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ENCAPSULATION OF CEPHALOPOD EMBRYOS: A SEARCH FOR FUNCTIONAL CORRELATIONS

SIGURD V. BOLETZKY
C.N.R.S.
LABORATOIRE ARAGO
66650 BANYULS-SUR-MER
FRANCE

ABSTRACT

This article considers basic traits and group-typical modifications of egg encapsulation in the molluscan class Cephalopoda, emphasizing evolutionary aspects of the coordinated organization of capsule production by the adult and structural adaptation of the embryo, especially with regard to hatching mechanisms. Particular attention is given to the modifications observed in octopods, in which nidamental glands lying outside the terminal oviduct are lacking. All material secreted around the egg chorion (cirrate octopods) or chorion stalk (incirrate octopods) is produced by the complex oviducal gland, which thus fulfills the function of both oviducal and nidamental glands of decapods. The incirrate octopods are unique in that the protective function of encapsulation is entirely replaced by the active protection of naked eggs by the female (brooding or ovovivipary).

Encapsulation of eggs appears to be a basic means of protecting developing embryos in the class Cephalopoda. The presence of a large nidamental gland complex in *Nautilus*, the only living representative of the ectocochlean cephalopods, and its positional, structural and supposed functional similarity to endocochlean (coleoid) nidamental glands indeed suggest that encapsulation of eggs is a common ancestral character of the class.

Within the coleoid cephalopods, there are various modifications in capsule structure, although these capsules are produced by a largely uniform apparatus of capsule formation. Evidently these modifications reflect adaptive "strategies" responding to extrinsic (ecological) and intrinsic (development *s.l.*) constraints. They can be viewed in the evolutionary context of coordinated organisation (i.e. intrasystemic coadaptation of functional components). Although many gaps in our knowledge of encapsulation of cephalopod eggs remain to be filled, the available data already permit a framework of questions to be raised in approaching functional correlations within the mechanism of encapsulation. This brief survey attempts to outline the subject using data available in the literature and unpublished observations.

As with many other areas of cephalopod research, an historical résumé of published observations could start out with the written report (on the eggs and their capsules) given by Aristotle. Here it is sufficient to recall the thorough analysis of encapsulation in decapods (orders Sepioidea and Teuthoidea) published by Jecklin (1934) with a careful survey of the older literature. Jecklin provides a detailed description of the structure of the mucinous egg cases in cuttlefish, sepiolid and teuthoid squids, analyzes the changes they undergo during embryonic development, and finally studies hatching mechanisms. More recent data are reviewed in vol. IV of "Reproduction of marine invertebrates" edited by Giese and Pearse (1977), in both volumes of "Cephalopod Life Cycles" edited by Boyle (1983, 1986), and in vol. VII of "The Mollusca" (Reproduction) edited by Tompa, Verdonk and van den Biggelaar (1984) where cephalopods (Arnold, 1984) are reviewed along with gastropods and bivalves.

ORGANS PRODUCING CAPSULE MATERIAL

The mature cephalopod ovum (ovarian egg) is surrounded by the chorion, a product of the follicular cells. Although in chronological terms this is the primary egg cover, it is generally called the secondary envelope; the fertilization membrane (vitelline membrane), which forms a temporary cover of the embryo at early developmental stages, is termed the primary envelope. All additional material added to the outside of the chorion may be called tertiary envelopes. It is indeed of little use to call the more or less distinct outer coat or shell quaternary, as it is not distinguishable by its mode of production. Within the so-called jelly coats lying inside the outer coat, there are again two different components laid sequentially, as shown by Jecklin (1934). Probably in all cephalopods, some jelly is produced by the distal part of the oviduct, which forms a more or less compact glandular ring both in paired (oegopsid squids, incirrate octopods) and unpaired, unilateral oviducts (*Nautilus*, cuttlefish, sepiolid and myopsid squids, cirrate octopods).

In *Nautilus* and in most of the decapods, a pair of nidamental glands lies in the mantle cavity, with their open-

ings situated close to the oviducal outlet(s). Although the process of nidamental jelly release has so far not been observed *in situ*, it seems most likely that eggs leaving the oviduct are immediately enveloped by the mucinous material "flowing" out of the nidamental glands (Arnold and Williams-Arnold, 1977). Eggs leaving the oviduct intermittently, one by one, are apparently enveloped individually; eggs leaving the oviduct serially are enveloped in a capsule enclosing a series of eggs.

Whether the paired accessory nidamental gland regularly provides secretions (Arnold and Williams-Arnold, 1977), e.g. for the formation and/or hardening of an outer coat, is not yet clear. The presence of bacteria in the winding ducts of this organ (Bloodgood, 1977), and the presence of clustered bacteria in the outer coat of *Rossia* eggs (Boletzky and Boletzky, 1973) suggest that the accessory nidamental gland may have a more complex role in the physiology of encapsulation than merely a function of finishing the capsule surface, but nothing is really known.

Finally it has been suggested that the salivary glands also contribute to the finishing of capsular structures (Jecklin, 1934). Similar suggestions concerning an intervention of salivary gland secretions in egg string formation by *Octopus* females are summarized by Prezant (1985) who quotes from earlier papers (Wood, 1963, Gennaro *et al.*, 1965). However, the oviducal gland secretion of octopus females provides most, if not all, of the "cement" material for the chorion stalks typical of the eggs of incirrate octopods (Froesch and Marthy, 1975). This oviducal gland secretion corresponds to the capsule material forming the outer envelope of cirrate eggs (Boletzky, 1978-79, 1982a). The complex structure of the octopodan oviducal gland, and in particular of the clearly bipartite gland of cirrate octopods (Meyer, 1907, Aldred *et al.*, 1983) ultimately raises the evolutionary question of the developmental pathways of structural modifications concerning both nidamental and oviducal glands (see Discussion). Here it can only be stated that the oviducal gland of cirrate octopods does indeed produce capsule material forming an envelope very similar to certain decapodan egg capsules, especially to those of *Rossia* eggs.

CAPSULE ARCHITECTURE IN DIFFERENT GROUPS

To use the term architecture of "slimy" secretions making up largely gelatinous coats that go through changes of size and structure during development of the embryos may appear inappropriate. However, in most instances, there is indeed a well-defined combination of volume, consistency and "packaging" in the secretory product that pre-programs the living conditions of the encapsulated embryos for the entire time of their development, which may last from a few days to more than one year depending upon species. In this section the capsule architectures typical of the different cephalopod groups are briefly described.

SUBCLASS NAUTILOIDEA

Nautilus eggs were described by several authors, beginning with Willey (1897). A peculiar feature of these very large eggs is that the hard outer coat is drawn out into a series of prominent folds each ending in an opening (cf. Haven, 1977). Thus the inner capsule only is entirely sealed from the outside. In preserved egg capsules I found the inner envelope to be continuous with the outer at the "attachments" described by Willey (1897). Thus the outer capsule appears to be an overturned bell-shaped ruffle, the edge of which is drawn over the apex of the inner capsule (leaving the resulting folds to form the open channels) before the egg is attached to the substratum.

As live observations of developing *Nautilus* embryos have become possible only very recently (Arnold and Carlson, 1986), it is too early to attempt functional interpretations of these structures, especially with regard to the hatching mechanism.

