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Embryology of the Mecoptera (Panorpidae, Panorpodidae, Bittacidae and Boreidae)*

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Synopsis

The comparative embryology of seven species of Mecoptera, representing four families, is studied. Especially the entire embryonic development is described in *Bittacus laevipes*, *Panorpodes paradoxus* and *Panorpa pryeri*, and the early and middle development of *Boreus westwoodi*.

The embryonic period is divided into nine stages in *P. pryeri*, and eight in *Pd. paradoxus* and *B. laevipes*. The germ rudiment formation of *Pd. paradoxus* is of the "immersed" type, and the germ band formation of *B. laevipes*, *Bo. westwoodi* and *P. pryeri* is of the "superficial" type. The germ band of *B. laevipes*, *Bo. westwoodi* and *Pd. paradoxus* is classified as the short germ type, and that of *P. pryeri* is the semilong germ type.

The mesoderm (inner layer) formation is of the "proliferating" type in *B. laevipes*, and of the "invaginating" type in *Bo. westwoodi*, *Pd. paradoxus* and *P. pryeri*. In the cephalognathal region, there are six pairs of ectodermal invaginations. A single invagination appears on the median line of the labrum and on the medioventral line at the intersegmental region between the ninth and tenth abdominal segments.

The ventral nerve cord consists of sixteen pairs of ganglia, one pair for each segment from the mandibular to the tenth abdominal. The median cord has the neuroblasts that participate in ganglion formation. The perineurium originates from the outer cells of the lateral cord. The formation of the larval compound eyes is described in *B. laevipes* and *P. pryeri*. The optic plate of *Pd. paradoxus* becomes vestigial during the embryonic stage, and the first-instar larva has no eyes.

Eighteen pairs of coelomic sacs are formed, one pair for each of the labral,

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antennal, three gnathal, three thoracic, and ten abdominal segments.

The stomodaeum and proctodaeum are formed in the usual manner as ectodermal invaginations. The anterior and posterior midgut rudiments are derived from proliferating cells of the stomodaeal and proctodaeal blind ends, respectively. Therefore the midgut epithelium is entirely ectodermal in origin.

The phylogenetic relationship of the four mecopteran families is discussed, and a phylogenetic tree is given, based on the embryological data presented.

Key words: Egg, organogenesis, *Bittacus*, *Boreus*, *Panorpodes*, *Panorpa*, phylogeny.

Introduction

The Mecoptera are known first from the Permian (WILLMANN, 1987), and this fossil record is the oldest among the holometabolous insects. Based on the fossil record, comparative morphology, and wing-venation, the Mecoptera are considered to be one of the most primitive groups in the Holometabola (TILLYARD, 1935; HENNIG, 1981).

It is thought that the embryology of the Mecoptera shows the basic developmental pattern of the embryology of the panorpoid orders and other Holometabola. There are six papers concerning the embryology of the Mecoptera, in addition to an unpublished thesis by WOLF (1961) in *Panorpa communis*. Two of these papers are dealing with the early embryology of *P. pryeri* (ANDO, 1960) and of *P. japonica*, *Panorpodes paradoxus*, *Bittacus mastrillii* and *B. marginatus* (ANDO, 1973). The remaining four are those on the development of the larval abdominal legs in *P. pryeri* and *B. mastrillii* (ANDO and HAGA, 1974), of the larval eye (ANDO and SUZUKI, 1977) and alimentary canal in *P. pryeri* (SUZUKI and ANDO, 1981), and of the larval eye in *Pd. paradoxus* (SUZUKI, 1985). However, the entire embryonic development of any of the Mecoptera has remained unknown.

Furthermore, on the relationship of families of Mecoptera, there are some arguments. For example, TILLYARD (1935) divided the Mecoptera into two suborders, *i.e.*, the Protomecoptera including the Notiothaumidae and Meropeidae, which have many cross veins on the wings as a primitive character, and the Eumecoptera including the Nannochoristidae, Bittacidae, Boreidae and Panorpidae, which have more evolved characters. HINTON (1958) advocated, however, that the Boreidae should be separated from the Mecoptera, as a new order Neomecoptera, based on the comparative anatomy of the larvae of the panorpoid orders. From the comparative morphology of the external genitalia, MICKOLEIT (1978) and WILLMANN (1981) suggested that the Nannochoristidae and Bittacidae are the most primitive mecopteran families, and they expressed some doubts about the views of TILLYARD (1935) and HINTON (1958). Recently, WILLMANN (1987) proposed a phylogenetic system for both the extinct and extant Mecoptera.

I have investigated the comparative embryology of the four families Panorpidae, Panorpodidae, Boreidae and Bittacidae. In this paper I describe the embryonic development

of these families in detail, and discuss the phylogenetic relation of these families from the comparative embryological views.

Materials and Methods

Specimens used in the present study include four families, four genera and seven species of Mecoptera: *Panorpa pryeri*, *P. japonica*, *P. nipponensis*, and *P. helena* of the Panorpidae; *Panorpodes paradoxus* of the Panorpodidae; *Bittacus laevipes* of the Bittacidae; and *Boreus westwoodi* of the Boreidae.

Pregnant females of *P. pryeri* were collected at Sugadaira, Sanada, in Nagano Prefecture, Japan, in June and July 1980 to 1981. Each insect was kept in a small plastic cage with soil or quartz sand, and fed on killed dipteran or lepidopteran insects. Under these conditions the females laid their eggs. Newly laid eggs were transferred to another plastic case with a humid plaster bottom and kept at 21°C.

Matured insects of *Pd. paradoxus* were captured at Sugadaira and Kakuma in Sanada, Seni in Suzaka, and Usuda in Saku, all in Nagano Prefecture, in June and July 1976 to 1982. Many pairs of the insects were kept in a screened cage with humid tissue papers, and fed on the pollen and nectars of various flowers (*Chrysanthemum leucanthemum*, *Euonymus sieboldiana*, *Cornus controversa*, etc.). Newly laid eggs on or in the tissue papers were kept in conditions similar to those described for *P. pryeri*.

Mated females of *B. laevipes* were collected at Kakuma in August and September during 1978 to 1981. Each female was kept in a small plastic cage with a humid filter paper. The oviposited and dropped eggs on the filter paper were placed in a plastic cage with a humid plaster bottom and kept at room temperature.

Pregnant females of *Bo. westwoodi* were obtained at Tübingen, West Germany, in December 1979, and reared with mosses in a laboratory, and eggs were collected from the mosses. All of these procedures and fixation of eggs were done by Drs. G. MICKOLEIT and E. MICKOLEIT. Larvae and eggs of this species also were collected and fixed by Dr. H. ANDO at Freiburg and Tübingen in early April 1975.

Eggs and larvae were fixed in alcoholic Bouin's fluid warmed to 40°C (80°C or room temperature in some cases) for 30 min. Some newly laid eggs of *B. laevipes* were fixed in 2.5% glutaraldehyde in 0.05M cacodylate buffer at pH 7.2 for 12 hr and postfixed in Carnoy's fluid for 30 min at room temperature. After fixation, materials were preserved in 70% ethyl alcohol.

In the case of observations of external features of embryos, the chorion, serosa and serosal cuticle were removed by forceps, then the bare eggs were stained with Mayer's hematoxylin, and observed in distilled water. In order to observe the thoracic and abdominal musculature of the first-instar larva of *P. pryeri*, the head and posterior half of the abdomen were removed, and the fixed larvae were stained with eosin, and observed in terpineol after dehydration.

For sectioning eggs and larvae, the materials were embedded in paraffin, paraffix or paraplast, after dehydration through an ethyl, normal or tertiary butyl alcohol series. They were cut into 6-10μm sections and stained with Delafield's hematoxylin and eosin, and borax carmine in some cases.

Drawing were made with the aid of an Abbe camera lucida.

Materials for the scanning electron microscope were prepared as follows. Materials fixed by the methods mentioned above were rinsed in distilled water to which was added a little detergent. After dehydration through an ethyl or tertiary butyl alcohol-isoamyl acetate series, they were dried by the critical-point drying method and coated with gold. In the case of *B. laevipes* eggs, surface granules of the chorion were removed in 5-10% sodium hypochlorite solution. Preparations were examined under the scanning electron microscope (model JSM T-200 of JEOL).

Observations

I. Oviposition and organization of newly laid eggs

Panorpa pryeri

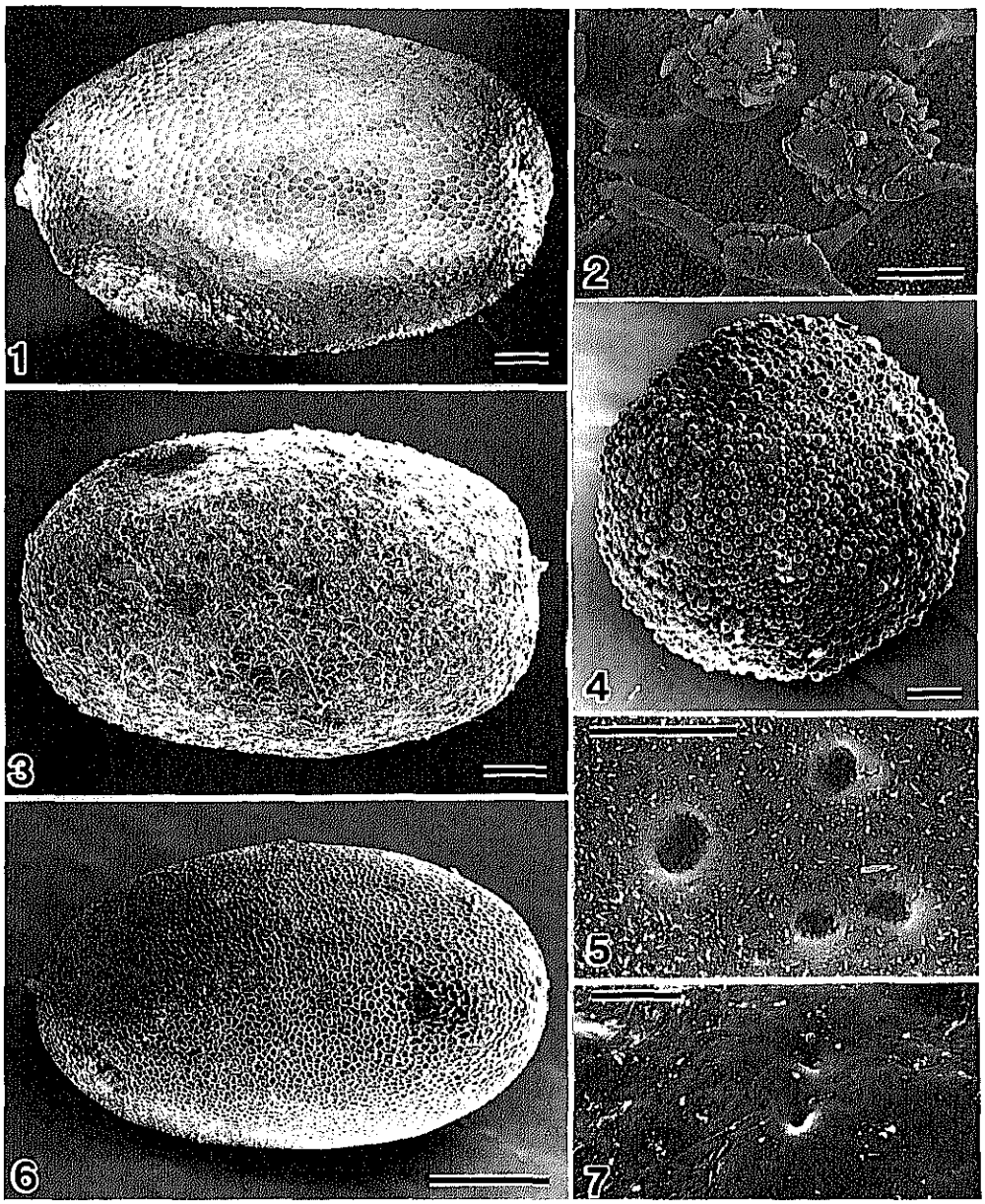
Twelve females reared in 1981 laid eggs thirteen egg clusters, and the number of eggs in a cluster was 18 to about 100 (average, 50.7). Only one of the females oviposited twice (39 and 70 eggs). It seems that a female oviposits several times in nature. A cluster of laid eggs is held together by a sticky substance that covers the surface of each egg. The color of the newly laid egg is creamy white, but it turns dark gray in half a day.

