

[REVIEW]

Evolutionary Changes in Developmental Potentials of the Embryo Proper and Embryonic Membranes along with the Derivative Structures in Atelocerata, with Special Reference to Hexapoda (Arthropoda)

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1. Introduction

The evolution of embryonic membranes is one of the most interesting comparative embryological subjects concerning Atelocerata (= Antennata = Tracheata = four myriapod classes + hexapods; cf. Heymons, 1901; Sharov, 1966), and has been discussed by many authors (*e.g.*, Heymons, 1901; Heymons and Heymons, 1905; Johannsen and Butt, 1941; Sharov, 1966; Jura, 1972; Anderson, 1973). Among them, Heymons (1901) and Heymons and Heymons (1905) advanced a theory focusing on the functional specialization between the embryo proper and embryonic membranes, as an approach to atelocerate evolution.

Heymons' view, despite its high potential value, has not been further developed, although, in relation to this, some comparisons in original papers (*e.g.*, Tiegs, 1940) and in reviews such as those quoted above have appeared, but no comprehensive argument concerning atelocerate evolution has been attempted. Much new information has been accumulated after Heymons, and it is timely to re-examine the functional specialization of the embryo proper and embryonic membranes in Atelocerata in regard to evolution.

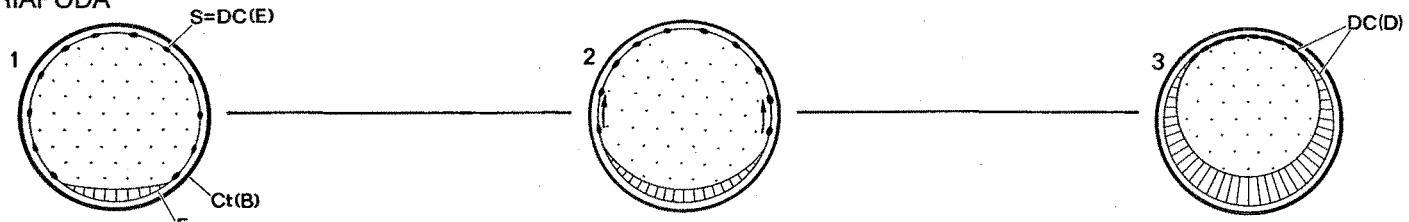
In the present paper, we reviewed the evolutionary changes of the embryo proper and embryonic membranes, and evolutionary transition of functional specialization between them in Atelocerata, focusing on the dorsal closure and secretion of the cuticular egg envelope (blastoderm and serosal cuticles), and we showed that a deeper insight to the amnioserosal fold in Ectognatha can be given in context of the transition of functional specialization between the embryo proper and embryonic membranes.

2. Developmental potentials of the embryo and embryonic membranes in each group of Atelocerata

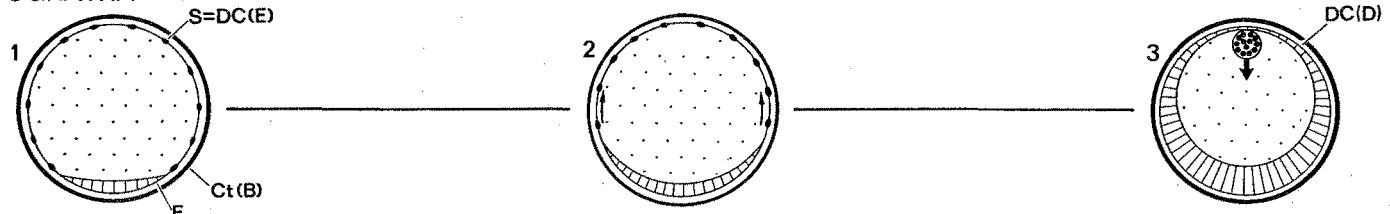
First, we define the terms used. In atelocerate embryogenesis, the germ disc or germ band forms, and the embryonic and extraembryonic areas differentiate. The term embryo can be applied to the whole egg at any developmental stage, but we defined it here in a narrow sense, *i.e.*, to the embryo proper which is directly derived from the germ disc/germ band.

The embryonic membranes are the serosa and amnion, and are the cellular membranes that occupy the extraembryonic area. The serosa is a cellular membrane directly derived from the extraembryonic blastoderm (Fig. 1a-1-d-1). The amnion is a second embryonic membrane, which develops in ectognathan hexapods and is produced from the margin of the embryo in association with the formation of the amnioserosal fold in the middle developmental stages (Fig. 1c-3, d-2). The embryonic membrane of the myriapods and entognathan hexapods, in which the amnion is not formed, is represented only by the serosa.

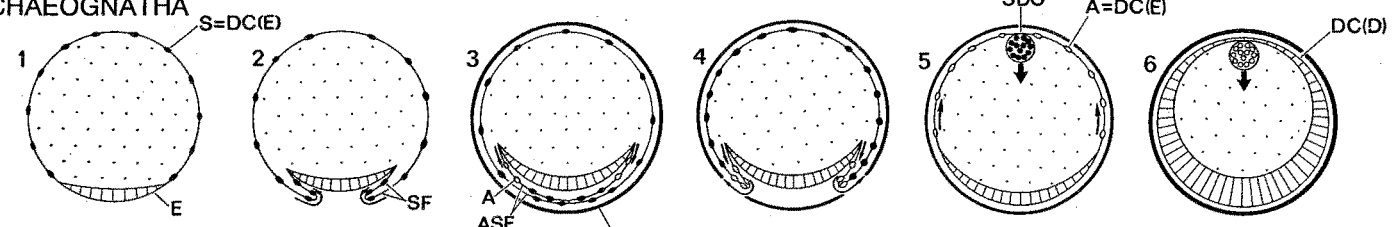
a. MYRIAPODA



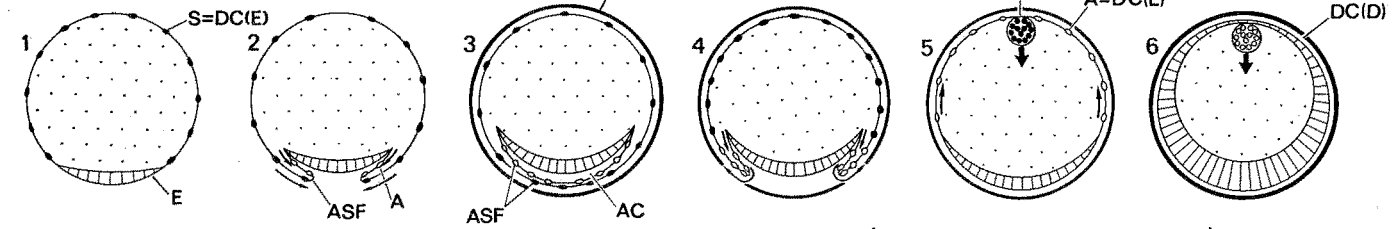
b. ENTOGNATHA



c. ARCHAEOGNATHA



d. ZYAGENTOMA-PTERYGOTA



▤ = embryo ● = serosa ○ = amnion

[It is known that the embryonic structure called 'primary dorsal organ' is formed in the extraembryonic area of the Symphyla, Collembola and Diplura (Uzel, 1898; Tiegs, 1940, 1942a, b; Jura, 1972). In the present paper, the structure is simply dealt with as a structure derived from the serosal area, and further reference to it is not given].

