Placenta (2004), **25**, Supplement A, Trophoblast Research, Vol. 18, S10–S15
doi:10.1016/j.placenta.2004.01.006

Evolutionary Conflicts in Pregnancy and Calcium Metabolism—A Review

D. Haig*

Department of Organismic and Evolutionary Biology, Harvard University, Cambridge, MA 02138, USA

Paper accepted 2 January 2004

The maternal-fetal unit contains three distinct haplotypes at each locus: the maternally derived fetal haplotype (MDFH) that is shared by the mother and fetus, the paternally derived fetal haplotype (PDFH), and the non-inherited maternal haplotype (NIMH). The evolutionary forces acting on these haplotypes are distinct. The NIMH is absent from the offspring and could benefit from early abortion if this enhances the probability of the mother conceiving again and producing an offspring that inherits the NIMH. This raises the possibility that some forms of recurrent spontaneous abortion may be caused by non-inherited haplotypes. Such 'selfish' behaviour would be opposed by other components of the maternal genome. Natural selection acting on genes expressed in fetuses (or their placentae) favours greater maternal investment in the fetus than does natural selection acting on genes expressed in mothers. Furthermore, in the presence of genomic imprinting, the PDFH favours greater levels of investment in the fetus than does the MDFH. These conflicts are illustrated using the example of maternal-fetal conflicts over the supply of calcium. Inactivation of the paternal copy of *GNAS* in proximal renal tubule is interpreted as a measure to maintain fetal bone mineralization in times of calcium stress at the expense of the maternal skeleton.
Placenta (2004), **25**, Supplement A, Trophoblast Research, Vol. 18, S10–S15

GENETIC FACTIONS IN THE MATERNAL-FETAL UNIT

Pregnancy is a joint venture involving three sets of genes. These are genes shared by the mother and fetus (inherited maternal haplotypes); genes present in the mother but not in the fetus (non-inherited maternal haplotypes); and genes present in the fetus but not in the mother (paternally derived fetal haplotypes). Each set has distinct interests in the outcome of a pregnancy because of a fundamental evolutionary trade-off that exists in the provision of maternal care: extra resources or effort put into enhancing the survival of any particular offspring is associated with an opportunity cost of less resources or effort available for other maternal activities. In evolutionary terms, the opportunity cost of extra care for a particular offspring has translated into a cost to a mothers' expected fitness from other offspring. The existence of this trade-off does not require the action of obscure processes. In fact, its expressions may be rather prosaic. When a human mother holds a baby, she is less able to grab a toddler who runs across a road. When a squirrel mother depletes her fat reserves during lactation, she is more likely to succumb to disease during the coming winter. Nor does the existence of an evolutionary trade-off require that everytime a mother does something to enhance the fitness of one offspring, another one suffers. All that is required is that this relation has existed on average.

* To whom correspondence should be addressed. Fax: +1-617-495-5667; E-mail: dhaig@oeb.harvard.edu

0143-4004/$–see front matter

Evolutionary conflicts arise during pregnancy because the reallocation of maternal investment from the current offspring to other offspring has had different fitness consequences for the three sets of genes in the maternal-fetal unit. How then does natural selection weigh benefits to one of a mother's offspring against costs to her other offspring? I will first consider genes expressed in mothers and then consider genes expressed in offspring (genes on inherited maternal haplotypes can belong to either category).

Genes expressed in mothers

A critical factor in determining the selective forces acting on an allele expressed in mothers is whether or not the allele's effects on offspring depend on whether the offspring also possesses a copy of the allele. If an allele has different effects when an offspring has and has not inherited a copy of the allele, then the allele can be figuratively said to 'recognize' itself in offspring or to have 'information' about its own pattern of inheritance. Haig [1,2] has discussed simple ways in which genes in mothers could acquire such information. These mechanisms involve epistatic interactions between closely linked loci, where one of the loci is expressed in mothers and the other is expressed in offspring. By analogy to systems of meiotic drive, I will refer to these loci as the *Distorter* locus (expressed in mothers) and the *Responder* locus (expressed in offspring). Epistasis ensures that the effects of an allele at the *Distorter*

locus will depend on the offspring's genotype at the *Responder* locus. Therefore, if *Responder* and *Distorter* loci are closely linked and a mother is heterozygous at both loci, then the effects of an allele at the *Distorter* locus will differ depending on whether or not the offspring inherits the allele. Natural selection will favour two-locus haplotypes that benefit offspring that inherit the haplotype, even if this is at the expense of siblings that do not. Haig [1,3] used the term 'gestational drive' to refer to such systems in which maternal care is preferentially directed to offspring who inherit one of the alleles at a heterozygous locus.

