The evolution of the placenta

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Abstract

The very apt definition of a placenta is coined by Mossman, namely apposition or fusion of the fetal membranes to the uterine mucosa for physiological exchange. As such, it is a specialized organ whose purpose is to provide continuing support to the developing young. By this definition, placentas have evolved within every vertebrate class other than birds. They have evolved on multiple occasions, often within quite narrow taxonomic groups. As the placenta and the maternal system associate more intimately, such that the conceptus relies extensively on maternal support, the relationship leads to increased conflict that drives adaptive changes on both sides. The story of vertebrate placentation, therefore, is one of convergent evolution at both the macromolecular and molecular levels. In this short review, we first describe the emergence of placental-like structures in nonmammalian vertebrates and then transition to mammals themselves. We close the review by discussing the mechanisms that might have favored diversity and hence evolution of the morphology and physiology of the placentas of eutherian mammals.

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Introduction

Viviparity has evolved independently and seemingly on multiple occasions across a diverse array of animal groups, including invertebrates (Kalinka 2015). It is a phenomenon whereby developing embryos are retained within the reproductive tract, leading to release of live offspring as an alternative to the more fecund egg laying or spawning. One consequence of viviparity is that the retained, fertilized egg must either survive off its own reserves, usually yolk, or obtain some or part of these resources from the mother. The latter situation, of necessity, is expected to lead to increased conflict over how provisions are partitioned between the supplier and the recipient, thus potentially sparking a genetic arms race. In turn, such conflict is expected to drive adaptive changes that lead to a more intimate and possibly more felicitous relationship between offspring and the reproductive tract of the mother that favors the transmission of both maternal and paternal genes to the next generation, despite a loss of overall fecundity.

The transition from oviparity to viviparity and the subsequent emergence of placentation within some vertebrate taxa clearly required major changes in the morphology and physiology of the reproductive tract and has its origins well before the advent of mammals (Van Dyke et al. 2014, Blackburn 2015). A placenta, as defined originally by Mossman, is the ‘apposition or fusion of the fetal membranes to the uterine mucosa for physiological exchange’ (Burton & Jauniaux 2015). It is a specialized organ whose purpose is to provide continuing support to the developing young through the provision of water, nutrients and gases, and to regulate maternal–fetal interactions often through hormone production. By this definition, placentas have evolved within every vertebrate class other than birds. Although placentation arose once in the common ancestor of mammals, it has arisen independently multiple times within other classes, and even families. The story of vertebrate placentation, therefore, is one of convergent evolution at both the macromolecular and molecular levels. In this short review of placental evolution, we first describe the emergence of placental-like structures in nonmammalian vertebrates and then transition to mammals themselves. We close the review by discussing mechanisms that might have favored diversity and hence evolution of the morphology and physiology of the placentas of eutherian mammals. Of necessity, many important references cannot be cited in a short review of this kind. Instead, we have attempted to direct the reader to scholarly articles that do list the primary source material.

Nonmammalian vertebrates

Cartilaginous fish

Viviparity is the most common mode of reproduction in elasmobranchs. However, there is a wide range in the degree of maternal provisioning of the embryo after...
ovulation (Wourms & Lombardi 1992), with embryos of some species depending entirely on the yolk sac for all of pregnancy, e.g. the spiny dogfish, which at birth weighs 40% less than the yolk-filled fertilized egg from which it develops. The tiger shark, by contrast, solves such limiting provisioning; in which the dominant embryo eats all of its littermates and any unfertilized eggs before birth (Korsgaard & Weber 1989).

In the elasmobranchs, two placental types are observed. Among stingrays, finger-like projections of the uterine wall, termed trophonemata, provide histotrophic nutrition to the developing embryo (Hamlett et al. 1993), while the epithelium overlying uterine blood vessels thins, lessening the barrier to exchange. The mature embryo of the cow nose, for example, weighs 3000 times as much as the egg as a result of reliance on the mother rather than the egg yolk (Hamlett et al. 1993). A different placental type, the yolk sac placenta, has evolved multiple times in the ground sharks (Wourms & Lombardi 1992). After the yolk has been consumed, the yolk sac becomes modified into an umbilical cord region and a placental region (Hamlett et al. 1993), which become closely apposed to the epithelium of a highly vascularized oviduct.