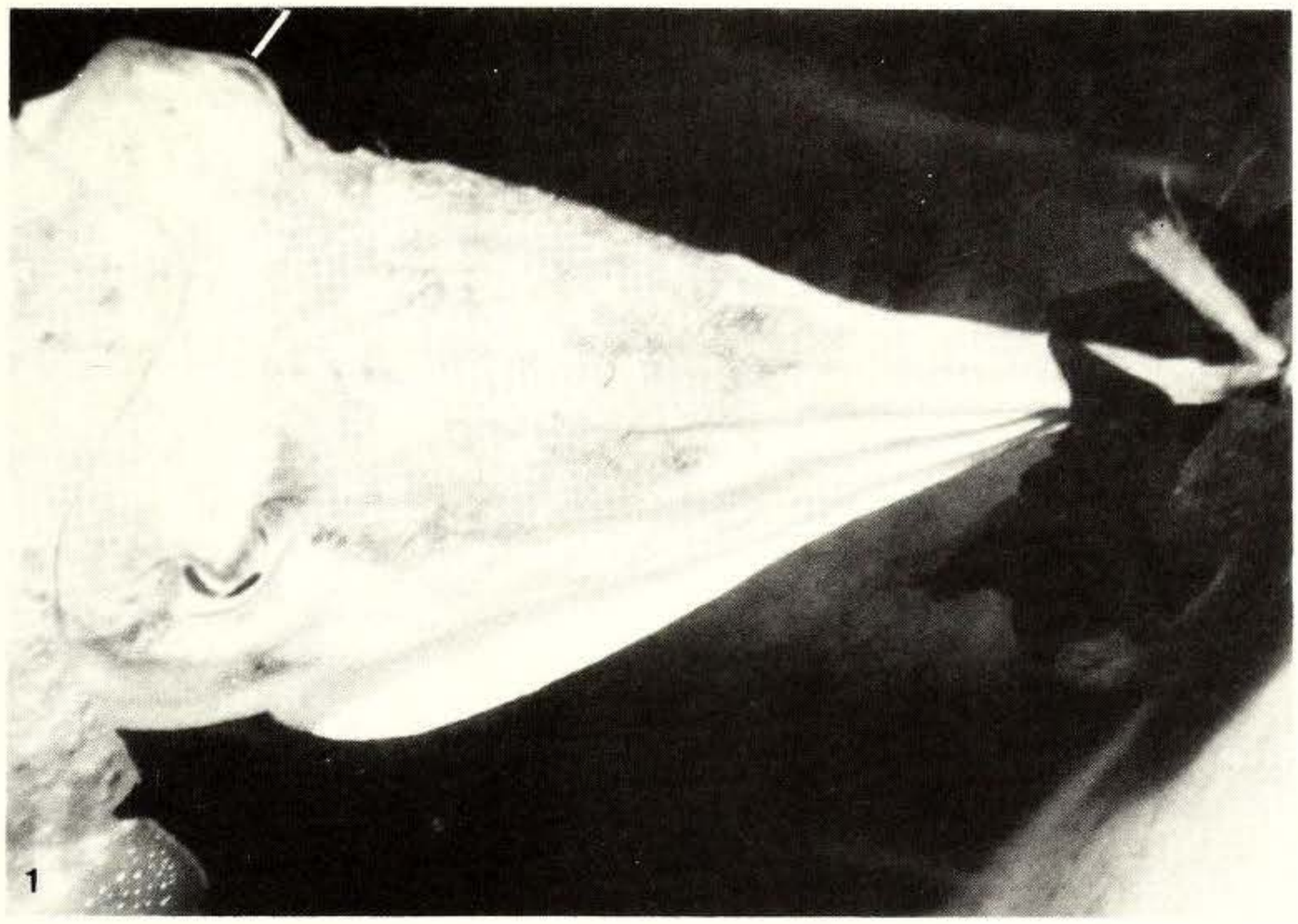
SUBCLASS COLEOIDEA

ORDER SEPIOIDEA

No observations are known on spawning in the pelagic genus *Spirula*. In the genus *Sepia*, the chorion of each egg is surrounded by spirally coiled oviducal jelly (Jecklin, 1934), plus a spirally coiled envelope of nidamental gland jelly, which in turn is surrounded by a soft outer coat (Figs. 2, 3). In *Sepia officinalis* Linnaeus, 1758, these envelopes are normally coloured by ink released with the jelly at spawning (Grimpe, 1926). At the moment of spawning, the female approaches an appropriate substrate for egg fixation, aims at the target site with binocular vision (Fig. 1), and at the same time uses the arm tips to draw out the very soft jelly coats into two filaments. Once she has made contact with the chosen substrate (any rod-like object or eggs already laid), she winds these filaments around the support so that they stick together and form a fixating ring. In aquaria, females unable to find an appropriate substrate for the fixation of their eggs drop them without producing filaments (for *Sepia orbignyana* and *S. elegans* see Ecological aspects of encapsulation).

The eggs of the Sepiolidae are rather similar to *Sepia* eggs, but they are always simply glued to a substrate, no matter whether it is flat or has prominent structures that would allow fixation by a ring. In the subfamily Sepiolinae, the outer coat is leathery and somewhat elastic (Fig. 6), whereas in *Rossia* eggs (and probably in the eggs of all Rossiinae), it is perfectly rigid. This outer case is ca. 200 μm thick (Fig. 5); it is made of several layers, which at the moment of laying are still very soft (Boletzky and Boletzky, 1973). Hardening into a true shell takes several hours. *Rossia* females space out their eggs on a substrate in regular intervals. When the egg capsules of this ground layer are firm, the spawning animal lays subsequent eggs on top of them. A typical egg mass of *Rossia* finally shows a fairly regular three-dimensional network, the eggs being piled up around large interstices (Fig. 4). They are most often found in empty bivalve shells (especially *Pinna pectinata* Linnaeus, 1767 in *Rossia macrosoma* [Delle Chiaje, 1829]), in which they are fixed to the ceiling of the shelter formed by the empty shell lying on the ground.

