

CHAPTER 8

Imprinted Genes and Human Disease: An Evolutionary Perspective

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Abstract

Imprinted genes have been associated with a wide range of diseases. Many of these diseases have symptoms that can be understood in the context of the evolutionary forces that favored imprinted expression at these loci. Modulation of perinatal growth and resource acquisition has played a central role in the evolution of imprinting and many of the diseases associated with imprinted genes involve some sort of growth or feeding disorder. In the first part of this chapter, we discuss the relationship between the evolution of imprinting and the clinical manifestations of imprinting-associated diseases. In the second half, we consider the variety of processes that can disrupt imprinted gene expression and function. We ask specifically if there is reason to believe that imprinted genes are particularly susceptible to deregulation—and whether a disruption of an imprinted gene is more likely to have deleterious consequences than a disruption of an unimprinted gene.

There is more to a gene than its DNA sequence. C. H. Waddington used the term “epigenetic” to describe biological differences between tissues that result from the process of development.^{1,2} Waddington needed a new term to describe this variation which was neither the result of genotypic differences between the cells nor well described as phenotypic variation. We now understand that heritable modifications of the DNA—such as cytosine methylation—and aspects of chromatin structure—including histone modifications—are the mechanisms underlying what Waddington called the “epigenotype.” Epigenetic modifications are established in particular cell lines during development and are responsible for the patterns of gene expression seen in different tissue types.

In contemporary usage, the term epigenetic refers to heritable changes in gene expression that are not coded in the DNA sequence itself.³ In recent years, much attention has been paid to a particular type of epigenetic variation: genomic imprinting. In the case of imprinting, the maternally and paternally inherited genes within a single cell have epigenetic differences that result in divergent patterns of gene expression.⁴ In the simplest scenario, only one of the two alleles at an imprinted locus is expressed. In other cases, an imprinted locus can include a variety of maternally expressed, paternally expressed and biallelically expressed transcripts.^{5–10} Some of these transcripts produce different proteins through alternate splicing, while others produce noncoding RNA transcripts.^{11–15} Genomic imprinting can also interact with the “epigenotype” in Waddington’s sense: many genes are imprinted in a tissue-specific manner, with monoallelic expression in some cell types and biallelic expression in others.^{16–20}

Other chapters in this volume cover our current understanding of the mechanisms of imprinting, the phenotypic effects of imprinted genes in mammals and what we know about imprinting in plants. In this chapter we discuss the link between imprinted genes and human disease. First, we consider the phenotypes associated with imprinted genes and ask whether the disorders as-

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sociated with these genes share a common motif. Second, we consider the nature and frequency of mutations of imprinted genes. We ask whether we should expect that imprinted genes are particularly fragile. That is, are they more likely to undergo mutation and/or are mutations of imprinted genes particularly likely to result in human disease? In general we consider how the field of evolutionary medicine—the use of evolution to understand why our body’s design allows for the existence of disease at all²¹—might contribute to our comprehension of disorders linked to genomic imprinting.

Do Disorders Linked to Imprinted Genes Share a Common Motif?

Many disorders linked to imprinted genes (see Table 1) are related to growth.²² The Kinship Theory of Imprinting^{23,24} explains why genetic loci that influence growth (and particularly the allocation of maternal resources) are prone to evolving imprinted gene expression. However, not all of these diseases are obviously growth related. In some cases, it might be possible to reconcile these disease phenotypes with the more general version of the Kinship Theory. In other cases, these disorders might be related to the mechanism of imprinting, rather than the gene function responsible for the evolution of imprinted expression.

According to the Kinship Theory, the pattern of expression shown by imprinted genes is a consequence of an evolutionary conflict between the maternally inherited (MI) and paternally inherited (PI) alleles at a locus. The theory relies on the notion of the inclusive fitness of an allele,²⁵ which includes not only the fitness of the individual carrying the allele, but also the fitnesses of other, related individuals who may have inherited an identical copy of that allele. That is, natural selection favors those alleles that maximize the number of copies passed on to future generations, regardless of whether those copies are passed on directly, or through the reproductive success of one’s kin. Which other individuals qualify as “relatives” can differ for the MI and PI alleles at a locus. In fact, in an outbred population, the only individuals to whom my MI and PI alleles are equally related are my direct descendants, my full siblings and their direct descendants.