Non-inherited maternal haplotypes (NIMHs) gain no direct benefit from the survival, health and eventual reproduction of an embryo in which they are absent. Worse than that, NIMHs have an interest in the embryo's early demise because this frees maternal energies for the care of existing offspring or speeds the conception of future offspring that may inherit the NIMH of the current pregnancy. The potential selective advantage to a gene of eliminating embryos that do not inherit its copies is so great that I would be surprised if systems of spiteful abortion have not evolved. However, I know of no unambiguous examples in mammals, and raise the possibility here as something to keep in mind when a common genetic variant appears to be associated with inexplicably high reproductive costs. Recurrent spontaneous abortion would be one place to look for such systems. In such cases, the genetic culprit may have been interpreted as 'protective' because the haplotype that is responsible for recurrent abortion is present in the surviving offspring of affected mothers, but not the aborted offspring. Geneticists should be aware of the danger of blaming the victim. (Pregnancy losses so early that they fail to be detected provide an even stronger selective advantage, but are unlikely to come to clinical attention).

If non-inherited alleles at every heterozygous locus in a mother's genome were actively conspiring against each and every embryo, it is hard to see how a successful pregnancy would ever be possible. Embryos are probably saved by a lack of reliable information available to genes expressed in their mothers (i.e., by the absence of the necessary forms of nepotistic epistasis). In this view, most maternal genes lack information about whether they are inherited by an offspring and so are unable to discriminate against their non-inheritors. A maternal gene that makes investment decisions behind this meiotic 'veil of ignorance' has an equal likelihood of being present in each of a mother's offspring [4]. Therefore, such genes will be selected to maximize the mother's total number of surviving offspring and to oppose the nepotistically epistatic conspiracies of the minority of well-informed genes.

Inherited maternal haplotypes (IMHs) can also conspire against the rest of the maternal genome, but here the conspiracy is one of favouritism rather than malice, of attempting to give the current fetus a little extra advantage (relative to its sibs) in the struggle for life [1]. Once again, the principal barrier to the evolution of such systems is the absence of reliable information about whether or not an offspring has inherited a particular allele.

In summary, most genes in mothers probably do not discriminate among offspring and are selected to maximize a mother's lifetime reproductive success. These genes will be described as residing on non-discriminating maternal haplotypes (NDMHs). However, a subset of genes (on discriminating NIMHs) may have been selected to cause the demise of their non-inheritors, and a different subset (on discriminating IMHs) may have been selected to direct preferential care to their inheritors.

Genes expressed in offspring

The question of whether an offspring has inherited a copy of a gene does not arise for genes expressed in offspring. Therefore, such genes will tend to favour maternal investment in their particular offspring (a sure bet) at the expense of maternal investment in sibs (a risky bet). By contrast, there are no sure bets for non-discriminating genes expressed in mothers: each offspring has one chance in two of inheriting a particular maternal allele. This asymmetry of information is the basis of the theory of parent–offspring conflict [5]. As a result of the additional information that genes in offspring possess about one toss of the meiotic coin, they will have been selected to demand more resources from mothers than mothers will have been selected to provide.

Questions of information do arise for genes expressed in offspring but, in this case, the question is not whether a gene sits on an NIMH or an IMH but whether the gene sits on a maternally derived fetal haplotype (MDFH) or a paternally derived fetal haplotype (PDFH). Genes on an MDFH have one chance in two of being present in each of a mother's other offspring, whereas genes on a PDFH have less than one chance in two because of the possibility of multiple paternity. Therefore, if genes were informed about their parental origin, genes on PDFHs would be selected to discount opportunity costs to mothers more heavily than would genes on MDFHs [6,7]. Information about a gene's parental origin is provided by epigenetic modifications (genomic imprinting) established in parental germ lines. Genes that are expressed in offspring, but that are uninformed about their parental origin, would be selected to discount opportunity costs at a rate intermediate between the rates favoured by MDFHs and PDFHs. Such uninformed genes will be described as residing on non-imprinted fetal haplotypes (NIFHs).

