

Reflections on the Evolution of Piscine Viviparity¹

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SYNOPSIS. Viviparity first makes its evolutionary appearance within the craniate-vertebrate line among fishes. We estimate that it has independently evolved at least 42 times in five of the nine major groups of fishes. Viviparity is the dominant mode of reproduction among the cartilaginous sharks and rays, *i.e.*, 55% of approximately 900 living species. It is less prevalent among the five major groups of bony fishes, *i.e.*, 2-3% of an estimated 20,000 or more species. The evolution of viviparity from oviparity involves: 1) a shift from external to internal fertilization; 2) retention of embryos in the female reproductive system; 3) utilization of the ovary or oviduct as sites of gestation; 4) structural and functional modification of the embryo and the female reproductive system and; 5) modification of extant endocrine mechanisms controlling reproduction. Viviparity offers selective advantages to parents and offspring, such as: 1) enhanced survival of offspring; 2) compensation for low fecundity; 3) amplification of reproductive niches to reduce competition; 4) exploitation of pelagic niches; 5) colonization of new habitats; and 6) increased energetic efficiency in viviparous matrotrophes. Its principal disadvantages include: 1) reduced fecundity; 2) cost to the female; and 3) risk of brood loss through maternal death. Acquisition of viviparity establishes new maternal-embryonic relationships, namely: 1) trophic; 2) osmoregulatory and excretory; 3) respiratory; 4) endocrinological; and 5) immunological. In sharks, rays, and the coelacanth, gestation takes place in the oviduct, but in teleosts gestation occurs either in the ovarian follicle or ovarian lumen. The cystovarian teleostean ovary is hypothesized to function both as ovary and oviduct. Oviductal, ovarian luminal, and follicular epithelial cells are the maternal sites of metabolic exchange. Metabolic exchange in embryos takes place across the epithelia of the general body surface and its derivatives or across the gut epithelium and its derivatives. Four patterns of piscine placentation have evolved, namely: 1) yolk sac; 2) follicular; 3) branchial; and 4) trophotaenial placentae. The pericardial amniochorion, the embryonic portion of the follicular placenta, occurs in poeciliids and several other teleostean groups. Developmentally, it is nearly identical to the anterior amino-chorionic fold of tetrapod vertebrates. Trophotaeniae are external rosette or ribbon-like structures that have evolved in four orders of teleosts by heterochrony, *i.e.*, accelerated outgrowth and differentiation of the embryonic hind gut. With the possible exception of the coelacanth, the yolk sac placenta occurs only in sharks. We estimate that it has independently evolved between 11 and 20 times. It displays considerable diversity. Evolution of the yolk sac placenta entails retention of the yolk sac and secondary differentiation of its distal portion for implantation and maternal tissue-embryonic tissue metabolic exchange and its proximal portion for oviductal fluid-embryonic tissue exchange. The yolk stalk lengthens, is modified into an umbilical stalk, and establishes a site of autotomy at the embryo-umbilical stalk junction. The luminal wall of the oviduct becomes competent to function as a site of implantation.

INTRODUCTION

Viviparity within the vertebrate line (or craniates, if one includes the hagfish) first evolved among the fishes. Viviparous reproduction has also evolved among mem-

bers of all other vertebrate groups except birds. Fishes are pivotal to our understanding of the evolution of vertebrate viviparity not only because they were the first viviparous vertebrates, but also because they manifest the greatest diversity in the degree of development of the maternal-embryonic relationship. Viviparity has been the subject of numerous reviews, several of which are solely concerned with the phenomenon as it occurs in fishes (Amoroso, 1960; Hoar,

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1969; Hogarth, 1976; Wourms, 1977, 1981; Amoroso *et al.*, 1979; Dodd, 1983; Wourms *et al.*, 1988).

Oviparity and viviparity are well recognized as two contrasting, successful modes of reproduction that occur among both vertebrates and invertebrates. Oviparity, or egg-laying, refers to the release of propagules that are enclosed within an egg envelope (*i.e.*, a "shell," jelly-coat, or egg-case) from the female reproductive tract into the external environment. A young organism hatches from the egg envelope. There are three major categories of oviparity, namely *ovuliparity*, *zygoparity*, and *embryoparity* (Blackburn *et al.*, 1985; Wourms *et al.*, 1988) that encompass conditions ranging from release of an ovum from the female reproductive tract to the release of a well-developed embryo surrounded by an egg envelope. Ovuliparity refers to the release of ova from the female reproductive tract followed by their subsequent fertilization or activation within the external environment. All organisms that undergo external fertilization, including most teleosts, are ovuliparous. Zygoparity refers to the oviparous reproductive mode in which fertilized ova (*i.e.*, zygotes, the product of egg-sperm fusion) or gynogenetically activated ova are retained within the female reproductive tract for short periods. Zygoparous reproduction characterizes all skates, some sharks and some teleosts. Embryoparity is the pattern of oviparous reproduction in which a definitive embryo is formed and may develop to an advanced state prior to its release from the female reproductive tract and subsequent eclosion (=hatching) from the egg envelope. The extreme limits of embryoparity overlap with viviparity (see Wourms *et al.*, 1988).

Some species have reproductive modes that lie within an area of overlap between embryoparity and viviparity. In advanced stages of embryoparity, if the time at which eclosion from the egg envelopes is accelerated so that it takes place while the embryo is still residing within the female reproductive tract, then a free-living neonate will be released from the maternal organism rather than an embryo surrounded by an egg envelope. Timing of eclosion relative to the overall program of development appears

highly variable among viviparous taxa, suggesting that the timing of eclosion is plastic and readily subject to evolutionary change. The various sequences in which the four critical events of early life history; ovulation, fertilization, eclosion from an egg envelope, and release from the female reproductive tract, can occur, as well as their significance in the transition between oviparity and viviparity, have been discussed at length by Wourms (1981) and Wourms *et al.* (1988).

We have defined viviparity in the context of the fishes (Wourms *et al.*, 1988, p. 32) as a reproductive mode, "in which eggs are fertilized internally and are retained and undergo development in the maternal reproductive system. Hatching (that is, eclosion from an egg envelope if one is present) precedes or coincides with parturition, and the result is a free-living fish." Other forms of internal-brooding in abdominal folds of pipefishes and oral cavities of teleosts (see Balon, 1975, 1977, 1984) are excluded by our definition of viviparity. Thus, we focus our investigations of piscine viviparity on the phenomenon of embryo-retention within the female reproductive tract and a consideration of its variations among contemporary taxonomic groups. Because the great majority of animals including fishes reproduces by laying eggs and because piscine viviparity seldom occurs among the primitive species within a taxonomic group, we consider it to be a more specialized and derived mode of reproduction that has independently evolved from oviparity within numerous, taxonomically divergent, contemporary, piscine groups.

