defects in wild-type grandprogeny, and the appearance of congenital malformations independent of maternal environment that persist for five generations, likely through transgenerational epigenetic inheritance [49]. The data on maternal folate status are less clear, with some evidence that maternal folate supplementation can lead to insulin resistance and transgenerational transmission of respiratory disorders in offspring [50, 51].

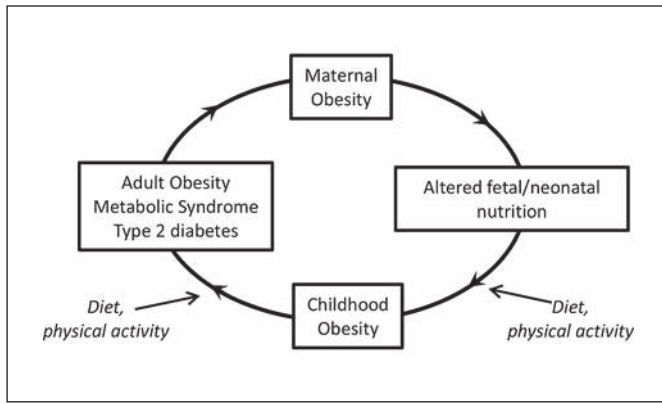
In addition to epigenetic effects, the contribution of

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***Suboptimal nutrition in utero causes  
DNA damage and accelerated aging of  
the female reproductive tract.***

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the uterine tract environment or maternal adaptations to pregnancy may be critical to programming inheritance via the maternal line. Suboptimal nutrition in utero causes DNA damage and accelerated aging of the female reproductive tract [52]. Aiken and Ozanne [2] suggested that developmental programming effects could be propagated through the maternal line de novo in generations beyond F2 as a consequence of development in a suboptimal intrauterine tract and not necessarily through directly transmitted epigenetic mechanisms. Further, as the effects of age exacerbate the programmed metabolic phenotype, advancing maternal age may increase the likelihood of developmental programming effects being transmitted to future generations.



**Fig. 3.** The so-called vicious cycle of obesity where obesity begets obesity. Maternal obesity results in obesity in offspring during their reproductive years, and thus the obesity cycle is perpetuated. The developmental programming effects can be exacerbated (or potentially ameliorated via lifestyle interventions) by neonatal nutrition and later diet and activity levels.

The *de novo* regeneration of the programming phenotype via the maternal line has been referred to as the ‘vicious’ cycle of developmental programming, e.g. obesity begets obesity (fig. 3). It is well established from epidemiologic and experimental studies that offspring of obese mothers are at increased risk for obesity and metabolic disorders in later life [53, 54]. Thus, increasing rates of maternal obesity translate to birth of offspring that are themselves predisposed towards obesity in their reproductive years, hence perpetuating the obesity cycle. Models in mice with a genetic tendency for obesity show that the effects of maternal obesity accumulate over successive generations and shift the population distribution towards an increased adult body weight. These data suggest that epigenetic mechanisms are involved in this process, and it has been shown that methyl donor supplementation, i.e. to induce DNA hypermethylation during development, can potentially prevent this transgenerational amplification of obesity [55].

### Summary

The experimental and human evidence to date suggests that developmental programming should be regarded as a transgenerational phenomenon. The transgenerational epigenetic transmission of traits allows future generations to be maximally competitive in their environment [33]. Under this assumption, adaptive gene programs acquired during the parental life span persist in the subse-

quent generation, enabling future generations to better exist in a potentially adverse environment. However, evidence suggests that environmental exposures such as poor early-life nutrition result in maladaptive parental responses that can be passed to offspring. These epigenetic traits have the potential to result in a population-wide manifestation of a phenotype over several generations – such transmission can exacerbate the rapid onset of phenotypes such as obesity currently observed in human populations [33].

To date, very few studies have examined the transgenerational transmission of obesity in the context of developmental programming, with most focussing on glucose tolerance, cardiovascular outcomes or methylation status. Further, few studies have examined a phenotype in the F3 generation, argued to be a true marker of transgenerational inheritance. For example, confirmation of a germline-based mechanism requires both the analysis of the F3 generation to rule out any direct effects of programming via maternal diet and the transmission through the paternal lineage to avoid the confounding contributions of maternal factors such as altered in utero environment or behaviour [33]. Those F3 data that have been reported have found varying and, in some cases, conflicting outcomes. These outcomes are confounded by the range of models and interventions used (e.g. low-protein high-energy diet, zinc deprivation) and the application of interventions in the F1 and subsequent generations [2].

Many rodent models have investigated the ‘parent of origin’ question, i.e. are the F2 effects produced via the maternal line, the paternal line, or both. This is an important component given the known sex differences programmed in the F1 generation in many developmental programming models [56]. Human epidemiological evidence and rodent studies suggest that transgenerational effects can be passed down the paternal line [18, 22, 33]. These effects are proposed to act via germline epigenetic modification despite evidence of extensive demethylation during germ cell formation and zygotic development. A maternal high-fat diet has been shown to program a true germline-based transgenerational phenotype in the male gametes [33]. Studies in F1 sperm have shown a role for altered IGF2 and H19 expression in the transmission of a phenotype to the F2 offspring [22]. However, not all studies reporting a paternal line transmission have reported epigenetic alterations in the F1 sperm [57]. Work by Radford et al. [58] failed to find any evidence that the epigenetic reprogramming of imprinting control regions in the germline was susceptible to nutritional restriction, thus



implying that mechanisms other than direct germline transmission are responsible.

Transgenerational programming via the maternal line is more complex to define as there are a number of possible interacting mechanisms by which the mother can exert programming effects on the subsequent generation [2]. These include the role of the intrauterine environment, effect of age of pregnancy, somatic epigenetics and ooplasmic (mitochondrial) programming.

Although a number of studies have now reported transmission through to the F2 lineage, transmission up to the F3 or subsequent generation is less clear, with some studies reporting a resolution of the phenotype by the F3 generation. In the meta-analysis by Aiken and Ozanne [2], of 9 studies carried through to F3, 5 failed to show any effect. Defining the mechanisms underpinning the transmission of developmental programming is an area urgently requiring further research and is particularly relevant to populations in transition between traditional and Western lifestyles. The fact that some traits appear to be resolved where others persist suggests that divergent mechanisms of transmission are involved and that those

metabolic traits that do persist are capable of being transmitted via the male germline [33]. However, human evidence remains largely unsubstantiated with the strongest argument for transgenerational epigenetic inheritance in humans being data derived from the rodent [11]. Understanding the mechanisms of transgenerational inheritance is essential for the development of future intervention strategies to modulate not only that of the immediate adult phenotype but also that of the offspring, grandoffspring and beyond.

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