Non-genomic transgenerational inheritance of disease risk

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Summary

That there is a heritable or familial component of susceptibility to chronic non-communicable diseases such as type 2 diabetes, obesity and cardiovascular disease is well established, but there is increasing evidence that some elements of such heritability are transmitted non-genomically and that the processes whereby environmental influences act during early development to shape disease risk in later life can have effects beyond a single generation. Such heritability may operate through epigenetic mechanisms involving regulation of either imprinted or non-imprinted genes but also through broader mechanisms related to parental physiology or behaviour. We review evidence and potential mechanisms for non-genomic transgenerational inheritance of 'lifestyle' disease and propose that the 'developmental origins of disease' phenomenon is a maladaptive consequence of an ancestral mechanism of developmental plasticity that may have had adaptive value in the evolution of generalist species such as *Homo sapiens*. *BioEssays* 29:145–154, 2007.

Introduction

A few diseases, such as cystic fibrosis, are monogenic in origin and show classical Mendelian inheritance, but for most of the chronic non-communicable diseases, such as type 2 diabetes mellitus, a range of familial factors operates, from multiple polymorphisms to lifestyle and socioeconomic effects. Dissecting the etiological significance and inheritance patterns of these factors, and the underlying mechanisms, is complex. Further, recent experimental modelling of such disease states has demonstrated that environmental influences impacting on one generation, particularly in early development, can have effects on the phenotype of subsequent generations, and there are hints from epidemiological data that such processes also operate in humans. These effects do not result from classical genetic inheritance—they may be mediated by intergenerational transmission of epigenetic marks or by more indirect mechanisms.

In the comparative biological sciences, the concept of 'maternal effects', whereby environmental influences on one generation can have significant impact on the next generation, and potentially on subsequent generations, is well recognized and the adaptive advantage of such processes has been discussed.[1] The relevance of such mechanisms for mammalian biology has received only cursory attention until recently, but there is growing realization that a range of nutritional, hormonal, xenobiotic and behavioural cues affecting parents (the F_0 generation) can have consequences for the next generation (F_1) and in some instances for subsequent generations (F_2 onwards), even if they did not experience the same cue.

In this review, we consider the evidence for such non-genomic inheritance in mammals, focusing on its importance for our understanding of the strong familial influences on susceptibility to the metabolic syndrome characterized by type 2 diabetes, obesity, disordered blood lipid levels and cardiovascular disease. Here we are not concerned with the well-documented effects of altered maternal physiology on the fetus but with effects that persist well after birth and particularly with those that persist to the F_2 generation and beyond (Table 1). This discussion is placed in the broader context of the potential adaptive value of transgenerational inheritance of non-genomic information.[23]

Experimental evidence for non-genomic inheritance

There is growing evidence that a number of challenges can induce transgenerational non-genomically determined phenotypic changes in mammals. Such challenges, generally of F_0 females during pregnancy, include imposed exercise (resulting in effects on F_2 litter size and weight[24]), streptozotocin-induced gestational diabetes (effects on F_2 pancreas[25]), uterine artery ligation (impaired F_2 glucose homeostasis[26]) and a variety of drug and hormone exposures.[27]

However, most studies have involved nutritional or endocrine manipulation. Feeding a low-protein or unpalatable diet

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Funding agencies: P.D.G. and A.S.B. are supported by the National Research Centre for Growth and Development. M.A.H. is supported by the British Heart Foundation.
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DOI 10.1002/bies.20522
Published online in Wiley InterScience (www.interscience.wiley.com).

Table 1. Diseases or physiological states in mammals reported to show non-genomic transgenerational inheritance (to F_2 generation or beyond)