We examined the functions of the embryo and embryonic membranes, focusing on the secretion of the cuticular egg envelope and the formation of the dorsal closure. The cuticular egg envelope is a cuticular structure secreted during embryonic development beneath the chorion by the embryo and embryonic membrane or by the latter only: in some atelocerate groups, the cuticular egg envelope, called the blastoderm cuticle, is secreted by both the embryo and embryonic membrane (serosa) or by the whole blastoderm that is the precursor of the embryo and serosa (cf. Fig. 1a-1, b-1), whereas in others, it is secreted only by the embryonic membrane or serosa, and is called the serosal cuticle (cf. Fig. 1c-3, d-3).

The dorsal closure is the dorsal covering of the embryo or the hatched individual. The term 'dorsal closure' is currently used for dual meanings, *i.e.*, the dorsal covering (as here defined) and the dorsal-closing process (cf. Anderson, 1972a); we sometimes use this term under the latter sense. The dorsum of the developing embryo, which is at the egg surface, is covered by the embryonic membrane: *i.e.*, the serosa as in Figure 1a, b and the serosa or amnion as in Figure 1c, d. The dorsal covering of the embryo is here called the 'dorsal closure in the embryonic period'. The dorsal covering, *i.e.*, the dorsal body wall, of the hatched individual is called the 'definitive dorsal closure'. In some atelocerates the dorsal closure in the embryonic period participates in the definitive dorsal closure (cf. Fig. 1a-3), while in others, the dorsal closure in the embryonic period degenerates without participating in the definitive closure, and the definitive dorsal closure is exclusively derived from the embryo, *i.e.*, by the extension of lateral parts of the embryo (cf. Fig. 1b-3, c-6, d-6). In the latter, the dorsal closure in the embryonic period is a provisional, and is called the 'provisional dorsal closure'.

2.1. Myriapoda

In myriapods, *i.e.*, the Pauropoda, Diplopoda, Symphyla and Chilopoda, the most primitive state in Atelocerata is evident in regard to functional specialization between the embryo and embryonic membranes.

The blastoderm differentiates into the embryo and the embryonic membrane or the serosa: the embryonic membrane is represented only by the serosa. Firstly, the embryo is able to secrete the cuticular egg envelope (at the presumptive stage), in cooperation with the (presumptive) serosa (Fig. 1c-1) (Pauropoda: Tiegs, 1947; Diplopoda: Dohle, 1964; Symphyla: Tiegs, 1940; Chilopoda: Knoll, 1974; cf. Anderson, 1973). Secondly, the embryonic membrane or the serosa not only functions as a dorsal closure in the embryonic period (Fig. 1a-1-2), but also definitively differentiates into, and takes part in, the definitive dorsal closure (Pauropoda: Tiegs, 1947; Symphyla: Tiegs, 1940; Chilopoda: Heymons, 1901; no data for Diplopoda): the serosa has an ability to form a definitive dorsal closure in cooperation with the embryo (Fig. 1c-3). [A part of serosa in the Symphyla and Chilopoda degenerates without participating in definitive dorsal closure (Heymons, 1901; Tiegs, 1940; Knoll, 1974), accompanied by the formation of a secondary dorsal organ in chilopods. In Chilopoda, a wide membranous area 'membrana ventralis' appears along the embryonic median line in the course of the ventral flexure (Einkrümmung) of the embryo, and also takes part in the formation of the larval body (Heymons, 1901)].

Fig. 1 Embryonic development of different atelocerate groups: Myriapoda (a), Entognatha (b), Archaeognatha (c) and Zygentoma-Pterygota (d): successive stages, 1-3/6. Outermost egg envelope, the chorion, is omitted. Downward heavy arrows imply degeneration. A: amnion, AC: amniotic cavity, ASF: amnioserosal fold, Ct: cuticular egg envelope, Ct(B): blastoderm cuticle, Ct(S): serosal cuticle, DC: dorsal closure, DC(D): definitive dorsal closure, DC(E): dorsal closure in the embryonic period, E: embryo, S: serosa, SDO: secondary dorsal organ, SF: serosal fold. Concerning the development of the amnioserosal fold in Archaeognatha and Zygentoma-Pterygota, details are given in Figure 2. See text.

Thus, the embryo and embryonic membrane (serosa) are not separately specialized in regard to function in Myriapoda, as they function together, suggesting that the difference between the embryo and the embryonic membrane might not be critical.

2.2. Hexapoda

2.2.1. Entognatha

Aspects concerning the embryo and embryonic membrane in Collembola and Diplura (the Protura on which we have no available embryological knowledge is excluded from the discussion) may be similar to that of Myriapoda. In these hexapods as in myriapods, the embryonic membrane is represented by the serosa, and the secretion of the cuticular egg envelope is performed by both the embryo and serosa (Fig. 1b-1) (Collembola: Jura, 1972; Uemiya and Ando, 1987, 1991; cf. Tiegs, 1940; Diplura: Uzel, 1898; Tiegs, 1942b). However, in these hexapod groups in contrast to myriapods, the embryonic membrane or the serosa functions only as a dorsal closure in the embryonic period (Fig. 1b-2), and does not participate in the definitive dorsal closure (Fig. 1b-3) (Collembola: Philpitschenko, 1912; Uemiya, personal communication; Diplura: Tiegs, 1942b).

Thus, in Entognatha the contribution of serosa to the dorsal closure is restricted to the provisional one, and the formation of the definitive dorsal closure is exclusively fulfilled by the embryo. Accordingly, the functional specialization between the embryo and embryonic membrane in Entognatha may be recognized as an advanced step when compared to that in Myriapoda.