Sepiola and *Sepietta* eggs may also be laid in several layers, but they never form a loose three-dimensional network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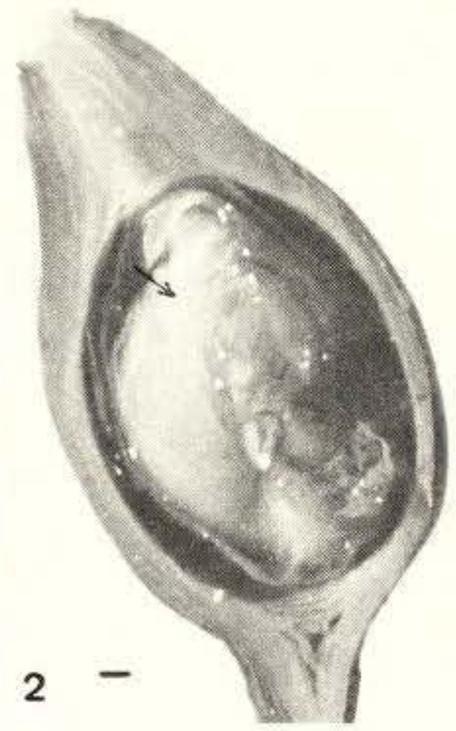
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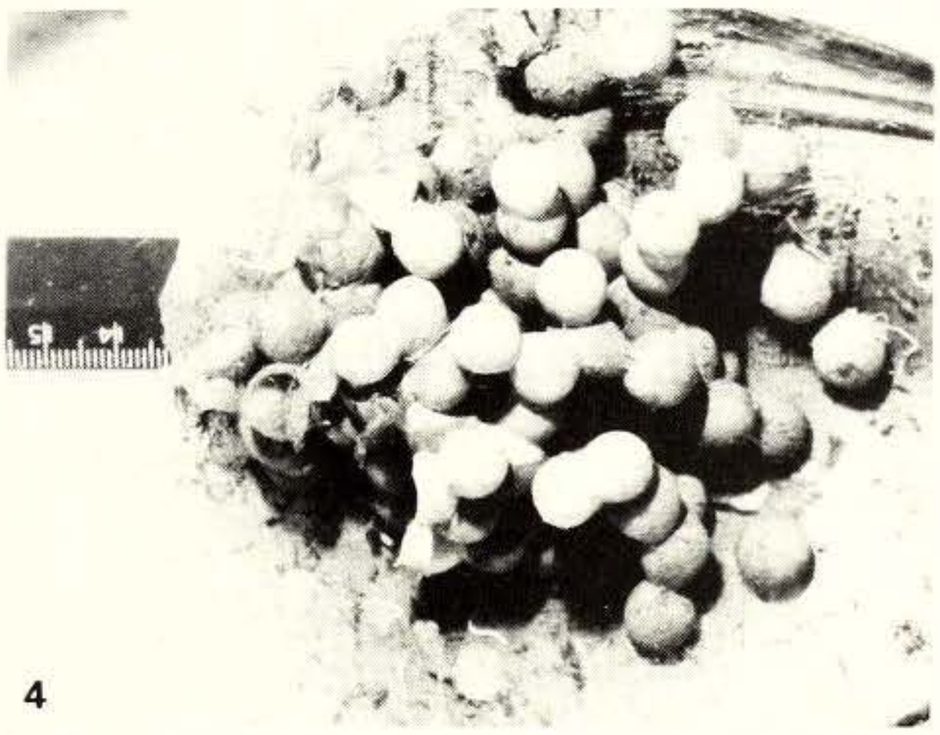
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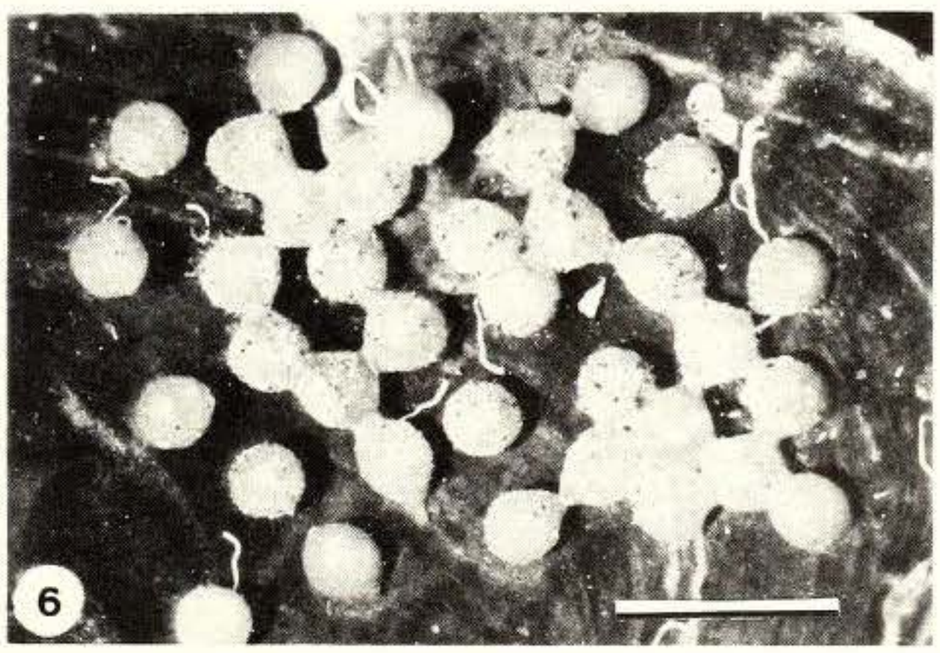
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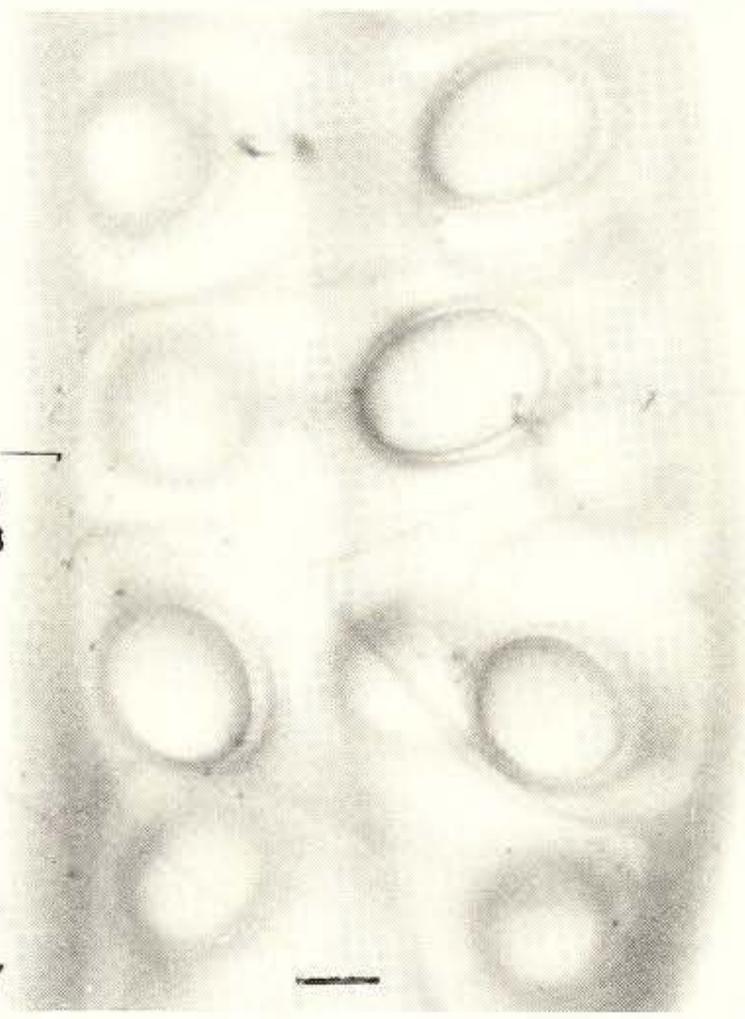
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Fig. 1. Female *Sepia officinalis* seen from the water surface while attaching an egg to a shackle suspended in the tank. The two bars indicate the middle axis of the eye ball to show convergent orientation for binocular vision; note also the arm tips stretched out towards the egg support. **Fig. 2.** An egg envelope of *Sepia officinalis* (laid empty, without an ovum), cut open. Inside the shrunken outer envelopes, the cavity normally containing the ovum is filled with a spirally coiled sheet of softer jelly, probably corresponding to the coiled oviducal jelly described by Jecklin (1934). Arrow indicates area enlarged in Fig. 3. Scale bar = 1 mm. **Fig. 3.** Detail of Fig. 2 at higher magnification. **Fig. 4.** Egg mass of *Rossia macrosoma* on a *Pinna pectinata* shell. **Fig. 5.** Semithin section through the outer shell of an egg of *Rossia macrosoma*. Note the very dense layers at the surface (above) and the alveolated inner layer (below). Scale bar = 10 μm. **Fig. 6.** Eggs of *Sepiola* sp. on a *Pinna* shell. Arrow head points to an elongated junction (see text). Scale bar = 10 mm. **Fig. 7.** Egg capsule of *Loligo vulgaris* shortly after laying, showing the spiral arrangement of the string of eggs embedded in oviducal jelly. Note the inversion of coiling direction in the lower right (this is close to the end of the capsule). Scale bar = 1 mm. **Fig. 8**. Enlargement of the area indicated in Fig. 7, after removal of the outer coat.

like egg masses of *Rossia*. As a consequence, the embryonic development of eggs covered by others is slowed due to poor oxygenation (Boletzky, 1983, Bergström and Summers, 1983).

The eggs of *Idiosepius*, the pygmy cuttlefish of the Indo-Pacific, are rather similar to the eggs of Sepiolinae, but there seems to be no distinct outer coat (Natsukari, 1970).

At the moment of laying, the spirally coiled nidamental coats always form a thick, but very soft capsule. In the course of early embryonic development, they lose water and progressively shrink until they form a rather thin compound ("multilayered") membrane (Fig. 2). Especially in *Sepia* eggs, this shrinkage is easily recognizable when one compares newly laid and moderately advanced eggs, the latter having a smaller size and a firmer consistency. In *Sepiola* and *Sepietta* eggs attached to one another, the shrinkage becomes clearly visible in the elongating junctions uniting eggs that stick together with their outer coats (Figs. 6). With the uptake of water by the chorionic contents, which are hypertonic against sea water (Russell-Hunter and Avolizi, 1967, De Leersnyder and Lemaire, 1972), the outer egg diameter then increases progressively so that the nidamental envelopes are stretched and grow ever thinner (Mangold-Wirz, 1963).

In *Rossia* eggs, the rapid hardening of the outer coat blocks the envelopes from stretching beyond the original diameter. The increase of the chorionic space related to the shrinkage of the soft envelopes thus ends when the inner egg shell diameter (minus the thin condensed nidamental layers) is attained. Although the outer coat of the eggs of Sepiolinae is elastic and allows some expansion at late embryonic stages, the size increase is rather limited. This is important for hatching, because the young animal has to prop its arms against the chorionic wall opposite to the hatch opening, as shown by Arnold *et al.*, (1972) in *Euprymna*.