The newly laid egg is oval in shape, though the anterior half is slightly sharp, and their size is about 1000 by about 600μm (Fig. 1). The surface of the chorion is decorated with a honeycombed network (Figs. 1, 2). On corners of these minute polygons there appears a brushy process which is associated with an aeropyle (Fig. 2). The honeycombed pattern of the chorion on and near the anterior pole is different from that of other regions, and I could not find the micropyle in this region even by use of the SEM and the light microscope. There is a very thin vitelline membrane below the chorion.

The periplasm occupying 5-6% of the egg volume is 4-5μm thick (Fig. 13), but ca. 10μm-thick near the both poles of the egg. The maturation of the female pronucleus can be observed in the cytoplasmic island that lies a little anterior from the center of the egg length (Fig. 13). The cytoplasmic reticulum or reticuloplasm is also found between the yolk spherules and vacuoles (Figs. 13, 14). A disk-shaped polar granule (40 to 50μm in diameter) is found in the periplasm on the posterior pole (Fig. 14).

Yolk spherules are homogeneously well stained with eosin, and their diameter varies from 5 to 15μm. Small yolk spherules are distributed mainly in the peripheral zone. These eosinophilic yolk spherules occupy about 80% of the egg volume.

Panorpodes paradoxus



Figs. 1, 3, 4, 6. Newly laid eggs of *P. pryeri* (1), *Pd. paradoxus* (3), *B. laevipes* (4) and *Bo. westwoodi* (6).

Fig. 2. Close-up of Fig. 1.

Fig. 5. Anterior pole of egg of *B. laevipes* showing micropyles. Granular substance on egg surface is removed.

Fig. 7. Micropyles of *Bo. westwoodi* on anterior pole of egg.

Scales=100μm (Figs. 1-4, 6) and 10μm (Figs. 5, 7).

During 1979 to 1981, I obtained about 100 egg clusters containing 50 to 60 eggs in each cluster. The cluster is held together with a sticky substance as in *P. pryeri*.

The shape of the newly laid egg is oval and its size is 750 to 800 by about 500μm (Fig. 3). The chorion bears a honeycombed pattern that becomes weak near both poles, but I failed to find the micropyles in either of these regions.

The chorion is very thin and creamy white just after oviposition, and turns dark after several hours.

A relatively large quantity of periplasm, 5-6% of the egg volume, and cytoplasmic reticulum is found in the egg (Fig. 15). Yolk spherules, about 10μm in diameter, are stained homogeneously with eosin and occupy about 60% of the egg volume. There are also many vacuoles, considered to be traces of dissolved lipid yolk spherules (Fig. 15).

The maturating female pronucleus is situated in the cytoplasmic island of the periplasm near the middle of the egg. The polar granule is not found in this species.

Bittacus laevipes

I obtained eggs from 62 females during 1978 to 1981. A female oviposits 20-30 eggs (about 10 eggs per day). The oviposition almost always started between 4 and 5 o'clock p. m. and continued for a few hours. The hanging females oviposit eggs, and an egg appears at the tip of the abdomen and drops as soon as the next egg is extruded.

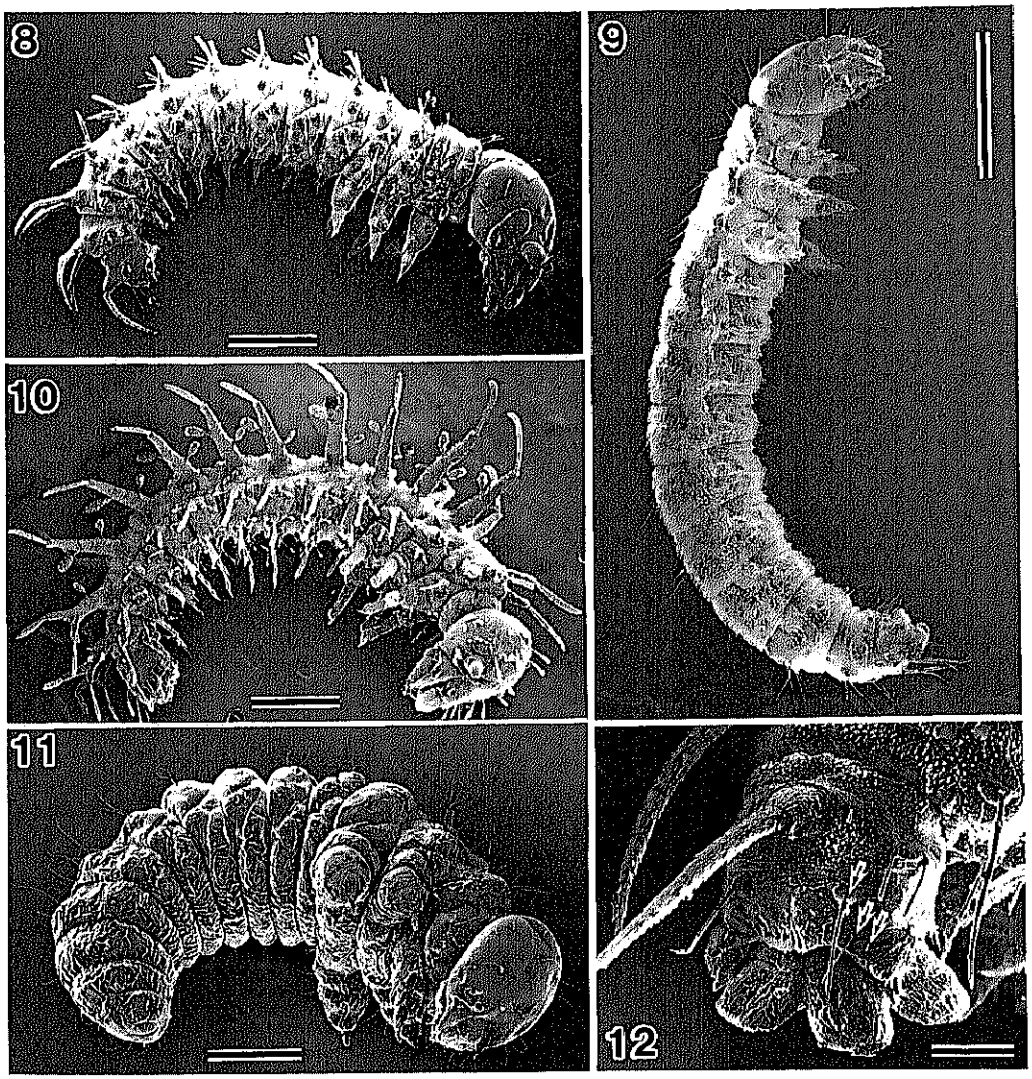
The newly laid egg is almost spherical, about 700μm in diameter (Fig. 4). The egg color is yellow just after oviposition and turns dark brown after several hours. The chorion is covered by numerous granular substances, and a polygonal surface pattern appears after these granules are removed. Two to five micropyles (Fig. 5) are observed on both poles of the egg. The chorion is very tough and thick (about 25μm), and the endochorion of 2 to 3μm thickness stains faintly with eosin (Fig. 16).

The periplasm occupying about 2% of the egg volume is very thin (2 to 3μm, Fig. 16) and stained lightly with hematoxylin. Little cytoplasmic reticulum is observed. Yolk spherules are homogeneously stained with eosin; their diameter is about 20μm, though there are smaller spherules at the periphery. The ratio of total volume of eosinophilic yolk spherules to the egg volume is approximately 65%. No polar granule is found in this species.