2.2.2. Ectognatha

In Ectognatha comprising Archaeognatha, Zygentoma and Pterygota, the functional specialization between the embryo and embryonic membranes is more advanced in comparison with Myriapoda and Entognatha, and new aspects have been added to the embryonic membranes (Archaeognatha: Heymons and Heymons, 1905; Larink, 1969; Machida *et al.*, 1992, 1994; Machida and Ando, 1994; Zygentoma: Heymons, 1897; Sharov, 1953; Wellhouse, 1953, 1954; Woodland, 1957; for review on 'ectognathous apterygotes' see Jura, 1972; numerous works on Pterygota, for reviews see Johannsen and Butt, 1941; Anderson, 1972a, b, 1973; Schwalm, 1988).

It should be emphasized that a second embryonic membrane or the amnion differentiates from the embryonic margin (Fig. 1c-3, d-2). [Especially in parasitic or holometabolan hexapods, there exist some exceptions where the amnion is not produced, but these may be considered as secondary modifications (cf. Johannsen and Butt, 1941; Anderson, 1972b; Ivanova-Kasas, 1972; Ando, 1988)]. As in the serosa, the amnion functions only as a dorsal closure in the embryonic period, and is never involved in the definitive dorsal closure: the larval body wall is exclusively formed by the embryo (Fig. 1c-6, d-6). In that the functional role of the embryonic membranes is restricted to the provisional role in regard to dorsal closure, the situation in ectognathous hexapods may be similar to that of entognathous. However, in ectognathous hexapods, the temporal specialization concerning provisional dorsal closure occurs between the serosa and amnion: that is, the serosa functions as the provisional dorsal closure during the first half of embryogenesis, and the amnion replaces the serosa, after the latter has secreted the cuticular egg envelope and degenerates.

It is noteworthy that the embryo entirely resigns from the secretion of the cuticular egg envelope in ectognathous hexapods, and that the functional role is exclusively allotted to the serosa (Fig. 1c-3, d-3). This is in sharp contrast to the situation in myriapods and entognathous hexapods.

In addition, in Ectognatha, a new structure derived from embryonic membranes, *i.e.*, the amnioserosal fold, is formed, and new functional aspects are assigned to the embryonic membranes or the serosa and amnion. A discussion concerning the amnioserosal fold is in Chapter 3. Here, we have reviewed the transition of functional specialization between the embryo and embryonic membranes along the atelocerate evolution, to which Heymons (1901) and Heymons and Heymons (1905) have provided much information. Table I summarizes the functional specializations between the embryo and embryonic membranes in different groups of Atelocerata: it is recognizable that the transition of functional specialization should have

Table 1 Functions of the embryo and embryonic membranes in different atelocerate groups.

	Embryo	Embryonic membranes	
		Serosa	Amnion
Myriapoda	1,3	1,2,3	—
Ectognatha	1,3	1,2	—
Archaeognatha	3	1,2'	2"
Zygentoma-Pterygota	3	1,2',4	2",4

Numerals imply functions shared: 1: secretion of the cuticular egg envelope, 2: dorsal closure in the embryonic period, 2': dorsal closure in the embryonic period, until relieved by the amnion, 2", dorsal closure in the embryonic period, after degeneration of the serosa, 3: definitive dorsal closure (formation of body wall or participation in the definitive dorsal closure), 4: formation of the amnioserosal fold—amniotic cavity system. For papers consulted, see text.

occurred along the atelocerate evolution. We believe that Ectognatha should have acquired the amnioserosal fold according to its background of being closely linked to such progressive functional specializations as we have reviewed, in particular regarding the secretion of the cuticular egg envelope.

3. Amnioserosal fold in Ectognatha

It is generally accepted that the amnioserosal fold is one of the most outstanding embryological features in Ectognatha. Some mechanical advantages, such as protection, have been assumed (Sharov, 1966; Ando, 1970, 1988; Zeh *et al.*, 1989), but nothing definite is known about its functional role (cf. Anderson, 1972a). However, the viewpoint put forward above should deepen our understanding concerning the amnioserosal fold in Ectognatha.

3.1. Archaeognatha

The development of the amnioserosal fold in Archaeognatha is summarized here, based on the descriptions of Machida *et al.* (1992, 1994) in machilid *Pedetontus unimaculatus*. First, a thickened area *ca.* 200 μm in diameter of the blastoderm appears at the posterior pole of the egg (*ca.* 1.3 \times 0.7 mm) which is the anlage of the embryo: the remaining area other than this is the serosa (Figs. 1c-1, 2a-1). The cells of the embryonic anlage concentrate to the center, to form a germ disc of *ca.* 100 μm in diameter. At the same time the serosal cells adjacent to the embryonic anlage participate in the concentrative movement of the cells involved in the formation of the germ disc, and move ventrally to the germ disc and roll up there, forming the serosal fold (Figs. 1c-2, 2a-2–3). The serosal cells then cover the entire surface of the egg. The serosa, including the serosal fold, starts to secrete the serosal cuticle (taking the form of a process beneath the center of the embryo, as a cuticular plug), and the continuous cuticular egg envelope covering the entire egg surface is completed.

As the embryo grows, cells move individually from the embryonic margin to form the amnion, which lies between the embryonic margin and serosal fold (Figs. 1c-3, 2a-4): the embryonic membrane fold is transformed from the serosal to the amnioserosal. The amnioserosal fold is, however, an ephemeral structure, and is soon withdrawn in the reverse order to form the serosal fold (Figs. 1c-4, 2a-5). Finally, the amnion and then the embryo reach the egg surface (Fig. 1c-5). The serosa starts to move on the egg surface and is condensed dorsally, forming the secondary dorsal organ, and then degenerates (Fig. 1c-5). Replacing the serosa, the amnion provides the dorsal closure in the embryonic period (provisional dorsal closure).

One of the most noticeable aspects of the archaeognathan embryonic membrane fold (serosal or amnioserosal fold) is that it is of short duration, and furthermore it varies in the length of its existence.

That is, generally, the withdrawal of the embryonic membrane fold varies in time from the stage of the early germ band (stage 2 in Machida, 1981) to the stage in which segmentation proceeds in the anterior body half and the ventral flexure deepens (stage 4 in Machida, 1981). In addition, there is a rare but extreme example that the fold regresses at the germ disc stage (stage 1 in Machida, 1981), in which the amnion is yet to be produced.