ORDER TEUTHOIDEA

Most observations on spawning reported in the literature deal with myopsid squids of the family Loliginidae (Roper, 1965). The few available data on oegopsid squid egg masses nevertheless permit some generalizations. It seems reasonable to suppose that a nidamental apparatus comprising both nidamental and accessory nidamental glands represents the primitive condition of decapods. Such a common ancestral condition easily accommodates the supposedly derived teuthoid mode of serial egg encapsulation (maintaining the spiral enveloping mechanism). Instead of wrapping a single egg in a sheet of mucinous secretion, a string of eggs united by oviducal jelly is rolled into a common sheet of nidamental jelly (Figs. 7-9). The precise "cork screw" or "spiral stair" arrangement achieved by this process suggests that the wrapping occurs very rapidly at the outlet of the nidamental glands, i.e. before the jelly bands take up additional water to swell to the final size observed in the capsule when it leaves the mantle cavity. Evidently the number of eggs that can be enclosed in a single capsule is limited by both the size of individual eggs and the size of the nidamental apparatus, which in turn depends on the body size of the spawning female.

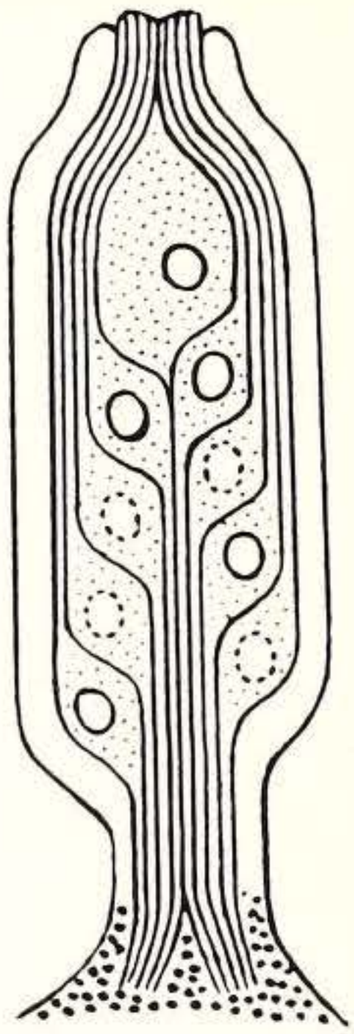


Fig. 9. A schematic presentation of a squid egg capsule (after Jecklin, 1934, modified). The coarse stippling corresponds to the fixating jelly, which grades into the (white) outer coat. The eggs are shown embedded in oviducal jelly marked by fine stippling. The lines represent the dense layers of nidamental jelly.

Not all squid egg masses are made of spirally coiled jelly layers, however. Ommastrephid squids produce extremely watery jelly masses that show no internal structure (Hamabe, 1961, Boletzky *et al.*, 1973, O'Dor, 1983). In some enoploteuthid squids, there are no nidamental glands at all, but the oviducal glands are extremely large (Naef, 1923).

ORDER VAMPYROMORPHA

Vampyroteuthis has no nidamental glands. The observed absence of jelly on pelagic eggs thought to be those of *Vampyroteuthis infernalis* Chun, 1903 (Pickford, 1949) is no proof that these eggs are released without any gelatinous material surrounding the chorion. It seems indeed more likely that the well developed oviducal glands produce a fragile jelly (providing some buoyancy?) that easily disintegrates when eggs are collected with nets.

ORDER OCTOPODA

The living octopods fall into two very distinct groups, the Cirrata (finned octopods) and the Incirrata.

SUBORDER CIRRATA

These deep sea animals encapsulate their very large eggs in a hard shell. The few eggs so far described (Boletzky, 1982a) show some variation in the structure of the egg shell, and also in its size relative to the chorion size. In only one case was the chorion surface separated by a wide, jelly-filled space from the outer shell (Fig. 10a). The surface of the shell, which is produced by the large oviducal gland (Aldred *et al.*, 1983), is smooth in some species, coarse (Fig. 10b) or distinctly sculptured in others. Such sculpturing sug-