Here, we reflect on some of our emerging thoughts on the evolution of piscine viviparity and its implications. This paper is a transitional statement to which we shall return in the near future (Lombardi and Wourms, in preparation). Our inquiries into the evolution of piscine viviparity possess a threefold significance because: 1) they provide an overview of the evolution of viviparity from oviparity among fishes; 2) they should contribute to an understanding of the evolution of viviparity among tetrapod vertebrates, and 3) they can be used as a vehicle for studying the evolution of devel-

opmental pathways and the role of ontogeny in evolution. Due to constraints of space and our desire to avoid undue duplication, we frequently and extensively refer to information contained in our previous three reviews of this series, *viz.*, chondrichthyan reproduction and development (Wourms, 1977), a systematic and comparative overview of piscine viviparity (Wourms, 1981), and an analysis of the maternal-embryonic relationship in viviparous fishes (Wourms *et al.*, 1988). These publications contain many references to pioneering and contemporary bodies of work on piscine viviparity and related subjects.

PISCINE VIVIPARITY AND ITS CONSEQUENCES

Viviparity has evolved in five of the nine major groups of fishes (Atz, 1985, Fig. 1). No evidence of viviparity has ever been reported among the jawless fishes, including the lampreys and the hagfishes. In contrast, viviparity is the dominant mode of reproduction among the cartilaginous sharks and rays with 513 species, representing 40 families, known to give birth to living young. A minority of the sharks and all of the skates are oviparous, however. Their cartilaginous sister group, the chimaeras or ratfishes, are oviparous with the possible exception of a single fossil form (Lund, 1980). Viviparity appears to be more dominant among the chondrichthyans than it is among the bony fishes. Of the approximately 900 living species that comprise these two groups of cartilaginous fishes, 55% are viviparous. Among the five major groups of bony fishes, viviparity is widespread but far less prevalent; only about 510 of an estimated 20,000 or more species (about 2–3%) have been described as viviparous. Nearly all of these occur in 13 or 14 families of teleosts, which are the most advanced, or modern, of fishes. Neither the lungfishes (Dipnoi) nor the bichirs (Cladistia) show any signs of viviparity, and only a few fossil relatives of the sturgeons and paddlefishes (Chondrostei) give any evidence of being viviparous (Bürgin, 1990). *Latimeria*, the living coelacanth, is viviparous, and so were at least some of its fossil relatives (Smith *et al.*, 1975; Wourms *et al.*, 1991). We estimate that

viviparity evolved from ovoparity on as many as 18 occasions in the chondrichthyans and on as many as 24 occasions in the osteichthyans. Thus, it appears that piscine viviparity may have arisen approximately 42 times in the fish-like vertebrates.² Among the various taxonomic assemblages of viviparous sharks, rays, and teleosts there is an array of species that display intermediates between a primitively viviparous state (lecithotrophy) in which embryos are essentially metabolically autonomous, to a specialized viviparous state (matrotrophy) with a high degree of maternal trophic dependency (Amoroso, 1960; Hoar, 1969; Wourms, 1981; Wourms *et al.*, 1988). Among the vertebrates, only extant mammals are represented by more viviparous species than are the fishes.

We believe that fishes provide a key to understanding the evolution of vertebrate viviparity. Our operating hypothesis is that two critical evolutionary steps took place in fishes. First, there was the innovation of the basic structural and functional characteristics of the vertebrate female reproductive system, namely, 1) the vertebrate pattern of genital tract development and sex differentiation, 2) neuroendocrine mechanisms involved in the control of reproduction, and 3) the vertebrate ovary and oviduct. Secondly, the basic developmental program of the vertebrate lineage was established in the fishes. Inasmuch as these features are critical for reproductive success, once they were established, they imposed constraints on possible pathways of further successful evolutionary innovation. Thus, the evolution

² These estimates should be regarded as a tentative first approximation of the number of independent events in the evolution of piscine viviparity. To arrive at this number, some major assumptions were made. We employed principles of cladistic analysis and treated viviparity as an irreversible character. We have assumed that if a taxon is represented only by viviparous forms, then it was derived from a viviparous ancestral stock. We have also assumed that the occurrence of both oviparity and viviparity within a taxon is evidence of independent evolutionary events within that taxon. Both of us believe that such analyses may be limited by the absence of robust phylogenies for many groups, and one of us (J.L.) believes that analyses may be further limited by not considering some forms of embryoparity as being secondarily derived from viviparity.

of vertebrate viviparity involves: 1) a shift from external to internal fertilization; 2) embryo retention within the female reproductive system; 3) utilization of the ovary or oviduct as a site of gestation; 4) structural and functional modification of the embryo and the female reproductive system; and 5) modification of extant endocrine reproductive control mechanisms (see Hoar, 1969; Wourms, 1981; Wourms *et al.*, 1988; Amoruso *et al.*, 1979; Dodd, 1983; Nagahama, 1983; Shine, 1985; Callard and Ho, 1987; Callard *et al.*, 1988, 1989).

EVOLUTION OF VIVIPARITY IN FISHES

Initial steps in the evolution of viviparity from oviparity involve a shift from external to internal fertilization followed by retention of developing eggs and embryos within the female reproductive system. In fishes, the acquisition of internal fertilization is the primary factor that limits the evolution of viviparity from ovuliparity. Although internal fertilization is considered a prerequisite for viviparity, it is important to realize that internal fertilization has evolved independently among many different groups of oviparous fishes. All contemporary cartilaginous fishes undergo internal fertilization. Introduction of male gametes into the female genital tract is effected by claspers, paired intromittent organs that arise from specialized regions of the male pelvic fins (Compagno, 1990). Approximately 55% of the extant chondrichthyans are viviparous. Among the teleosts, approximately 3% of all families undergo internal fertilization and 57% of those are viviparous (Wourms, 1981; Gross and Shine, 1981; Gross and Sargent, 1985). Internal fertilization is facilitated by numerous independently evolved modifications of the male anal fin (*e.g.*, gonopodia in poeciliids). Gonopodial structures are described in three genera of fossil actinopterygian fishes from the middle Triassic, one of which is known to be viviparous (Bürgin, 1990).

Contemporary species of sharks exhibit modes of reproduction that could very well represent stages in an evolutionary sequence from zygoparous-oviparity to viviparity (Nakaya, 1975; Dodd and Dodd, 1986; Wourms *et al.*, 1988). All sharks undergo

internal fertilization. Species representing intermediate stages in the evolutionary sequence from zygoparity to viviparity are especially evident within the carpet sharks (Orectolobidae) and the catsharks (Scyliorhinidae). Such transitional species are characterized by: 1) retention of developing embryos in the oviduct for extended periods, 2) a tendency toward the presence of several eggs within the oviduct, each of which contains an embryo at a stage of development corresponding to its place in the temporal sequence in which the eggs were ovulated (=multiple oviparity) (Nakaya, 1975), 3) reduction in thickness of the egg envelope, and 4) loss of egg envelope ornamentation used for attachment of the egg-case to the substrate. Among the different species within the catshark genus *Halaehur*, a nearly complete set of intermediates between zygoparity and viviparity is evident (see also Wourms *et al.*, 1988).