Thus, the archaeognathan amnioserosal fold ought to be functionally independent of, or little related to, the term of its existence and the participation of the amnion. Accordingly, we suggest that the functional role of the embryonic membrane fold in Archaeognatha should be represented by the serosal fold, and that it should lie in the formation of the serosal cuticle beneath the embryo: the formation of the cuticular layer is fulfilled just by the invasion of serosal cells under the embryo in the form of a serosal fold, and this leads to the completion of the continuous cuticular egg envelope covering all of the egg surface. The participation of the amnion may not be a requisite for the formation of the archaeognathan embryonic membrane fold: the addition of amnion to the serosal fold might be recognized as a passive change of the embryonic margin by the excessive stretching of serosal cells of the fold caused by the growth of the embryo. Thus, probably the principal functional role of the amnion is very simply that of a dorsal closure in the embryonic period (provisional dorsal closure), which replaces the serosa that is in the course of degenerating after secreting the serosal cuticle.

Our suggestion that the functional role of embryonic membrane fold in Archaeognatha lies in secreting the serosal cuticle beneath the embryo may seem more realistic in context regarding the transition of functional specialization between the embryo and embryonic membranes in Atelocerata. Archaeognatha may have evolved at the stage when the hexapods started to exploit terrestrial habitats (cf. Kukulová-Peck, 1987). There, the cuticular egg envelope which protects the egg from outside influences must have been a prerequisite for hexapods as well as for the myriapods and entognathous hexapods which had been endeavouring to find their way to similar environments (cf. Zeh *et al.*, 1989). In contrast, however, in the transition of the functional specialization between the embryo and embryonic membranes in the Atelocerata, the Archaeognatha has been at the stage the embryo discards the ability for secreting the cuticular layer as an egg envelope. Here, unless a member of Atelocerata, the Archaeognatha, had developed the embryonic membrane fold (serosal or amnioserosal one), it could not acquire a cuticular egg envelope covering the entire egg surface and maintain the continuity of a cuticular egg envelope, as the cuticular element could not be secreted beneath the embryo.

The most noticeable embryological transformation from Entognatha to primitive Ectognatha may be the 'long germ' of the myriapod type (cf. Jura, 1972) to the short germ (cf. Sander, 1984). This transformation implies the relative expansion of the serosal area maintaining the ability for cuticular secretion, and this must have been advantageous in terms of the loss of cuticular secretion by the embryonic area: that is, the rolling up of serosa linked to the formation of the serosal fold, for example in Archaeognatha, may have been minimized.

3.2. Zygentoma-Pterygota

For the examination of the zygentoman amnioserosal fold, we refer to the studies on *Lepisma saccharina* by Heymons (1897), Sharov (1953) and Larink (1983) (Heymons and Sharov reported that the amniotic pore which remains as a result of incomplete fusion of the amnioserosal fold is filled by a cuticular plug, but Larink's re-examination revealed that the amniotic pore is completely fused and closed as in the higher hexapods), and on *Ctenolepisma lineata* by Woodland (1957). Regarding Pterygota, there are numerous reports on the amnioserosal fold, and reviews such as Johannsen and Butt (1941), Anderson (1972a, b) and Schwalm (1988) can be referred to. There are also some examples that deviate from the generalization on the amnioserosal fold as addressed below, (including the cases in which the amnioserosal fold undevelops), especially in the parasitic or holometabolous pterygotes, but they may be considered as secondary modifications (other than the reviews quoted above, Ivanova-Kasas, 1972; Ando, 1988; Kobayashi and Ando, 1988).

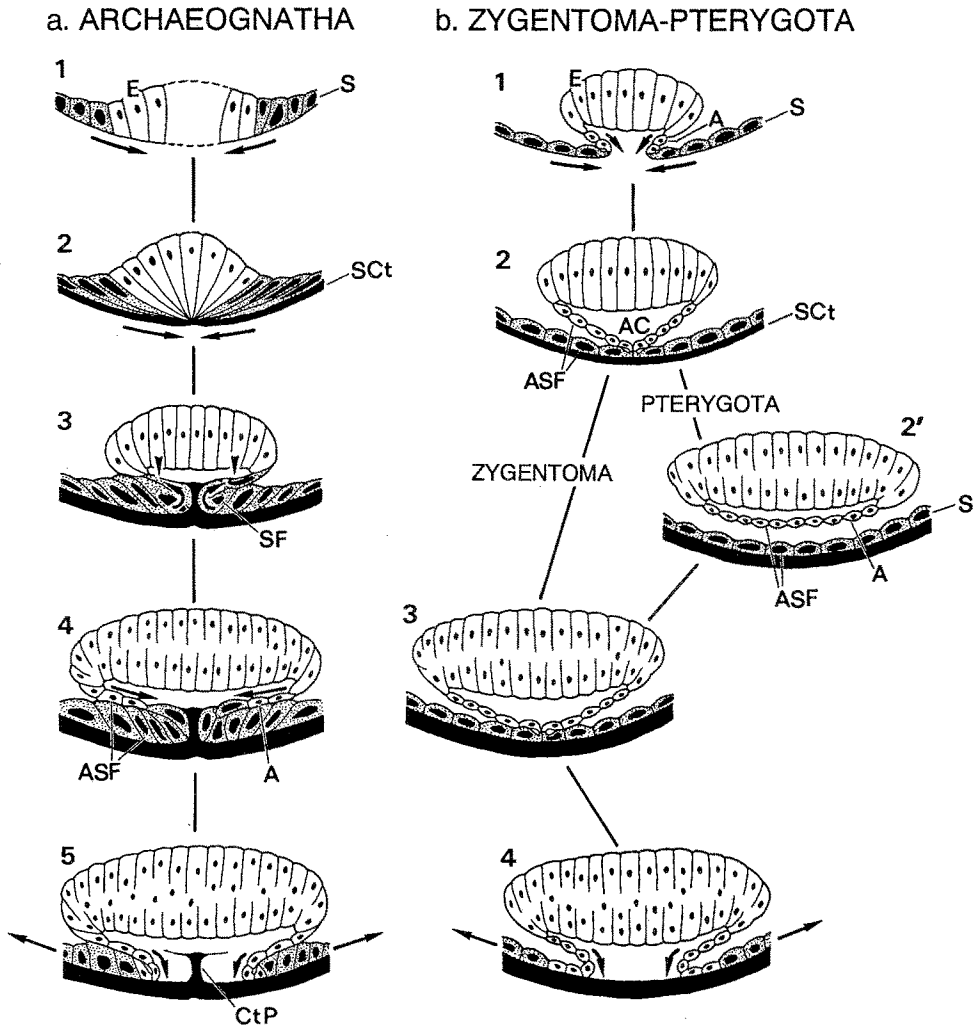


Fig. 2 Development of amnioserosal fold in Archaeognatha (a) and Zygentoma-Pterygota (b): successive stages, 1-4/5. A: amnion, AC: amniotic cavity, ASF: amnioserosal fold, CtP: cuticular plug, E: embryo, S: serosa, SCt: serosal cuticle, SF: serosal fold. See text (for arrowheads in a-3, see Chapter 3.2.1).