This section and the previous section have identified five sets of genetic interests in the maternal-fetal unit (Table 1). These are discriminating NIMHs, NDMHs, discriminating IMHs (equivalent to imprinted MDFHs), NIFHs, and imprinted PDFHs (arranged in order from lowest to highest interest in the current fetus relative to other maternal offspring; [6].

BONES OF CONTENTION

As thou knowest not what is the way of the spirit, nor how the bones do grow in the womb of her that is with child: even so thou knowest not the works of God who makest all.

Ecclesiastes 11:5, King James Version

Table 1. The probability that a particular haplotype is present in the current offspring (P_1) or another offspring (P_2) of the same mother (probabilities calculated for identity by immediate common descent; p is the probability that other offspring have the same father as the current offspring). P_1/P_2 is a measure of a haplotype's degree of 'preference' for maternal investment in the current offspring rather than other offspring

Haplotype	P_1	P_2	P_1/P_2
Non-inherited maternal	0	1/2	0
Non-discriminating maternal	1/2	1/2	1
Inherited maternal	1	1/2	2
Non-imprinted fetal	1	$(1+p)/4$	$4/(1+p)$
Paternally derived fetal	1	$p/2$	$2/p$

The discussion, so far, has been in the abstract. Theories of gestational conflict will be of little use unless they can be used to illuminate the organization of physiological processes during pregnancy. Haig [3,4] has described various aspects of human pregnancy when viewed under this light. One system I have not previously considered is that of potential conflicts over the allocation of calcium during pregnancy. This section will present such a discussion as a prelude to a highly speculative hypothesis to explain why the stimulatory G protein α-subunit ($G_s\alpha$) is translated only from transcripts of its maternally derived allele in proximal renal tubules [8,9]. For the most part, I will assume that the physiological processes I discuss are determined by interactions between non-discriminating maternal haplotypes (NDMHs) and non-imprinted fetal haplotypes (NIFHs). I will not address possible effects of discriminating maternal haplotypes (either NIMHs or IMHs) because I have been unable to think of plausible ways in which well-informed maternal haplotypes could bias the supply of maternal calcium. In the context of the effects of fetal genes, my default assumption is that genes are unimprinted unless there is evidence to the contrary (as exists for *GNAS*, the gene that encodes $G_s\alpha$). My focus will be on human pregnancy, supplemented with evidence from laboratory rodents. Calcium metabolism is complex and it is with some trepidation that I take on a subject that is far from my own area of expertise. I can only hope that the interest of a somewhat idiosyncratic perspective can partially compensate for what will undoubtedly be a naïve presentation.

Calcium exists in the body in two compartments: (i) a large insoluble pool, stored in bone in fixed stoichiometric ratio with phosphate; (ii) a much smaller soluble pool in extracellular and intracellular fluids. The calcium concentration of the soluble pool must be tightly regulated: short-term increases of soluble calcium can be compensated by increased excretion of calcium in urine; short-term deficiencies can be compensated by releasing calcium (with phosphate) from bone and by increased reabsorption of calcium in the kidney [10].

Two hormones, parathyroid hormone (PTH) and 1,25-dihydroxyvitamin D ($1,25(OH)_2D$), are principally responsible for the short- to medium-term regulation of serum calcium. PTH has a half-life in plasma of 2–4 min [11] and is released when serum calcium falls below its set point. PTH causes the withdrawal of calcium from bone and increases reabsorption of calcium in the distal renal tubule. These effects serve to restore serum calcium to its set point. PTH also promotes the formation of $1,25(OH)_2D$ from 25-hydroxyvitamin D in the proximal renal tubule. $1,25(OH)_2D$ has a half-life in circulation of about 6 h [12]. Thus, its concentration integrates the degree to which calcium withdrawals have been made from bone over the course of a few hours. $1,25(OH)_2D$ increases intestinal absorption of calcium and prepares bone for future withdrawals [13].

The fixed stoichiometry of calcium and phosphate in bone means that their regulation is coupled. Phosphate is required to deposit calcium in bone and is released when calcium is mobilized from bone. Despite their fixed ratio in bone, calcium is dear but phosphate is cheap. That is, phosphate is more readily available in the diet, relative to requirements, than is calcium [14]. Therefore, the body can afford to be more profligate with phosphate than with calcium: approximately 99 per cent of the filtered load of calcium is reabsorbed by renal tubules [15] whereas only 80–85 per cent of the filtered load of phosphate is reabsorbed [16]. One of PTH's action is to decrease the reabsorption of phosphate in the proximal renal tubule [17]. By this means, PTH can increase serum calcium without a concomitant rise in serum phosphate.