**Teleosts**

In Poeciliopsis fish, eggs are retained within the follicle after fertilization and throughout embryonic development (Wourms & Lombardi 1992). Most species are lecithotrophic, relying on yolk, but in others, a highly folded, follicular epithelium forms a ‘follicular placenta’ with the embryonic pericardial sac. This epithelial layer is lined with microvilli, while the surrounding tissue is highly vascularized, presumably to facilitate maternal–fetal exchange (Kwan et al. 2015). This type of placental structure has arisen several times during Poeciliopsis evolution (Kwan et al. 2015) and has been subsequently lost and regained many times, making it a model for the evolution of placentation (Pollux et al. 2014).

Among syngnathid fish (pipefish and seahorses) the males, and not the females, become pregnant, after the female transfers the fertilized eggs to a brood pouch on the ventral side of either the trunk (Gastrophori) or the tail (Urophori) of the male’s body. The degree of contact and exchange between developing embryos and the father range from minimal to situations that display all the defining features of placentation (Wilson et al. 2001). For example, in the straight-nosed pipe fish Nerophis ophidion, eggs stick to the epithelium of the father’s pouch, which is open to the sea environment (Carcupino et al. 2002), while in the pot-bellied seahorse Hippocampus abdominalis, the pouch is sealed and its epithelium highly folded, thinned and vascularized (Carcupino et al. 2002), thereby allowing exchange of nutrients.

**Amphibians**

Viviparity has arisen multiple times in amphibians, accompanied in some cases with the evolution of placentas. In frogs, there are examples in which the tadpoles develop in the father’s mouth, in the mother’s stomach, or on the skin of the back (Wake 1993). In the marsupial frogs, development occurs inside a specialized maternal pouch on the back of the animal. Following ovulation, secretory cells of the vascularized pouch enlarge. The pouch epithelium is closely apposed to specialized fetal gills, allowing exchange between maternal and fetal circulations (Savage 2002). This, as A.M. Carter wrote, ‘would satisfy most people’s definition of a placenta’ (Carter 2016). Arguably, however, a structure found outside the reproductive tract, especially following external fertilization that occurs in these frogs and the sea horses described earlier, does not fit the typical definition of a placenta. Nor have any of the ‘gill placentas’ of these endangered frog species yet been shown to be the primary source of fetal nutrient support.

**Reptiles**

Viviparity occurs in more than 25 families of lizards and snakes (squamate reptiles). Although estimated by some to have arisen independently more than one hundred times, it is conceivable that there was a single origin of viviparity and multiple subsequent reversions to oviparity (Pyron & Burbrink 2014, Van Dyke et al. 2014, Blackburn 2015). In many species, the embryo relies on egg contents for nutrition, but in others a range of adaptations of the female reproductive tract provides a means to exchange gases and nutrients with the conceptus (Stewart 2015). In squamates the placenta is chorioallantoic, but unlike mammals it does not develop from an early arising, extraembryonic trophoblast layer. Most squamate placentas demonstrate a simple interdigitation of the chorioallantois with the uterine epithelium (Stewart 2015), but in some skinks the interface is more intimate (Fig. 1). In the case of the African skink Trachylepis ivensi, there is even a degree of invasive implantation, with direct contact between chorionic projections and maternal capillary endothelium (Blackburn & Flemming 2012).

The chorioallantoic placenta of viviparous lizards can be highly efficient in facilitating exchange of nutrients and gases. Offspring of the Mabuya lizard weigh 500 times more at birth than the egg at fertilization, indicating a nearly complete reliance for growth on maternally supplied materials (Thompson & Speake 2002). There is also evidence for the expression of active transporters at the sites of maternal–fetal apposition in some species (Murphy et al. 2011) and for placental production of steroid hormones (Painter & Moore 2005). Overall, where placentation occurs in squamates, it bears a superficial resemblance to the epitheliochorial...
placenta. The evolutionary significance of this unique placental structure in marsupials has been the subject of much research.