Trait affected	Inducing stimulus	Generations affected	Suggested mechanism	Reference
Human observations				
Mortality (diabetes-related and overall)	Food supply during specific growth periods	F_2	Reprogramming of imprinted genes during gametogenesis	(2,3)
Reproductive abnormalities and tumours	Exposure to diethylstilbestrol during pregnancy	F_1–F_2	Epigenetic	(4,5)
Prader-Willi and Angelman syndromes (some cases)	Imprinting defect	F_2	Failure to erase grandparental imprint	(6)
Birth size	Nutrition in the matriline	Multiple	Maternal nutritional status or altered development of maternal reproductive tract	(7,8)
Type 2 diabetes	'Nutritional stress'	Multiple	Intrauterine hyperglycaemia caused by maternal diabetes	(9)
Animal studies				
Agouti phenotype (mouse)	Natural variation, ? maternal nutrition or endocrine exposure	F_1–F_2	Phenotype determined by DNA methylation status of metastable epiallele. Mechanism of inheritance unclear	(10,11)
Fertility, various abnormalities in adulthood (rat)	Exposure to endocrine disruptors during pregnancy	F_1–F_4	Epigenetic change in male germline	(12,13)
Reproductive tumours (mouse)	Exposure to diethylstilbestrol during pregnancy	F_1–F_2	Epigenetic change in female germline	(14)
Behaviour (rat)	Maternal attention to neonate	F_1–F_2	Phenotype determined by DNA methylation status of hippocampal glucocorticoid receptor. Inheritance may be epigenetic or behavioural	(15,16)
Dysregulation of glucose metabolism, liver enzyme levels (rat)	Exposure to glucocorticoid during pregnancy	F_1–F_2	Transmission by F_1 male and female lineages, lost in F_3; epigenetic effect on embryonic germ cells?	(17)
Endothelial dysfunction (rat)	Protein restriction during pregnancy	F_1–F_2	Uterine vascular function?	(18,19)
Growth, glucose/insulin metabolism (mouse, rat)	Protein restriction during pregnancy	F_1–F_3	Altered intrauterine environment resulting from dysregulation of maternal glucose/insulin metabolism	(20–22)

to rats for twelve generations progressively reduced birthweight, which did not return to control values until three generations after reinstating a balanced diet.[28] Offspring of rat dams exposed to a low-protein diet showed effects on weight and DNA content of the cerebral hemispheres in the F_2 generation that were not prevented by cross-fostering pups to dams fed the control diet.[29] In rats fed a low-protein diet, reduced organ weights in F_2 were potentiated by undernutrition of F_1 females.[30] F_2 offspring of the female lineage from protein-restricted dams show elevated blood pressure, endothelial dysfunction[18] and increased insulin resistance[20] despite normal nutrition of the F_1 generation, and the effects on glucose metabolism extend to the F_3 generation.[21]

Endocrine or behavioural challenges can also result in effects on the F_2 generation. Offspring of rat dams exposed to dexamethasone in late pregnancy develop a phenotype with similarities to the human metabolic syndrome (e.g. elevated hepatic phosphoenolpyruvate carboxykinase) in F_1 and F_2,[17] the latter effect transmitted via both the male and female F_1 lineages irrespective of the lineage of the mate and the non-exposure of the F_1 generation. Effects on F_2 behaviour, reflected in altered expression of the glucocorticoid receptor in the hippocampus, can be induced by F_0 maternal behaviour such as grooming and by handling the pups.[15] Exposure of pregnant F_0 mice to diethylstilbestrol increases susceptibility to tumour formation in both female and male mice in the F_2 generation, transmitted via the maternal lineage, observations that are strikingly similar to those in humans. This effect is associated with persistent changes in proto-oncogene expression and concomitant alterations in DNA methylation.[14]

Of particular interest is the report that transgenerational effects through the male lineage can be induced by endocrine disruptors such as vinclozolin (antiandrogenic) or methoxychlor (estrogenic). Maternal exposure of F_0 rats during the period of offspring sex determination and testis development, but not exposure later in pregnancy, induced defects in offspring sperm formation and fertility transferred through the male line to the fourth generation.[12] These defects correlated with specific alterations in DNA methylation patterns in the testis.[12] Offspring of vinclozolin-treated animals also displayed heightened susceptibility to abnormality or disease in a number of systems during late adulthood, a susceptibility that was heritable to the fourth generation without exposure of intervening generations.[13]