Natural selection favors strategies that increase an allele’s inclusive fitness. When the gene affects the fitness of individuals to whom the MI and PI alleles have different degrees of relatedness, an allele’s optimal expression strategy will depend on its parental origin.²⁶ This can lead to silencing of the allele favoring the lower expression level and expression of the other allele at the level that maximizes its inclusive fitness.²⁷ For example, consider a locus at which an increase in level of expression (which we denote by X) enhances the fitness of the individual carrying the gene, but reduces the fitness of that individual’s matrilineal kin (relatives to whom one is related through one’s mother), henceforth referred to as resource enhancer. The level of expression that maximizes the inclusive fitness of the PI allele, \hat{X}_p , will be higher than that maximizing the inclusive fitness of the MI allele, \hat{X}_m . That is, $\hat{X}_p > \hat{X}_m$. Any intermediate level of expression $\hat{X}_p > X > \hat{X}_m$ results in conflict between the MI and PI alleles. If the locus becomes imprinted (acquires the ability to independently regulate the expression level of the MI and PI alleles) this conflict will result in the silencing of the MI allele. Expression of the PI allele will evolve to \hat{X}_p , the level that maximizes the patrilineal inclusive fitness. Analogous results apply to a locus where increasing the level of expression, Y , benefits matrilineal kin at the expense of the individual (a resource inhibitor). In this case, however, it is the PI allele that becomes silenced.^{28,29}

Most work on imprinted genes has focused on their effects on fetal growth. In this context, the relatedness asymmetries between the maternally and paternally derived alleles are well understood. A gene that enhances fetal growth places a resource demand on the mother, presumably reducing the availability of resources for her other offspring. The magnitude of this fetal demand will be limited by the fact that the MI alleles in the fetus have a fifty per cent chance of being inherited by any one of those other offspring and excessive demand could actually reduce the allele’s inclusive fitness (even while increasing the fitness of the individual offspring). Because the mother’s other offspring may have a different father, the PI alleles in the fetus care less than the maternally derived alleles about the consequences of increasing resource demand.

The taxonomic and functional distribution of imprinted genes suggest that conflicts over maternal resources have played an especially important role in the evolution of imprinting. Many

imprinted genes have been associated with prenatal growth effects.³⁰ Furthermore, mammalian imprinting appears first to have evolved in the common ancestor of marsupials and eutherian (placental) mammals, coinciding with the origination of viviparity.^{31,32} Viviparity and particularly the placental interface, provides an opportunity for the offspring to actively manipulate the availability of maternal resources. In oviparous (egg-laying) species, the mother has unilateral control over the distribution of resources among her offspring. While an intragenomic conflict might, in principle, exist within these offspring, there is no arena in which this conflict can play out.

Genomic imprinting in plants is not yet as well understood, but appears to follow a similar pattern: imprinting has evolved independently in angiosperms (flowering plants) where offspring (seed/fruit) develop in physical contact with the maternal parent. As in the case of mammals, the imprinted genes of angiosperms appear to modulate an offspring's access to maternal resources (see ref. 33 and the chapter by Spillane et al). When the tools of molecular genetics are applied to other plant groups, such as ferns and mosses, we might expect to find a similar set of phenomena.

There is obviously a strong correlation between prenatal growth effects and imprinting. However, similar reasoning applies to any trait where changes in gene expression affect the fitness of matrilineal and patrilineal kin differently.²⁶ In fact, many imprinted genes have effects on behavior that are difficult to interpret as straightforward extensions of parental conflict. Some of these behavioral effects include maternal care, reactivity to novel environments and social behaviors.^{5,34-38} Similarly, viviparity alone is not sufficient to drive the evolution of imprinting. Many viviparous species lack imprinting, including many species of fish.³⁹

Three features of mammalian pregnancy are likely responsible for its central role in the evolution of mammalian imprinting. First, there is a large asymmetry of parental resource contribution (maternal, but not paternal, pregnancy). Second, through the placental interface, the offspring plays an active role in soliciting maternal resources. Third, viviparity appears to have been maintained consistently in mammals since its introduction (in contrast to viviparity in other vertebrates, which is more evolutionarily labile).⁴⁰⁻⁴²

The existence of an inclusive-fitness asymmetry is not unique to mammalian pregnancy. In fact, there may be no single locus in any (biparental) organism for which the optimal expression pattern for the MI and PI alleles are exactly identical. The difference in mammalian pregnancy (and some plant reproductive systems) is a quantitative one. These systems have evolved many imprinted genes because the inclusive-fitness asymmetry is large. Furthermore, the systems are relatively easy to manipulate and have persisted in something like their present form for many millions of years.

Growth and Resource Acquisition

Our discussion of growth-related disorders in pregnancy follows that of Haig.⁴³ Many of these disorders likely involve the action of imprinted genes (see Table 1), but they should not necessarily be viewed as a consequence of imprinted gene expression. In addition to the conflict between the MI and PI alleles in the offspring, pregnancy is characterized by parent-offspring conflict.⁴⁴ The same sorts of inclusive-fitness considerations that underlie the evolutionary explanation for imprinting suggest that the fetus should favor a higher degree of resource demand than the mother. This reasoning applies even in the absence of imprinting. Of course, the set of genes most centrally involved in this conflict should significantly overlap with the set of genes most prone to evolving imprinted gene expression.