**Mammals**

All mammals except the egg-laying platypus and the five species of echidnas, the only surviving monotremes, rely on a placenta for their reproduction. Indeed, the first lineage decision made during embryonic development of Mammalia is the segregation of cells that are destined to become the external tissue layer of the placenta. This lineage, usually called trophoblast (or, because it forms the exterior of the conceptus, trophectoderm), diverges early from the pluripotent lineage that advances to form the inner cell mass. The cells that give rise to the hypoblast and epiblast are unique to mammals. The term trophoblast was first coined by Hubrecht (1904). Its roots are ‘tropho’ meaning nourishment, and ‘blast’ for embryonic. It is therefore questionable whether embryo-derived placental cells of other animals, which unlike mammals do not form an initiating trophectoderm, should be termed ‘trophoblast’, despite many of their shared functions. When and how trophoblast emerged in the presumed metatherian ancestors of the Class Mammalia is mysterious and unlikely ever to be revealed from the fossil record.

**Marsupials**

Although eutherians are often referred to as ‘placental mammals’, marsupials do possess placentas. However, the placentas are short-lived and marsupials give birth to relatively underdeveloped young. In marsupial blastocyst, blastomerleres adhere to the inner surface of the zona pellucida, resulting first in a cup-shaped blastocyst, and then a hollow sphere with no discernible inner cell mass (Selwood & Johnson 2006). Both the embryonic and extraembryonic lineages emerge from this single cell layer, with trophoblast materializing from the pole of the blastocyst opposite the position of the future embryo (Frankenberg et al. 2013, Morrison et al. 2013, Familari et al. 2016). There are also indications that early lineage specification may be controlled by a similar set of transcription factors as in eutherians (Frankeberg et al. 2013, Morrison et al. 2013, Familari et al. 2016).

The trophoblast cells originating from the marsupial blastocyst combine with the yolk sac endoderm and extraembryonic mesoderm to form a yolk sac placenta, which can be bi- or trilaminar (Renfree 2010). However, there are a species of koalas, wombats and bandicoots, in which a chorioallantoic placenta forms, but it appears to be supplementary to the main yolk sac placenta (Freyer et al. 2003). Although the placentas of marsupials are generally regarded as noninvasive and of the epitheliochorial type (see next section), an area of syncytiotrophoblast forms in the yolk sac placenta of the gray short-tailed opossum and possibly related species. This tissue makes direct contact with maternal blood vessels (Zeller & Freyer 2001). Additionally, trophoblast syncytialization (syncytial trophoblast; STB), where adjacent cells fuse to produce cells with more than a single nucleus, is accompanied by the expression of an endogenous, retrovirus-derived protein, analogous to what occurs in many eutherians (Cornelis et al. 2014). Another similarity to the eutherians is that the placenta of the tammar wallaby secretes a complement of hormones that include IGF2 and relaxin, which, in that species, may be responsible for the phenomenon of maternal recognition of pregnancy (Renfree 2010).