Epigenetic mechanisms

Apart from the infrequent occurrence of mosaicism, organisms carry the same genotype in all their somatic cells, but time-, location- and environment-dependent modulation of gene expression, often by 'silencing' of expression,[31] is fundamental to tissue differentiation and development, X chromosome inactivation and genomic imprinting. Such modulation is a result of epigenetic processes, a term that today implies alteration of gene expression by chemical modification ('marking') of chromatin—either of DNA without change in the underlying nucleotide sequence or of DNA-binding proteins leading to alteration of DNA packing around the histone core—or by specific binding of small RNA molecules. The term 'epigenetic' was coined by Waddington[32] to refer to the ways in which the developmental environment can influence the mature phenotype. His work stemmed from observations that environmental influences during development could induce alternative phenotypes from a genotype, some of the clearest examples being polyphenisms in insects.[33] Such processes can however also induce a gradation of phenotypes, constituting a population reaction norm.[34] Waddington showed in *Drosophila melanogaster* that a developmentally plastic response, alteration in wing vein pattern, could be induced by heat-shock treatment of the pupae. Breeding of flies particularly susceptible to this environmentally induced change selected for genetic variants that exhibited the phenotype without the environmental stimulus. Waddington termed this 'genetic assimilation'.[35] Other processes of phenotypic accommodation have also been suggested and a large body of work in developmental plasticity is based on these concepts.[36] Such work, largely overlooked by proponents of the Modern Synthesis of genetic and evolutionary biology,[37] demonstrates a dynamic interaction between the genome and the environment during the plastic phase of development, producing effects that could be heritable.[36]

Methylation of the cytosine residues of DNA, particularly in CpG dinucleotides present in regions termed CpG islands within or near to promoters at the 5′ end of genes, is the best characterized form of epigenetic marking. DNA methylation is generally associated with reduced transcriptional activity, not only through decreased binding of transcription factors but also by attracting methyl-CpG-binding proteins that act as transcriptional repressors.[38] The small proteins called histones are involved in packaging of DNA into chromatin, and chemical modification of histones by, for example, lysine residue methylation or acetylation causes relaxation or condensation of the local chromatin structure, thereby adding another layer of control of gene expression by regulating access to the underlying DNA.[39] The enzymes that control these processes are only recently becoming understood. DNA methyltransferases are responsible for the initiation of DNA methylation and for its replication during mitotic cell division,[38] whereas demethylation may occur actively and specifically, a process for which the details remain controversial but may involve base excision repair enzymes or a specific DNA demethylase,[40] or passively via failure to maintain methylation during cell division. Histone modification appears to involve pairs of enzymes that add (for example, acetyltransferases and methyltransferases) or remove (deacetylases and demethylases) the corresponding marks.

Beyond these chemical modifications, emerging evidence for transcriptional gene silencing by binding of small non-coding RNA molecules adds a further dimension. The best-studied mammalian example is X-chromosome inactivation in females, but it is now also becoming apparent that RNA-based mechanisms can mediate other types of gene silencing.[41,42] Indeed, the binding specificity conferred by base-pairing interactions between DNA and small RNAs may control epigenetic marking of specific regions of the genome, providing a mechanism for the precision of DNA and histone modification in spite of the very small set of enzymes that appears to be involved.[43]

Epigenetic mechanisms underlying non-genomic inheritance

Epigenetic inheritance is defined as acquired information that can be passed to progeny through the genome without changing its DNA sequence.[23] Epigenetic variation generated by epigenetic inheritance systems[44] can be random with respect to the environment and has been termed epimutation[45] or can be induced by specific environmental cues and may therefore be directional. Inheritance of epigenetic states is common in plants,[46] where germ cells are formed from somatic cells at a late stage in development. DNA hypermethylation underlies the epimutation responsible for radial symmetry of toadflax flowers, a variant described by Linnaeus in the eighteenth century.[47]

The discovery of genomic imprinting,[48] in which expression of certain genes is determined by the gender of the parent that contributed the allele, established that patterns of gene expression could be inherited without changes in the sequence of genomic DNA through silencing of one set of alleles depending on parental origin. In mammals, imprinted genes are frequently involved in fetal and placental growth.[49] Disease resulting from dysregulation of imprinting is well recognized, e.g. Beckwith-Weidemann syndrome, but rare, although its incidence is increased in offspring of assisted reproduction.[50] Imprinting is mediated by allele-specific DNA methylation of imprinting control regions, although the precise mechanisms of how imprinting is established and maintained remain unclear.[51] Following fertilization, similar mechanisms can silence non-imprinted genes throughout development.[31]