Calcium metabolism in pregnancy

The human neonatal skeleton contains about 30 g calcium, most deposited during third trimester [18]. During pregnancy, calcium is pumped across the placenta to establish a higher concentration of ionized calcium in fetal serum than in maternal serum. A particular maternal-fetal gradient is not maintained. Rather, the fetus has a higher set point for serum calcium that is maintained independently of the ambient maternal concentration [19]. Pregnancy therefore entails a flux of calcium from the maternal soluble pool through the fetal soluble pool into fetal bone, with the maternal soluble pool being topped up from maternal dietary intake and release from maternal bone. Except in exceptional circumstances, this flux is maintained independently of maternal dietary intake, with any deficit between fetal accretion and maternal intake occurring at the expense of the mother's skeleton.

Parathyroid hormone-related peptide (PTHrP) stimulates calcium transport across the basal membrane of human syncytiotrophoblast [20] and the maternal-fetal calcium gradient is reduced in PTHrP knockout mice [21]. These results suggest that fetal serum calcium levels are maintained by parathyroid hormone-related peptide (PTHrP) regulating the placental calcium pump. The release of PTHrP by human cytotrophoblasts is regulated by extracellular calcium, but is inhibited by a higher concentration of extracellular calcium than that which inhibits PTH release from the parathyroid [22]. This difference could account for the higher set point of fetal calcium. If PTHrP activates the calcium pump at a higher calcium

concentration than the concentration at which PTH activates calcium release from maternal bone, then short-term deficits in serum calcium will be compensated by calcium release from the maternal skeleton, rather than from the fetal skeleton.

The principal maternal response to the increased calcium demands of pregnancy is to increase the extraction of calcium from diet. Intestinal uptake of calcium is increased during pregnancy, probably in response to elevated levels of $1,25(OH)_2D$ in maternal serum. The gestational increase in intestinal uptake is usually in excess of fetal needs and is accompanied by increased urinary excretion of calcium [23]. Despite this, there is evidence of increased maternal bone turnover, in both pregnant women and rats [18], and evidence for a decline in maternal bone mineral density during pregnancy (women: [24,25]; rats: [26]).

There is some evidence that a fetus will withdraw calcium from its own skeleton if the flux across the placenta is inadequate. Thus, fetuses of mothers with untreated hypoparathyroidism exhibit symptoms of hyperparathyroidism, including bone resorption and reduced bone mineralization [27,28]. In such pregnancies, the inability of a mother to maintain normal levels of serum calcium, because of her untreated PTH-deficiency, appears to result in activation of fetal PTH and mobilization of calcium from fetal bone. Similarly, knockouts of the calcium-sensing receptor cause elevated PTH and increased bone resorption in fetal mice [19].

A maternal-fetal phosphate gradient is also maintained across the placenta [10]. However, I conjecture that the opportunity cost of phosphate transfers during pregnancy is usually less than the opportunity cost of calcium transfers because of the greater availability of phosphate in the diet (relative to needs). If it were metabolically possible, mothers would be prepared to spend phosphate to save calcium.

In summary, the fetus appears to withdraw calcium freely from maternal serum, dictated solely by its own needs and without regard to the needs of its mother. If maternal dietary intake is adequate, the potential for conflict is largely limited to costs associated with the insensitivity of fetal demands to short-term fluctuations in maternal calcium supply. However, if maternal dietary intake is inadequate, the fetal skeleton will develop largely at the expense of the maternal skeleton. Why does one need to invoke evolutionary conflict to explain this organization of calcium metabolism? A skeptic might argue that the fetal needs are given first priority because this maximizes maternal fitness. In the next section, I propose that imprinting of *GNAS* in proximal renal tubules has evolved because marginal reallocations of calcium from the fetus to the mother have been associated with enhanced maternal fitness.

$G_s\alpha$ and the phosphate-depletion hypothesis

GNAS is perhaps the most complex of all imprinted loci, with multiple gene products transcribed from multiple promoters in a complex tissue-specific pattern. Some transcripts are expressed only from the maternally derived allele, some only from the paternally derived allele, and some show imprinted or biallelic expression depending on cell type. Some transcripts are translated, whereas others function solely as RNAs. One transcript is antisense to an oppositely imprinted transcript. Another encodes two interacting proteins in overlapping reading frames [29–32]. In this section, I will attempt an evolutionary explanation for a small part of this complexity. Specifically, I will present a hypothesis to account for the pattern of imprinting of transcripts encoding $G_s\alpha$ in the renal tubule.