While the existence of growth-related disorders does not rely on imprinting, the existence of imprinting might be expected to exacerbate these disorders. In the absence of imprinting, the conflict will be between the maternal interests, on the one hand and the fetal interests (some average of the interests of the MI and PI alleles) on the other. When a growth enhancer becomes imprinted, the MI allele is transcriptionally silenced. At this locus, the parent-offspring conflict then shifts: on one side we still have the maternal interests; on the other, we now have the interests of the PI alleles, which favor a higher level of resource demand than do the fetal genes taken as a

Table 1. Diseases linked to imprinted genes

Disorder	Phenotype	Genes
Growth and resource acquisition		
Beckwith-Wiedemann Syndrome	Excessively large organs; Fetal and postnatal overgrowth; Low blood sugar in the newborn; Predisposition to tumors	<i>IGF2</i> (LoF K) <i>LIT1</i> <i>ARHI</i>
Growth Related Defects and Metabolic Abnormalities	Placental, pre and post-natal growth excess or deficiency	<i>ESX1L</i> (LoF K) <i>GNAS1</i> <i>H19</i> (LoI) <i>TSSC3</i> (LoF K)
Hyperinsulinism	High insulin levels in blood; Resistance to insulin resulting in its overproduction to compensate	
Pre-eclampsia	Elevated blood pressure and sometimes protein in the urine during pregnancy; Swelling of the face and hands	<i>STOX1</i> (LoF)
Pseudohypoparathyroidism 1A	Lack of response to parathyroid hormone; Low calcium and high phosphate levels in blood; Round face, short stature and hand bones	<i>GNAS1</i> (LoF K)
Silver-Russell Syndrome	Pre and post natal growth retardation; Predisposition to developmental and motor delays as well as learning disabilities; Body asymmetry	<i>SGCE</i> (LoF)
Transient Neonatal Diabetes	Inability to use blood glucose for energy resulting in hyperglycemia	<i>PLAGL1</i> <i>LOT1</i> , <i>HYMA1</i>
Post-natal behaviour		
Angelman Syndrome	Feeding problems; Noticeable developmental delays; Pronounced speech impairment; Hyperactivity; Severe movement and balance disorders; Bouts of laughter	<i>UBE3A</i> (LoF K) <i>ATP10C</i>
Reduced Maternal Nursing	Reduced post-natal maternal care. Growth related defects	<i>PEG3</i> (LoF K)

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Table 1. Continued

Disorder	Phenotype	Genes
Prader-Willi Syndrome	Obesity; short stature; decreased muscle tone; hypogonadism; decrease mental capacity	<p>NDN SNRPN PWCR1 (LoF D) IPW MAGEL2 MKRN3 (LoF K)</p>
Cancer		
Adrenal Cortical Carcinoma		H19 (LoI)
Breast Cancer		<p>PEG1 (LoI) PLAGL1</p>
Hepatoblastoma		H19 (LoI)
Hydatidiform Mole	Uncontrolled growth of the tissue that is supposed to develop into the placenta; Often, there is no fetus at all	
Hyperplasia	Increased cell production in normal tissue or an organ	IGF2 (ΔE)
Nonfunctioning Pituitary Adenomas	Pituitary tumor that does not result in the enhanced production of pituitary hormones	PLAGL1
Retinoblastoma	Retina cancer occurring in children	H19 (LoI)
Wilms' Tumor	Kidney cancer occurring in children	CDKN1C

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Table 1. Continued

Disorder	Phenotype	Genes
Other		
Autism	Impaired social interactions; Impaired verbal and nonverbal communication; restricted and repetitive patterns of behavior	<i>ATP10C</i>
Bipolar Affective Disorder	Mood swings from mania (exaggerated feeling of well-being, stimulation and grandiosity) to depression (overwhelming feelings of sadness, anxiety and low self-worth)	
McCune-Albright Syndrome	Premature puberty mainly in girls; abnormal fibrous development in the bone; Café-au-lait spots on the skin	<i>GNAS1</i>
Myoclonus Dystonia Syndrome	Obsessive compulsive disorder; panic attacks	<i>SCCE</i> (LoF)
Schizophrenia	Severe problems with thoughts, feelings, behavior, and use of language; delusions often paranoid and persecutory in nature; hallucinations	
Williams-Beuren Syndrome	Mild mental retardation; problems with calcium balance; blood vessels defects	

We indicate the chromosome or chromosomal region involved, whether it is a maternal or paternal UPD and a sketch of the clinical phenotype in each case. (Sources: Imprinted Gene Catalogue, MedlinePlus; Online Mendelian Inheritance in Man).

whole. One of the consequences of imprinting may be an intensification of the pre-existing conflict between mother and fetus.