Observations that environmental influences acting in the F_0 generation can have consequences for later generations suggest epigenetic effects on either imprinted or non-imprinted genes. Unfortunately, most mechanistic studies of epigenetic inheritance have focused on F_0 to F_1 transmission, with much more limited information beyond. The best-characterized examples in mammals involve two retrotransposon-associated metastable epialleles in the mouse, *agouti*[10] and *Axin-fused*,[52] both of which display epigenetic inheritance from F_0 to F_1 in association with, but for *agouti* at least not directly mediated by,[11] the DNA methylation status of the retrotransposon sequences. Importantly, the level of DNA methylation of the epialleles in the F_1 generation, and the resulting phenotype, can be altered by the F_0 maternal environment as reflected by dietary manipulation[53,54] or, for *agouti*, endocrine exposure.[55] Although it remains unknown whether the environmentally induced phenotypes can be transmitted to the F_2 generation, naturally occurring variation in the *agouti* trait does indeed show a grandmaternal effect.[10]

There are an increasing number of studies showing that maternal nutritional or hormonal manipulations in the rat typical of those known to induce transgenerational non-genomic inheritance also induce specific epigenetic changes in DNA methylation or histone acetylation in the offspring. These in turn correlate with altered gene expression. For example, in rats subject to maternal protein restriction during pregnancy, Lillycrop et al.[56] confirmed that reciprocal changes occur in the F_1 offspring in gene promoter methylation and expression of the peroxisome proliferator-activated receptor (PPAR) α and glucocorticoid receptor genes in the liver, associated with changed expression of downstream genes such as acyl-CoA oxidase. The changes appear to be targeted to specific genes, as they are not observed for hepatic PPARγ.[56] All these effects were absent from offspring of dams given the low-protein diet with folate supplementation, suggesting the importance of methyl group provision. Prenatal under-nutrition also induces changes in histone H3 and H4 acetylation, consistent with facilitated transcription, at the glucocorticoid receptor gene in the liver.[57] Recent work shows that changes in methylation of the glucocorticoid receptor promoter in the liver are also reflected in the F_2 generation in the absence of dietary manipulation of F_1 female offspring;[58] as in the *agouti* model,[11] it may not be the methylation mark itself that is transmitted. Using the more severe challenge of uterine artery ligation, Pham et al. showed changes in promoter methylation of the gene for the pro-apoptotic protein p53 in the kidney of the offspring,[59] suggesting a mechanism for the reduced nephron number seen.

As reviewed recently,[60] changes in mitochondrial copy number and function have been shown in various tissues of F_1 offspring of rat dams fed a high-fat or low-protein diet, and mitochondrial DNA synthesis during preimplantation development is particularly susceptible to environmental stress. Although mitochondrial DNA is inherited via the maternal lineage, the mitochondrial genome encodes only a small proportion of mitochondrial proteins, the remainder being encoded by nuclear DNA. Transmission of mitochondrial dysfunction could therefore involve either maternal or paternal nuclear epigenetic processes, such as that by which expression of a nuclear-encoded binding factor that stabilises mitochondrial DNA is regulated by promoter methylation of a related transcription factor.[61]

An additional mechanism concerns effects on primary oogenesis, which occurs only in fetal life.[62] Any maternal exposure that modifies oocyte development will specifically affect F_2 offspring but will not necessarily be transmitted further. This intriguing possibility may underlie the observation described above of transmission of elevated enzyme levels to F_2 but not F_3 offspring after F_0 maternal glucocorticoid treatment.[17]

Although epigenetic memory of active transcription can be inherited through mitotic divisions after somatic cell nuclear transfer,[63] it is not clear how epigenetic marks on imprinted or non-imprinted genes could be transmitted intergenerationally through normal fertilization. Widespread reprogramming of epigenetic marks, involving both active and passive demethylation and reorganisation of histone modifications, occurs in early post-fertilization mammalian development to ensure totipotency of the developing zygote, followed by establishment of different sets of marks associated with various cell lineages.[64] However, imprinted genes and some types of retrotransposon appear to be resistant to demethylation. In the primordial germ cells of the developing fetus, erasure of methylation is followed by sex-specific de novo methylation during gametogenesis. In the mouse, demethylation occurs at about E12 in both sexes and remethylation of the male genome begins at about E16 and is complete shortly after birth. In contrast, in the female, remethylation occurs during postnatal oogenesis, with some imprints being acquired relatively late, although the process is complete before metaphase II arrest of the oocyte.[65] The timing of re-acquisition of imprinted methylation marks during gametogenesis is parental allele-specific;[65,66] this indicates that, although methylation is erased, the two alleles retain some form of epigenetic memory of their origin, possibly involving small RNAs or modification of DNA-associated proteins such as histones, that is able to direct re-methylation. The period between erasure and complete re-acquisition of oocyte methylation in the mouse spans prenatal and postnatal development, providing a wide window of opportunity for environmental modulation of the imprinting process.