Intermediate stages between ovuliparous oviparity and viviparity occur within the teleostean family Scorpaenidae which include rockfishes and scorpionfishes (Wourms, 1991). Most families of scorpaenoid fishes are oviparous and spawn pelagic eggs, except those within the subfamily Sebastinae which are mostly viviparous. Fishes with the least specialized reproduction pattern, such as *Inimicus* are ovuliparous, broadcast spawners of individual eggs that hatch into altricial larvae. Fertilization is external. In *Scorpaena* and *Sebastobus*, there is a shift from a primitive to specialized mode of spawning. Fertilization is still external, development is ovuliparous, but eggs are embedded in a gelatinous matrix. Larvae are altricial. Transitional stages from the specialized mode of scorpaenid oviparity, to viviparity occur within the genus *Helicolenus*. At one extreme, species such as *H. dactylopterus* display a zygoparous or embryoparous form of oviparity and fertilization is internal. Fertilized eggs at stages of development from the late blastoderm through embryogenesis are deposited in a pelagic, gelatinous matrix in which development is completed, hatching thus occurring some time after oviposition. The advent of viviparity occurs in fishes such as *Helicolenus percoides* and some species of

Sebastes, e.g., *S. schlegeli*. In *H. percoides*, fertilization is internal and all of embryonic development takes place within the female reproductive system. During parturition, females extrude a large gelatinous matrix that encloses 80,000 or more embryos which leave the matrix within 20 min. The definitive stage of rockfish viviparity is found in most species of *Sebastes*. The gelatinous matrix is absent and there is a shift in embryonic nutrition from lecithotrophy to a modified form of matrotrophy.

SELECTIVE ADVANTAGES

The success of various forms of oviparity in fishes is evident by the diversity of extant species which exhibit this reproductive mode. During the transition from oviparity to viviparity in many groups, the advantages of prolonged embryo-retention and nutritional dependency by embryos on the maternal organism must have outweighed the disadvantages. Models of life history strategies (see Stearns, 1976, for discussion) predict that viviparity, usually characterized by the production of a few well-developed offspring, is adaptive when the environment is stable and competition is intense (K-selection), or when a fluctuating environment encourages hedging. Attempts to account for the evolution of viviparity through this approach have met with limited success because of the difficulty in finding clear cut examples. Having said this, we wish to emphasize that the major advantage of the life history model is that it forces an examination of the reproductive biology of each viviparous species in terms of its overall life history and ecology (Shine, 1985). Valid generalizations may be drawn through this approach by comparing and contrasting reproductive strategies and life histories between species. Thibault and Schultz's (1978) study of reproductive adaptations in viviparous poeciliid fishes is an example of the utility of this approach. Regrettably, attempts to analyze the evolution of piscine viviparity have been hindered by a tendency toward over-generalization and oversimplification because of an inadequate data base of the characters involved. The task is complicated because one must consider all the different kinds of sharks and rays, the

coelacanth, and a number of very dissimilar teleosts. Not only do these fishes represent three distinct evolutionary lineages, but they are considerably different in size.

For viviparous reproduction to evolve, it must offer selective advantages. Most obviously, retention of embryos in the female's reproductive tract affords them increased protection from predation. Predation of eggs and larvae developing in the water column can result in a high mortality rate, i.e., 30–40% per day for planktonic marine eggs and larvae (Hunter, 1981). Survival of offspring may also be enhanced because the physiological homeostasis maintained by the maternal fish serves to regulate the environment of embryos within her genital tract. This relationship is highly efficient because the mass of the maternal fish greatly exceeds that of single embryos and is always much greater than the total mass of the developing embryos. However, this arrangement is costly to the female (*vide infra*). Another advantage may be realized through the release of precocial young from the maternal reproductive tract. The degree of precocity varies considerably. On the one hand, new-born young of the viviparous rockfish, *Sebastes*, are at a developmental stage only slightly more advanced than young of oviparous scorpaeniform fishes (Washington *et al.*, 1984; Wourms, 1991); on the other hand neonatal sand-tiger sharks and most other oophagous lamniform sharks are experienced predators in excess of a meter in length, and new-born males of certain surperches (Embiotocidae) are sexually mature (see Wourms, 1981; Wourms *et al.*, 1988). The advantages accruing to precocial offspring are those of overall greater fitness as indicated by a higher trophic level, increased swimming speed and locomotor control, and the fully differentiated state of their sense organs and other organ systems. Precocity also avoids the biological and physiological demands associated with various degrees of metamorphosis through which altricial fish larvae must pass (Moser, 1981; Youson, 1989). Finally, because the young of viviparous and brooding species are almost always larger than those of oviparous congeners, a number of advantages may accrue to large-sized neonates. Large size in neo-

nates may reduce their risks from predation and also increase the food-resource base by raising the offspring's trophic level in the food chain (Wourms, 1977). Matrotrophic provision of nutrient reserves within the body of precocial neonates may also serve to increase the survival of offspring within environments with spotty resource distribution.

A set of advantages accrue to viviparous species that have evolved from oviparous congeners that have low fecundity. Viviparous species, because of enhanced survival of individual offspring, are able to produce as many, if not more, surviving young than do their oviparous congeners from egg clutches equivalent in number. Because of the difference in absolute size and evolutionary constraints, cartilaginous fishes and teleosts must be treated separately. Elsewhere (Wourms, 1977, 1981), we have postulated that the high incidence of viviparity and its repeated occurrence in divergent taxa of chondrichthyan fishes have taken place because viviparous species produce as many, if not more, offspring than oviparous species. Many oviparous chondrichthyans produce a moderate number of relatively large eggs (100–200 eggs/year in the most fecund species such as skates and catsharks) and do not exercise parental care. Incubation times can be quite long (from 1–2 months in warm-water skates to an estimated 12 months in deep-water catsharks), thus presenting a long-term risk of predation (Dodd, 1983). Viviparous chondrichthyans do not produce significantly fewer eggs than their oviparous congeners (brood sizes as high as 50–100 are reported in blue sharks and cow sharks [Ballinger, 1978]) but the probability of survival of the offspring that result from these eggs is much greater.

Among teleost fishes, a different set of circumstances is encountered. Teleost egg size tends to be relatively uniform, falling within a 1–10 mm range, and clustered about a mean value of about 1–2 mm. Large egg size is both a primitive condition as well as a derived, specialized feature (*e.g.*, in mouth brooders [Marshall, 1953; Balon, 1975, 1977, 1984; Elgar, 1990]). As a group, teleosts, both oviparous and viviparous, produce eggs of small absolute size. Among

fishes, fecundity is a function of body size so that larger individuals are more fecund than smaller ones of the same species. Thus, it follows that among different species of teleosts that have eggs of similar size, individuals of fish species that reproduce at a small size are intrinsically less fecund than individuals of a given species that reproduces at a large size. Size is an important but neglected factor because the average length of all the 20,000–25,000 species of teleosts is 150 mm (Marshall, 1971). In the case of ovuliparous broadcast spawners, one would predict that under idealized conditions, the larger, more fecund species would produce more surviving offspring. Under these circumstances, it would be selectively advantageous to the individuals of small species to evolve reproductive strategies that would significantly increase offspring survival. A variety of strategies have evolved (Breder and Rosen, 1966; Balon, 1975, 1981, 1984). Viviparity, brooding, and other forms of parental care must significantly enhance offspring survival. We predict that viviparity should be more common among small teleosts than large ones. In general, this prediction holds true. Of the 13 or 14 families of viviparous teleosts, only the Anablepidae and Scorpaenidae are moderately large to large fishes (300 mm and longer). In both of these cases, viviparity offers other advantages (*vide infra*).