One of the phenotypes associated with the fetal manipulation of maternal resources is placental invasion. The placenta comprises a fetal portion derived from trophoblasts and a maternal portion derived from the inner layer of the uterine wall. Placental trophoblasts modify maternal arteries to allow greater blood flow through the intervillous space. The greater the penetration of arterial modification into the myometrium, the greater the blood flow and maternal resource transfer to offspring.⁴³ The higher-than-normal concentration of Insulin-like growth factor type 2 (IGF2)—encoded by the paternally expressed *IGF2* gene—in invasive trophoblasts suggests that IGF2 may influence the extent of placental invasion.⁴³

Genetic conflict has also been related to deregulation of maternal blood pressure.⁴³ The higher the maternal blood pressure the greater the blood flow through the intervillous space and the transfer of resources to the offspring. Paternally inherited genes would favor greater gestational hypertension than their maternally inherited homologs. One of the most common complications of pregnancy (fatal in developing countries) is pregnancy-induced hypertension and its extreme form pre-eclampsia. Pre-eclampsia can be caused by mutations at one of several loci, at least one of which is known to be imprinted—the maternally expressed *STOX1* gene.⁴⁵⁻⁴⁷ While the exact role of *STOX1* remains unclear, the Kinship Theory would predict that increased expression of *STOX1* would reduce maternal blood pressure.

Increasing the flow of blood to the placenta is one mechanism of fetal resource acquisition. A second is to increase the concentration of nutrients in the maternal circulation. After each meal, maternal insulin prompts the uptake of glucose by maternal cells. During pregnancy, the placenta antagonizes the action of insulin by secreting human placental lactogen (hPL) into the mother's system. This placental hPL generates resistance to insulin in the maternal cells, thereby elevating the level of glucose in the maternal circulation. This manipulation may be the cause of gestational diabetes, which occurs late in pregnancy, but generally resolves quickly following delivery. Imprinting of a locus involved in the placental regulation of hPL could exacerbate this effect and potentially increase the frequency or severity of disorders such as gestational diabetes.

Post-Natal Behavior

After birth, mammals continue to rely heavily on maternal resources (breast milk and supplemental food), although they are no longer transmitted by means of the placenta. The conflict between mother and offspring—and between the PI and MI alleles in the offspring—then shifts primarily into the behavioral arena. Genes expressed in the brain will be under selection to maximize their inclusive fitness, just as they were during pregnancy. However, in this behavioral context, it is often much more difficult to understand the nature of the inclusive-fitness asymmetries that underlie imprinting. Two other chapters in this volume (by Goos and Ragsdale and by Davies et al) focus specifically on behavioral effects associated with imprinted genes and we will discuss the topic only briefly here.

Some of the postnatal behavioral effects of imprinted genes are easily interpreted as the natural extension of the prenatal conflict over maternal resources, such as those affecting suckling and weaning behaviors.⁴³ In this context, the MI alleles would be expected to favor weaning at an earlier age than the PI alleles. Similarly, PI alleles are expected to more strongly favor behaviors that elicit maternal care. This reasoning has been invoked to explain at least some aspects of the phenotype of two disorders associated with different parental inheritance of deletions or mutations on the long arm of chromosome 15: paternally inherited Prader-Willi syndrome (PWS) and maternally inherited Angelman syndrome (AS).⁴⁸

Each of these disorders exhibits a complex phenotype. AS is associated with enhanced activity, prolonged but poorly coordinated suckling, bouts of laughter, sleeping problems and developmental disorders (speech impairment, movement and balance disorders). Prior to weaning, PWS is associated with reduced activity, poor suckling, weak cry, sleepiness and decreased mental capacity. Following weaning, the child develops an insatiable appetite and becomes obese.⁴⁸ Viewed in the light of the

Kinship Theory, the increase in the duration of suckling found in AS may result from the loss of MI alleles that have been selected to reduce the demand for maternal resources. Conversely, PWS is associated with poor food uptake prior to weaning and ravenous food uptake after. The pre-weaning phenotype may result from the loss of PI alleles that been selected to increase the demand of maternal resources. The post-weaning phenotype is more difficult to explain but still is consistent with the Kinship Theory if (a) the offspring's voracious appetite is satisfied primarily by its own foraging efforts and translates in a reduced consumption of breast milk,⁴⁸ and (b) the paternal contribution to resource provisioning increases after weaning (F. Úbeda, manuscript in preparation).

Two imprinted genes, *MEST/Peg1* and *Peg3*, show paternal-specific expression in the brains of adult mice. These genes appear to affect the quality of care that mothers provide to their offspring, as knockouts of these genes result in defects in maternal behaviors of nest building, pup retrieval and placentophagy.^{37,38} Although the phenotype involves the provisioning of maternal resources to offspring, the conflict in this case is between the mother's two alleles, rather than those of the offspring. The source of this conflict is not obvious, however, since each of the mother's alleles has an equal chance of being passed to each of her offspring. However, if there is some inbreeding in the population (the mother mates with a related male), the offspring could inherit an allele from the father that is identical to one of the mother's alleles.