The early separation of somatic and germline tissues in mammals suggests that examples of transgenerational epigenetic inheritance are most likely to be found when environmental factors have generated epialleles in the germ line.[67] Indeed, most examples of confirmed or suspected epigenetic inheritance result from environmental exposures during F_0 pregnancy or other periods of gametogenesis (Table 1). That environmentally induced changes in gene expression in somatic tissues of adult mammals could result in heritable modifications of the gametes appears less plausible, although the heritability of systemically applied small RNA signals in the roundworm *Caenorhabditis*[68] suggests a possible mechanism by which such an effect might occur.

The epigenetic activity of small RNAs in the gametes is receiving increasing attention. RNA-mediated epigenetic inheritance through both paternal and maternal lineages has been demonstrated in the mouse,[69] and there is growing appreciation that such a mechanism may at least in part explain non-genomic inheritance via the paternal line.[70] Human spermatozoa contain a number of microRNAs complementary to genes involved in early development[71] as well as an abundant and germline-specific class of small RNAs.[72] Small non-coding RNAs in the gametes may have a direct role in the differentiation and development of the early zygote or may play a part in post-fertilization epigenetic reprogramming; the mechanisms by which environmental factors could affect such processes remain speculative.

Evidence for non-genomic inheritance in humans

Human studies are much more limited but provide a number of lines of evidence suggesting transgenerational non-genomic inheritance, although it is inevitably difficult to define the relative contributions of genetic, epigenetic and common environmental or learned behavioural factors. For example, patterns of smoking, diet and exercise can affect risk across more than one generation by several mechanisms.[73] The strongest evidence for transgenerational non-genomic inheritance comes from dietary and endocrine exposure.

Historical records from Överkalix in northern Sweden for individuals born in the late nineteenth and early twentieth centuries have shown that diabetes mortality increased in men if the paternal grandfather was exposed to abundant nutrition during his prepubertal slow growth period.[2] A similar effect on overall mortality was later extended to paternal grandmother/granddaughter pairs and shown to be transmitted in a gender-specific fashion.[3] During the winter famine of 1944/45 in the Netherlands, previously adequately nourished women were subjected to low caloric intake and associated environmental stress. Pregnant women exposed to famine in late pregnancy gave birth to smaller babies[7] who later developed an increased risk of insulin resistance,[74] and F_2 offspring of females exposed in utero in the first trimester also had reduced birth size,[7] independent of any effect on F_1 maternal birth weight.

It is well documented that exposure of pregnant women to diethylstilbestrol, a synthetic estrogen previously used to prevent miscarriage, led to a marked increase in reproductive abnormalities and cancers in their children,[75] an effect mediated by transplacental passage of the drug. Evidence is now emerging for third-generational effects of diethylstilbestrol in both males and females, transmitted through the maternal line without further exposure of the intervening generation.[4,5]

At the mechanistic level, human studies are very limited. Differences in environmental exposure lead to different

patterns of epigenetic marking in the somatic tissues of individuals, as shown by recent studies on twins in which DNA methylation and histone acetylation patterns diverged more strongly in older twin pairs with more marked life history differences,[76] and there is some evidence for inheritance of tissue-specific DNA methylation patterns.[77] An epimutation in the gene for a DNA mismatch repair enzyme was found in both somatic and germline tissues of patients with multiple primary tumours, suggesting potential heritability.[78] Analysis of the paternally derived imprinting defect in some patients with Prader–Willi syndrome shows inheritance from the paternal grandmother, suggesting failure to erase the maternal imprint during spermatogenesis in the fathers of these patients.[6] Pembrey[79] has speculated that the transgenerational effects of food supply in the Swedish cohorts described above[2,3] are a result of environmental influences on reprogramming of imprinted genes during gametogenesis. The environmental sensitivity of epigenetic marking of metastable alleles associated with transposable elements[53–55] suggests a mechanism for such reprogramming; these elements are common in protein-coding genes of the human genome[80] and may have roles in developmental plasticity and in evolution of the genome.[81]

Non-epigenetic mechanisms of non-genomic inheritance

Other organic processes can cause aspects of the environment during development to affect the phenotype of subsequent generations, for example through changes in the maternal reproductive tract or in her adaptations to pregnancy or lactation, and such processes can help to explain intergenerational correlations in birth size[8] or propensity to develop type 2 diabetes.[9] In addition, so-called 'cultural' inheritance can operate when two or more generations share a common environment or societal conditions.[23] This can produce heritable disease risk—examples range from alcohol consumption or smoking to the funeral rituals of the Fore people which led to familial clusters of kuru.[82] Heritable characteristics are sometimes termed 'social' when they involve elements of learning and choice in each generation and 'biological' when other processes operate. Here we focus on the latter.