**Eutherians**

**Origin of trophoblast:** Eutherians consist over 5000 species in roughly 20 phylogenetic orders. Most of our knowledge about their early development and the events that lead to the initial specification of trophoblast has been obtained only in a handful of these species, especially the mouse. In mouse (Mus musculus), the
first visible differentiation event, called compaction, occurs between the 8 and 16 cell stage of embryonic development, when the blastomeres polarize to form extended contact zones with their neighbors and an outward-facing apical surface (Posfai et al. 2014). Further symmetrical and asymmetrical divisions follow, so that by the 16 cell morula stage a population of polarized cells exist on the cell surface, which becomes trophectoderm, and another population of cells are positioned inside, which is the precursor of the inner cell mass (ICM). By the time of blastocyst formation, when the mouse conceptus comprises about 32 cells, about two-thirds of which are trophectoderm, the precursors of extraembryonic endoderm (hypoblast) have begun to segregate toward the base of the ICM. From this location, they migrate outward to line the trophectoderm, forming the yolk sac cavity. Later, extraembryonic mesoderm joins with the trophectoderm to form what at this stage is a bilayered chorion. Although there are clear morphological similarities in the steps leading to blastocyst formation in rodents, humans and domestic mammalian species, there are differences in how these events are timed and subsequently played out. In many species, for example, mesoderm differentiates to vascularize the yolk sac, forming a yolk sac placenta that is eventually replaced by the chorioallantoic placenta. Such a placenta does not form in humans, but persists to term in rodents and lagomorphs. Similarly, compared with the mouse, conceptuses from many species undergo at least one further round of cell division before the blastocoel cavity becomes visible. For details on how placentas subsequently develop and begin to diverge in morphology, the reader is directed to the still germane reviews by Amoroso (1952) and Renfree (1982).

Range of placental morphologies: Despite the spectacular diversity encountered in placental structure at both the gross and microanatomical levels (Enders & Carter 2004), there have been attempts over the years to classify placentas and (somewhat unsuccessfully) to relate the outcomes to mammalian phylogeny. For example, placentas come in a range of shapes and sizes, and have been placed in groups based on the general gross morphologies of the sites where the chorion attaches to the endometrium (Fig. 2). They have also been classified according to how many cell layers separate the maternal and fetal circulations (Fig. 2). A simplified version of this system recognizes three main placental types (Renfree 1982): epitheliochorial, in which there is no erosion of the uterine epithelium; endotheliochorial, where the invading trophoblast reaches but does not penetrate maternal capillaries within the endometrium; and hemochorial, where the trophoblast surface is in direct contact with maternal blood (Fig. 3). A fourth type, synepitheliochorial, has been recently used to describe the placentas of ruminants (Wooding 1992). Here, specialized trophoblast binucleated cells fuse with uterine epithelial cells to form trinucleated cells, a syncytium, or both. The placentas of ruminants, with the exception of the phylogenetically more primitive tragulids, are also characterized as cotyledonary (Fig. 2). Cotyledons are the primary sites of attachment of the chorion to the endometrium and comprise tufts of villous trophoblast that interdigitate and interlock with complementary structures on the endometrium, called caruncles. It is within these sites, often known as placentomes, that binucleated cells, placental and maternal blood capillaries, and placental interchange are most concentrated. Therefore, this kind of placenta is a derived form, secondary to the more superficial, diffuse epitheliochorial type typical of most other artiodactyls.

Superficial vs invasive placentation: Intuitively, it might be inferred that the least invasive types of placenta, the kind where multiple cell layers separate the two blood supplies (Fig. 2B), would be the most inefficient and also the most primitive of all placental types because there are more apparent physical barriers to limit the movement of nutrients and dissolved gases. Also, such placentas have a resemblance to placentas described earlier for nonmammalian species, such as skinks (Fig. 3). However, this is clearly not the case.
The evolution of the placenta (Wildman et al. 2006). Compare, for example, the relative maturity and independence of a newborn horse, pig or whale, supported during their gestations by diffuse epitheliochorial placentas, to a hapless pup born to a mouse or a baby born to a human, species where the placenta is hemochorial and discoid. Nor is the more superficial form of placenta evolutionarily more ancient than the kinds that are invasive and make direct contact with maternal blood. On the contrary, careful analyses, in which placental features were mapped to mammalian phylogenies, have clearly dispelled this myth and demonstrated that the epitheliochorial placenta is not only a derived form that has evolved from a more invasive placental type, but has arisen independently in three distinct mammalian lineages, including primates (Wildman et al. 2006) (Fig. 4). Moreover, this and similar trees based on other features of the placenta strongly imply that the placenta of the last common eutherian ancestor was discoid, either hemochorial or endotheliochorial (Mess & Carter 2007), and possessed a labyrinth-type of placental interdigitation.