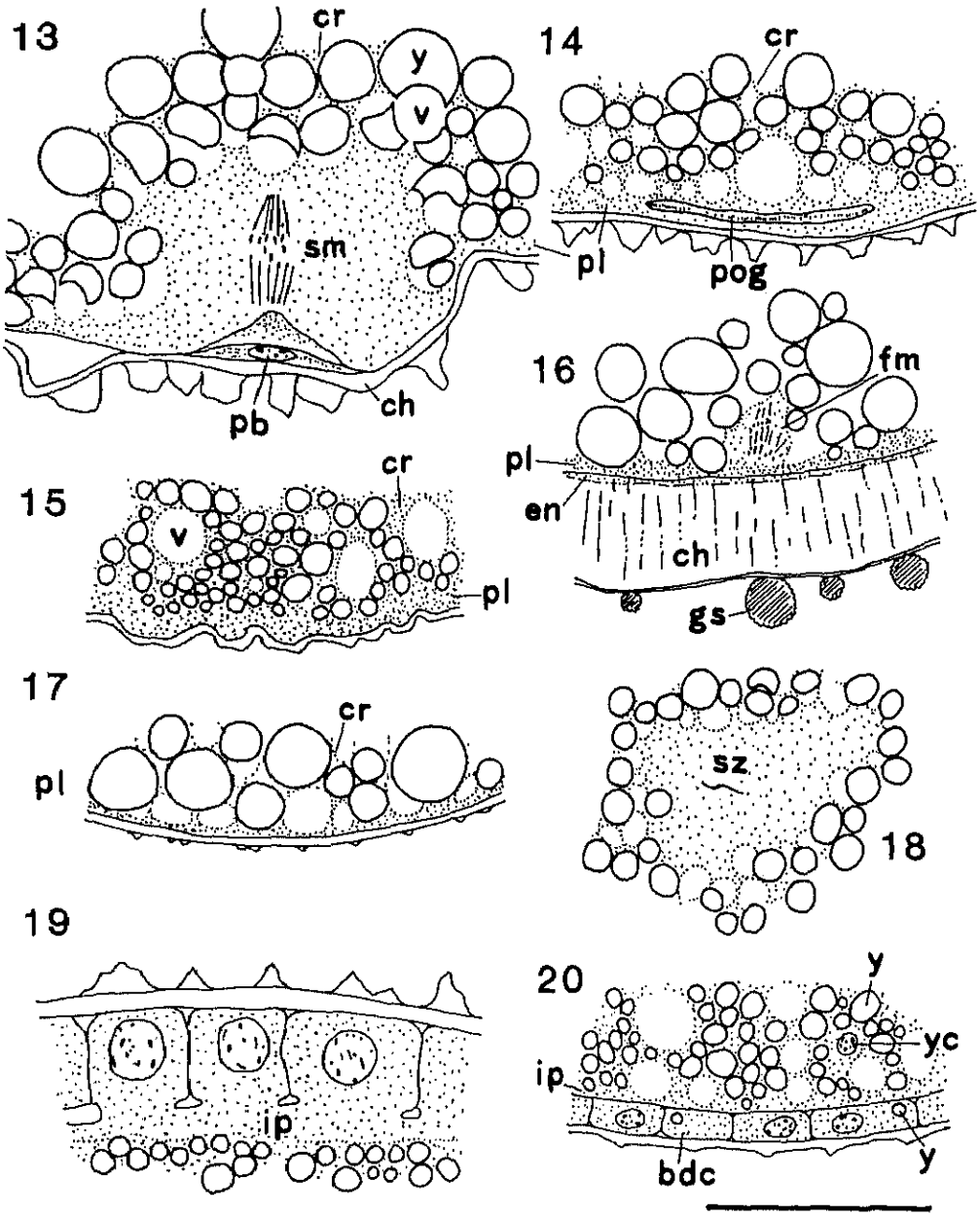
Boreus westwoodi

The shape of mature ovarian eggs and newly laid eggs is oval, and their size is 450 to 500 by 250 to 300μm (Fig. 6). On the surface of the chorion there appears a honeycomb network, which becomes faint near both poles of the egg, as in *Pd. paradoxus*. Two micropyles are found separately on both poles (Fig. 7). The chorion is about 2μm-thick and thickens near both poles. The chorion is transparent even if embryogenesis is proceeding.

The periplasm occupying about 35% of the egg volume and cytoplasmic reticulum are poor (Fig. 17), though richer than those of *B. laevipes*. The maturating female pronucleus in the cytoplasmic island of the periplasm is located one-third of the egg length from the anterior



Figs. 8-10. First-instar larvae of *P. pryeri* (8), *Pd. paradoxus* (9) and *B. laevipes* (10).
Fig. 11. Larva of *Bo. westwoodi*.
Fig. 12. Tenth abdominal segment of first-instar larva of *P. pryeri*. Arrow, spine belonging to vestigial eleventh abdominal segment.
Scales=500μm (Figs. 8-11) and 100μm (Fig. 12).



Figs. 13, 15-18. Cross sections of eggs of *P. pryeri* (13, 18), *Pd. paradoxus* (15), *B. laevipes* (16) and *Bo. westwoodi* (17) in Stage 1.
Figs. 14, 19. Longitudinal sections of eggs of *P. pryeri* in Stage 1.
Fig. 20. Blastoderm cells of *Pd. paradoxus* late in Stage 1.
Scale=50μm.

pole. The yolk spherules stain homogeneously with eosin, and their maximum size is about 20μm in diameter. The eosinophilic yolk spherules occupy about 60% of the egg volume. I could find no polar granule in this species.

Discussion of oviposition and newly laid eggs

Eggs are laid in soil crevices in *P. pryeri* and *Pd. paradoxus*, and in mosses in *Bo. westwoodi*, as in *Panorpa* spp. (MIYAKE, 1912; YIE, 1951; ANDO, 1960, 1973; BYERS, 1963), *Panorpodes* (ANDO, 1973), and *Boreus* spp. (WITHYCOMBE, 1922; COOPER, 1974). It seems also general that the oviposition of *B. laevipes* occurs while the female is hanging, as in *B. nipponicus* (ISHII, 1937), though *B. stigmaterus* deposits the eggs while alighting on the ground (SETTY, 1931).

As observed in the present study, the shape of the newly laid eggs is oval in *Panorpa* (MIYAKE, 1912; YIE, 1951; ANDO, 1962, 1973; BYERS, 1963), *Panorpodes* (ANDO, 1973), and *Boreus* spp. (WITHYCOMBE, 1922; COOPER, 1974). In *Bittacus*, however, the shape varies from cuboidal (*B. stigmaterus*, SETTY, 1931; *B. nipponicus*, ISHII, 1937) to spherical (*B. apicalis*, HINTON, 1981; *B. mastrillii* and *B. marginatus*, ANDO, 1973). ANDO (1973) observed that the shape of the oöcyte in ovarioles of *B. mastrillii* during chorion formation is spheroidal.

As for the micropyles, ANDO (1973) described those on the posterior pole of the egg in *B. mastrillii*, and HITON (1981) on the anterior pole in *B. pilicornis*. It is possible that they failed to find the micropyles on the other pole, because the micropyles are observed on both poles in *B. laevipes*.

Eosinophilic yolk spherules of *P. pryeri* and *B. laevipes* may correspond to the protein yolk of *P. communis* (RAMAMURTY, 1964a). In *B. laevipes* they are often indicated by dissolving features when fixation is unfavorable, and this characteristic of the yolk spherules tends to lead to overestimation of the amount of the yolk spherules. ANDO (1973) mentioned that the very thin periplasm of *B. mastrillii* is observed to be discontinuous.

Polar granules observed in the eggs of *P. pryeri* were described also in *P. japonica* (ANDO, 1973) and *P. communis* (RAMAMURTY, 1964b), so that the existence of polar granules is considered to be a common characteristic of the egg of *Panorpa*.

II. Duration of egg period

The egg periods (egg stages) of the species investigated in the present study is shown in Table 1 for comparison with those of other species reported by earlier authors.

Discussion of duration of egg period

There are not many reports on the life history and egg period of mecopteran insects, and the life histories of panorpids have been examined most frequently in this order. The egg period of the Panorpidae seems to vary from one to two weeks, and that of the Panorpodidae, that is, *Pd. paradoxus*, from three to six weeks.

The egg period of several species of the Bittacidae is more than 200 days, and they pass the

Table 1. Egg period of Panorpidae, Panorpodidae, Bittacidae and Boreidae.

Panorpidae	
Panorpa pryeri	ca. 150 hr (21°C)
Panorpa pryeri	150-160 hr (ca. 21°C) (ANDO, 1960)
Panorpa nipponensis	ca. 140 hr (25°C)
Panorpa japonica	ca. 160 hr (21°C), ca. 150 hr (25°C)
Panorpa japonica	168-180 hr (18-24°C) (ANDO, 1973)
Panorpa klugi (=*japonica*)	6-8 days (MIYAKE, 1912)
Panorpa bicornuta	ca. 7 days (22°C) (ANDO, 1973)
Panorpa helena	ca. 8 days
Panorpa nuptialis	8 days (BYERS, 1963; GASSNER, 1963)
Panorpa communis	144 hr (21°C) (WOLF, 1961)
Panorpa falsa	9-13 days
Panorpa akasakai	15-16 days
Panorpa laiwanensis	15-16 days
Panorpa shibatai	15-16 days
Panorpa pectinata	15-16 days
Panorpa longiramina	14 days (YIE, 1951)
Panorpa ochraceocauda	7 days
Neopanorpa makii	7 days
Neopanorpa ophthalmica	6 days
Neopanorpa formosana	6-8 days
Neopanorpa sauteri	5-14 days
Panorpodidae	
Panorpodes paradoxus	28-40 (mean, 32.6) days (21°C)
Panorpodes paradoxus	ca. 26 days (20-24°C) (ANDO, 1973)
Bittacidae	
Bittacus laevipes	250-300 (mean, 270) days
Bittacus mastrillii	ca. 245 days (ANDO, 1973)
Bittacus marginatus	ca. 240 days (ANDO, 1973)
Bittacus nipponicus	ca. 290 days (ISHII, 1937)
Bittacus stigmaterus	216-256 (mean, 226) days (SETTY, 1931)
Bittacus puncliger	20-37 days (SETTY, 1940)
Bittacus pilicornis	29-78 days (SETTY, 1940)
Boreidae	
Boreus hyemalis	9-10 days (8.8°C) (WITHYCOMBE, 1922)
Boreus hyemalis	21 days (21°C) (STRÜBING, 1950)
Boreus notoperates	24 days (20°C) (COOPER, 1974)

winter in the egg stage, though *B. punctiger* and *B. pilicornis* overwinter in the larval stage (SETTY, 1940). SETTY (1931) observed the exceptionally short egg period of 58 days in *B. stigmaterus*, which suggests that egg hibernation is not fixed in this species.

Prolongation of the egg period in *Bo. hyemalis* is known accompanied with a rise in incubation temperature (WITHYCOMBE, 1922; STRÜBING, 1950). This phenomenon seems to be correlated with the fact that oviposition of the species occurs in winter, and low temperature may be favorable for the embryonic development.

III. Early embryonic development

In this section I will describe embryonic development from immediately following oviposition to completion of the cellular blastoderm as Stage 1.

Panorpa pryeri

Stage 1 (0-28 hr)

Just after oviposition, the metaphase of the first maturation division is found in the cytoplasmic island. The second maturation division occurs at 1 hr after oviposition (Fig. 13). At that time, the spermatozoon is observed near the anterior pole of the egg (Fig. 18), and soon transforms into the male pronucleus at that site. The female pronucleus moves to fuse with the male pronucleus, and fertilization is completed and followed by the first cleavage.