Viviparity may also enhance reproductive success by amplifying the number of reproductive niches within a shared habitat. By avoiding the competition involved in the establishment and maintenance of spawning or nesting sites and territories, viviparity may be advantageous in a habitat with a high diversity of species and a high density of individuals and where reproduction tends to be confined to an optimal season of the year. Where the number of conventional reproductive niches is limited, smaller teleosts might be expected to evolve reproductive strategies that involve parental care, such as nest-tending, brooding, or viviparity. Most forms of parental care occur within a fixed environmental space shared with other individuals of the same or different species. Viviparity may serve to decrease competition for space by permitting

embryos to escape in space; the site of reproduction is transferred from the commonality of an external niche to the monopoly of an internal one.

Reduction of reproductive competition in shared habitats, a consequence of viviparity, may be one of the selective advantages involved in the evolutionary origin and geographical spread of the viviparous rockfish *Sebastes*. *Sebastes* originated in the Northwest Pacific as part of the fauna associated with the expansion of cold water that occurred about 13 million years ago (Kendall, 1991). Nearly all members of the genus have successfully adopted a reproductive strategy that combines the fecundity of oviparity with the enhanced survival of viviparous young. In cottids, reproduction is oviparous and females lay clusters of adhesive demersal eggs. Parental care is common (Washington *et al.*, 1984). *Sebastes*, cottids, and other cold-water scorpaeniform fishes spread into the North and Northeast Pacific and occupied niches made available by displacement of the original warm-water fauna. Lack of reproductive competition between the two colonizing groups permitted them to exploit available niches fully. As a result, *Sebastes*, other scorpaenids and cottids underwent extensive speciation and now dominate the fish fauna of the coastal waters of the northeastern Pacific. Similar scenarios may have occurred during the radiation of surfperches (embiotocids) of the north Pacific, the comephorids of Lake Baikal, and possibly the viviparous coral reef ophidioids and clinids of rocky in-shore marine habitats as well (see Wourms, 1981, 1991).

The evolution of viviparity may also have facilitated the exploitation of pelagic niches. Chondrichthyans provide the best examples of a shift from benthic, oviparous species to pelagic, viviparous species (Tortonese, 1950; Wourms, 1977). According to Compagno (1990), the littoral ecomorphotype is perhaps the most primitive among chondrichthyans and may be a predecessor to specialist ecomorphotypes including several pelagic types. Littoral, benthic species of sharks and skates (a taxon that evolved from oviparous sharks or a shark-like ancestral form) are oviparous. Pelagic sharks and pelagic species of electric rays (Torpedini-

formes) and stingrays (Myliobatiformes) are viviparous. Viviparity which appears to have evolved independently in some sharks and all of the rays allows successful reproduction in a pelagic habitat. Viviparity and the production of precocial offspring is a successful reproductive strategy in pelagic organisms such as sharks and rays where both adult and newborn young must maintain their position in the water column. However, it is not the most common way of becoming reproductively pelagic. Nearly all oviparous teleost fishes with generalized reproductive strategies lay either demersal or pelagic eggs. A similar situation also occurs among marine invertebrates. The adaptive value of a pelagic reproductive strategy to a pelagic organism is obvious. The two egg types represent: 1) a simple way of partitioning reproductive niches in the aquatic environment, even in shallow littoral waters, and 2) a means of invading deep pelagic waters. Chondrichthyan fishes appear to lack the option of producing buoyant, pelagic eggs. In them, the combination of internal fertilization and the oviposition of large, non-buoyant fertilized eggs in egg-cases are primitive, unspecialized features that evolved early and have been retained in extant species with unspecialized modes of reproduction. They pose strong developmental constraints that do not favor the production of pelagic eggs but do favor egg-retention and viviparity. Unable to alter their reproductive patterns in order to partition the available reproductive niches, sharks and rays invaded the pelagic realm through evolution of a new reproductive niche, viviparity. The evolution of viviparity along with the exploitation of pelagic niches has also occurred among a number of teleostean families (for example, comephorids and hemirhamphids).

Exploitation of unoccupied ecological niches in a pelagic habitat is part of a broader issue of colonization of new habitats. Viviparity may facilitate dispersal of a population and colonization because a single gravid female has the potential to colonize new habitats. If one takes into account the fact that storage and the production of multiple broods from single matings are well developed in several viviparous species, then it

is clear that the reproductive potential of individual females becomes even greater. Previously, we noted that the evolution of viviparity in *Sebastes* may have facilitated their colonization of newly available habitats by amplifying the number of reproductive niches and thus avoiding reproductive competition. One additional factor is that viviparity in highly fecund fish such as *Sebastes* (where brood sizes of up to 5 million are attained) may aid in colonization. Similar factors appear to affect the colonizing abilities of sharks. Barlow (1981) suggests that viviparity in chondrichthyans may facilitate dispersal and notes that small benthic sharks, which are often oviparous, are conspicuously absent from coral reefs.

Finally, viviparity may be advantageous in species whose embryos derive metabolites from the maternal organism during gestation (=matrotrophic viviparity) because of energetic considerations. In matrotrophic species, nutrients required for embryonic development are provided on demand, rather than being sequestered in the eggs prior to the initiation of development. Matrotrophy also serves to reduce the energetic losses that would be incurred through gamete wastage and embryonic death. The ability to provide developing embryos with nutritional resources during either oogenesis or gestation gives the maternal organism an additional advantage in allocating environmental resources into reproduction (Thibault and Schultz, 1978; Thibault, 1979). Evidence of an organism's ability to balance oogenetic and gestational nutrient allocation is now emerging (Cheong *et al.*, 1984; Reznick and Miles, 1989; Trexler, 1985, 1990) and may be a significant factor in the evolution of superfetation in poeciliids and clinids. Matrotrophic viviparity also frees the developing embryo from the constraints imposed by the fixed metabolic content of the egg and thus permits it to grow to larger size. Most notable, in this regard, are the viviparous lamniform sharks in which oophagy facilitates neonatal gigantism.