3.2.1. General features of the amnioserosal fold in Zygentoma-Pterygota

During the invagination of the early embryo or anatrepsis, the amnioserosal fold is formed by the production of the amnion from the embryonic margin and by the invasion of serosa underneath the embryo (Figs. 1d-2, 2b-1). The amnioserosal fold then fuses beneath the embryo, and the closed amniotic cavity appears between the embryo and amnioserosal fold (Figs. 1d-3, 2b-2). For a time (the diapause), embryogenesis continues in the same way (Fig. 2b-2-3). In Pterygota, during diapause, the amnioserosal fold separates into individual amniotic and serosal layers (recognizable as a more advanced condition than in Zygentoma), but it returns to the initial condition just before katrepsis (Fig. 2b-2'-3). In katrepsis, the amnioserosal fold is withdrawn (Figs. 1d-4, 2b-4), and the amnion and then the embryo appear at the egg surface (Fig. 1c-5). The serosa begins to move on the egg surface and is condensed, forming the secondary dorsal organ, and then degenerates (Fig. 1d-5). The amnion, replacing the serosa, provides a dorsal closure in the embryonic period (provisional dorsal closure).

The amnioserosal fold in Zygentoma-Pterygota may be characterized as follows. First, the formation of the amnioserosal fold is directly linked to the production of the amnion, the amnion being a requisite for the embryonic membrane fold (amnioserosal fold). Second, the amnioserosal fold fuses beneath the embryo to form a closed amniotic cavity, and the elaborate 'amnioserosal fold-amniotic cavity system' appears. This system should be understood within the framework that: first, the system is so firmly established in structure and manner of formation that this should be recognized as a synapomorphy of the Zygentoma-Pterygota; second, this system is maintained at a temporarily fixed embryogenetic stage for each group, and therefore must be an important structure integrated as part of its embryogenesis. A contrast between the amnioserosal folds of the Archaeognatha and Zygentoma-Pterygota is evident: in the former, the participation of the amnion in the embryonic membrane fold may not be so critical, and the fold does not provide such an established and durable system as in the latter.

Thus, the established and evolutionarily conservative amnioserosal fold-amniotic cavity system in Zygentoma-Pterygota should be recognized as having a special role in embryogenesis, and might be related to the protection of the embryo, as has been suggested (e.g., Sharov, 1966). Whatever the function is, some special functional aspects concerning this system have been newly allotted to the amnion and serosa constituent (Table 1). However, the ancestral Zygentoma-Pterygota should have maintained the continuity of a cuticular egg envelope covering the entire egg surface, assuring the further radiations in terrestrial habitats, as did the ancient Archaeognatha. That is, the original functional role of this system may have been the secretion of a cuticular layer as an egg envelope beneath the embryo, the same as in the archaeognathan embryonic membrane fold. Although not directly substantiating this hypothesis, we can provide two interesting points: 1) it is after the completion of anatrepsis and before katatrepsis that the cuticular egg envelope (serosal cuticle) is secreted in Zygentoma-Pterygota: namely, it is secreted during the diapause when the embryo is completely concealed with the amnioserosal fold (see original papers, e.g., Tojo and Machida, 1997: cf. Haget, 1977); 2) the cuticular egg envelope is not secreted in pterygotes in which the amnioserosal fold is under- or undeveloped, that is, the fold does not cover the embryo (e.g., *Drosophila melanogaster*, cf. Johannsen and Butt, 1941, Campos-Ortega and Hartenstein, 1985). [It is known that the serosal cuticle is not secreted in the eggs of some hexapod groups which have well-developed amnioserosal folds. It might be thought that the loss of cuticular secretion in some hexapods should have been brought about by the secondary modification, probably in association with the alteration in reproduction strategy and the high elaboration of egg membranes (chorion etc.) (cf. Hinton, 1981; Zeh *et al.*, 1989)].

The addition of the amnion to the serosal fold in Archaeognatha may be read as the 'preadaptation' to the amnioserosal fold or the amnioserosal fold-amniotic cavity system shown in Zygentoma-Pterygota: for the archaeognathan embryonic membrane fold, the participation of amnion is not so essential. If so, what was the cause of the close linkage of the amniotic production to the formation of the embryonic membrane folds, which may be one of the significant features of this system in Zygentoma-Pterygota? It might be possible to speculate as follows. Generally, the relative size of the embryo when the embryonic membrane fold forms may be larger in Zygentoma-Pterygota than that in Archaeognatha, for example, in zygentoman *Lepisma saccharina*, the embryo of $150 \times 300 \mu\text{m}$ versus the egg of 0.8–1.1 mm (Sharov, 1953 and after his Fig. 9), and in archaeognathan *Pedetontus unimaculatus* the embryo of $100 \mu\text{m}$ in diameter versus the egg of $0.7 \times 1.3 \text{ mm}$ (Machida *et al.*, 1990, 1994). To encompass the larger embryo by the serosal fold, as a matter of course, the serosal cells would be required to roll up more and to stretch and attenuate more (for example, the serosal parts as indicated by the arrowheads in Fig. 2a-3 should be more stretched): and it is likely that against such a problem the differentiation of the amnion occurred in parallel with the formation of the embryonic membrane fold; which in turn might ensure the further immersion or invagination of the embryo into the egg inside, thereby allowing the elaboration of the embryonic membrane fold into the amnioserosal fold-amniotic cavity system.

We have mentioned that the amnioserosal fold of Zygentoma-Pterygota fuses beneath the embryo to form the closed amniotic cavity and that it leads to the acquirement of an established and elaborate amnioserosal fold-amniotic cavity system in this group. We also concluded that the original functional role of

the fold or the system must have lain in the secretion of a cuticular layer (cuticular egg envelope) beneath the embryo, in lieu of the embryo which has lost the ability. We find, however, that the embryogenesis of zygotoman *Thermobia domestica* may be in contradiction.

3.2.2. Amnioserosal fold–amniotic cavity system in a zygotoman *Thermobia domestica*

According to Wellhouse (1953, 1954) and Woodland (1957), in *Thermobia*, the amnioserosal fold does not fuse, and the amniotic pore remains open. This is a deviation from our above-mentioned generalization on the amnioserosal fold–amniotic cavity system. Furthermore, according to Woodland (1957), the germ disc of this zygotoman secretes a cuticular layer ‘subchorionic cuticle’ at the stage the embryo is still on the egg surface: after anatrepsis and the formation of the amnioserosal fold, the serosa lays the subchorionic cuticle, and the continuous cuticular egg envelope covering the entire egg surface is completed. As reviewed (Chapter 2), however, the secretion of the cuticular layer as an egg envelope by the embryo (embryonic area) should be seen only at levels less advanced than Archaeognatha in regard to functional specialization of the embryo and embryonic membranes, and such an ability of the embryo should have been discarded at levels more advanced than Entognatha.