The second cleavage occurs at 4 hr, and the seventh cleavage occurs at 8 hr after oviposition. The polar granule, which is located at the posterior pole of the egg, disappears by the ninth cleavage (ca. 10 hr after oviposition). At 12 hr after oviposition, cleavage cells reach into the periplasm, and the syncytial blastoderm is formed, though they reach to both poles a little later. The penetrating cleavage cells or syncytial blastoderm cells divide mitotically again.

At about 20 hr after oviposition, formation of cell membranes of syncytial blastoderm cells begins, and periplasm which does not participate in blastoderm formation becomes the inner periplasm by the completion of blastoderm cells (Fig. 19). The cleavage cells that did not participate in blastoderm formation remain in the yolk and become primary yolk cells (in the present paper, the term *yolk cell* has been adopted instead of *yolk nucleus*). The aggregate composed of some dozens of primary yolk cells is situated at the center of the egg, and the nuclei of these cells disintegrate later.

The blastoderm is completed at 22 hr after oviposition. The blastoderm cells are about 20μm in width and length, and the diameter of their nuclei and yolk cells is 10 to 15μm.

The present observation on the early embryonic development agrees with the detailed study of ANDO (1960).

Panorpodes paradoxus

Stage 1 (0-32 hr)

Just after oviposition, the female nucleus shows the metaphase of the first maturation division in the cytoplasmic island. After fertilization, the first cleavage occurs near the egg center. At 12 hr after oviposition, the sixth cleavage commences and the cleavage cells are located in a zone of 150 to 200μm inward from the egg surface.

At 1 day after oviposition, the cleavage cells reach the egg periphery. At this time the syncytical blastoderm appears to contain yolk spherules. The syncytial blastoderm is soon partitioned into cells by the formation of the cell membrane, and the cellular blastoderm is completed. Several yolk spherules are seen in the blastoderm cells, and there exists an inner periplasm (Fig. 20) as in *P. pryeri*.

Bittacus laevipes

Stage 1 (0-3 day)

Just after egg deposition, the female nucleus indicates the metaphase of the first maturation division (Fig. 16). The second maturation division follows 3 hr later. I could not observe fertilization.

At 1 day after oviposition, the fifth cleavage occurs, and the cleavage cells reach the egg periphery at 2 days after oviposition. Nuclei of the syncytial blastoderm divide mitotically, and at this time some nuclei divide tangentially against the egg surface and seem to be concerned with the formation of secondary yolk cells.

At 3 days after oviposition, the nuclei of the syncytial blastoderm divide again, and the cellular blastoderm is completed.

Boreus westwoodi

In *Bo. westwoodi*, I could not know how old the fixed eggs were, and therefore could describe only their histological features in early and middle embryonic development.

Stage 1

Maturation of the female pronucleus and fertilization were not observed. When the fifth to the sixth cleavage occurs, the cleavage cells are located in a zone about 100μm inward from the egg surface.

As development proceeds, cleavage cells reach into the egg periphery (Fig. 21) and increase in number. Then the cell membranes appear and the blastoderm is completed.

Discussion of early embryonic development

In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, the female nucleus of the newly laid egg exhibits the metaphase of the first maturation division. This fact is known in many insects, for example, in the odonatan *Calopteryx atrata* (ANDO, 1962), blattarian *Blattella germanica* (WHEELER, 1889), megalopteran *Sialis mitsuhashii* (SUZUKI *et al.*, 1981), trichopteran *Stenopsyche griseipennis* (MIYAKAWA, 1973), lepidopteran *Amata fortunei* (TANAKA, 1985a), and hymenopteran *Athalia proxima* (FAROOQI, 1963).

The inner periplasm found in *P. pryeri* and *Pd. paradoxus* is also reported in some other

holometabolan insects, *e.g.*, *Stenopsyche griseipennis* (MIYAKAWA, 1973), *Athalia proxima* (FAROOQI, 1963), dipteran *Dacus tryoni* (ANDERSON, 1962), etc. As ANDO (1960) described, the existence of the inner periplasm is probably related to the thickness of the periplasm.

The intake of yolk spherules by the blastoderm cells, seen in *Pd. paradoxus*, is not so general, although similar phenomena occur in the coleopteran *Epilachna vigintioctomaculata* (MIYA and ABE, 1966) and lepidopterans *Endoclita signifer* and *E. excrecens* (ANDO and TANAKA, 1980).

Generally speaking, the early embryonic development of *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi* seems to be similar to that of many other insects.

IV. External features of embryos during middle and late development

Panorpa pryeri

Stage 2 (28-51 hr)

At 28 hr after oviposition, the embryonic and extraembryonic areas differentiate in the uniform blastoderm. The cells of the embryonic area (=ventral plate) are small, about 12μm in height, with nuclei about 10μm in diameter (Fig. 22). Those of the extraembryonic area are 20 to 25μm in height, and their nuclei, about 10μm in diameter, are placed at the distal ends of the cells. The cells of the extraembryonic area contain many vacuoles (Fig. 23). At 32 hr after oviposition, the mitotic figures appear only in the embryonic area.

At 36 hr after oviposition, cells in the embryonic area are somewhat rounded and become 18 to 20μm in height, and their nuclei become smaller (ca. 8μm in diameter). Only slight changes are found in the extraembryonic area. Almost all of the mitotic figures are found in the embryonic area.

At 45 hr after oviposition, the amniotic folds appear at the anterior and posterior margins of the embryonic area (Fig. 24). At this time, the cells of the extraembryonic area are about 30μm in height. Their nuclei are 15 to 20μm in diameter, and many nuclei are situated at the inner or proximal ends of the cells. The cells of the embryonic area are scarcely altered morphologically except for the increase in their number.

At 46 hr after oviposition, the amniotic fold appears around the entire margin of the embryonic area and the embryonic area diminishes in size. Along the median line of the embryonic area the primitive groove progresses from the caudal end (Fig. 25).

At 47 hr after oviposition, the embryonic area becomes a pear-shaped germ band and much of its outer surface is covered with the amnion (Figs. 26, 27). It takes only ca. 2 hr from the appearance of the amniotic fold to the completion of the pear-shaped germ band.

At 49 hr after oviposition, the amniotic fold spreads further, and the protocorm of the germ band elongates backward along the egg surface and attains to the level indicated by the arrow in Fig. 28.

Stage 3 (51-54 hr)

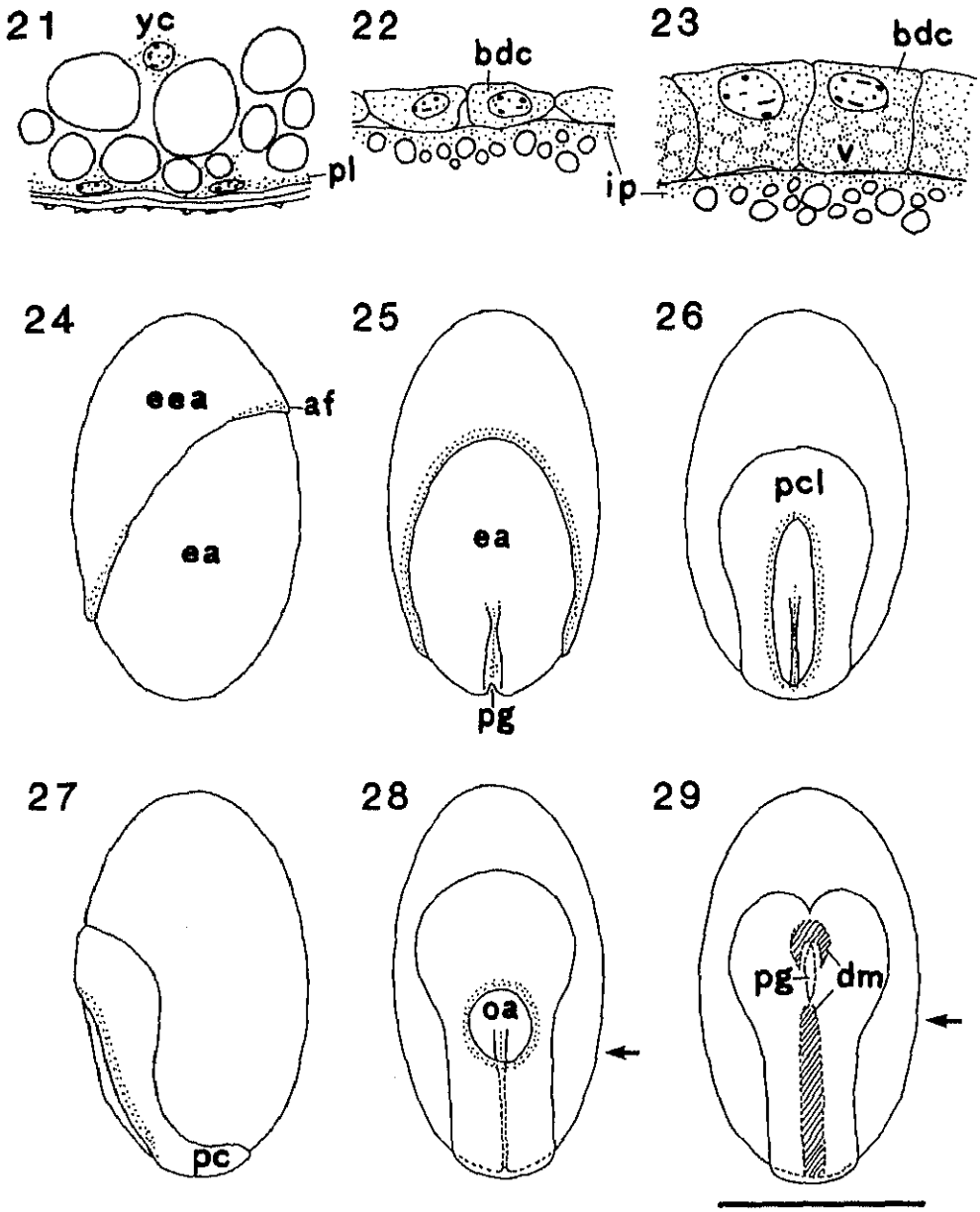


Fig. 21. Cross section of egg of *Bo. westwoodi* late in Stage 1.
Figs. 22, 23. Cells of embryonic area (22) and extraembryonic area (23) of *P. pryeri* early in Stage 2.
Fig. 24. Lateral view of egg of *P. pryeri* in Stage 2.
Fig. 25. Ventral view of egg of *P. pryeri* late in Stage 2.
Figs. 26-29. Embryos of *P. pryeri* late in Stage 2 (26, 27), at the end of Stage 2 (28) and early in Stage 3 (29). Arrow, caudal end of embryo.
Scale=50μm (Figs. 21-23) and 500μm (Figs. 24-29).