It would be inappropriate to discuss advantages of viviparity without pointing out some of its possible disadvantages. First, it must not be forgotten that although ovi-

parity is the primitive and, at least in theory, the unspecialized mode of reproduction, it is nonetheless a highly successful mode evident among a great majority of teleost fishes including most of the more advanced taxonomic groups. We consider the disadvantages of viviparity to be related to the following: 1) brood loss resulting from maternal death; 2) energetic cost to the female; and 3) reduced fecundity. The first point is obvious, death of the maternal fish almost always results in the death of her brood. Exceptions may occur when term young are spontaneously released from the female in response to trauma. Energetic costs to the maternal organism fall into two categories: a) increased costs associated with increased maternal bulk and mass (*i.e.*, increased risk of predation of the discommoded gravid fish), and b) the energetic cost of supporting metabolism of an entire brood (see Boehlert and Yamada, 1991). If brood size is reduced to lessen these constraints, the female must spend a disproportionate part of her maturity in the production of each smaller brood. Indeed, fecundity is typically lower in viviparous fishes than in comparably sized oviparous ones. Fecundity may be limited by the relative size of the maternal organism and there may exist a trade-off between increased survival of viviparous young and absolute fecundity. In the most fecund of all viviparous teleosts, the rockfishes, neonates appear to be only slightly more developed than their newly hatched oviparous relatives.

NOVEL MATERNAL-EMBRYONIC RELATIONSHIPS

The transition from oviparity to viviparity is marked by the establishment of novel maternal-embryonic relationships involving the: 1) trophic, 2) osmoregulatory and excretory, 3) respiratory, 4) endocrinological, and 5) immunological needs of the developing young. The most obvious and up to now, the most studied of these relationships is the trophic one. Patterns of embryonic nutrition evident among viviparous fishes range from strict lecithotrophy to extreme forms of matrotrophy. Lecithotrophic embryos, whether oviparous or viviparous, derive their nutrition solely from

yolk reserves. Matrotrophic embryos supplement endogenous yolk reserves by sequestering maternally derived nutrients during the course of gestation (Wourms, 1981; Wourms *et al.*, 1988). We will not discuss the other relationships in detail because of spatial limitations, the fact that they have been reviewed recently, and because the relative sketchiness of data available in the other four categories precludes a meaningful comparative analysis (see Wourms *et al.*, 1988, for an extensive review of maternal-embryonic relationships as well as recent pertinent reports in Boehlert and Yamada, 1991; Callard *et al.*, 1988; Callard *et al.*, 1989; Fasano *et al.*, 1989; Ingermann, 1991; Kormanik, 1991; Nakanishi, 1991).

EVOLUTION OF SITES OF MATERNAL-EMBRYONIC METABOLIC EXCHANGE

The morphology of the female reproductive system determines the sites of gestation in viviparous fishes and it, in turn, is the end-result of the evolutionary history of major piscine taxonomic units (*vide infra*). In fact, the basic, unspecialized pattern that distinguishes the female reproductive system of all the vertebrates first manifested its definitive character in chondrichthyans. This pattern, which has been retained by the chondrichthyans, also occurs in some primitive actinopterygians (*e.g.*, sturgeons) and has been carried through the sarcopterygian lineage (lungfishes and coelacanth) to the tetrapods (*e.g.*, amphibians, reptiles, birds, and mammals).

In chondrichthyan fishes as well as the sarcopterygian fishes, the female reproductive system consists of single or paired ovaries with either one or two oviducts. The oviducts are embryological derivatives of the Müllerian ducts (Wourms *et al.*, 1988). In viviparous chondrichthyans and the living coelacanth, the posterior part of the oviduct is highly modified and called a uterus. Fertilized eggs are retained and develop to term within the uterus. Teleost fishes, however, are an apparent anomaly. The ovary of viviparous teleost fishes differs from other vertebrates inasmuch as it is the site of both egg production and gestation. Gestation in

viviparous teleosts occurs either in the ovarian lumen (intraluminal gestation) or in the ovarian follicle (intrafollicular gestation) (Wourms *et al.*, 1988). Intraluminal and intrafollicular gestation are the consequences of the specialized condition of the teleost female reproductive system. The currently accepted view is that teleosts lack an oviduct, the term "oviduct" being restricted to a structure derived from the Müllerian duct (Hoar, 1969; Wake, 1985). There is a consensus that the Müllerian ducts, which are present in primitive actinopterygian fishes, were lost during teleostean evolution for reasons as yet unknown. In both instances, the supporting evidence is weak (*vide infra*).

Intraluminal gestation is the prevalent mode of development in viviparous teleosts. It occurs in slightly more than half of the viviparous species and in 10 of the 14 or 15 families in which viviparity is known. In most teleosts with intraluminal gestation, fertilization and embryonic development commence in the ovarian follicle and proceed to completion in the ovarian lumen. Only in viviparous zoarcids and scorpaenids does ovulation precede fertilization. Diverse specializations of maternal and embryonic tissues have evolved to facilitate physiological exchange (Wourms, 1981; Wourms *et al.*, 1988). Intrafollicular gestation occurs in clinids, some labrisomids, the poeciliids, and the anablepid *Anableps*. In these forms, ova are fertilized within the ovarian follicle and the developing embryos are retained within it until they are released at the time of parturition. The most remarkable specialization associated with intrafollicular gestation is the post-fertilization follicle. In most oviparous and viviparous teleosts, ovulation occurs prior to or soon after fertilization, and the follicle then degenerates. In species with intrafollicular gestation, however, the follicle remains intact after fertilization and undergoes changes to accommodate the requirements of embryonic development within its confines. During gestation, the follicle wall must function in gas exchange, nutrient transfer, maintenance of the osmotic environment of the embryo, and possibly the protection of embryos from immunological rejection. In

species that have been examined in detail, functional change is clearly reflected by the structural differences between the pre- and post-fertilization follicles (Wourms *et al.*, 1988).

Viviparous teleosts possess a cystovarian ovary in which the germinal epithelium lines the internal surface of the ovarian lumen. The teleostean ovary is formed by the folding of the embryonic gonad into a sac so that the inner (luminal) epithelium is derived from the coelomic (peritoneal) mesodermal epithelium. In most teleosts, the luminal epithelium and associated germinal tissue undergo extensive folding to form ovigerous folds or sheets that project into the ovarian lumen. In all teleosts, unfertilized ova, fertilized ova, developing embryos, or highly developed young are released into the lumen of the ovary unlike most other vertebrates in which unfertilized ova are released into the coelomic cavity. During development, the teleostean ovary is enclosed within a capsule formed by peritoneal (coelomic) folds. In viviparous teleosts, the wall of the ovary is continuous with a gonoduct that extends posteriorly and opens to the exterior at the genital pore. The gonoduct is formed by the posterior growth of the ovarian capsule (Amoroso *et al.*, 1979; Dodd, 1977; Hoar, 1969; Wourms, 1981; Nagahama, 1983; Wallace and Selman, 1990). The state of knowledge concerning the embryonic origin and morphogenesis of the teleost ovary and its associated duct systems is not satisfactory since the handful of older studies is limited to only a few species (reviewed in Goodrich, 1909, 1930; Nelsen, 1953). The problem needs to be reinvestigated using modern conceptual and technical approaches like those of Begovac and Wallace (1987). The critical issue is whether the gonoduct of the cystovarian ovary in viviparous teleosts is derived from the Müllerian duct and is homologous to the oviduct of other vertebrates. Recently, Wake (1985), who has concluded that the gonoduct is not homologous to the Müllerian duct, has suggested an homology between teleost viviparity and that of other vertebrate groups. In principle, we agree with her but take a somewhat different approach.