Under several plausible patterns of inbreeding, the allele inherited from the father is more likely to be identical to the mother's paternally derived allele than to her maternally derived allele. For example, if the mother mates with her own father (i.e., the father and maternal grandfather are the same individual), the mother's paternally derived allele will be more closely related to her own offspring than her maternally derived allele will. If the pattern of inbreeding changes over the course of the female's life, an intra-genomic conflict will arise over the distribution of maternal resources to present and future litters.⁴⁹ The conditions under which this selective force might exist are fairly general, but a test of whether this is the factor that is actually responsible for imprinting of these "maternal-care" loci will require close study of multiple species with different patterns of inbreeding.

Cancer

There is mounting evidence that somatic mutations at imprinted loci are associated with a variety of cancers.^{50,51} The silencing of one of the alleles turns the imprinted locus functionally haploid. It has been argued that the functional haploidy might increase the risk of cancer by exposing the phenotypic consequences of deleterious recessive mutations. As we discuss in the following section, there are good reasons to believe that deleterious mutations at an imprinted locus are less likely to be recessive than deleterious mutations at other loci (see also the chapter by Moore and Mills).

While we doubt that functional haploidy is the reason for the association of imprinted genes with cancer, there are other features of imprinting that may be relevant. Many genes have evolved imprinting due to a role in modulating fetal growth.^{30,52} It is not surprising that many of these same genes influence mitogenic activities in adult somatic tissues. In this context, we suspect that it is not the fact of imprinting that makes these genes associated with tumor growth. Likewise, we doubt that the tumor-suppressing activity of imprinted genes is directly responsible for the evolution of imprinting (but see ref. 53).

Rather, there are a set of genes that affect growth and cell division; this set of genes is more likely to become subject to imprinting and is more likely to be associated with tumorigenic mutations. Imprinting also allows the evolution of antagonistic growth suppressors. Once these exist, they may take on a tumor suppressor role, essentially reducing the selection on other tumor suppressor mechanisms. Here, functional haploidy may be relevant, but not for the obvious reason. In this case, imprinting may replace a biallelically expressed tumor suppressor with a monoallelically expressed tumor suppressor at a different locus. The result may be a system that is less robust to somatic mutation.

Are Imprinted Genes Particularly Fragile?

There are two reasons why an imprinted gene might be either more likely to express a mutant (sick) phenotype or more susceptible to mutations. First, as mentioned above, imprinted genes

are functionally haploid. While a recessive mutation has no phenotypic consequences on unimprinted genes, it is exposed on imprinted genes. Second, the expression of imprinted genes, being conditioned by epigenetic factors, is susceptible not only to mutations but also to epimutations. Interestingly, epimutations can be influenced by the environment and do not need to be transient; they might revert after one generation, after a few generations or otherwise become permanent. This opens up a wide range of mutational possibilities.

Mutations

We will distinguish “mutation” (a change in the DNA sequence) from “epimutation” (a heritable change not coded in the DNA sequence). At an unimprinted autosomal locus, loss-of-function mutations are often recessive—a single functional copy of the gene is sufficient to maintain an approximately normal phenotype. At an imprinted locus, one of the two copies is transcriptionally silent and the loss-of-function phenotype depends not on dominance, but on parental origin. A mutation on the silenced allele will have no phenotypic effect. A loss-of-function mutation on the active copy will be equivalent to a homozygous knockout in the absence of imprinting.

Consequently, deleterious recessive mutations, which would have no phenotypic consequences when heterozygous at an unimprinted locus, may have severe phenotypic and fitness effects at an imprinted one. More specifically, the mutant phenotype will be fully revealed half of the time (see Fig. 1). If we assume that most deleterious mutations are recessive, this suggests the functional haploidy associated with genomic imprinting introduces fragility, by increasing the phenotypic and fitness effects of deleterious mutations.

However, there is reason to question the assumption that deleterious mutations of imprinted genes would predominantly be recessive (although this may be true for the genome as a whole). Consider a loss-of-function mutation at an unimprinted locus. This results in a fifty percent reduction in the expression level of the gene. If this deleterious mutation is recessive—the phenotype of a heterozygous carrier of this allele is identical to that of the wild-type—we can infer one of two things. Either there is little or no phenotypic consequence to a fifty percent reduction in gene expression, or there are regulatory feedback mechanisms in place that increase expression from the wild-type allele to compensate for this reduction.

In either case, we should not expect this locus to become imprinted. If a two-fold reduction in expression has no phenotypic consequence, there is little opportunity for an allele to gain an inclusive fitness benefit through changes in the expression level. Similarly, if appropriate feedback mechanisms exist, there will be no selective benefit to silencing the allele favoring lower expression, since the other allele will maintain the overall expression from the locus at a constant level. In fact, the loci where imprinted gene expression will most easily evolve are those loci for which some phenotype is very sensitive to changes in gene expression and where the expression level for each allele is set independently of the total level of gene product. The lack of feedback mechanism might bias imprinted genes towards those whose gene products are exported from the cell (such as growth factors and hormones), making intracellular feedback difficult. The sensitivity to dosage changes implies that deleterious mutations at these loci will not be recessive.