Heterozygous inactivating mutations of $G_s\alpha$ in humans result in a suite of characters known as Albright's hereditary osteodystrophy (AHO). Of particular significance, these include mutations in an exon (exon 1) that is present only in spliced transcripts encoding $G_s\alpha$ [33]. Therefore, AHO appears to be caused by haploinsufficiency for $G_s\alpha$ in tissues where transcripts are normally expressed from both parental alleles. Individuals with a mutation of their maternally derived allele display resistance to the effects of PTH in the proximal renal tubule in addition to AHO, a combination of symptoms known as pseudohypoparathyroidism type Ia (PHP-Ia). By contrast, individuals with a mutation of their paternally derived allele exhibit AHO without hormone resistance, a combination known as pseudopseudohypoparathyroidism (PPHP) [34]. Both PHP-Ia and PPHP can be caused by mutations in exon 1 [35]. Therefore, hormone resistance appears to be caused by an absence of $G_s\alpha$ function in cells in which the protein is normally produced predominantly, or exclusively, from transcripts of the maternally derived allele. Resistance to PTH appears to be restricted to proximal tubules because distal tubules [36] and the skeleton [37] maintain their responsiveness to PTH. Evidence from mice that are heterozygous for inactivating mutations suggest that $G_s\alpha$ is translated only from the maternally derived allele in proximal renal tubules, but is translated from both alleles in the distal tubule [8].

Individuals with PTH-resistance of the proximal tubule, but without AHO, are described as having pseudohypoparathyroidism type Ib (PHP-Ib). Most patients with PHP-Ib show biallelic expression of transcripts containing exon 1A (an alternative first exon) and a lack of methylation of both copies of exon 1A. The maternally derived allele of exon 1A is normally methylated and transcriptionally silent [9] Transcripts containing exon 1A are probably untranslated [38]. One individual with PHP-Ib has been described with paternal uniparental disomy for *GNAS* [39]. Three brothers have been reported with selective resistance to PTH, but without AHO, caused by a mutation in exon 13 that prevents $G_s\alpha$ interacting with the PTH receptor. These siblings inherited the mutation from their unaffected mother, who inherited it from her unaffected father [40]. A hypothesis that unites these diverse clinical data is that transcription of exon 1A in proximal renal tubules somehow results in a failure to produce functional $G_s\alpha$ in *cis*.

$G_s\alpha$ is released when PTH binds to PTH receptors at the surface of epithelial cells of renal tubules. After its release, $G_s\alpha$

stimulates the production of cAMP by activating adenylyl cyclase. Elevated cAMP appears to mediate two of the principal effects of PTH in the proximal tubule. The first is the retrieval of NPT2a (the principal sodium/inorganic phosphate cotransporter) from apical brush border membranes, thus reducing phosphate reabsorption [17]. The second is the stimulation of 1α-hydroxylation of vitamin D to produce the active metabolite $1,25(OH)_2D$ [10]. In the distal tubule, PTH promotes increased reabsorption of calcium [15].

The kinship theory of genomic imprinting predicts that paternally derived alleles are inactivated when matrilineal inclusive fitness is maximized by a higher level of gene product than that which maximizes patrilineal inclusive fitness [41]. Thus, the phenotypic silence of the paternal allele encoding $G_s\alpha$ in proximal renal tubules suggests that patrilineal inclusive fitness would be increased, and matrilineal inclusive fitness decreased, by reduced retrieval of NPT2a and hence increased reabsorption of phosphate. Conversely, matrilineal inclusive fitness would be enhanced by increased excretion of phosphate. It seems reasonable first to consider possible selective forces that act during gestation, and involve the opportunity costs that care for a fetus imposes on its mother, before considering more complicated models that invoke postnatal interactions among kin. (PTH also causes increased production of $1,25(OH)_2D$ in proximal renal tubules. However, conflict over the level of production of $1,25(OH)_2D$ is not a promising candidate to explain imprinted expression of $G_s\alpha$ because mice with targeted ablation of 25-dihydroxyvitamin D 1α-hydroxylase appear grossly normal before weaning [42].)