The cystovarian teleostean ovary func-

tions as both an ovary and oviduct. We choose to avoid the unresolved issue of homology. During intraluminal gestation the teleostean luminal epithelium serves the same function as the luminal uterine epithelium of chondrichthyans, sarcopterygians, and tetrapods. We suggest that the luminal epithelium of the cystovarian teleost ovary and the uterine luminal epithelium, are evolutionary parallelisms because although their morphogenesis differs, both epithelia are derived from adjacent regions of the urogenital plate (*i.e.*, coelomic mesoderm). Thus, it is not surprising that the ovarian luminal epithelium of viviparous fishes with luminal gestation can undergo a major increase in its surface area, and vascularization for gas exchange, and establish transport and secretory functions. In point of fact, even the ovarian luminal epithelium of oviparous teleosts is capable of responding to endocrine stimulation by acquiring transport and secretory functions (Nagahama, 1983). In the case of the oviduct and the teleostean ovary, explorations of the issues of homology, parallelism, and convergence must go beyond the organ and tissue level to a consideration of homologous gene expression during the differentiation of functionally equivalent cell types (Kosswig, 1948; Rensch, 1960).

In fishes with intrafollicular gestation, the luminal epithelium of the ovary assumes a less important role and the ovarian follicle functions both in egg production and as the site of gestation. Intrafollicular gestation may be considered a prime example of evolution-by-tinkering; inasmuch as follicular function has been merely co-opted and two relatively minor changes have been introduced into the reproductive program. First, the morphology of the follicle-ovum unit has been structurally modified to permit intrafollicular fertilization. It is entirely possible that the initial evolutionary stages of these modifications may have been associated with internal fertilization and sperm storage. Moreover, in some chordates (*e.g.*, ascidians and mammals) the ovulated egg is invested by follicle cells that pose no serious barrier to fertilization (Wourms, 1987). Secondly ovulation has been repressed during gestation and replaced by parturition at

term (Turner, 1942). Finally, in matrotrophic species, the basic function of follicle cells in transporting essential metabolites to the differentiating oocyte has been retained and co-opted for embryonic growth and development in matrotrophic species (Wourms, 1987; Wallace and Selman, 1990). Efficiency of the trans-follicular carriage of metabolites should not be underestimated; it is responsible for the dramatic growth of ova in some sharks and the living coelacanth (*e.g.*, ova with dry masses of up to 185 g and diameters of up to 90 mm).

Embryonic uptake of maternally-derived nutrients takes place across two major types of embryonic epithelial surfaces, the integument and the gut. Integumental or dermotrophic transfer takes place across the epithelium of the general body surface and its derivatives. Transfer sites include: 1) general body surface (*e.g.*, in clinids and early *Heterandria* embryos), 2) gill filaments (*e.g.*, in sharks and rays), 3) finfolds and fin epaulets (*e.g.*, in some goodeids and surfperches), 4) yolk sac surfaces (*e.g.*, in sharks, rays and the living coelacanth), 5) pericardial sac surfaces (*e.g.*, in some poeciliids and goodeids), 6) surfaces of the pericardial amniochorion (*e.g.*, in *Heterandria* and other poeciliids), and 7) the pericardial trophoderm (*e.g.*, in the four-eyed fish, *Anableps*). Gut-associated or enterotrophic transfer takes place across the epithelium of the gut and gut derivatives. Transfer sites include: 1) the gut (*e.g.*, in sharks, rays, surfperches, and *Anableps*), 2) the branchial portion of the branchial placenta (*e.g.*, in *Jenynsia*, some rays), and 3) trophotaeniae (*e.g.*, in perianal extensions of the intestinal epithelium present in goodeids and a parabrotulid, a surfperch, and two ophidioids (see Wourms *et al.*, 1988, for discussion).

Three transfer sites are of particular interest: the pericardial amniochorion, the yolk sac placenta, and trophotaeniae. The pericardial amniochorion which occurs in the embryos of poeciliid fishes, some goodeids and in a modified form (=pericardial trophoderm) in *Anableps* is of particular interest because of its striking similarity to the anterior amnio-chorionic fold of amniotes. This is an apparent anomaly in that the amnion and chorion first appear in the tetrapod lineage as one of the major steps in the evo-

lution of the terrestrial cleidoic egg (Needham, 1942; Mossman, 1937, 1987). The pericardial amnio-chorion is derived from "extra-embryonic" ectoderm and mesoderm of the pericardial sac that expands by differential growth to form a hood-like fold that grows in a dorsal and posterior direction and envelops the head of the embryo during early and mid-phases of development. Subsequently, the structure regresses. It appears to function in the uptake of nutrients and other metabolites as well as gas exchange (Grove, 1985; Knight *et al.*, 1985; Grove and Wourms, 1991; see Wourms *et al.*, 1988, for a review and summary of earlier work). Besides being a remarkable example of evolutionary convergence in an extraembryonic membrane, it helps one to understand the developmental basis for the evolution of the amnion and chorion in amniotes. Poeciliids, which are modern teleosts, are highly modified and evolutionarily as far removed from the basic piscine-vertebrate lineage as are the amniotes. By definition, no *direct* homology can be established between the two amniochorionic structures; however, teleosts and amniotes share large portions of a basic developmental program that is common to all vertebrates. The occurrence of an amniochorionic membrane in fishes and amniotes is clearly a convergence but its fundamental form is unique to the vertebrates and is found nowhere else. This fact suggests that although vertebrates can freely adapt their embryonic epithelia for the uptake and exchange of metabolites, the patterns that these adaptations assume are narrowly constrained. Such constraints are inherent in the vertebrate design, are phylogenetically established, and are manifested in individual life histories as fixed algorithms in the developmental program. Stated alternatively, the developmental anlagen and phylogenetic antecedents of both extraembryonic membranes are mutual homologues. Because the homologues are complex integrated structures, subsequent modification of them is restricted to a few, narrow functional pathways. Thus, the evolution of what appears to be the same developmental adaptation in two distinct lineages in response to two different sets of selective forces, *i.e.*, viviparity or terrestriality, sug-

gests that the shared propensity to undergo this change in the developmental program may be established by developmental constraints and is promoted by epigenetic facilitation (Kauffmann, 1983; Maynard Smith *et al.*, 1985; Müller, 1990). Selective factors leading to the evolution of vertebrate extraembryonic membranes are discussed elsewhere.