It should be stressed that the perceived disadvantages of extended diffusion distances for dissolved gases and solutes encountered with a superficial placenta, like that of the pig, are largely illusory. As pregnancy proceeds, the surfaces of the uterine and trophoblast epithelial layers become interlocked (Fig. 5B). Capillaries on the maternal and fetal sides come within a few microns of each other (Fig. 5A), often squeezing down to the tight junctions between the trophoblast cells on the one side and uterine epithelial cells on the other, probably allowing efficient exchange of small molecules (Friess et al. 1980). Movement of macromolecules that carry essential components such as metals and vitamins is more problematic, but an alternative strategy has been employed, namely the delivery of these components not in the blood but in uterine secretions – a process called histotrophic nutrition (Fig. 5C) (Renegar et al. 1982). The uterine glands of the pig, for example, release uroteroferrin, a bi-iron containing acid phosphatase to supply iron. Uroteroferrin and other proteins with a similar provisioning function are taken up by specialized regions of endocytic trophoblast cells congregated in cup-like structures called areolae, which develop opposite the mouths of each uterine gland (Fig. 5C). From there, the maternal factors are transported via the fetal bloodstream to the liver and other organs for processes such as hematopoiesis (Renegar et al. 1982).

The likely advantages of a noninvasive placenta over the
more primitive hemochorial and endotheliochorial types include reduced exposure to the potential threats of the maternal immune system, less damage to the uterus associated with birth and elimination of the placenta, and minimizing the transmission of fetal cells into the maternal blood circulation and vice versa. Finally, superficial placenta may limit disease transmission between fetus and mother.

As the nature of the maternal–fetal interface evolves, so does the nature of the biochemical communication possible between conceptus and mother. This is particularly true in early pregnancy where there is an urgency to interrupt estrous cyclicity and maintain continued production of progesterone, if the pregnancy is to continue (Bazer et al. 2009). As far as is known, it is invariably the trophoblast that is responsible for generating this signal for maternal recognition of pregnancy, although the process is understood in only a handful of species and even then rather poorly. What is clear is that the mechanisms are highly variable, evolving rapidly, and no one approach is used widely across taxa. A couple of examples illustrate the kind of strategies used in apes, where implantation occurs immediately after the blastocyst attaches, and in artiodactyls, where implantation is superficial and delayed. In the human and most probably higher primates, but not in strepsirrhines (lemurs and related species) that have a noninvasive trophoblast, chorionic gonadotrophin (CG) provides luteotropic support to the progesterone-producing cells of the corpus luteum soon after implantation. In women, hCG becomes measurable in blood and even in urine by the second week of pregnancy, most likely as a result of being released directly into maternal capillaries even before the villous placental bed has formed (Roberts et al. 1996). In ruminant species, the pregnant mother responds to the presence of the conceptus even before the trophoblast has attached to the uterine wall. The factor responsible is not a gonadotropin, but instead a type 1 interferon-τ (IFNT), which acts on the uterine epithelium, where it modulates the release of the luteolytic factor, prostaglandin F2α, that would normally cause the corpus luteum to regress (Bazer et al. 1997, Roberts et al. 1999). In pig, where maternal recognition of pregnancy is also achieved during the period when the conceptuses are elongating within the uterine lumen, the anti-luteolytic factor appears not to be a protein, but the steroid hormone estrogen (Spencer et al. 2007)