I propose that $G_s\alpha$ acts to reduce retention of phosphate by the proximal tubule because of the indirect effect this has on the calcium flux across the placenta. This mechanism is proposed to be activated when mothers are calcium-stressed. To a first-order approximation, the calcium flux in third trimester should equal the rate of accretion of calcium in fetal bone. Therefore, if bone mineralization could be slowed by a relative scarcity of phosphate, then the placental pump for calcium would be down-regulated to prevent fetal hypercalcemia. In this scenario, the paternally derived allele of *GNAS* is silent in proximal tubules because patrilineal interests are promoted by maintaining a higher rate of fetal bone mineralization. By contrast, the maternally derived allele is expressed because marginal increases in the excretion of phosphate reduce the rate at which calcium can be deposited in fetal bone and thus, indirectly, reduce the rate at which calcium is lost from the maternal skeleton. Under conditions of maternal calcium stress, paternally-derived alleles of the fetus favour retention of both calcium and phosphate to maintain the calcium flux into fetal bone, whereas maternally-derived alleles favour depletion of phosphate (but retention of calcium) to reduce the calcium flux across the placenta. This provides an adaptive explanation of why the paternal copy of $G_s\alpha$ is silent in the proximal tubule but remains active in the distal tubule where its effect is to promote uptake of calcium. Phosphate (and calcium) voided in fetal urine can be recovered from amniotic fluid, but the hypothesis predicts that the marginal effect of increased excretion of phosphate is reduced bone deposition and down-regulation of the placental calcium pump.

The hypothesis predicts that a marginal decrease in expression of $G_s\alpha$ in proximal tubules would result in increased fetal bone mineralization under conditions of calcium stress, accompanied by increased loss of calcium from the maternal skeleton. Testing this prediction would be challenging. A test would be somewhat less daunting if the predicted effects of a marginal decrease in expression under calcium stress can be extrapolated to the effects of complete absence of expression of $G_s\alpha$ under normal availability of calcium. If so, the hypothesis predicts greater mineralization of the neonatal skeleton in PHP-Ia (and PHP-Ib) than in PPHP. Knockouts of $G_s\alpha$ in mice (e.g., [8]) would allow these predictions to be tested in calcium-stressed mothers.

The phosphate-depletion hypothesis is a first attempt to explain the imprinting of *GNAS* in proximal renal tubules. Among its weaknesses, the hypothesis assumes that the (largely unstudied) functions of PTH and $G_s\alpha$ in the fetal kidney resemble their functions in the postnatal kidney. These assumptions are, of course, testable. Zheng et al. [43] found no evidence of monoallelic expression of exon 1 transcripts in samples of kidney cortex from a human fetus with PHP-Ib. This suggests that expression of $G_s\alpha$ may not be imprinted in fetal kidney, although it is possible that evidence of imprinting may have been obscured by the samples containing a mixture of cells with monoallelic and biallelic expression [44]. Moreover, Zheng et al. [43] did not provide evidence on the expression of exon 1A transcripts, nor whether exon 1 transcripts of both alleles were translated. The phosphate-depletion hypothesis proposes that the selective force favouring imprinting is the calcium demand on mothers during pregnancy, even though there is an even greater calcium demand during lactation [45]. However, the ability of genes expressed in offspring to influence the maternal supply of calcium appear much more limited during lactation than during gestation.

REFERENCES

[1] Haig D. Gestational drive and the green-bearded placenta. Proc Natl Acad Sci U S A 1996;93:6547–51.
[2] Haig D. Altercation of generations: genetic conflicts of pregnancy. Am J Reprod Immunol 1996;35:226–32.
[3] Haig D. Genetic conflicts in human pregnancy. Q Rev Biol 1993; 68:495–532.
[4] Haig D. Genetic conflicts of pregnancy and childhood. In: Stearns SC, editor. Evolution in health and disease. Oxford: Oxford University Press; 1999, p. 77–90.
[5] Trivers RL. Parent–offspring conflict. Am Zool 1974;14:249–64.
[6] Haig D. Genomic imprinting and the theory of parent–offspring conflict. Semin Devel Biol 1992;3:153–60.
[7] Haig D. Genomic imprinting and kinship. New Brunswick (NJ): Rutgers University Press, 2002.
[8] Yu S, Yu D, Lee E, Eckhaus M, Lee R, Corria Z et al. Variable and tissue-specific hormone resistance in heterotrimeric G_s protein α-subunit ($G_s\alpha$) knockout mice is due to tissue-specific imprinting of the $G_s\alpha$ gene. Proc Natl Acad Sci U S A 1998;95:8715–20.