The juxtaposition of maternal and embryonic sites of metabolic exchange and the selective advantage of more efficient transport processes has led to the evolution of different kinds of placentae among diverse groups of viviparous fishes. Placenta is used here in its modern sense, as proposed by Mossman (1937, p. 156) and subsequently adopted by other authorities, among them Amoroso (1952, 1960): "An animal placenta is an intimate apposition or fusion of the fetal organs to the maternal (or paternal) tissues for physiological exchange." We recognize four classes of placental relationships among viviparous fishes: 1) yolk sac placenta, occurring in some sharks; 2) follicular placenta, an intimate apposition between follicular epithelium and embryonic surface in poeciliid and other fishes; 3) branchial placenta, an association between villi of oviductal luminal epithelium and the branchial region of embryonic rays; and 4) trophotaenial placenta, *i.e.*, perianal extensions of the embryonic intestinal epithelium that lie in proximity to the maternal ovarian luminal epithelium of goodeids, a parabrotulid, a surfperch, and two ophidioids (Wourms *et al.*, 1988). Mossman's definition of placentation shifts the emphasis from criteria that depend on the stereotyped patterns of extraembryonic membranes in the amniote vertebrates and toward criteria based on the functional role of parental and embryonic tissues in physiological exchange. His definition extends the placental concept to include many of the wide variety of parental-embryonic exchange systems occurring in both viviparous invertebrates and vertebrates.

EVOLUTION OF THE TROPHOTAENIAL PLACENTA

Trophotaeniae, literally "growth ribbons," are external rosette- or ribbon-like structures that project from the embryonic

hindgut into the fluid-filled ovarian lumen of several different viviparous teleosts (Turner, 1937; Wourms *et al.*, 1988). Their structure and function have been most extensively investigated among the goodeid fishes. Trophotaeniae and the ovarian luminal epithelium constitute a trophotaenial placenta that facilitates substantial nutrient transfer to developing embryos. Among goodeids, two types of trophotaeniae occur: rosette trophotaeniae that consist of a series of short, lobulated processes and ribbon trophotaeniae that consist of long, slightly flattened ones. Trophotaeniae of both types consist of a simple surface epithelium surrounding a highly vascularized core of loose connective tissue. Trophotaenial epithelial cells are derived from the embryonic hindgut and exhibit the structural and functional characteristics of intestinal absorptive cells (Lombardi and Wourms, 1985*a, b*; 1988*a*). Cells of ribbon trophotaeniae take up macromolecules, possess an apical endocytotic complex, and are structurally organized in the open configuration characteristic of neonatal mammalian, intestinal, absorptive cells. Rosette cells transport small molecules, lack an apical endocytotic complex, and are structurally organized in the closed configuration characteristic of adult mammalian, intestinal, absorptive cells. Intestinal cells of both embryonic and adult goodeids endocytose macromolecules and are in the open configuration (Hollenberg and Wourms, 1986). This condition is so common as to be the rule in adult teleosts (Iida and Yamamoto, 1985; LeBail *et al.*, 1989; Vernier and Sire, 1989).

Any embryonic adaptation as spectacular as trophotaeniae, that has appeared independently in four distantly related orders of teleosts, invites inquiry into its evolutionary origin. We have proposed that trophotaeniae represent one culmination in an evolutionary sequence of adaptations of the piscine embryonic gut (Wourms, 1981; Lombardi and Wourms, 1988*a*). Here, we draw on comparative and developmental studies to propose a scenario for the evolution of trophotaeniae in the light of gut ontogeny and the evolution of other gut derived specializations. As regards both their ontogeny and phylogeny, trophotaeniae have

originated from a simple tubular embryonic gut. Once they have differentiated, their cells can absorb macromolecules and are in an open configuration. This condition prevails in the adult intestine of most fishes as well as larval amphibia and neonatal mammals (Colony and Neutra, 1985; Govani *et al.*, 1986; Stevens, 1988; Vernier and Sires, 1989). Two adaptations for viviparity are derived from the embryonic gut. In the first, hypertrophied villi develop as in the enlarged embryonic gut of surferperches, the eelpout, *Zoarces*, and the four-eyed fishes, *Anableps*. These facilitate the uptake of imbibed follicular or luminal fluid (Wourms *et al.*, 1988). In the second, trophotaeniae develop and by means of a process of differential growth, the hind-gut epithelium becomes externalized and forms short, projecting, ribbon-like prototypic trophotaeniae whose cells are initially in the open configuration. This represents the terminal state in species of surferperches, *e.g.*, *Rhacochilus vacca*, that exhibit trophotaeniae. The evolution of trophotaeniae appears to have been heterochronic, probably involving an accelerated expression of genes that regulate the onset, rate, and extent of intestinal morphogenesis and cell differentiation. As a result of non-allometric hypertrophy of gut tissue the epithelial surface of the terminal end of the hindgut that formerly faced the intestinal lumen becomes externalized through a process of growth-based eversion. Prototypic ribbon trophotaenia of goodeids had a dual evolutionary fate, one leading to the definitive ribbon trophotaenia and the other to the rosette trophotaenia. The definitive number of ribbon trophotaeniae arose from an increase in number of prototypic trophotaeniae as a result of branching and considerable axial elongation. The rosette trophotaenia first appeared as a prototypic ribbon trophotaenia. While retaining the bud-like morphology of a prototypic trophotaenia, its cells hyperdifferentiated from an open configuration, with an apical endocytotic complex which is presumed to be able to endocytose macromolecules, to a closed configuration which lacked an apical endocytotic complex and thus could not endocytose (Hollenberg, unpublished observations). Prototypic rosette trophotaenia

underwent moderate growth in a radial pattern to assume its definitive morphology. Its cells remain in the closed configuration. The closed configuration is a specialization for the transport of small molecules; it represents an evolutionary innovation. If one considers trophotaenial cells to be a type of intestinal epithelium, then the evolutionary innovation of the closed configuration in the rosette trophotaeniae of goodeid fishes does not put in any evolutionary appearance again until adult amphibians reptiles, birds and mammals. Among the amphibians, mammals, and apparently birds as well, a similar transition occurs either in their embryonic or neonatal stages (Luppa, 1977; Dauca and Hourdry, 1985; Stevens, 1988). Since trophotaeniae are efficient in the uptake of maternal nutrients, they presumably confer a selective advantage on the embryos that exhibit them.

EVOLUTION OF THE YOLK SAC PLACENTA

The yolk sac placenta of fishes is formed by the apposition of a modified embryonic yolk sac to the uterine mucosa. With the exception of the living coelacanth (*Latimeria*), the yolk sac placenta occurs only in sharks. Placental viviparity in sharks is far more widespread than generally thought. At least 70 of the 250 species of viviparous sharks (about 28%) are placental. Placental species occur in 21 genera belonging to 5 families (estimates derived from Compagno, 1988, 1990). Within a single genus, such as *Mustelus*, 13 species are placental and 10 aplacental (see Teshima, 1981). We estimate that among living selachians, the yolk sac placenta has independently evolved between 11 and 20 times.