The case of *Thermobia* may be treated as follows. *Thermobia* embryo secretes a subchorionic cuticle which has an extension corresponding to the embryo’s size: according to Woodland (1957), the embryo at the stage of cuticular secretion is more than 850 μm in length, and it is presumed from his description and Figure 20, to be 200–350 μm in width. On the other hand, also according to Woodland, the amnioserosal folds of *Thermobia* are never closer together than 800 μm longitudinally and 150 μm transversely. That is, the embryonic subchorionic cuticle and the amniotic pore in *Thermobia* approximately agree in extension with each other. Re-examination is needed, but in *Thermobia* it is likely that the embryonic subchorionic

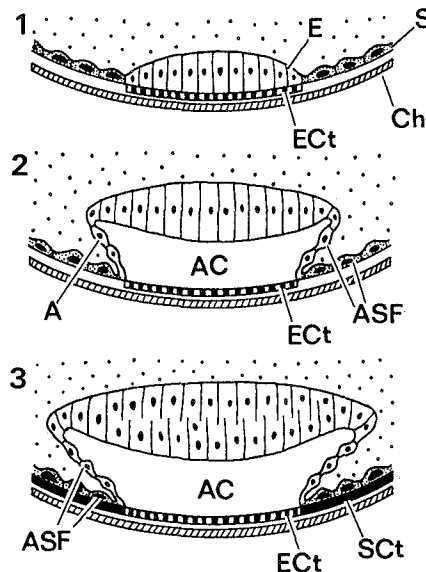


Fig. 3 A supposed amnioserosal fold–amniotic cavity system for *Thermobia*, reconstructed from Woodland (1957), successive stages 1–3. 1: Secretion of subchorionic cuticle by embryo. 2: Formation of amnioserosal fold, linked to anatrepsis. Amniotic cavity bounded with embryo and amnion is formed, and the opened amniotic pore is plugged and closed by the embryonic subchorionic cuticle. 3: Secretion of the subchorionic cuticle by the serosa, which leads to the completion of a continuous cuticular layer covering the egg surface in cooperation with the embryonic subchorionic cuticle previously secreted. A: amnion, AC: amniotic cavity, ASF: amnioserosal fold, Ch: chorion, E: embryo, ECt: embryonic subchorionic cuticle, S: serosa, Sct: serosal subchorionic cuticle.

cuticle enters between the wide-apart amnioserosal folds, formed just the same way as in other Zygentoma-Pterygota, to form the closed amniotic cavity, as Schwalm (1988) suggested. It is possible that the amnioserosal fold-amniotic cavity system should be acquired also in this zygentoman, although in a different way from in the other zygentomans and pterygotes. This speculation on *Thermobia* is diagrammatically represented in Figure 3 (details are given in its legend). Wellhouse's (1953) photographs (Figs. 9, 10) seem to support our speculation.

Thus, we consider that the amnioserosal fold-amniotic cavity system should exist also in *Thermobia*. Using a phylogenetic tree (Fig. 4, after Mendes, 1991), we examined the regular amnioserosal fold-amniotic cavity system of zygentomans *Ctenolepisma* (Woodland, 1957) and *Lepisma* (Larink, 1983) and pterygotes of which the amniotic pore is closed by the complete fusion of the amnioserosal folds [Fig. 2b; amnioserosal fold-amniotic cavity system type I (ASF-AC I), I in Fig. 4], and that of *Thermobia* in which the amniotic pore is plugged and closed by the embryonic cuticular egg envelope [Fig. 3; amnioserosal fold-amniotic cavity system type II (ASF-AC II), II in Fig. 4]. The following interpretation may be the most parsimonious. First, the ASF-AC I is acquired in the stem of the Zygentoma-Pterygota as an autapomorphy, and the ASF-AC I is shared in the zygentoman and pterygote lineages as a synapomorphy. Then, inherited as a symplesiomorphy by the zygentoman and pterygote descendants, the ASF-AC I is handed over to the zygentomans *Lepisma* and *Ctenolepisma* and the pterygotes, but is transformed into the ASF-AC II in zygentoman *Thermobia* to be its autapomorphy.

The unique condition found in *Thermobia*, which may seemingly represent the primitive state, proves to be secondarily derived as the specialization of amnioserosal fold-amniotic cavity system. As part of this specialization, the secretion of the cuticular egg envelope by the embryo newly appears or revives, and is not the direct inheritance from lower atelocerates. In addition, the widely opened amniotic pore is one of the specialized features in *Thermobia*. Hence, the foregoing generalization on the amnioserosal fold-amniotic cavity system for Zygentoma-Pterygota may be taken as valid, as well as the foregoing review on the functional specialization of the embryo and embryonic membranes in Atelocerata.

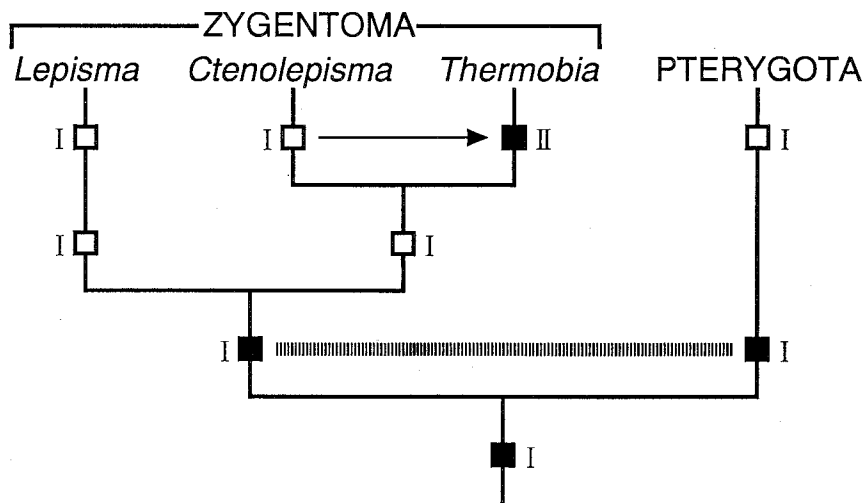


Fig. 4 Examination on the character state of amnioserosal fold-amniotic cavity systems in Zygentoma. Only three genera, i.e., *Lepisma*, *Ctenolepisma* and *Thermobia*, are dealt with: phylogenetic relationships between them are after Mendes (1991). The sister group of Zygentoma-Pterygota is supposed to be Archaeognatha which possess the embryonic membrane (serosal/amnioserosal) fold regarded as a prototype of the system. See text.