The amnion covers the ventral surface of the embryo completely (Fig. 29), and there appears a notch in the center of the anterior margin of the protocephalon, at 51 hr after oviposition. Yolk cleavage begins.

At 52 hr after oviposition, a low protuberance, the rudimental labrum, becomes recognizable at the anteromedial part of the protocephalon. The germ band continues to elongate and reaches three-quarters of the egg's circumference (Fig. 30).

Stage 4 (54-60 hr)

At 54 hr after oviposition, the labral anlage begins to divide. A pair of rudimental antennae appears, and the intercalary segment, mandible, maxilla and labium differentiate (Fig. 31).

At 57 hr after oviposition, the germ band becomes longer than at 52 hr, and a shallow invagination, the developing stomodaeum, is found in the center of the protocephalon. The three thoracic segments become distinguishable at this stage (Fig. 32).

Stage 5 (60-80 hr)

At 60 hr after oviposition, the germ band attains the length of five-sixths of the egg's circumference (Fig. 34). The rudimental abdomen divides into ten metameres, and small, paired thoracic appendages arise (Fig. 33).

At 70 hr after oviposition, the cephalic lobes of the embryo occupy a position at the anterior pole of the egg, and the gnathal segments begin to come together (Fig. 35).

Stage 6 (80-100 hr)

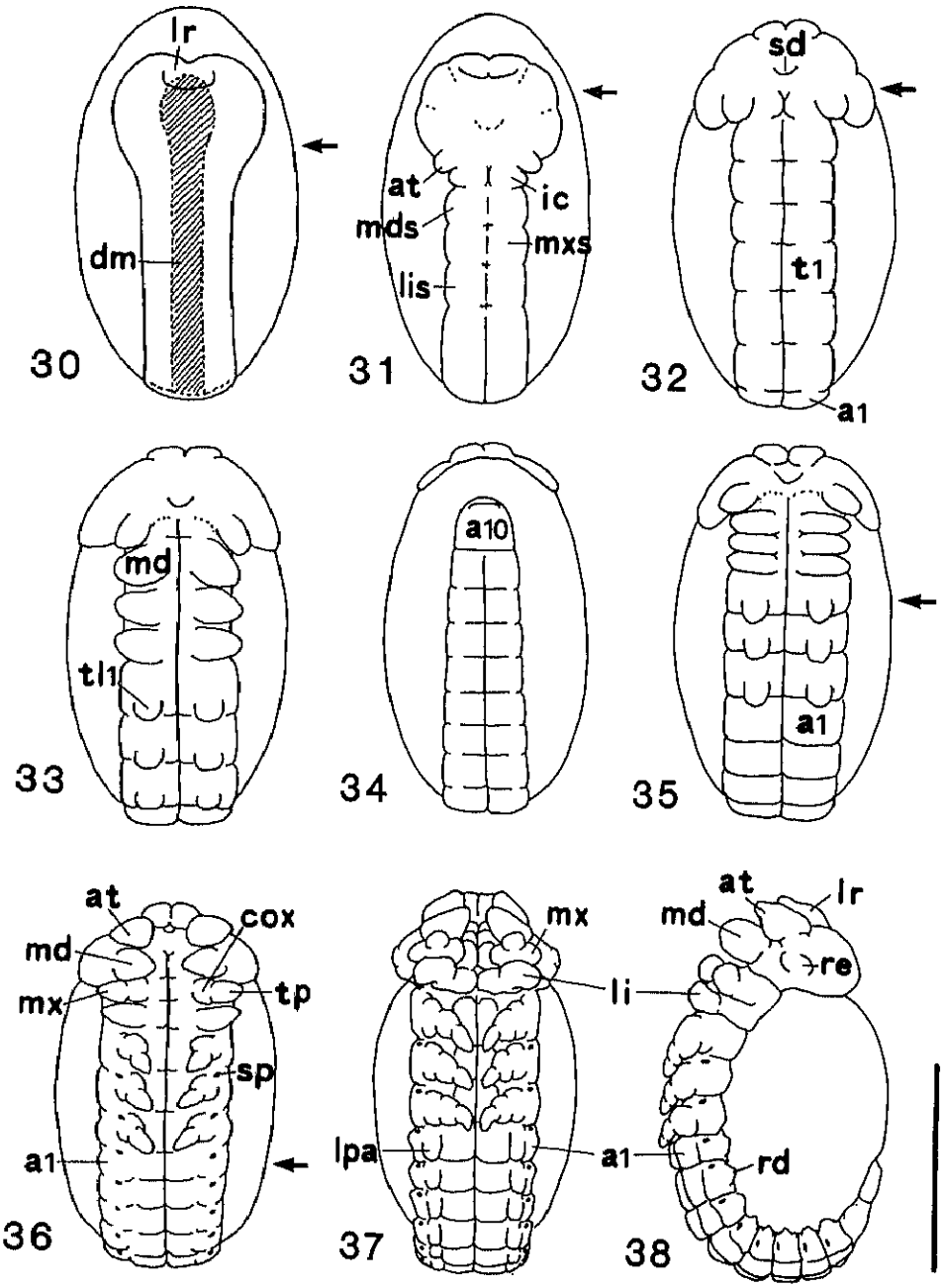
At 80 hr after oviposition, the forward shift of the gnathal segments proceeds further, and the rudimentary maxilla divides into a coxopodite and telopodite (Fig. 36). Paired tracheal invaginations appear anterolaterally on the second thoracic segment to the eighth abdominal segment.

A 90 hr after oviposition, the future galea and lacinia are formed from the inner side of the coxopodite of the maxilla (Fig. 37). The thoracic appendages divide into three segments each, and small paired processes appear on the first eight abdominal segments (Fig. 37). They are arranged in a row with the anlagen of the thoracic appendages and increase slightly in size during embryonic development. On the lateral margins of the first nine abdominal segments rudimental paired dorsal processes are formed (Fig. 38), and the future larval compound eyes begin to appear on the sides of the embryonic head.

Stage 7 (100-116 hr)

At 100 hr after oviposition, the head of the embryo approaches morphological completion (Fig. 39). Each thoracic appendage consists of four segments as in the first-instar larva. The spiracles on the second and third thoracic segments disappear, and a pair of new spiracles appears on the posterolateral bases of the first thoracic appendages (Fig. 39).

Paired processes, *i.e.*, rudiments of so-called abdominal legs, develop below the ganglia of



Figs. 30-38. Embryos of *P. pryeri* late in Stage 3 (30), early in Stage 4 (31), late in Stage 4 (32), early in Stage 5 (33, 34), late in Stage 5 (35), early in Stage 6 (36) and late in Stage 6 (37, 38). Arrow, posititon of caudal end of embryo. Scale=500μm.

the first eight abdominal segments (Fig. 40). The dorsal processes of the first nine abdominal segments elongate, and a tip of the process of the tenth abdominal segment can be seen beyond the anal legs.

At 110 hr after oviposition, the embryo elongates further, its caudal end almost touching its head (Fig. 41). From this time on, the yolk is consumed rapidly.

Stage 8 (116-130 hr)

At 116 hr after oviposition the embryo twists in such a way that the posterior half of its abdomen curves around the rest of its body (Fig. 42). The dorsal closure of the embryo is completed except for the first to fifth abdominal segments.

At 120 hr after oviposition, the embryo finishes its rotation (Fig. 42), and the dorsal closure of the anterior half of the abdomen is completed.

Stage 9 (130-150 hr)

At 130 hr after oviposition, the embryo has almost the appearance of the first-instar larva. An egg tooth is formed on the frons (Fig.44).

Stage 10 (First-instar larva)

The first-instar larva hatches at ca. 150 hr after oviposition. Its body length is approximately 3mm. Characters of the first-instar larva of *P. pryeri* include the following: a pair of compound eyes which consist of about thirty ommatidia each, paired abdominal prolegs located on the first eight abdominal segments, paired dorsal processes on the first nine abdominal segments and a dorsal process on the tenth abdominal segment (Fig. 8).

Panorpodes paradoxus

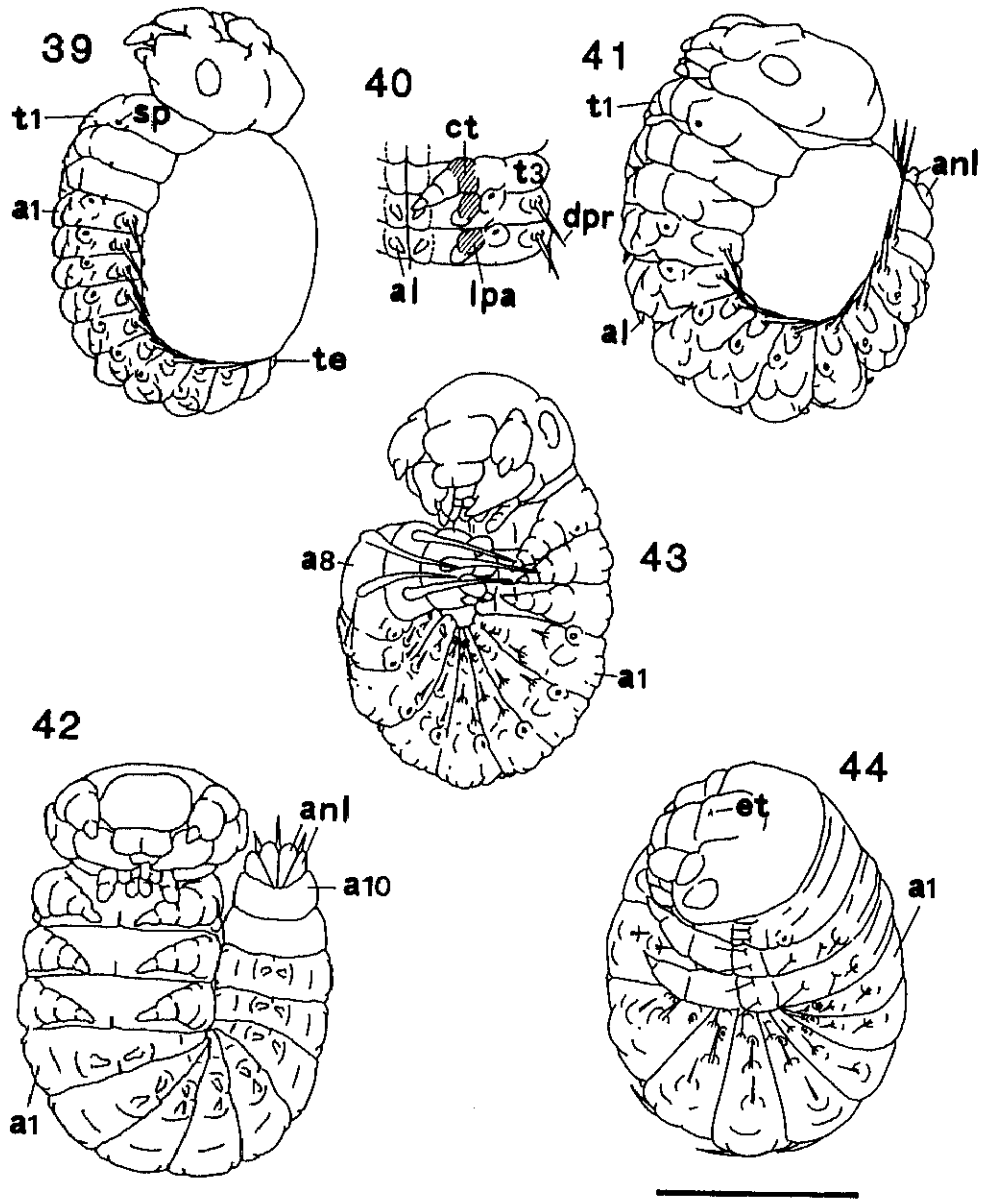
Stage 2 (32 hr-4 days)