The repeated independent evolution of the shark yolk sac placenta in various taxa of sharks has generated considerable structural and presumably functional diversity. Recently, a classification of yolk sac placenta into six categories has been proposed, based on a sequential reduction in the number and thickness of the cellular layers and the amount of extracellular matrix that constitutes the maternal-embryonic placental barrier, along with an increase in the area of contact between maternal and fetal tissues. Reduction of the maternal-fetal

barrier in shark yolk sac placenta parallels that found in mammalian chorionic placenta, namely an epitheliovitelline-to-hemovitelline progression in sharks compared with an epitheliochorial-to-hemochorial progression in mammals (Mossman, 1937, 1987; Wourms *et al.*, 1988). Presumably, reduction of the placental barrier increases the rate of metabolic exchange and results in both quantitative and qualitative differences in molecular transport between parent and offspring. Investigation of the structure, physiology, and morphogenesis of the selachian yolk sac placenta is an actively emerging field (see Wourms, 1981; Teshima, 1981; Dodd, 1983; Hamlett *et al.*, 1985a, b, c; Dodd and Dodd, 1986; Wourms *et al.*, 1988; Hamlett, 1987, 1989; Lombardi *et al.*, in prep.). Even so, the relatively limited number of investigations reveal a surprising diversity of placental structure and function, for example: 1) early vs. late implantation; 2) superficial vs. interdigitated implantation; 3) variation with respect to thickness and number of intervening tissue and matrix layers; 4) retention or loss of the egg envelope; 5) occurrence of epitheliovitelline, hemovitelline, and possibly endotheliovitelline placenta; 6) presence or absence of umbilical stalk appendiculae; and 7) haemotrophic and paraplacental modes of nutrient transfer.

The frequent, independent evolution of the shark yolk sac placenta suggests that the shark yolk sac is developmentally plastic and can easily evolve into the more differentiated state of the yolk sac placenta. Moreover, the shark uterus and maternal physiology appears to easily co-evolve with the yolk sac placenta. Although, it may be somewhat premature, in view of the limited morphological studies and dearth of experimental studies, we propose a scenario for the evolution of the yolk sac placenta. Inasmuch as it is a working hypothesis, we only outline it here and comment on certain critical points of interest.

The evolution of lecithotrophic viviparity from oviparity in elasmobranch fishes, along with the concomitant establishment of new maternal-embryonic relationships and the alteration of maternal physiological regulatory processes, probably sets into place

the basic maternal and embryonic morphology and physiology that facilitates the evolution of placentation (Wourms *et al.*, 1988; Callard and Ho, 1987; Callard *et al.*, 1989). It is necessary to distinguish between two sets of events, one leading to the establishment of the maternal and embryonic portions of the placenta and a second set that involves placental specialization. During the early evolution of viviparity, the lecithotrophic embryo probably possessed essentially the same vascularized yolk sac as did oviparous embryos. The elasmobranch yolk sac, unlike that of teleosts, is a trilaminar extraembryonic membrane that communicates with the embryonic gut (Mossman, 1937, 1987). It is a specialized structure that facilitates gas exchange and yolk processing. During development, it diminishes in size as its yolk content is used. An unspecialized form of the yolk sac placenta occurs among some sharks and possibly the living coelacanth (Wourms *et al.*, 1991). In such forms, the vascularized cortical layer of the yolk sac retains its full size throughout development and its epithelial surfaces presumably function in gas exchange and nutrient uptake after yolk reserves are diminished. The flaccid and yolk-depleted yolk sac can be considered a "floating placenta" because, though it may lie in loose contact with the uterine mucosa, it neither interdigitates nor adheres to it. The definitive yolk sac placenta, in which adhesion between maternal and embryonic tissues occurs, forms a two component system. Three sets of events are involved in its establishment. First, there is the differentiation of the yolk sac into a proximal and distal portion. The proximal yolk sac is specialized for gas exchange with luminal fluids and the distal portion will form a component that is specialized for the exchange of metabolites. Second, there is an interdigitation and adhesion between the distal yolk sac, the intervening egg envelope, and the uterine mucosa. Finally, epithelial cells of the uterine mucosa differentiate at the site of adhesion in order to facilitate maternal-embryonic transport of metabolites and gas exchange. The key element in this process is the establishment of an intimate association of maternal and embryonic tissues. If

interdigitation and adhesion involved only the yolk sac placenta and the uterine mucosa, the underlying mechanisms would seem to be straightforward (i.e., the production of cell-surface adhesion molecules on the distal yolk sac and the elaboration of adhesion molecule receptor sites at localized sites on the uterine mucosa). However, the tertiary egg envelope intervenes in most species of placental sharks. We suggest that the adhesion of the yolk sac to the tertiary egg envelope acts as a specific adhesive locus that elicits a generalized adhesive interaction from the implantation-competent uterine mucosa. Once it was established, the unspecialized form of the shark yolk sac placenta underwent further modification of maternal and embryonic tissues to facilitate more effective metabolic exchange, e.g., by reduction in number or thickness of tissue layers and reduction or loss of the tertiary egg envelope.

Establishment of a definitive placental association is accompanied by a lengthening of the juxtaembryonic yolk sac junction to form a yolk stalk or umbilical stalk (Castro and Wourms, 1992). In both oviparous and non-placental viviparous elasmobranchs, the yolk stalk is short (about 10 mm in length) whereas in placental forms, the umbilical stalk attains a length of 150–250 mm or more. It connects the embryo to the proximal portion of the embryonic yolk sac placenta. Lengthening of the umbilical stalk affords the embryo a considerable degree of freedom of movement within the uterine lumen. Subsequently, a site of autotomy evolved at the embryo-umbilical stalk junction. Its differentiation late in gestation and activation at the time of parturition results in shedding of the placenta. The umbilical stalk is bounded by a sheath of yolk sac cortical tissue and contains the vitelline vein and vitelline artery, and yolk or vitello-intestinal duct. In most sharks, the umbilical stalk is an unadorned, smooth-surfaced cylindrical tube. Some placental sharks possess vascular, finger-like, or branched, processes termed appendiculae (Budker, 1953). These epithelial specializations extend outward from the umbilical stalk and greatly amplify the effective surface area available for paraplacental exchange between the

embryo and the surrounding periembryonic or uterine fluid. Appendiculae occur in eight shark genera representing three families within the order Carchariniiformes: *Hemigaleus*, *Hemipristis*, *Paragaleus*, *Loxodon*, *Rhizoprionodon*, *Scoliodon*, *Eusphyrna* and *Sphyrna*. At least seven classes of appendiculae are recognized on the basis of morphological differences and they appear to function in nutrient absorption, gas exchange, and embryonic secretion (Wourms *et al.*, 1988; Lombardi and Wourms, 1988b; Lombardi and Wine, 1989; Hamlett, 1989; Castro and Wourms, 1992). Finally, it should be noted that the evolution of the selachian yolk sac placenta reaches its zenith in the spadenose shark, *Scoliodon laticaudus*, in which the ovum is microlecithal and reduced to a size of 1 mm. Implantation occurs very early in development and the umbilical stalk soon loses its yolk duct. The stalk is festooned with long appendiculae. A hemovitelline relationship is established between the embryonic yolk sac placenta and a highly specialized maternal implantation site. Extensive nutrient transfer is likely since this species exhibits a gestational dry mass increase of $120,000 \times$ (Wourms *et al.*, 1988).

EPILOGUE

We have provided a sketch of our emerging views on the evolution of piscine viviparity and hope that some of the issues that have been raised will serve as a stimulus for future research.

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