Therefore, even if most deleterious mutations in the genome are recessive, imprinted loci might be more fragile than unimprinted ones not because of their functional haploidy but because of intrinsic properties of the genes that are likely to evolve imprinting, namely, deleterious mutations tend not to be recessive more often than mutations at other loci. Within the group of ancestral genes where imprinting evolved, the functional haploidy of imprinting might actually provide a fitness advantage, by reducing the number of sick phenotypes by one half.

Genomic imprinting is caused by conflict between the two alleles at a single locus, but the outcome of this conflict—silencing of one of the two alleles—creates the potential for conflict among distinct loci.^{28,29} Genes with antagonistic effects can become oppositely imprinted and then engage in an arms race characterized by increased expression from both loci. After this escalation occurs, a mutation in either one of the loci can produce a large phenotypic effect. For example, consider an imprinted locus, paternally expressed, with level of expression X and an antagonistic imprinted locus, maternally expressed, with level of expression Y . Assume the difference $X-Y$ determines the

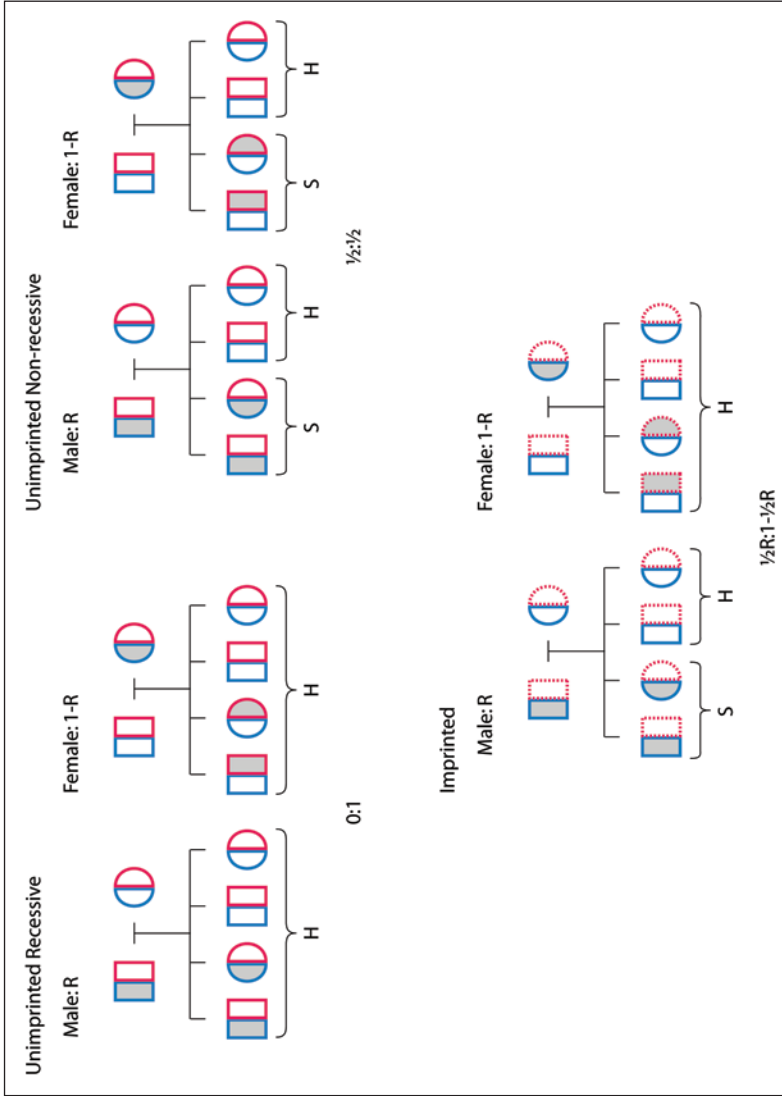


Figure 1. Expected proportion of descendants manifesting a mutant phenotype. We consider a loss-of-function mutation (in grey) in a population with a fraction R of males and $1-R$ of females. In each scenario we consider that the mutation arise in males (squares) and females (circles). The clinical phenotype, healthy (H) or sick (S), is indicated underneath together with its fraction in the population. The scenarios considered are recessive and non-recessive mutations at an unimprinted locus and mutation at an imprinted locus.

blood levels of certain hormone α . Suppose that prior to the acquisition of imprinting at either locus, the expression levels were $X = 2$ and $Y = 1$, resulting in a circulating hormone level of $\alpha = 1$. Now assume that these two loci evolve antagonistically, resulting in increased expression from each locus, say, $X = 20$ and $Y = 19$. In this example the normal phenotype is unchanged, since we still have $\alpha = 1$. However, the consequences of a loss-of-function mutation at either locus has been dramatically enhanced. In the original case, deletion of the maternally expressed gene resulted in an increase of α from 1 to 2. After the antagonistic coevolution, deletion of the same gene would increase α from 1 to 20.