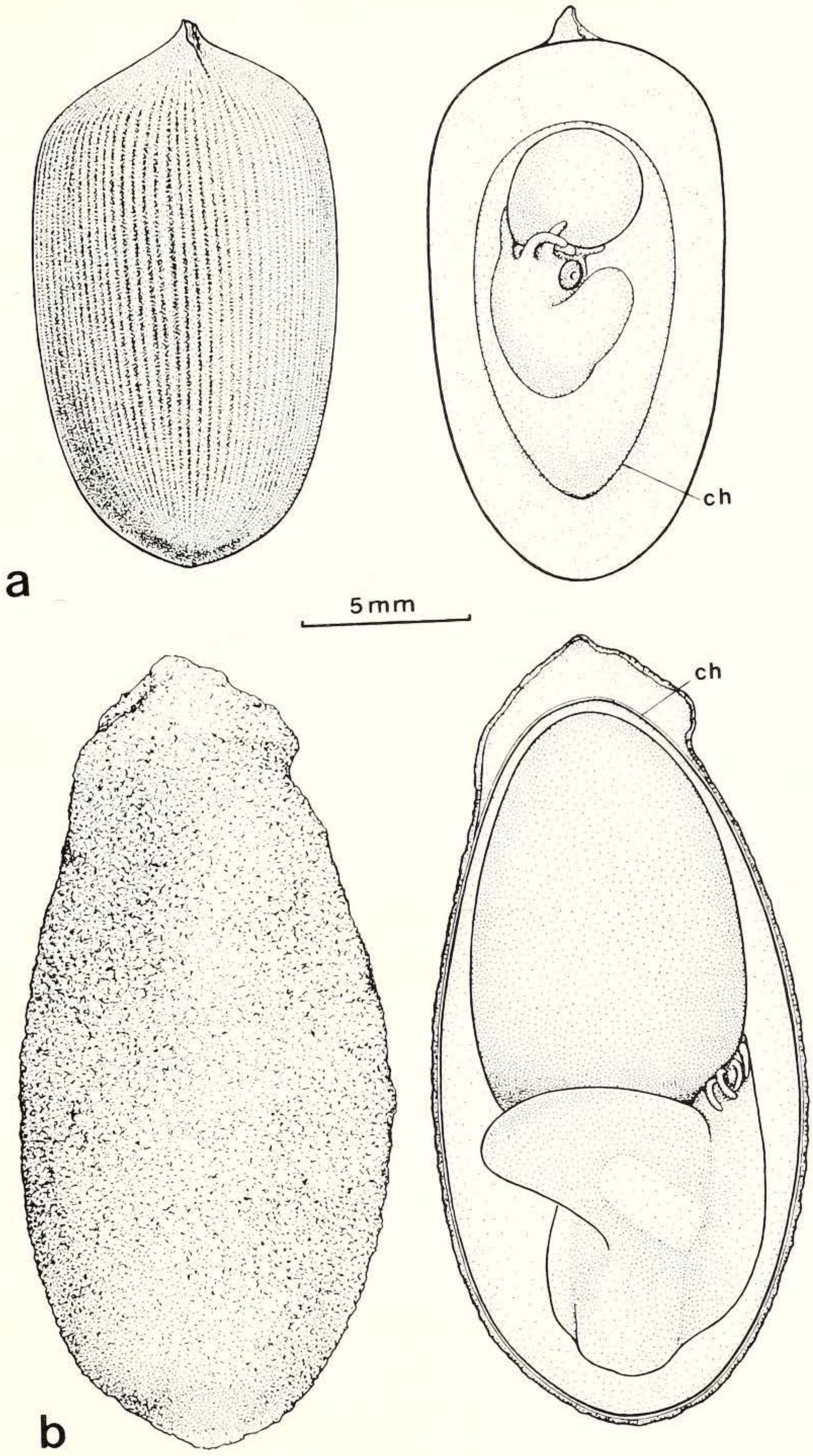
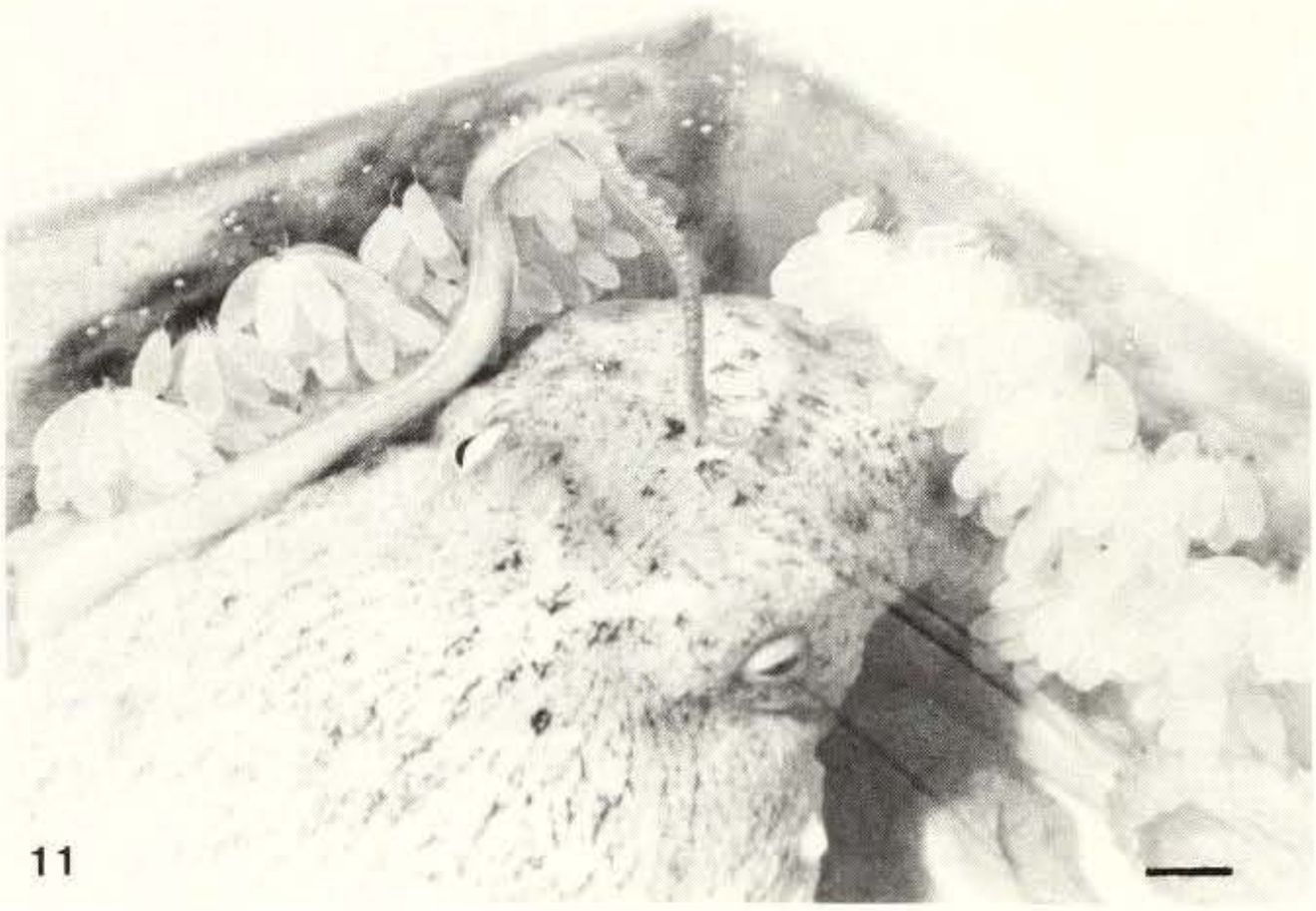
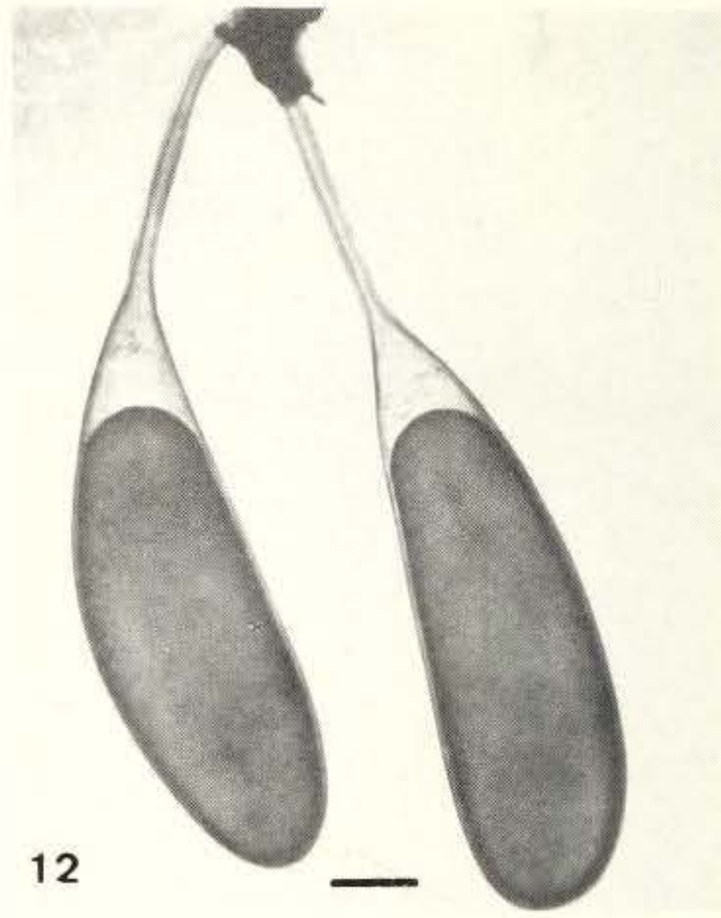
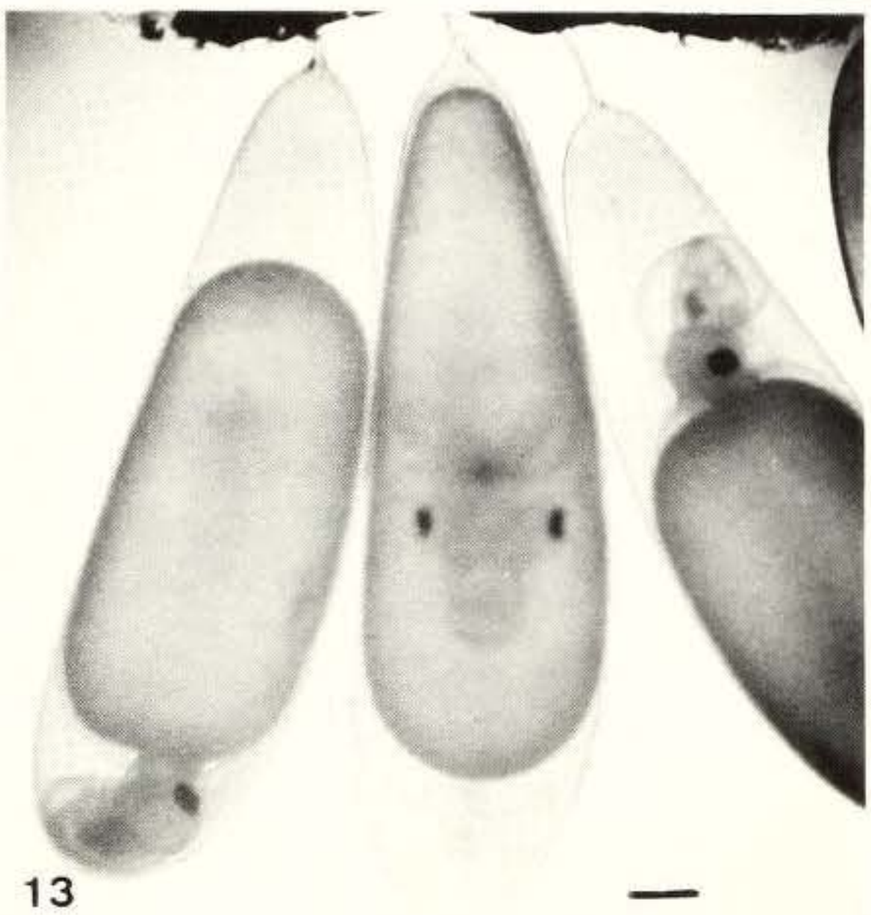
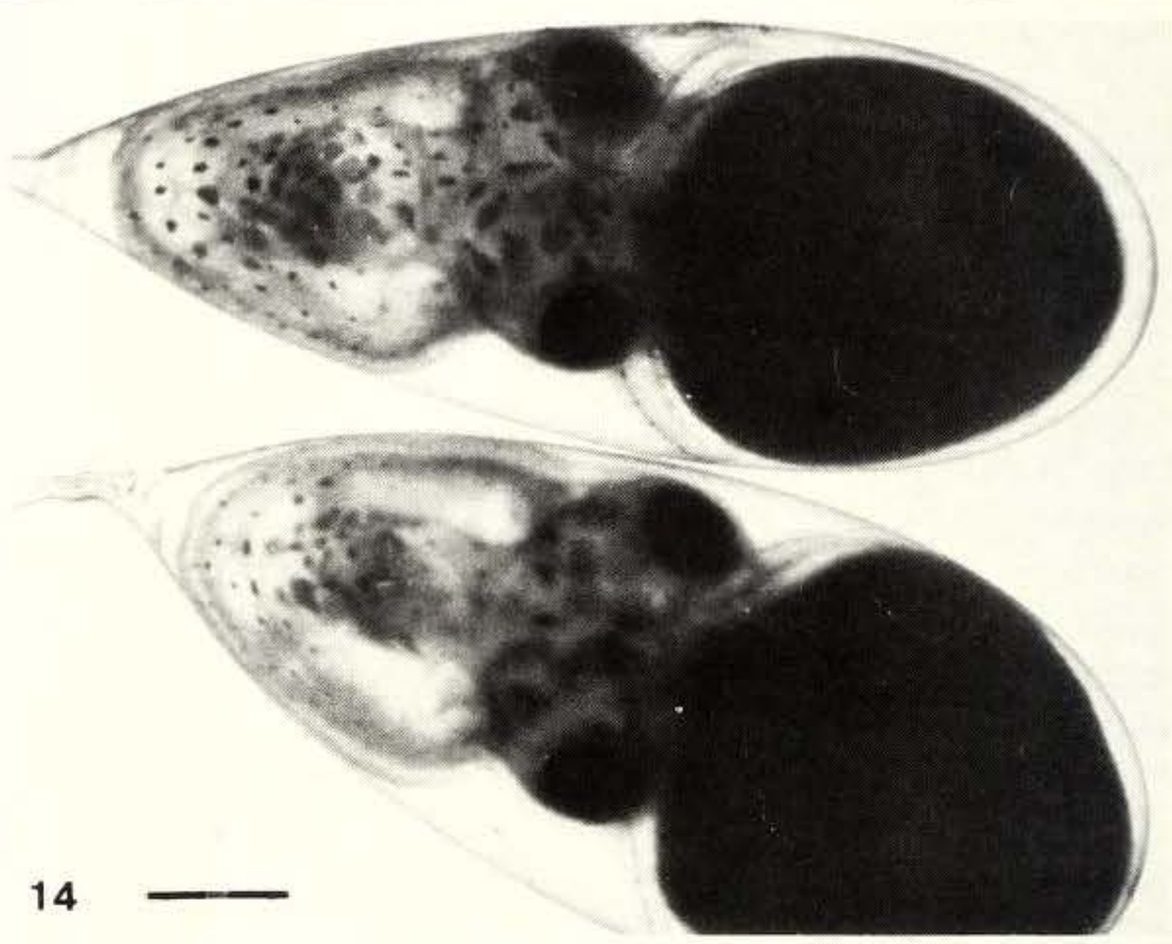