4. Conclusion

As we have pointed out, the atelocerate evolution has been proceeding, involving functional specialization between the embryo and embryonic membranes, accompanied by the differentiation of new embryonic membrane or the amnion, and the acquirement of a new structure or the amnioserosal fold or amnioserosal fold-amniotic cavity system (Table 1). We realize that the amnioserosal fold should have been acquired in Ectognatha, closely linked to the transition of functional specialization between the embryo and embryonic membranes along the atelocerate evolution.

In summary, we illustrate the events which should have happened to the embryo and embryonic membranes in the course of atelocerate evolution, on a phylogenetic tree (the 'Entognatha-Ectognatha system' of Hennig, 1969) (Fig. 5) [events mentioned in this figure are also applicable, without alteration, to other phylogenies currently proposed such as those of Kukalová-Peck (1987), Štys and Biliński (1990) and Štys *et al.* (1993), because these phylogenies agree in supporting the monophylies of the Hexapoda, Ectognatha and Dicondylia]. Figure 5 can be read as follows. 1) Plesiomorphic is the condition that the functional specialization between the embryo and embryonic membrane (serosa) concerning the dorsal closure and secretion of the cuticular egg envelope is not definite. Such a condition is seen in Myriapoda. 2) In Hexapoda, the serosa renounces participation in the definitive dorsal closure, and consequently it is exclusively performed by the embryo: this is an autapomorphy of this group. 3) Further, in Ectognatha, the following occurs as autapomorphies of this group: i) the renouncement of the ability to secrete the cuticular egg envelope by the embryo, ii) the formation of the embryonic membrane fold (serosal or amnioserosal) for the cuticular secretion beneath the embryo, iii) the differentiation of the amnion, and iv) the temporal specialization of provisional dorsal closure by the serosa and amnion. 4) Finally, Zygentoma-Pterygota acquire the following autapomorphies: i) the close linkage of the production of the amnion to the formation of the embryonic membrane fold, and ii) the amnioserosal fold-amniotic cavity system and the assignment of functional aspects concerned to the embryonic membranes.

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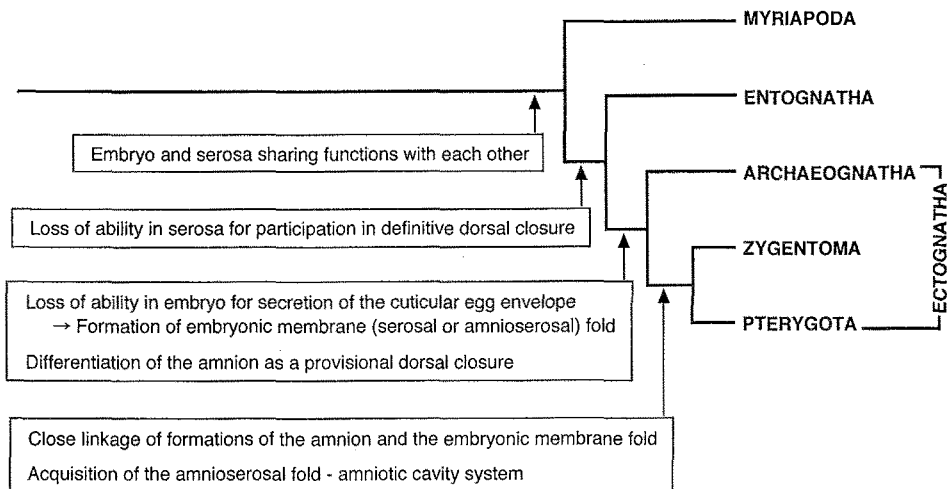


Fig. 5 Illustrating the events which should have happened to the embryo and embryonic membranes in the course of atelocerate evolution, on a phylogenetic tree. See text.