Epimutations

Imprinted gene expression is associated with differential epigenetic modifications on each of the chromosomes. These modifications include DNA methylation (on cytosines in CpG dinucleotides), as well as histone modifications (including methylation and acetylation). These modifications are established during gametogenesis and are often remodeled following fertilization. Molecular mechanisms exist that reproduce these modifications in the wake of DNA replication.⁵⁴ Further epigenetic reprogramming can occur in particular cell lineages during development, resulting in tissue-specific patterns of imprinting. In many cases imprinted gene expression involves the transcription of noncoding RNA transcripts that suppress the production of other transcripts *in cis*.^{3,22}

Any of these epigenetic systems is potentially susceptible to failure, resulting in phenotypes that bear a resemblance to genetic disorders, but are not associated with mutations in the DNA sequence. In principle, this sort of failure could occur at any point in the life cycle, including reprogramming errors in gametogenesis or early development, or maintenance errors in particular somatic cell lineages.

DNA methylation patterns are reproduced following DNA replication through the action of a particular DNA methyltransferase (DNMT1). In principle, this provides a passive mechanism by which an epigenetic state, once established, can be maintained across multiple cell divisions. Similarly, it appears that there are passive mechanisms to propagate aspects of chromatin structure, including patterns of histone modification. There is some evidence to suggest that these two systems interact to stabilize epigenetic marks against stochastic loss.⁵⁴

Failure to propagate these marks can result in silencing or reactivation of a particular allele. Of course, susceptibility to somatic epimutations is not limited to imprinted genes. However, as in the case of DNA sequence mutations, the functional haploidy of imprinted loci may make these epimutations particularly detrimental. For instance, inappropriate silencing of the active allele at an imprinted locus will be equivalent to a homozygous loss-of-function mutation in the DNA sequence.

We can distinguish between epimutations that are meiotically heritable and those that are not.⁵⁵ Meiotically heritable epimutations occur in the germline and are passed on to the offspring. In some cases (e.g., an error in germline reprogramming at an imprinted locus), we expect the epimutation to persist for a single generation. In other cases, these germline epimutations might be more stable, persisting for several generations, or even being assimilated into the genotype.⁵⁶⁻⁵⁸

There is speculation that some of these meiotically heritable mutations may actually respond adaptively to environmental conditions (e.g., nutrients and environmental contaminants), creating a form of trans-generational phenotypic plasticity.⁵⁵⁻⁵⁸ Dietary conditions at critical ontogenic stages may result in a shortage or an excess of methyl donors.^{56,57,59} As a consequence, particular DNA and/or histone methylations might be lost at particular loci, possibly altering their expression pattern. While this mechanism might provide an adaptive response to certain nutrient deficiencies during early development, there may be maladaptive consequences later in life—for instance, as the result of a change in environmental conditions or due to pleiotropic effects of the deregulated imprinted genes.

One candidate example of an environmentally driven epimutation with trans-generational consequences is the so called “metabolic syndrome.”⁵⁹ Mothers who experience nutritional constraints during pregnancy often have descendants who suffer from glucose and insulin metabolism disorders, weight problems, hypertension, diabetes and cardiovascular diseases.⁵⁹⁻⁶¹ Interestingly,

these health problems are not limited to the mother's children, but rather can persist in future generations of descendants.⁵⁹ The "Fetal Programming Hypothesis of Adult Disease"²² proposes that dysregulation of imprinted genes may play a role in the clinical phenotype of patients experiencing the metabolic syndrome.⁵⁹

"Metabolic syndrome" is one of many disorders thought to result from epimutations. It is possible that this represents an adaptive response to a nutritional deficiency early in development and that it develops into a disease only when that nutritional constraint is removed. This type of epigenetic response might allow greater adaptability to environmental changes. Epimutations occur frequently (10^{-2} in Tobacco plants⁵⁸) and have a duration of effect that may lend them to certain types of environmental variation. However, adaptive epimutation also has limitations. In particular, the developmental window during which the organism can assimilate environmental cues may be narrow, but the response to those cues may be long lasting. This may lead to maladaptive responses, particularly in the context of contemporary human cultures.