Fig. 10. Eggs of unidentified cirrate octopods. a. surface view of shell (left) and internal view after removal of one half of the shell. Note wide space between chorion (ch) and shell. b. Surface view of an egg, in which chorion (ch on the right) is rather tightly surrounded by the shell.









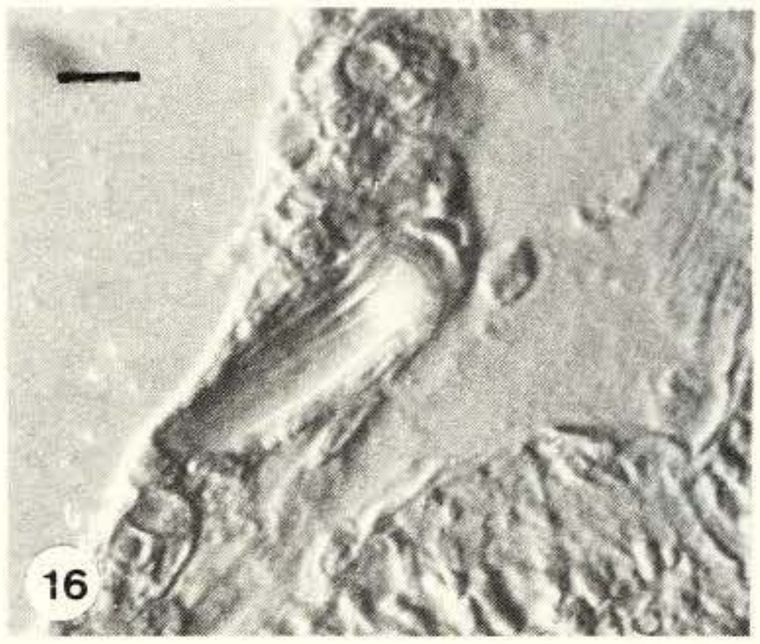


Fig. 11. A female *Eledone moschata* brooding her eggs in the corner of an aquarium tank. Scale bar = 1 cm. **Fig. 12.** Newly laid eggs of *Pteroctopus tetracirrhus*. Note the rather short, thick chorion stalks fixed to a common base of oviducal "cement". Scale bar = 1 mm. **Fig. 13.** Eggs of *Octopus briareus* attached by the chorion stalk to a common axis (above). Note the different positions of the embryos due to delayed inversion (middle) or absence of inversion (left). Scale bar = 1 mm. **Fig. 14.** Advanced embryonic stages of *Eledone cirrhosa*, with a still large outer yolk sac (at right). Note that the embryo is very tightly surrounded by the chorion. Scale bar = 1 mm. **Fig. 15**. Detail view of an embryo of *Octopus vulgaris* (cf. Fig. 17) hatching from the chorion (at left). The arrow points to the edge of the hatching slit. Arrow heads indicate organs of Koelliker seen through the transparent skin. Scale bar = 0.1 mm. **Fig. 16**. Histological section of the skin of an *Octopus vulgaris* hatchling, showing an organ of Koelliker with its setal core anchored in the basal cell (above) and its outer end lying under a very thin tissue membrane (below). Scale bar = 10 μm.

gests that the shell hardens before the egg is released from the oviduct. Nothing is known of the laying procedure. In particular it is not clear whether the eggs are fixed to a specific substrate.

SUBORDER INCIRRATA

The members of this suborder invariably produce eggs devoid of protective capsules. The chorion is always drawn out into a stalk, the length of which is very variable among species (Figs. 11-13). The material secreted by the oviducal glands (Froesch and Marthy, 1975) normally surrounds only the end of this stalk and serves to fix it either directly to a substrate (Figs. 11, 12) or to other egg stalks thus forming the central axis of a festoon-like egg string (Fig. 13). Eggs are always actively protected by the female throughout the time of embryonic development (Fig. 11). In the Octopodidae, which are the only bottom living incirrates, females generally attach eggs or egg strings to a hard substrate, inside a shelter, and remain with them for the entire brooding time; this may last a full year in certain coldwater species producing very large eggs, as for example *Bathypolypus arcticus* (Prosch, 1849) (cf. O'Dor and Macalaster, 1983).

In a few octopus species, the females carry their egg strings or clusters in their arms (e.g. Tranter and Augustine, 1973). This method closely resembles the brooding habits of pelagic incirrates. Among these, *Argonauta* produces an elaborate auxiliary apparatus in the form of a calcitic brood shell, in which the eggs are carried. A simpler type of egg carrier is produced by *Tremoctopus* females. Instead of secreting organic material and calcium carbonate in the form of a thin-walled shell, the dorsal arms of the female produce short rods to which the eggs are attached (Naef, 1923). In both forms, the release of eggs is delayed beyond the first cleavage stages. This delay is pushed to true ovovivipary in *Ocythoe*,in which the eggs remain in the very long oviduct until the embryos are ready to hatch (Naef, 1923). The observations of Young (1972) on a bathypelagic octopus of the family Bolitaenidae suggest the existence of a special adaptation of the arm crown of the female to function as a brood pouch in this particular species (probably *Eledonella pygmaea* Verrill, 1848).

A feature common to all incirrates is the rather limited expansion of the chorion during embryonic development (Figs. 14, 17). Although the volume of the egg may increase by more than 150% during embryonic development, the embryo remains tightly surrounded by the chorion, which is much tougher than the decapodan chorion.