Selective mechanisms that may contribute to placental diversity: It is generally recognized that there are additional driving forces promoting adaptive changes in the placenta and hence contributing to its rapid evolution. One is undoubtedly the struggle for control of maternal physiology in terms of nutrient allocation (Fowden & Constancia 2012). As the demands of the fetus increase, they will likely conflict with the ability of the mother to provide such resources. An extensive literature has developed around this topic, with particular emphasis on the role played by imprinted genes in controlling nutrient supply and growth of the fetus (Reik et al. 2003). Many placental gene products, including hormones and transporters, for example, likely play a role in raising the concentrations of nutrients in maternal blood and facilitating their uptake by the placenta. There is, thus, a drive to keep the paternal genes representing the fetus active, while silencing the corresponding maternal alleles. Maternal efforts to counteract the acquisitiveness of the fetus may lead to the evolution of oppositely imprinted genes. Again, most
speculation on the role of imprinted genes has come from the mouse, with roles inferred for the human, both species with an invasive placenta (Reik et al. 2003). Very little is known about imprinting in those species where there is little or no direct access of the trophoblast to maternal blood, although data are beginning to emerge (Chen et al. 2016).

A second force driving placental evolution is undoubtedly the avoidance of immune rejection (Hemberger 2013), a process still little understood, although speculation abounds. It is assumed that to minimize attention from various arms of the maternal immune system, the trophoblast must continue to evolve counter-measures for its own protection and even advantage. These measures will undoubtedly be subtle and complex and change as the degree of intimacy with maternal system itself evolves. Some clues may emerge by examining candidates encoded by rapidly evolving genes and gene families within individual mammalian clades, some of which are discussed in the next section.

Evolution of placenta-specific genes

Despite the observation that the initial lineage regulators for trophoblast cell fate decisions may be at least partially conserved across mammals, the resulting placental structures and their associated trophoblast cell populations appear not to be governed by particular master genes. There are a few coding genes, e.g., GCM1, a transcription factor that plays a role in controlling the formation of syncytial trophoblast (STB) (Cross et al. 2003) that appear to have an entirely placental function in the sense that they have not been implicated in developmental events outside the trophoblast lineage. However, it seems likely that much of the structural diversity and functional refinements associated with mammalian placentas depend on widely expressed transcription factors operating in a combinatorial, but trophoblast-specific manner and poorly understood processes linked to peculiarities in the epigenetic landscape of trophoblast. There have also been a large number of gene family expansions (Rawn & Cross 2008). These gene families may be performing roles in processes such as maternal recognition of pregnancy, nutrient acquisition and immune protection, thereby contributing to the unique functional needs of different placental forms. It is worth noting that ‘placenta-specific’ in this context means that such genes are disproportionately, but not necessarily uniquely, expressed in the placenta. For example, it is not unusual for some placenta-specific gene products to be elevated in cancer and germ cells.

Placenta-specific gene families

Gene duplication provides a useful opportunity for natural selection to operate (Hughes 1994) and allows the duplicated genes to acquire new functions, and, through the acquisition or loss of regulatory regions, changes in the temporal, spatial, or magnitude of transcription compared with the original gene. Gene family expansions associated with distinct placental types have arisen repeatedly in eutherians (Rawn & Cross 2008). Here, we provide four examples from the many known placenta-specific gene families. In general, assigning function of individual members within a family will be difficult. Indeed, knockouts in mice performed on single members of gene families have generally not been informative, possibly because phenotype is not obviously impaired within the friendly confines of a vivarium, although it may become so when the pregnant animal is under stress (Ain et al. 2004). Also, products of multigene families likely overlap in function, so that loss of one gene may be compensated by the presence of one or more of its orthologs. Finally, family members may even interact in an epistatic manner, providing a combinatorial benefit not achieved by a single member operating alone.

The growth hormone and prolactin-related proteins are found in primates, rodents and ruminants and, most likely, other taxa. Interestingly, gene family expansions have occurred independently in these different lineages, providing yet another example of convergent evolution. Growth hormone (GH) and prolactin (PRL) themselves arose from a common ancestral gene. In most species, the GH gene is a single copy, but in humans, there are five members, one restricted in expression to the pituitary and the other four to the placenta (Haig 2008). Their likely role is in resource allocation between the fetus and the mother. By contrast, there is only a single copy of the PRL gene in human, whereas there are 23 members in mouse, 24 in rat and 12 in cattle (Haig 1993). All except prolactin itself are expressed in the placenta. In mouse, they have diverse patterns of spatial and temporal expression (Simmons et al. 2008) and appear to target the maternal endometrium (Soares et al. 2007).