Altered uterine development and perfusion

Girls who are born small, presumably as a result of poor intrauterine nutrition, have reduced uterine volumes in late pre-puberty.[83] A reduced uterine size in adulthood will be reflected in reduced uterine vascular perfusion and can induce effects on F_2 offspring. The increase in uterine perfusion during pregnancy is associated with changes in vascular responsiveness, which are blunted in rats fed a low-protein diet and in their F_1 offspring.[84] Uterine vascular function can then be impaired in pregnant female F_1 offspring, affecting the development of F_2 progeny.[19]

Indirect effects mediated by maternal metabolism

There is considerable experimental and clinical evidence that an unbalanced diet or altered body composition before and during pregnancy produces altered metabolism in the offspring; unbalanced maternal nutrition, thinness or overweight and gestational diabetes are all associated with changes in metabolic control in the offspring, which then have a greater propensity to diabetes and/or obesity.[21,85–87] This may or may not be mediated by epigenetic change in the F_1 generation. But, irrespective of the mechanism leading to insulin resistance in the F_1 generation, when such individuals become pregnant, they are more likely to have gestational diabetes because pregnancy constitutes a state of moderate insulin resistance, produced by the action of placental somatogenic hormones.[88] Thus the F_2 generation is more likely to be exposed to hyperglycaemia in utero, which in turn predisposes them to a greater risk of metabolic compromise postnatally.[22]

Indirect effects mediated by maternal behaviour

Stress responses in adult rats are determined by the level of maternal attention that they receive during suckling, and these effects extend to the F_2 generation.[15] The behavioural changes are paralleled by changes in the pattern of DNA methylation of the glucocorticoid receptor $GR1_7$ promoter region in the hippocampus and can be mimicked by pharmacological manipulation of this epigenetic marking.[16] Although the mothering induces epigenetic change affecting behaviour, transmission to the next generation may be dependent on the behaviour in turn inducing epigenetic change rather than on direct epigenetic inheritance.

Adaptive value of non-genomic inheritance

The increasing evidence for non-genomic inheritance and particularly epigenetic inheritance raises the question of why the processes underpinning it (summarised in Table 2) have been preserved through evolution. Natural selection is viewed as the process by which a species and its environment become matched, whereas developmental plasticity utilizes environmental cues to fine-tune the individual phenotype to the current environment.[36] We have argued for a predictive component of developmental plasticity that would have been important for human evolution,[89] enhancing fitness during short-term environmental shifts and/or ensuring a greater match to a variable environment than selection alone can generate. Theoretical modelling suggests that such a strategy of 'phenotypic memory' would be advantageous to an extent governed by the fidelity of the transmission of environmental cues, the degree of predictability of environmental conditions and the costs of incorrect prediction.[90]

Table 2. Possible mechanisms involved in non-genomic transmission of risk of disease

Paternal transmission		Maternal transmission	
Epigenetic	**Non-epigenetic**	**Epigenetic**	**Non-epigenetic**
DNA methylation[a]	Behavioural and cultural effects	DNA methylation[a]	Altered growth of reproductive tract (e.g. small maternal birth size reflected in smaller uterine size at maturity)
Chromatin structure (e.g. specificity of histone-protamine replacement)[b]		Chromatin structure (e.g. specificity of histone modification such as acetylation or methylation)[b]	Impaired uteroplacental adaptations to pregnancy (e.g. vascular dysfunction)
Sperm RNAs		Oocyte RNAs	Maternal metabolic effects (e.g. fetal hyperglycaemia resulting from gestational diabetes causes insulin resistance)
		Mitochondrial defects?	Behavioural and cultural effects

During its life course, the F_0 generation will be subjected to environmental influences that cause accumulation of epigenetic change in somatic and germline tissues; somatic changes persist for only the life of the individual (and may be reversible) whereas germline changes may be transmitted to further generations. Additionally, the maternal environment during F_0 pregnancy could induce epigenetic changes in the developing primordial germ cells of the F_1 fetus that are transmissible to the F_2 generation. Finally, the maternal environment during F_0 pregnancy will affect the development of the somatic tissues of the F_1 fetus; any resulting changes in the adult phenotype of the F_1 generation may affect the phenotype of the F_2 generation by non-epigenetic mechanisms.