FERTILIZATION AND HATCHING

These two events mark the beginning and the end, respectively, of embryonic development. For both processes, egg capsules represent a barrier to be overcome as well as a substrate to be used for locomotory actions.

Except for octopods, in which fertilization is achieved in the oviduct or in the ovarian cavity (Mangold, 1983a, b), spermatozoa always have to cross some jelly material in order to arrive at the micropyle of the chorion. Depending on the

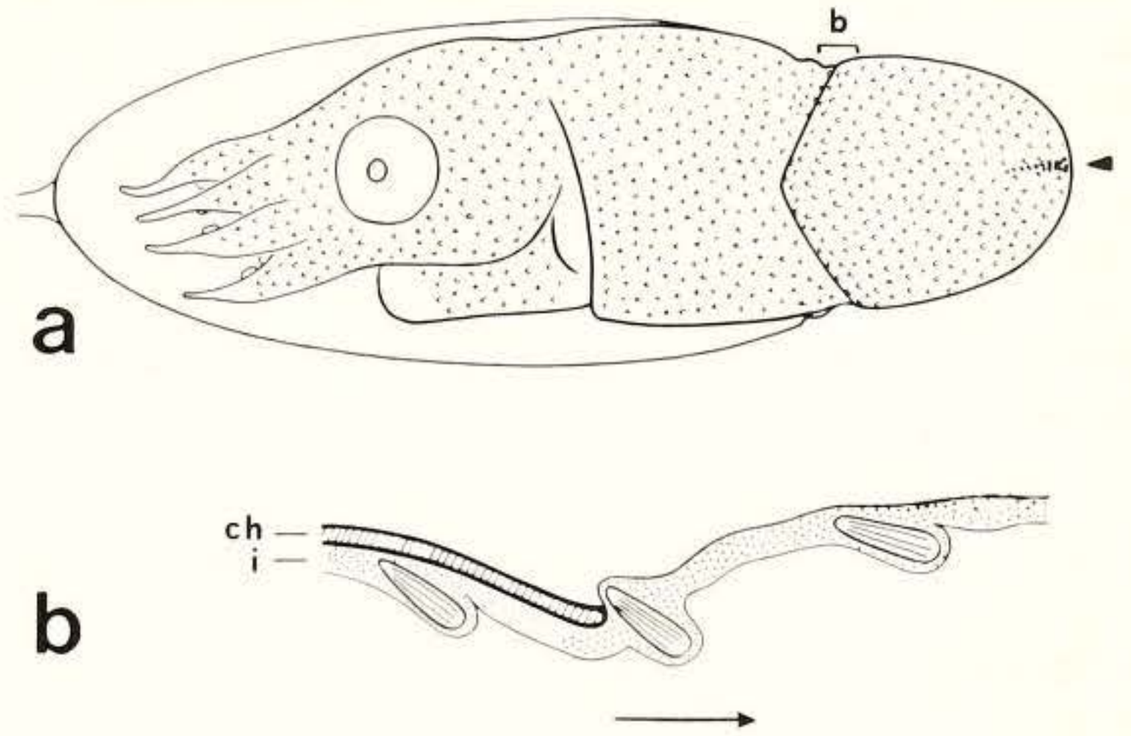


Fig. 17. A schematic presentation of hatching in *Octopus vulgaris* (or any other species of *Octopus* producing small-sized hatchlings with short arms). The arrow head points to the hatching gland (transversal bar of cells). The dense distribution of Koelliker organs is represented schematically in a. In b, a schematic longitudinal section of the skin at the hatching slit is given, corresponding to the area marked in a (see also Figs. 15, 16). The long arrow at the bottom indicates the direction of the hatching movement. ch = chorion, i = integument.

site where spermatophores are stored after copulation (infrabuccal pouch, mantle cavity, etc.), and according to the capsular structure, access routes to the individual egg are shorter or longer for the spermatozoa. In all events, the consistency of the capsule material, which is still very soft at that stage, would seem to be important for penetration by the spermatozoa. The functional morphology of the latter should therefore be viewed on the background of locomotory requirements defined by the mucinous envelopes, across which they have to move. This concerns the leading structure formed by the acrosome; the position of the flagellum (or flagella) in relation to the posterior part of the sperm head; and the structure of the spine-like posterior process of squid spermatozoa (Fields, 1965, Franzen, 1967, Millard de Montrion, 1984), also called mitochondrial spur (Fields and Thompson, 1976).

At the moment of hatching, similar constraints arise when the young animal has to move across the capsule material in a direction opposite to that of the spermatozoa. Meanwhile, however, the consistency of the capsule material has thoroughly changed. In all cephalopod hatchlings, the leading structure of the animal when moving across the capsule wall is the mantle end, which is equipped with a hatching gland (organ of Hoyle). This organ forms at late embryonic stages. It is made of special glandular cells of the epidermis which store proteolytic enzymes (cf. Denucé and Formisano, 1982). In the decapods, these cells are arranged in one dorsal and two lateral branches forming together an anchor-shaped complex. In the octopods, there is only one transversal band of glandular cells, which are less prominent than in the decapods (Fig. 17).

How hatching is triggered in cephalopods is still

obscure. Probably all cephalopod embryos are kept "quiet" by a tranquillizing factor contained in the perivitellin fluid (Marthy *et al.*, 1976) so that premature hatching is largely prevented. How the threshold set by this system is finally overcome in the absence of artificial stimuli (which easily trigger hatching in the aquarium) remains to be demonstrated. The hatching process generally starts with characteristic stretching movements of the mantle, which seem to rupture the apex of the gland cells (Orelli, 1959). The enzymes thus released onto the chorion wall immediately dissolve it locally. Indeed in all known cephalopods, the position of the hatchling inside the chorion and the limited expansion of the latter have the effect of bringing the hatching gland into very close contact with the wall. Recent experiments (Boletzky, unpubl. results) have shown that the enzymes of the hatching gland are not species-specific. Loliginid hatchlings artificially enclosed in envelopes of a different species *(Loligo vulgaris* Lamarck, 1799, *Alloteuthis media* [Linnaeus, 1758]) were able to hatch out, and hatchlings of both the above-mentioned species were able to open the thick chorion of newly laid eggs of *Sepia officinalis* Linnaeus, 1758.

The role of the organ of Hoyle has been known since Wintrebert (1928) demonstrated its function as a hatching gland. Furthermore Jecklin (1934) has shown that there is no preparatory softening of the chorion, and that perforation of the chorion and the surrounding membranes is achieved solely by the instantaneous action of the hatching gland secretion. However, the importance of auxiliary processes in hatching have largely been ignored. Indeed hatching depends on both the *perforating action* of the organ of Hoyle and the *locomotion* generated by other organs of the hatchling. A close correlation between the capsule architecture and the lay-out of the entire hatching apparatus is clearly recognizable in the representatives of the Sepiidae, Sepiolidae, Loliginidae and Octopodidae so far studied (Boletzky, 1982b).

In *Sepia officinalis*, as in all decapod embryos, the skin contains very numerous ciliary cells. The motile cilia all beat in anterior direction (i.e. the effective stroke is directed away from the posterior mantle end). Together with the ciliature of the outer yolk sac (which disappears only towards the end of embryonic development), these cilia maintain the perivitellin fluid in continuous circulation. The three branches of the hatching gland are surrounded by ciliary bands that are distinct from the ciliary tufts covering the rest of the body. At the moment of hatching, the cilia of these bands are the first to be in contact with the edges of the hatch opening and they probably assist in providing a slight locomotory effect (cf. loliginid squids, below).

In contrast, in the Sepiolidae, there are no ciliary bands. The skin is only covered with rather widely scattered ciliary tufts. The rear end of the hatching gland is underlain by a peculiar conical organ, the so-called terminal spine (Naef, 1928). The tip of the spine is made of very dense connective tissue grading into a muscular basis anchored on the mantle musculature. Artificially immobilized hatchlings exposed to certain tactile stimuli go through rapid stretching movements during which the terminal spine strongly projects over the mantle end, thus demonstrating the autonomous contraction of the muscular basis of the terminal spine (Boletzky, unpubl. obs.). The punctual pressure achieved by this autonomous contraction is no doubt important in breaking the hard outer shell of *Rossia* eggs. This action is possible only in limited space allowing the animal to prop its arms against the chorionic wall when pushing the mantle end through the hatch opening.