The pregnancy-specific glycoproteins (PSGs) are secreted molecules that are abundantly produced by primate and rodent placentas. They are related to the carcinoembryonic antigen cell adhesion molecules (CEACAM) found on the surface of certain tumors and selected normal somatic cells. There are 10 intact PSG genes in the human genome and 17 in mouse (McLellan et al. 2005), although it is almost impossible to assign orthologous members based on sequence identity. Moreover, the organization of the domains in human and mouse PSG proteins are quite different (McLellan et al. 2005), raising the possibility that their roles in primates and humans may not be identical. PSGs are able to enter the maternal circulation and, in human, can accumulate to extraordinarily high concentrations in blood (200–400 µg/mL at term), but their roles remain enigmatic.

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The interferon τ (IFNτ) proteins are products of the filamentous conceptus of ruminants and have been mentioned earlier in the context of maternal recognition of pregnancy. They have been described in no other taxa (Leaman & Roberts 1992). Within a species they are well conserved, but considerable divergence is evident in inter-species comparisons. They are transcribed from multiple, structurally conserved genes that have probably arisen by duplications and gene conversion events from another type 1 IFN gene, interferon omega (IFNW). Although builds of the bovine genome have suggested IFNτ to be a relatively small gene family (Walker & Roberts 2009), a large number of gene variants have been described in each species and even within single conceptuses, suggesting that the families continue to diverge rapidly. The IFNT proteins are secreted from mononucleated trophoblasts and act on the uterus by binding to type I interferon receptors, with the period of release occurring before the trophoblast has formed definite attachment to the uterine epithelium (Bazer et al. 1997, Roberts et al. 1999). Another notable feature of the IFNτ is that these proteins have retained typical features of other type I interferons, such as potent antiviral activity, although their primary role has shifted to a reproductive function. Conceivably, the progenitor gene product in the common ancestor of present-day ruminants served to protect the conceptus from viral infections.

The pregnancy-associated glycoproteins (PAGs) are products of a gene family present in the placental trophoblasts of swine and ruminants and most other even-toed ungulates (Wallace et al. 2015). They are related to aspartic proteinases, although some notably lack the capacity to cleave protein substrates due to modifications of normally conserved amino acids around the active sites. The PAGs are particularly complex in ruminants; in cattle, for example, there are approximately two dozen members along with some closely related paralogs. Most of the PAGs in ruminants are expressed by specialized polyploid trophoblast cells (binucleate giant cells) discussed earlier. After fusion, the contents of secretory granules within the binucleate cells are disgorged into the uterine stroma, and many of the released PAGs enter the maternal circulation and form the basis of useful pregnancy tests for various ruminants, including dairy cows. The role of PAGs is unknown, but their abundance suggests an important function during pregnancy.

Endogenous retroviruses and retroviral elements

Ancestral retroviral infections have provided another source of novel protein-coding genes that have played a role in placental evolution. To ensure that the viral genes do not replicate, the control elements of endogenous retroviruses (ERVs) are usually highly methylated and effectively silenced. As a result, the fate of most ERVs is gradual genetic degradation through mutation and homologous recombination. Nevertheless, a minority of ERVs retain a whole or partially intact open reading frame potentially capable of producing a functional protein (Weiss 2016). This point is particularly salient in regard to the placenta, which in many species express a range of ERVs that are involved in trophoblast function (Denner 2016). Of those ERVs expressed in the placenta, the most studied have originated from the envelope (env) elements of the integrated viral DNA, and have been called syncytins (Lokossou et al. 2014).