At 32 hr after oviposition, the blastoderm begins to thicken, especially near the posterior pole. This thick region is the embryonic area (=germ disk). In this area there are some cells which were divided inwards from the blastoderm cells; they seem to be future germ cells.

At 40 hr after oviposition, cells of the embryonic area become higher (Fig. 45) and those of the extraembryonic area lower or thinner. At this time, an aggregation composed of about a dozen primary yolk cells is often found in the center of the egg. As development proceeds, the embryonic area commences to sink into the yolk.

At 2 days after oviposition, the invaginating embryonic area develops into a sac-shaped germ rudiment (Figs. 46, 47). Nuclei of the extraembryonic area (serosa) are stained more deeply with hematoxylin than those of the germ rudiment (Fig. 47).

At 3 days after oviposition, the germ rudiment is almost separated from the egg's periphery (Fig. 48). It contains the developing amnion and the embryo, in which the mesoderm formation starts soon. Yolk cleavage can also be observed.



Figs. 39-44. Embryos of *P. pryeri* early in Stage 7 (39, 40), late in Stage 7 (41), early in Stage 8 (42), late in Stage 8 (43) and in Stage 9 (44).
Scale=500μm.

Stage 3 (4-15 days)

At 4 days after oviposition, the germ rudiment is completely immersed in the yolk. At 7 days the germ rudiment or embryo takes a position in the center of the egg (Fig. 49).

As development proceeds, the embryo elongates and approaches the egg periphery again, and at 10 days after oviposition the embryo touches the egg periphery with its protocephalon (Fig. 50).

At 14 days, the embryo comes out of the yolk except for the posterior half of the protocorm, but embryonic segmentation can not be seen at this stage (Fig. 51).

Stage 4 (15-19 days)

At about 15 days after oviposition, the gnathal segments and then following segments appear, and the embryo reveals its shape on the yolk except for the fourth and following abdominal segments (Figs. 52, 53). A shallow pit, the future stomodaeum, arises in the center of the cephalic lobes.

Stage 5 (19-22 days)

At 19 days after oviposition, anlagen of gnathal and thoracic appendages make their appearance (Figs. 54, 55). The embryo emerges completely from the yolk, and the developing abdomen can be seen to ten segments.

At 21 days, the gnathal segments of the embryo commence to move forward.

Stage 6 (22-26 days)

At 22 days after oviposition, paired spiracles are observed on the second thoracic to eighth abdominal segments (Fig. 56).

At 23 days, the gnathal segments become more compact than in the prior stage. The spiracles of the second and third thoracic segments disappear, and a pair of spiracles appears on the first thoracic segment (Fig. 57). Paired swellings arise on the ventral surface of the first eight abdominal segments, arranged in alignment with the developing thoracic appendages. These swellings do not develop further.

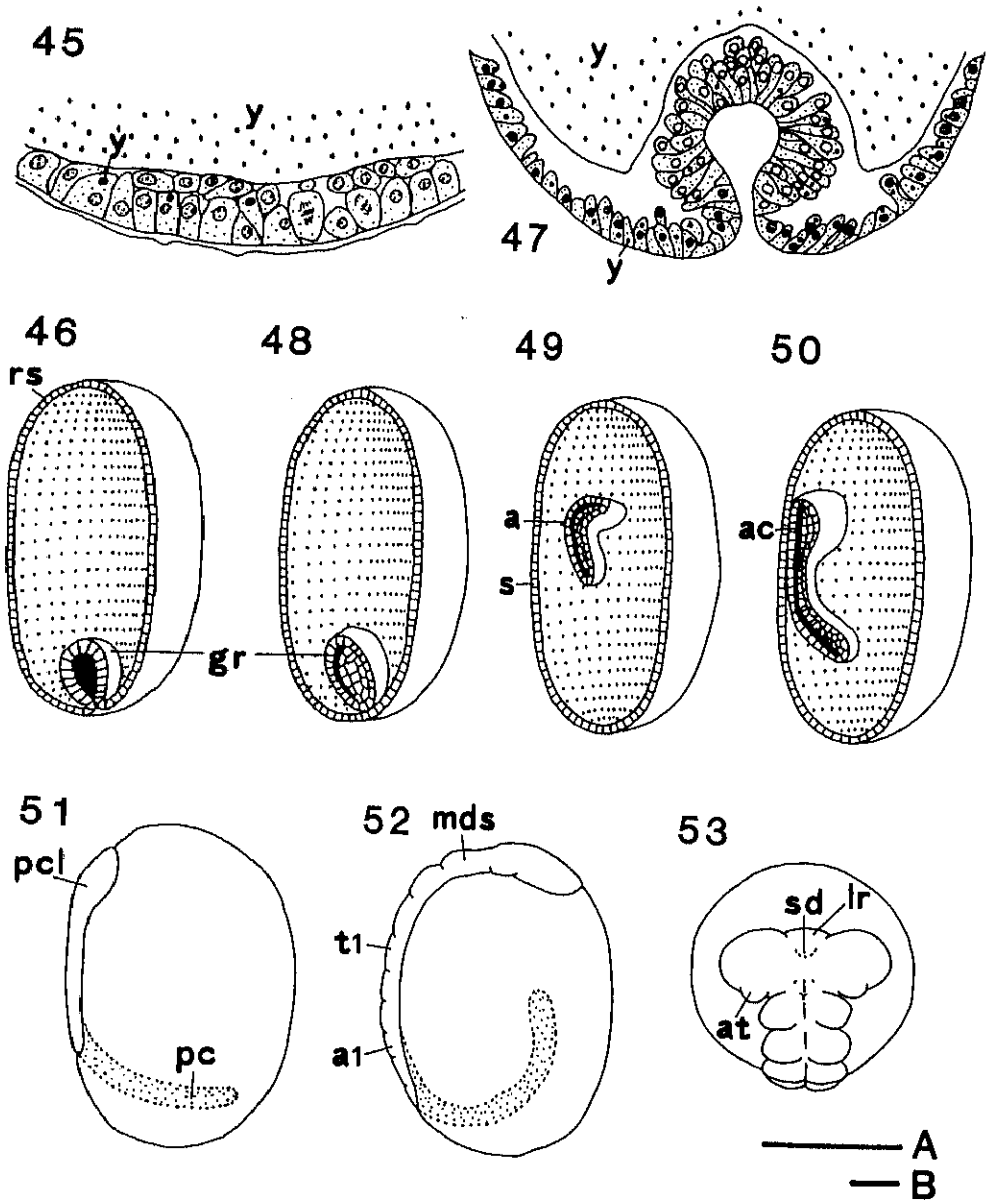
At 25 days after oviposition, the head and caudal end of the embryo almost touch each other (Fig. 58); this is just before rotation of the embryo.

Stage 7 (26-31 days)

At 26 days after oviposition, the turning occurs in the manner shown in Fig. 59. Dorsal closure is completed shortly after the rotation.

Stage 8 (31-33 days)

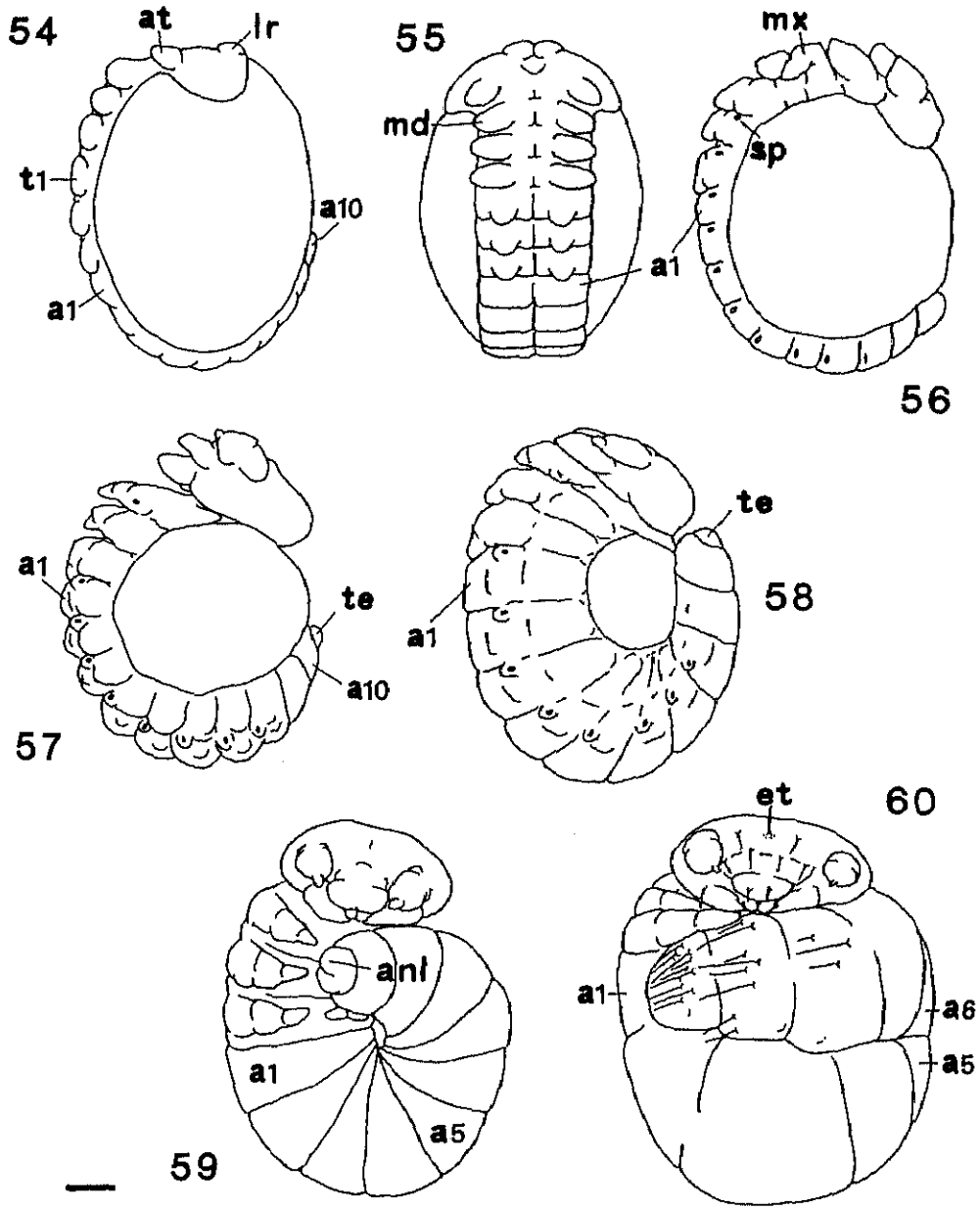
At 31 days after oviposition, the embryo looks almost like the first-instar larva (Fig. 60). An egg tooth appears on the frons, but the insect stays in the egg for one or two more days. As hatching approaches, the mandibular tips become fulvous.