[9] Liu J, Litman D, Rosenberg MJ, Yu S, Biesecker LG, Weinstein LS. A *GNAS1* imprinting defect in pseudohypoparathyroidism type IB. J Clin Invest 2000;106:1167–74.
[10] Bringhurst FR. Calcium and phosphate distribution, turnover, and metabolic actions. In: DeGroot LJ, editor. Endocrinology, 3rd ed. Philadelphia: W. B. Saunders; 1995, p. 1015–43.
[11] Potts JT, Bringhurst FR, Gardella T, Nussbaum S, Segre G, Kronenberg H. Parathyroid hormone: physiology, chemistry, secretion, metabolism, and mode of action. In: DeGroot LJ, editor. Endocrinology, 3rd ed. Philadelphia: W. B. Saunders; 1995, p. 920–66.
[12] Holick MF. Vitamin D: photobiology, metabolism, and clinical applications. In: DeGroot LJ, editor. Endocrinology, 3rd ed. Philadelphia: W. B. Saunders; 1995, p. 990–1014.
[13] Jones G, Strugnell SA, DeLuca HF. Current understanding of the molecular actions of vitamin D. Physiol Rev 1998;78:1193–231.
[14] Potts JT. Parathyroids. Introduction. In: DeGroot LJ, editor. Endocrinology, 3rd ed. Philadelphia: W. B. Saunders; 1995, p. 917–9.
[15] Friedman PA. Renal calcium metabolism. In: Seldin DW, Giebisch G, editors, 3rd ed. The kidney: physiology and pathophysiology Philadelphia: Lippincott, Williams & Wilkins; 2000;2:1749–89.
[16] Silve C, Friedlander G. Renal regulation of phosphate excretion. In: Seldin DW, Giebisch G, editors, 3rd ed. The kidney: physiology and pathophysiology Philadelphia: Lippincott, Williams & Wilkins; 2000; 2:1885–904.
[17] Murer H, Hernando N, Forster I, Biber J. Proximal tubular phosphate reabsorption: molecular mechanisms. Physiol Rev 2000;80:1373–409.
[18] Kovacs CS. Maternal-fetal calcium and bone metabolism during pregnancy, puerperium, and lactation. J Clin Endocrinol Metab 2001; 86:2344–8.
[19] Kovacs CS, Ho-Pao CL, Hunzelman JL, Lanske B, Fox J, Seidman JG et al. Regulation of murine fetal-placental calcium metabolism by the calcium-sensing receptor. J Clin Invest 1998;101:2812–20.
[20] Strid H, Care A, Jansson T, Powell T. Parathyroid hormone related peptide (38–94) amide stimulates ATP-dependent calcium transport in the basal plasma membrane of the human syncytiotrophoblast. J Endocrinol 2002;175:517–24.
[21] Kovacs CS, Lanske B, Hunzelman JL, Guo J, Karaplis AC, Kronenberg HM. Parathyroid hormone-related peptide (PTHrP) regulates fetal-placental calcium transport through a receptor distinct from the PTH/PTHrP receptor. Proc Natl Acad Sci U S A 1996;93:15233–8.
[22] Hellman P, Ridefelt P, Juhlin C, Åkerström G, Rastad J, Gylfe E. Parathyroid-like regulation of parathyroid-hormone-related protein release and cytoplasmic calcium in cytotrophoblast cells of human placenta. Arch Biochem Biophys 1992;293:174–80.
[23] Gertner JM, Coustan DR, Kliger AS, Mallette LE, Ravin N, Broadus AE. Pregnancy as a state of physiologic absorptive hypercalciuria. Am J Med 1986;81:451–6.
[24] Karlsson C, Obrant KJ, Karlsson M. Pregnancy and lactation confer reversible bone loss in humans. Osteoporos Int 2001;12:828–34.
[25] More C, Bettembuk P, Bhattoa HP, Balogh A. The effects of pregnancy and lactation on bone mineral density. Osteoporos Internat 2001; 12:732–7.
[26] Tojo Y, Kurabayashi T, Honda A, Yamamoto Y, Yahata T, Takakuwa K et al. Bone structural and metabolic changes at the end of pregnancy and lactation in rats. Am J Obstet Gynecol 1998;178:180–5.
[27] Aceto T, Batt RE, Bruck E, Schultz RB, Perez YR. Intrauterine hyperparathyroidism: a complication of untreated maternal hypoparathyroidism. J Clin Endocrinol Metab 1966;26:487–92.