Uniparental Disomies

Genomic imprinting gives rise to the possibility of another type of hereditary defect, uniparental disomies (UPDs). Many animals are sensitive to gene dosage effects and changes in chromosome copy number (e.g., monosomies and trisomies) can often have deleterious effects. For chromosomes containing one or more imprinted loci, parental origin can be as significant as copy number. For instance, an individual who inherits two paternally derived copies of a particular chromosome will have normal gene function at unimprinted loci. However, at a paternally silenced imprinted locus, the individual will functionally be a homozygous knockout. Similarly, each maternally silenced imprinted locus on the chromosome will be expressed at twice its normal level. Most imprinted genes appear to occur in clusters, so that a UPD will typically affect multiple imprinted genes. Not surprisingly, most UPDs are associated with growth abnormalities (see Table 2 and ref. 62). However, given the magnitude of the developmental perturbation typically associated with this type of chromosomal abnormality, it can be difficult to interpret the resulting phenotypes in an evolutionary context.⁶³⁻⁶⁵

The consequences of imprinting can also be seen in certain trisomies. The deleterious effects of trisomy are not fully attributable to imprinting, but the trisomic phenotype can vary systematically depending on parental origin. For instance, whole-genome triploidy can result in partial hydatidiform moles. While these moles do not go on to form viable offspring, they do undergo partial development and tissue differentiation. Triploid zygotes with an extra paternal genome produce large placentas and small heads. Conversely, zygotes with an extra maternal genome produce small placentas and large heads.⁴³

Implications for the Prevention and Treatment of Human Disease

The extension of evolutionary medicine to encompass epigenetic phenomena may prove valuable in the analysis, prevention and treatment of diseases associated with deregulation of imprinted genes. Perhaps the most general insight provided by the evolutionary analysis of imprinted genes is that natural selection does not necessarily act to optimize the fitness (or the health) of an individual organism. Genomic imprinting represents a case where selection to increase inclusive fitness can actually work to the detriment of the individual.⁶⁶

More specifically, the establishment, propagation and interpretation of the epigenetic marks at imprinted loci involve a complex set of mechanisms. Failure of any one of these mechanisms can result in a disease phenotype. In this sense, imprinted genes may represent particularly large mutational targets. Additionally, the escalatory conflicts to which imprinted genes are prone may generate conditions in which mutations (or epimutations) are particularly deleterious.

The reliance of these epigenetic mechanisms on chemical modifications (such as methylation) generates specific nutritional requirements. A deficiency in these or other nutrients can trigger epigenetic reprogramming of particular loci. In some cases, the reprogrammed marks may persist

Table 2. Uniparental disomies

Chromosome	UPD	Phenotype
5 U2AF1RS1	Maternal	Growth retardation
	Paternal	Growth enhancement
6	Maternal	Embryonic lethality; intra-uterine growth retardation
	Paternal	Transient neonatal diabetes mellitus. Growth retardation
7	Maternal	Silver-Russell syndrome (severe intrauterine growth restriction)
7 Grb10	Maternal	Growth retardation
	Paternal	Growth enhancement
11	Paternal	Beckwith-Wiedemann syndrome (fetal and postnatal overgrowth and low blood sugar in the newborn)
14	Maternal	Intra-uterine growth retardation; hypotonia, motor delay and precocious puberty
	Paternal	Growth retardation
15	Maternal	Prader-Willi syndrome (obesity, short stature, decreased muscle tone)
	Paternal	Angelman syndrome (feeding problems, noticeable developmental delays, hyperactivity)
16	Maternal	Intra-uterine growth retardation

We consider four categories: (a) disorders related to growth and resource acquisition (b) disorders related to post-natal behaviour (c) cancers and (d) other disorders. In each case we indicate the disorder, a sketch of the clinical phenotype, the imprinted genes involved (paternally expressed genes in first column and maternally expressed genes in second column). (Sources: Imprinted Gene Catalogue, MedlinePlus; Online Mendelian Inheritance in Man).

across one or more generations. While these changes could simply be a passive byproduct of certain nutrient deficiencies, it is also possible that they represent an adaptive response to environmental cues that are presented in early development. This insight may have implications for the treatment of nutrient deficiencies. In cases like these, simply supplying the missing nutrient at a later developmental stage may create a new set of disease conditions. Nutritional supplementation may have to be coupled with restoration of the original epigenetic state of the modified genes.

Diseases caused by mutations or epimutations at imprinted loci make intriguing candidates for gene therapy. In particular, clinically useable tools for activating or inactivating alleles could provide treatment for many of the disorders mentioned in this chapter. A loss-of-function mutation of the active allele at an imprinted locus might be treated by reactivation of the silent copy. A loss-of-imprinting mutation (inappropriate reactivation of the silenced copy) could be treated through downregulation of the locus as a whole.³ However, given the complex patterns of regulation and expression at many imprinted loci, this approach may prove technically challenging, is not without potential dangers. Unintended consequences such as a predisposition for tumor formation, will be a danger of any therapy that attempts to quantitatively modify the expression level of imprinted genes.³ Furthermore, while an intervention of this sort might be beneficial for the patient, the possibility exists that induced epigenetic changes could be passed on to offspring.

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