In loliginid squids, the hatching apparatus is more similar to the situation observed in *Sepia*. However, instead of being limited to the immediate vicinity of the hatching gland, the distinct ciliary bands cover a large part of the upper and lower mantle surface. Live observations have shown that the relatively short cilia of these bands have only a very limited effect in circulating the perivitellin fluid, in contrast to the long cilia of the tuft cells (Arnold and Williams-Arnold, 1980). These short cilia appear to provide most of the locomotory effect obtained on the gelatinous substrate made available to them by the action of the hatching gland. The latter indeed acts like a "bore head" opening a tunnel in the nidamental jelly layers. Regardless of the initial direction a squid hatchling takes within the common egg capsule, it automatically arrives at the capsule surface by purely ciliary locomotion (Boletzky, 1979). Observations on *Illex* hatchlings indicate that the same mechanism allows these extremely small animals to leave the large jelly mass typical of ommastrephid squids (O'Dor, 1983).

A completely different arrangement characterizes the incirrate octopods. The skin of the hatchling is devoid of motile cilia. The transversal band of cells forming the hatching gland (Orelli, 1959) produces a slit in the chorion (Fig. 17). Only the mantle end is extruded due to the release of pressure from the elastically stretched chorion. Its contraction is insufficient, as in decapods, to expulse the animal. Although the octopus hatchling has to free itself of only one simple membrane, it is momentarily stuck with the greater part of its body still inside the envelope. Two structures are important in overcoming this situation: 1. the simple slit produced in the vaulted end of the elongate, relatively tough chorion presents a relatively sharp edge (Figs. 15, 17), and 2. the skin of octopus hatchlings contains a dense set of hard rod-like structures (the setal core of the organs of Koelliker), which together form a shingle-like surface preventing the body end from slipping back into the chorion (Figs. 16, 17). Indeed the setal core of each organ of Koelliker lies in an oblique position, its outer end pointing anteriorly. Although it is covered by a thin tissue membrane (Fig. 16), it slightly projects under the external pressure exerted by the sharp edge of the hatching slit (Fig. 15), allowing gliding of the skin in only one direction: outward. Thus, one-way movement is generated by repeated, rapid stretching of the body (Boletzky, 1978-79).

Within the benthic family Octopodidae, many species produce large eggs from which large hatchlings develop that already have long arms with many suckers. These animals use their arms to crawl out through the hatch opening. In contrast, in most small-sized hatchlings with short arms, the arms are not used during hatching (but see Boletzky, 1984, for an exception to this rule).

ECOLOGICAL ASPECTS OF ENCAPSULATION

Cephalopods are found in virtually all marine environments, in inshore waters as well as in the open ocean from tropical to circumpolar latitudes, in surface waters and at great depths. At virtually all depths, cephalopods having different life styles coexist in the near-bottom water layer, so that eggs laid on the bottom may be those of nektonic or of demersal and benthic cephalopods. In contrast, in midwater only eggs of midwater species are found.

Hard egg shells appear to be typical of benthic and bentho-pelagic cephalopods laying large eggs at great water depths or at high latitudes (*Nautilus, Rossia*, cirrate octopods); together the size of the eggs and the low water temperatures result in long embryonic development. However, alternative solutions to the problem of long term protection of the embryos do exist. Thus *Sepia orbignyana* Férussac 1826, a species living in rather deep water, inserts eggs into the oscula of sponges (Naef, 1923). The elongate shape and the transparency of the egg case are reminiscent of large incirrate eggs, but in contrast to the statement of Naef (1923) saying that "complete jelly coats are not produced", it must be stressed that the chorion of these eggs is surrounded by the typical spirally coiled nidamental jelly and a rather tough outer membrane, all of which are unstained. Thus the sponges do not replace the protective function of capsules; they provide *complementary* protection against predators (camouflage), and they also maintain a steady water exchange around the egg capsule. Females of *Sepia elegans* Orbigny, 1835, generally fix their eggs on branches of octocorallians (Bouligand, 1961) so polyps completely surround the egg. Finally the incirrate habit of brooding the eggs has also proved successful at great water depths. However, under these conditions, apparently only the "holobenthic" mode is represented by octopodids producing large eggs, whereas in shallower waters, this mode coexists with the "meroben-thic" alternative that is characterized by a planktonic juvenile phase as shown by sympatric occurrence of octopodid sibling species distinguished by these adaptive strategies.

The *holopelagic* life cycle of the nektonic incirrate octopods, which produce large numbers of offspring of small individual size, is in many ways similar to that of squids producing floating eggs and egg masses, but in contrast to these, the nektonic incirrates invariably provide active protection in the form of "brooding" or ovovivipary.

Loliginid squids always fix their egg capsules to a substrate, either in such a way that the capsules hang from the point of fixation, or that they stick to sand particles or coarser substrata. The latter mode seems to be correlated with the production of very watery capsules having minimal weight in sea water, so that they move freely around the point of fixation and are thus continually flushed by water movement (Roper, 1965).

High water content of egg capsules clearly provides some protection against desiccation, as demonstrated by viable eggs collected on beaches above the water line, or from trawl nets that had been out of water for hours. Especially the eggs of *Sepia officinalis* are often washed ashore with the algae or grass weeds on which they are frequently laid. Under natural conditions, the embryos thus removed from their normal environment have a chance to survive only if they have not been exposed to high temperatures, and if they are again immersed, for example by a high tide. The apparently "wasteful" habit of many cuttlefishes and neritic squids of fixing their egg masses to easily detachable substrates must be counterbalanced by relatively high fecundity, i.e. high energy investment in both gamete and capsule production (Boletzky, 1981). In return, this behavior opens the possibility of "rafting" of eggs. Especially in *Sepia*, which remains close to the bottom and on the bottom throughout its life time, this may provide a means of dispersion of offspring.

DISCUSSION

Ecological aspects of encapsulation inevitably raise questions on adaptation, which can only be considered from the viewpoint of evolution. No matter to which particular theory of evolution one subscribes, the processes involved in adaptation appear complex. The present paper presents an attempt to find correlated processes in the life cycle of different cephalopods that have something to do with encapsulation. If particular features of encapsulation are viewed as the result of evolutionary change, it is legitimate to wonder which changes in adult, juvenile or embryonic morphology and physiology may be related to the former. Clearly some speculation is involved here, but it is perfectly acceptable as long as it is only used to handle established facts (not hypotheses).

In surveying different cephalopod groups, the foregoing sections have provided a number of arguments allowing one to suggest correlated changes in capsule structure, functional morphology of spermatozoa and skin structures of hatchlings. Within the decapods, the modifications are relatively clear, although several details remain to be clarified. As an example, the obscure phylogenetic position of the Sepiolidae (do they really belong to the Sepioidea, with which they share the character "eggs laid singly"?) raises a few problems; one is the questionable homology of "outer case" material in the egg capsules. Apart from this uncertainty, the homology of capsules and capsule-producing glands within the decapods is not called in question, however.

What can be said in this respect about the octopods? They lack nidamental glands in the mantle cavity, as do the Vampyromorpha, which are no doubt closely related to the Octopoda (see e.g. Young, 1977). Does their reproductive apparatus represent an ancestral condition? Assuming that it does would mean that nidamental glands in *Nautilus* and in decapods are analogous (evolved convergently), not homologous structures. This seems less likely than their homology. Consequently the absence of nidamental glands in the Vampyromorpha and Octopoda appears derived from a decapodan condition. Does this mean that nidamental glands have simply been *lost*?

Here speculation definitely has to come in if the question is to be pursued any further. But speculation can be firmly "based" on an embryological fact: the oviducal glands of both

the decapods and the octopods are formed by an ectodermal invagination (see Marthy, 1968). In decapods, the nidamental glands are formed later on by an adjacent ectodermic territory (Lemaire and Richard, 1970). In the early morphogenetic processes, synchronization of organogenesis in both these territories, and "lengthening" of the invagination would suffice to include the nidamental gland in the oviduct. My suggestion that such a process may have occurred at the outset of the vampyromorphan/octopodan line of descent is pure speculation. And yet, it may lead to a better understanding of the cirrate and incirrate modes of encapsulation, provided that the correlation between changes in organ development can be established.

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