[28] Stuart C, Aceto T, Kuhn JP, Terplan K. Intrauterine hyperparathyroidism. Postmortem findings in two cases. Am J Dis Child 1979; 133:67–70.
[29] Hayward BE, Moran V, Strain L, Bonthron DT. Bidirectional imprinting of a single gene: *GNAS1* encodes maternally, paternally, and biallelically derived proteins. Proc Natl Acad Sci U S A 1998; 95:15475–80.
[30] Wroe SF, Kelsey G, Skinner JA, Bodle D, Ball ST, Beechey CV et al. An imprinted transcript, antisense to *Nesp*, adds complexity to the cluster of imprinted genes at the mouse *Gnas* locus. Proc Natl Acad Sci U S A 2000;97:3342–6.
[31] Klemke M, Kehlenbach RH, Huttner WB. Two overlapping reading frames in a single exon encode interacting proteins: a novel way of gene usage. EMBO J 2001;20:3849–60.
[32] Holmes R, Williamson C, Peters J, Denny P, RIKEN GER Group and GSL Members, Wells C. A comprehensive transcript map of the mouse *Gnas* imprinted complex. Genome Res 2003;13:1410–5.
[33] Aldred MA, Trembath RC. Activating and inactivating mutations in the human *GNAS1* gene. Hum Mutat 2000;16:183–9.
[34] Weinstein LS, Chen M, Liu J. Gs_{α} mutations and imprinting defects in human disease. Ann N Y Acad Sci 2002;968:173–97.
[35] Fischer JA, Egert F, Werder E, Born W. An inherited mutation associated with functional deficiency of the α-subunit of the guanine nucleotide-binding protein G_s in pseudo- and pseudopseudohypoparathyroidism. J Clin Endocrinol Metab 1998;83:935–8.
[36] Yamamoto M, Takuwa Y, Masuko S, Ogata E. Effects of endogenous and exogenous parathyroid hormone on tubular reabsorption of calcium in pseudohypoparathyroidism. J Clin Endocrinol Metab 1988;66:618–25.
[37] Murray TM, Rao LG, Wong MM, Waddell JP, McBroom R, Tam CS et al. Pseudohypoparathyroidism with osteitis fibrosa cystica: direct demonstration of skeletal responsiveness to parathyroid hormone in cells cultured from bone. J Bone Miner Res 1993;8:83–91.
[38] Liu J, Litman D, Chen W, Weinstein LS. Identification of a methylation imprint mark within the mouse *Gnas* locus. Mol Cell Biol 2000; 20:5808–17.
[39] Bastepe M, Lane AH, Jüppner H. Paternal uniparental isodisomy of chromosome 20q—and the resulting changes in *GNAS1* methylation—as a plausible cause of pseudohypoparathyroidism. Am J Hum Genet 2001; 68:1283–9.
[40] Wu WI, Schwindinger WF, Aparicio LF, Levine MA. Selective resistance to parathyroid hormone caused by a novel uncoupling mutation in the carboxyl terminus of $G\alpha_s$. A cause of pseudohypoparathyroidism type Ib. J Biol Chem 2001;276:165–71.
[41] Haig D. Parental antagonism, relatedness asymmetries, and genomic imprinting. Proc R Soc Lond B Biol Sci 1997;264:1657–62.
[42] Panda DK, Miao D, Tremblay ML, Sirois J, Farookhi R, Hendy GN et al. Targeted ablation of the 25-hydroxyvitamin D 1α-hydroxylase enzyme: evidence for skeletal, reproductive, and immune dysfunction. Proc Natl Acad Sci U S A 2001;98:7498–503.
[43] Zheng H, Radeva G, McCann JA, Hendy GN, Goodyer CG. Gαs transcripts are biallelically expressed in the human kidney cortex: implications for pseudohypoparathyroidism type 1b. J Clin Endocrinol Metab 2001;86:4627–9.
[44] Weinstein LS. The stimulatory G protein α-subunit gene: mutations and imprinting lead to complex phenotypes. J Clin Endocrinol Metab 2001; 86:4622–6.
[45] Wysolmerski JJ. The evolutionary origins of maternal calcium and bone metabolism during lactation. J Mammary Gland Biol Neoplasia 2002; 7:267–76.