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References

- Anderson, D. T. (1972a) The development of hemimetabolous insects. In S. J. Counce and C. H. Waddington (eds.), *Developmental Systems: Insects, Vol. 1*, pp. 95-163. Academic Press, London.
- Anderson, D. T. (1972b) The development of holometabolous insects. In S. J. Counce and C. H. Waddington (eds.), *Developmental Systems: Insects, Vol. 1*, pp. 165-242. Academic Press, London.
- Anderson, D. T. (1973) *Embryology and Phylogeny in Annelids and Arthropods*. Pergamon Press, Oxford.
- Ando, H. (1970) Embryonic development. In T. Uchida (ed.), *Systematic Zoology, Vol. 7, Arthropoda IIIa, Insecta I*, pp. 37-130. Nakayama-Shoten, Tokyo. (in Japanese).
- Ando, H. (1988) Insecta. In K. Dan, K. Sekiguchi, H. Ando and H. Watanabe (eds.), *Embryology of Invertebrates, Vol. 2*, pp. 131-248. Baifukan, Tokyo. (in Japanese).
- Campos-Ortega, J. A. and V. Hartenstein (1985) *The Embryonic Development of Drosophila melanogaster*. Springer-Verlag, Berlin.
- Dohle, W. (1964) Die Embryonalentwicklung von *Glomeris marginata* (Villers) in Vergleich zur Entwicklung anderer Diplopoden. *Zool. Jb. Anat.*, **81**, 241-310.
- Haget, A. (1977) L'embryologie des insectes. In P. P. Grasse (ed.), *Traité de Zoologie, Vol. 8, Fasc. 5B*, pp. 1-387. Masson, Paris.
- Hennig, W. (1969) *Die Stammesgeschichte der Insekten*. Kramer, Frankfurt am Main.
- Heymons, R. (1897) Entwicklungsgeschichtliche Untersuchungen an *Lepisma saccharina* L. *Z. Wiss. Zool.*, **62**, 583-631.
- Heymons, R. (1901) Die Entwicklungsgeschichte der Scolopender. *Zoologica (Stuttg.)*, **13**, 1-244.
- Heymons, R. and H. Heymons (1905) Die Entwicklungsgeschichte von *Machilis*. *Verh. Dtsch. Zool. Ges.*, **15**, 123-135.
- Hinton, H. E. (1981) *Biology of Insect Eggs. Vols. I-III*. Pergamon Press, Oxford.
- Ivanova-Kasas, O. M. (1972) Polyembryony in insects. In S. J. Counce and C. H. Waddington (eds.), *Developmental Systems: Insects, Vol. 1*, pp. 243-271. Academic Press, London.
- Johannsen, O. A. and F. H. Butt (1941) *Embryology of Insects and Myriapods*. McGraw-Hill, New York.
- Jura, Cz. (1972) Development of apterygote insects. In S. J. Counce and C. H. Waddington (eds.), *Developmental Systems: Insects, Vol. 1*, pp. 49-94. Academic Press, London.
- Knoll, H. J. (1974) Untersuchungen zur Entwicklungsgeschichte von *Scutigera coleoptrata* L. (Chilopoda). *Zool. Jb. Anat.*, **92**, 47-132.
- Kobayashi, Y. and H. Ando (1988) Phylogenetic relationships among the lepidopteran and trichopteran suborders (Insecta) from the embryological standpoint. *Z. Zool. Syst. Evolut.-forsch.*, **26**, 186-210.
- Kukulová-Peck, J. (1987) New Carboniferous Diplura, Monura, and Thysanura, the hexapod ground plan, and the role of thoracic side lobes in the origin of wings (Insecta). *Can. J. Zool.*, **65**, 2327-2345.
- Larink, O. (1969) Zur Entwicklungsgeschichte von *Petrobius brevistylis* (Thysanura, Insecta). *Helgoländer Wiss. Meeresunters.*, **19**, 111-155.
- Larink, O. (1983) Embryonic and postembryonic development of Machilidae and Lepismatidae (Insecta: Archaeognatha et Zygentoma). *Entomol. Gen.*, **8**, 119-133.
- Machida, R. (1981) External features of embryonic development of a jumping bristletail, *Pedetontus unimaculatus* Machida (Insecta, Thysanura, Machilidae). *J. Morphol.*, **168**, 339-355.
- Machida, R. and H. Ando (1994) Reduction of lateral yolk folds in flattened eggs of a jumping bristletail, *Pedetontus unimaculatus* Machida (Microcoryphia: Machilidae). *Int. J. Insect Morphol. Embryol.*, **23**, 293-295.
- Machida, R., T. Nagashima and H. Ando (1990) The early embryonic development of the jumping bristletail *Pedetontus unimaculatus* Machida (Hexapoda: Microcoryphia, Machilidae). *J. Morphol.*, **206**, 181-195.
- Machida, R., T. Nagashima and H. Ando (1992) Amnioserosal fold of the jumping bristletail *Pedetontus unimaculatus* Machida (Hexapoda, Microcoryphia, Machilidae). *Proc. Arthropod. Embryol. Soc. Jpn.*, **27**, 13-15.
- Machida, R., T. Nagashima and H. Ando (1994) The embryonic development of the jumping bristletail *Pedetontus unimaculatus* Machida, with special reference to embryonic membranes (Hexapoda: Microcoryphia, Machilidae). *J. Morphol.*, **220**, 147-165.
- Mendes, L. F. (1991) On the phylogeny of the genera of Lepismatidae (Insecta: Zygentoma). In G. K. Veeresh, D. Rajagopal and C. A. Viraktamath (eds.), *Advances in Management and Conservation of Soil Fauna*, pp. 3-13. Oxford and IBH Publishing Co. Pvt. Ltd., New Delhi.

- Philipschenko, J. (1912) Beiträge zur Kenntnis der Apterygoten. III. Die Embryonalentwicklung von *Isotoma cinerea* Nic. *Z. Wiss. Zool.*, **103**, 519-660.
- Sander, K. (1984) Extrakaryotic determinants, a link between oogenesis and embryonic pattern formation in insects. *Proc. Arthropod. Embryol. Soc. Jpn.*, **19**, 1-12.
- Schwalm, F. E. (1988) *Insect Morphogenesis*. Karger, Basel.
- Sharov, A. G. (1953) Razvitje schetinokvostok (Thysanura, Apterygota) v svyzi s problemoi filogenii nasekomykh. *Trud. Inst. Morf. Zhivot.*, **8**, 63-127.
- Sharov, A. G. (1966) *Basic Arthropodan Stock with Special Reference to Insects*. Pergamon Press, Oxford.
- Štys, P. and S. Biliński (1990) Ovariole types and the phylogeny of hexapods. *Biol. Rev.*, **65**, 401-429.
- Štys, P., J. Zrzavý and F. Weyda (1993) Phylogeny of the hexapoda and ovarian metamorphism. *Biol. Rev.*, **68**, 365-379.
- Tiegs, O. W. (1940) The embryology and affinities of Symphyla, based on a study of *Hanseniella agilis*. *Q. J. Microsc. Sci.*, **82**, 1-225.
- Tiegs, O. W. (1942a) The 'dorsal organ' of collembolan embryos. *Q. J. Microsc. Sci.*, **83**, 153-169.
- Tiegs, O. W. (1942b) The 'dorsal organ' of the embryo of *Campodea*. *Q. J. Microsc. Sci.*, **84**, 35-47.
- Tiegs, O. W. (1947) The development and affinities of the Pauropoda, based on a study of *Pauropus silvaticus*. Part I. *Q. J. Microsc. Sci.*, **88**, 165-267.
- Tojo, K. and R. Machida (1997) Embryogenesis of a mayfly, *Ephemera japonica* McLachlan (Insecta: Ephemeroptera, Ephemeridae), with special reference to abdominal formation. *J. Morphol.*, **234**, 97-107.
- Uemiyama, H. and H. Ando (1987) Blastodermic cuticles of a springtail, *Tomocerus ishibashii* Yosii (Collembola: Tomoceridae). *Int. J. Insect Morphol. Embryol.*, **16**, 287-294.
- Uemiyama, H. and H. Ando (1991) Mesoderm formation in a springtail, *Tomocerus ishibashii* Yosii (Collembola: Tomoceridae). *Int. J. Insect Morphol. Embryol.*, **20**, 283-290.
- Uzel, H. (1898) *Studien über die Entwicklung der Apterygoten Insecten*. Friedländer & Sohn, Berlin.
- Wellhouse, W. T. (1953) The Embryology of *Thermobia domestica* Packard. Dr. thesis, Iowa State College.
- Wellhouse, W. T. (1954) The embryology of *Thermobia domestica* Packard. *Iowa St. Coll. J. Sci.*, **28**, 416-417.
- Woodland, J. T. (1957) A contribution to our knowledge of lepismatid development. *J. Morphol.*, **101**, 523-577.
- Zeh, D. W., J. A. Zeh and R. L. Smith (1989) Ovipositors, amnions and eggshell architecture in the diversification of terrestrial arthropods. *Q. Rev. Biol.*, **64**, 147-168.