In humans, ERVW1 (syncytin-1) facilitates the fusion of mononucleated cytotrophoblasts to form multinucleated STBs (Mi et al. 2000). This process involves the interaction of ERVW1 with a ‘receptor’, most probably the neutral amino acid transporter, SLC5A1, on a neighboring cell. The expression of ERVW1 is regulated by the placenta-specific transcription factor GCM1 discussed earlier, via an enhancer element present in the long-terminal repeat (LTR) of the gene (Lin et al. 2005). It is also notable that ERVW1 is not involved in trophoblast fusion in Old World monkeys, despite being present in their genomes (Cáceres et al. 2006). These animals make use of a distinct syncytin, ERVV2, for STB formation. Conversely, ERVV2 is expressed in human placenta but does not appear to be involved in cytotrophoblast fusion (Blaise et al. 2005). An important point illustrated here is that expressed retroviral envelope genes (env) often fulfill similar roles across species, but few represent orthologous genes. Instead, the majority has arisen as a result of independent integration events and coopted for similar functions by different species.

A second env gene from a different endogenous retrovirus (ERVFRD1; syncytin-2) is expressed in the villous trophoblast of the human placenta (Blaise et al. 2003, Malassine et al. 2007) and is also believed to be involved in trophoblast fusion events through interaction with another transporter protein, MFSD2. ERVFRD1 has been proposed to be immunosuppressive, an activity not shared by ERVW1 (Mangeney et al. 2007). Several other ERV proteins, e.g. ERV3-1, and ERVK family members, including an antagonist of cell fusion (Sugimoto et al. 2013) are expressed in the placenta, but the roles of most of them are even more obscure than that of ERVFRD1.

In mouse placenta, the labyrinth layer, which is the functional equivalent of human floating villous trophoblast, forms the transport surface in contact with the maternal bloodstream. It is hemochorial, i.e. it has a cytotrophoblast layer overlaid by two STB layers (Dupressoir et al. 2009). Two syncytin genes (Syna & Synb), products of separate integration events, are expressed in this tissue. The SYNb protein is primarily localized to the innermost STB layer, and, like ERVFRD in the human, may have immnosuppressive activity, as well as roles in facilitating cell fusion

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Neither of these gene products, however, is related to human syncytins, except for the fact they are derived from ERV genes and are expressed placentaally.

Finally, endogenous retroviruses, with no obvious homology to each other, have been described in the placentas of a number of other species, including marsupials, ruminants, carnivores and lagomorphs (Denner 2016). In ruminants, they are mainly associated with trophoblast binucleate cells that fuse with uterine epithelial cells, but are assumed in all these species to play roles that are somewhat similar to syncytins in the human placenta.

Conclusions

Placentas, whatever their anatomic features, share a number of common functions, all of which require cooperative interactions with the maternal system. Placentas deal with nutrient and oxygen import, disposal of fetal waste products and gases, help to physically retain the conceptus in the reproductive tract, commandeer the local maternal blood supply in some manner, release factors including metabolic hormones to adjust the needs of the fetus to the resources provided by the mother, and provide immune protection. Yet, despite these apparently conserved functions placentas are arguably the least conserved and the most rapidly evolving mammalian organs. These changes are speculated to be driven, at least in part by maternal–fetal conflict (Haig 1996). Placental divergence has, in turn, been promoted both mutations to become fixed and the placenta to relaxed silencing of gene control elements, including (1) duplications and gene conversion events to create large gene families that themselves are continuing to diverge extensively; (2) co-opting of endogenous retrovirus-derived genes and gene control elements; (3) rapid evolution of placenta-specific enhancers and promoter elements; and (4) imprinting. It is also possible that the hypomethylated state of the placental genome (Chuong 2013) has permitted relaxed silencing of gene control elements, including those of ERVs. Perhaps this feature of the placenta sets the stage for increased opportunity for epistasis – the interactions of genes that are not allelic. For example, a harmful mutation in one gene may occasionally become beneficial and confer increased fitness in the presence of a mutation in another gene (sign epistasis) (Weinreich et al. 2005), possibly allowing both mutations to become fixed and the placenta to adapt to the constraints of a changing environment.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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