Figs. 45, 47. Longitudinal sections through posterior part of egg of *Pd. paradoxus* early in Stage 2 (45) and in the middle of Stage 2 (47).

Figs. 46, 48-53. Embryos of *Pd. paradoxus* in the middle of Stage 2 (46), late in Stage 2 (48), in the middle of Stage 3 (49), late in Stage 3 (50), at the end of Stage 3 (51) and in Stage 4 (52, 53).

Scale (A) =50μm (Fig. 45) and 100μm (Fig. 47). Scale (B) =100μm (Figs. 46, 48-53).



Figs. 54-60. Embryos of *Pd. paradoxus* in Stage 5 (54, 55), early in Stage 6 (56), in the middle of Stage 6 (57), late in Stage 6 (58), in Stage 7 (59) and in Stage 8 (60). Scale=100μm.

Stage 9 (First-instar larva)

At ca. 33 days after oviposition, the first-instar larva hatches. Its body length is about 2mm (Fig. 9), and it has no eyes. On the medioventral line of the first eight abdominal segments there is a minute median process, those of the first and second abdominal segments being especially minute. The newly hatched larva bears relatively long setae but has no dorsal processes such as those in *P. pryeri*.

Bittacus laevipes

Stage 2 (3.5-9 days)

At 3 days after oviposition, or shortly after the blastoderm completion, part of the blastoderm thickens and forms a circular germ disk (about 200μm in diameter, Fig. 61).

At 4 days, the germ disk or germ band widens. It becomes pear-shaped at 5 days (Fig. 62), and as development proceeds the narrow germ band elongates.

Stage 3 (9-21 days)

At 9 days after oviposition, it has a length of one-fourth of the egg's circumference.

At 20 days after oviposition, cephalic lobes of the embryo become distinguishable (Fig. 63), but the segmentation of the embryo is not discernible.

Stage 4 (21-55 days)

At 21-24 days after oviposition, gnathal segments appear, and at 25 days, segments from the intercalary to the tenth abdominal can be seen though indistinctly, and a shallow pit or rudimental stomodaeum appears in the center of the protocephalon (Fig. 64).

At 35 days after oviposition, the labral rudiments increase in size and antennal anlagen occur (Fig. 65). The rudimental stomodaeum invaginates more deeply. The embryo elongates to half of the egg's circumference.

At 45 days, the paired labral rudiments begin to fuse, and anlagen of the mandible, maxilla and labium develop (Figs. 66, 67).

At 50 days after oviposition, the length of the embryo is equivalent to three-quarters of the egg's circumference. At the same time, anlagen of the thoracic appendages are discernible (Fig. 68).

Stage 5 (55-248 days)

At 55 days, the labral rudiments almost fuse and the opening of the developing stomodaeum is hidden by them (Fig. 69). The anlagen of the thoracic appendages elongate, and the gnathal segments move forward.

As 65 days after oviposition, the distal parts of both rudimental mandibles turn medioventrally, and the apices of each pair of developing thoracic appendages meet at the medioventral line of the embryo (Fig. 70). The embryo enters diapause at this stage.

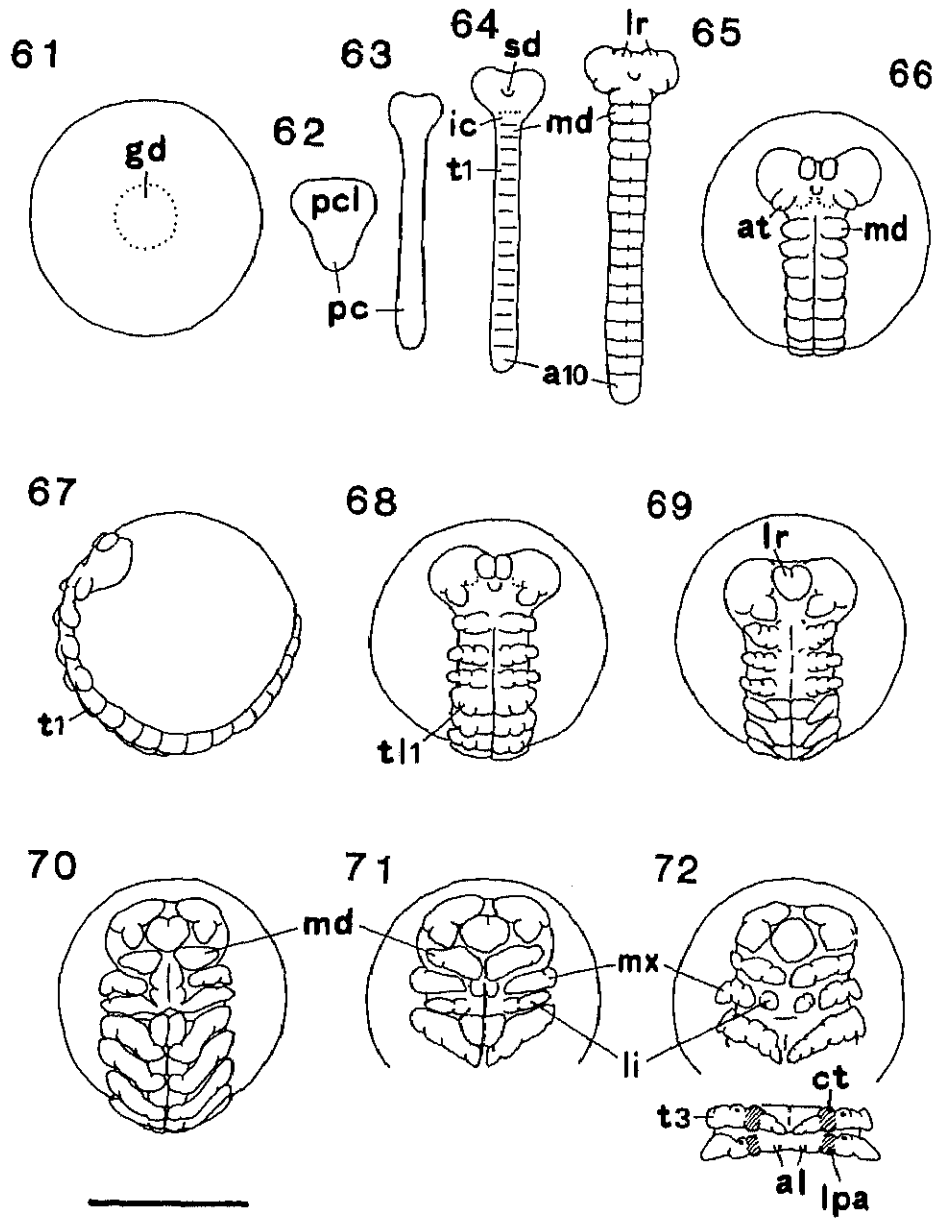


Fig. 61. Germ disk of *B. laevipes* early in Stage 2.

Figs. 62-72. Embryos of *B. laevipes* late in Stage 2 (62), in Stage 3 (63), in Stage 4 (64), in the middle of Stage 4 (65), late in Stage 4 (66, 67), at the end of Stage 4 (68), early in Stage 5 (69), in the middle of Stage 5 (70), early in Stage 6 (71) and in the middle of Stage 6 (72).

Scale=500μm.

Although almost all embryos go into diapause at about 65 days after oviposition, the time of the termination of diapause is different between embryos to some extent. The time from the termination of diapause to hatching, however, is approximately constant, so that I set the day of the termination of diapause as the 248th day from oviposition, for convenience in the following descriptions.

Stage 6 (248-263 days)

From 248 to 260 days after oviposition, the developing gnathal appendages shift their positions as shown in Figs. 71-74. The maxillary rudiment differentiates into two parts; the lateral one is the developing maxillary palp and the medial one is further divided into the future galea and lacinia. The labral rudiment dimishes in size and shifts medioventrally (Fig. 73).

At ca. 250 days after oviposition, paired spiracles appear on the second thoracic to eighth abdominal segments. On the ventral side of the abdomen there appear two pairs of small processes on each of the first eight abdominal segments. The outer processes are aligned with the rudimental thoracic appendages (Fig. 72), and the inner ones later become the so-called abdominal legs (prolegs) of the first-instar larva.

At 260 days after oviposition, a pair of new spiracles appears on the first thoracic segment (Fig. 74), but those of the second and third thoracic segments disappear. Dorsal processes elongate, and paired larval eyes are discernible (Fig. 74). At ca. 262 days after oviposition, the yolk diminishes in volume (Fig. 75).