[a]Some evidence (e.g. in the *agouti* model[11]) that DNA methylation may not be the epigenetic mark transmitted.

[b]Unclear how specificity could be achieved, but small non-coding RNAs may be involved.

Such intergenerational non-genomic inheritance, by which parents transmit information about their current environment to their progeny (a 'carry over' strategy[90]) might enhance the value of prediction by reflecting longer term trends rather than short-term fluctuations.[91] Such adaptive versatility may have been important in the evolution of mammalian generalist species[92] and would have been of particular value in coping with climatic variability occurring on a multigenerational time scale.[93]

Developmental plasticity is a variable, selectable and heritable trait,[94] indicating that the capacity for plasticity and the mechanisms that underlie it are encoded in the genome and have been retained for their adaptive advantage. Similar considerations are also likely to apply to transgenerational inheritance of the plastic response. Thus, memory of developmental states set by many phylogenetically ancient signalling or regulatory processes may be transmissable to offspring. Developmental plasticity in response to early cues about the anticipated nutritional environment, and transgenerational inheritance of that information, are likely to be adaptive processes,[89,91] but may also be maladaptive if the anticipated and actual environments are mismatched, for example because of rapid nutritional transition (see next section). But there is another class of plastic response to the modern environment that is likely to be predominantly maladaptive; here, some novel environmental chemical mimics a physiological ligand (as do, for example, the so-called 'endocrine disruptors') and induces disruptive and toxic changes. If the appropriate developmental response to the physiological ligand has evolved to display phenotypic memory, the deleterious response to the toxin might also be transmitted to subsequent generations.[12,13]

Conclusion

Relevance to the risk of metabolic disease

The first adaptive explanation of the widespread increase in incidence of non-communicable diseases in contemporary populations was advanced by Neel, who suggested that 'thrifty' alleles were selected in Palaeolithic and Neolithic ancestral hominin environments of poor or uncertain nutrition.[95] Such 'thrifty' genes would promote Darwinian fitness by increasing tolerance to nutritional uncertainty through reduced skeletal muscle mass, a tendency to deposit visceral fat, reduced capillary density and insulin resistance.[96] Such traits would however promote disease in the present 'energy dense' (abundant nutrition and low energy expenditure) environment in which longevity has increased.[97] Neel suggested that such genes were selected through the periods of feast and famine that he postulated hunter–gatherers were exposed to, although more recent evidence would suggest this was not the case.[98] Although there appears to have been positive selection in human populations over the past

10,000 years for genes associated with nutrition and fat metabolism,[99] no plausible thrifty alleles have been found in the 40 years since Neel's proposal and the experimental and clinical data suggest that greater attention should be paid to non-genomic possibilities.[97,100]

We now live much longer than our hominin ancestors.[101] Thus, mechanisms selected for their advantage in our earlier evolution may no longer be advantageous in post-reproductive life. Elsewhere we have pointed out that there are limits to the environment that the fetus can sense and use to adjust its development.[102] Non-genomic processes of transmitting environmental information between generations evolved to assist our evolution as we moved across changing environments. Such processes were not designed to deal with the massive mismatch between the generally constrained fetal environment and the modern postnatal environment of high energy intake and low energy expenditure.[103] Indeed, disease risk is amplified by a greater mismatch between prenatally predicted environment and actual adult environment, and so societies in rapid economic transition are particularly vulnerable.[104,105] Epigenetic and other non-genomic inheritance processes may have conferred survival advantage on evolving hominins; they now exacerbate risk of disease for several successive generations. We have proposed that transgenerational non-genomic factors thus play a major part in the current epidemics of metabolic and cardiovascular disease.[106] Additionally, the possibility that exposure to xenobiotics such as endocrine disruptors may have multigenerational effects through similar mechanisms cannot be ignored. Elucidation of the underlying mechanisms offers hope that early prognostic markers such as epigenetic marks can be identified and that interventions can be designed to counter the effects of adverse non-genomically inherited traits.

Acknowledgments

We thank Dr Graham Burdge and Dr Karen Lillycrop for their comments.

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