Stage 7 (263-268 days)

The embryo revolves at ca. 263 days after oviposition (Fig. 76), and after revolution dorsal closure is completed. The embryo puts its caudal end on either side of the head (Fig. 77) and puts it on the opposite side about a day later (Fig. 78).

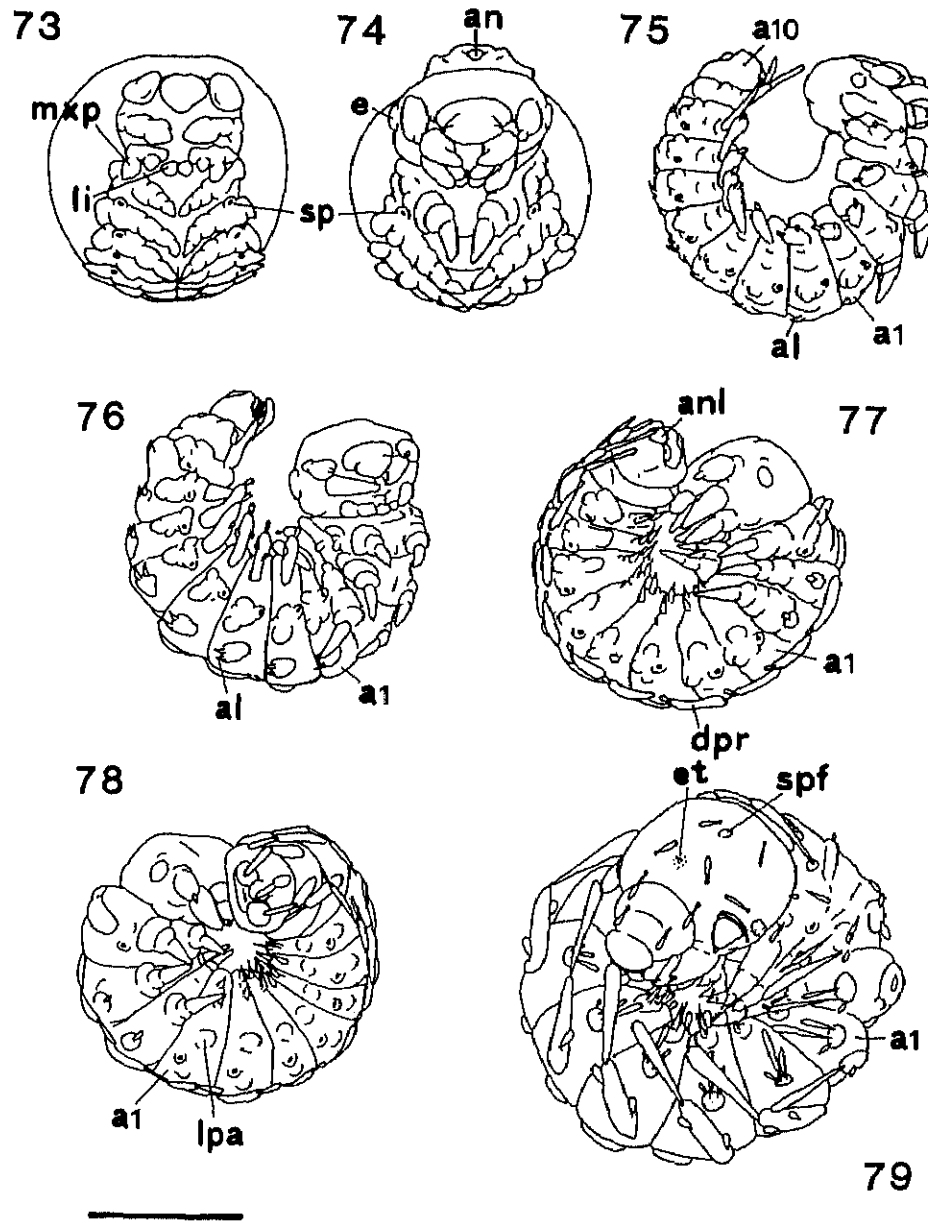
Stage 8 (268-270 days)

At 268 days after oviposition, the embryo is almost complete morphologically (Fig. 70). On the frons an egg tooth and a small process are found. The process is made of a thickening of ectoderm. I failed to find any innervation from the brain, so it seems doubtful that this process is a sensory organ, especially light-sensitive one.

At half a day later, tips of the egg tooth and mandibles become fulvous, and the antennae and dorsal processes bear fulvous pigment one or more days later.

Sage 9 (First-instar larva)

The first-instar larva hatches at ca. 270 days after oviposition and is about 3.5mm in length (Fig. 10). The larval eyes consist of seven ommatidia each. Paired prolegs are observed on the first eight abdominal segments. Long paired dorsal processes are situated on the abdominal segments, though that of the tenth segment is single. There are many clavate



Figs. 73-79. Embryos of *B. laevipes* late in Stage 6 (73, 74), at the end of Stage 6 (75), early in Stage 7 (76), late in Stage 7 (77, 78) and in Stage 8 (79).
Scale=500μm.

setae on the body wall.

Boreus westwoodi

Stage 2

After completion of the thin cellular blastoderm, the elliptical embryonic area or ventral plate, which is composed of vacuolated cells, differentiates (Fig. 80). The cells of this area are much thicker than those of the extraembryonic one. The lateral plates and middie plate, which later becomes the mesoderm, are distinguishable in the anterior part of the embryonic area (Fig. 81).

Stage 3

As development proceeds, a pear-shaped embryo appears (Fig. 82), and the ventral side of the embryo is covered with the thick amnion (Fig. 83). The embryo elongates and a notch appears in the center of the anterior margin of the protocephalon (Fig. 84).

Stage 4

A shallow invagination, the future stomodaeum, first becomes evident in the center of the protocephalon, and the gnathal and thoracic segments are differentiated (Fig. 85). The caudal end of the embryo is located at the posterior pole of the egg.

Stage 5

The embryo elongates and its abdomen consists of ten segments (Fig. 87). The last three abdominal segments are narrower than the prior segments. At the same time, a pair of labral and antennal rudiments appears on the protocephalon (Fig. 86). Paired anlagen of gnathal and thoracic appendages are also discernible.

As development proceeds, the gnathal segments shift forward.

Stage 6

Paired spiracles appear on the second thoracic segment to the eighth abdominal segment (Fig. 88), and the labial anlagen move medioventrally. The developing thoracic appendages have three visible segments, and paired small swellings are arranged in aligment with the thoracic appendages on the first eight abdominal segments (Fig. 89).

Larva

The larva is fat, scarabaeiform, resembling a weevil larva (Fig. 11); its first thoracic legs are smaller than the others. Although the larva has relatively long setae, there are no dorsal processes which occur in *P. pryeri* and *B. laevipes*.

The middle and late embryonic development of *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi* is summarized in Table 2.

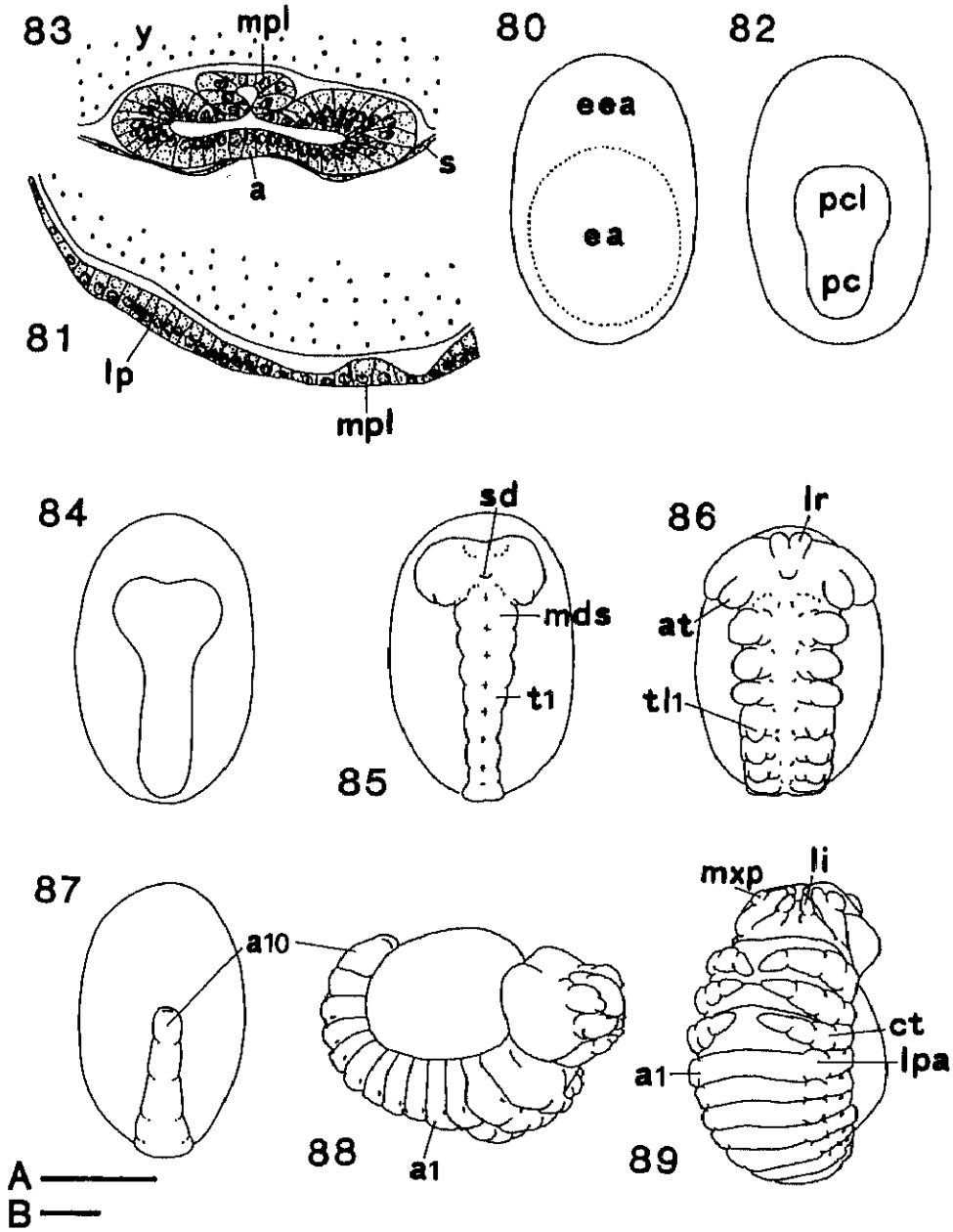


Fig. 80. Ventral view of egg of *Bo. westwoodi* early in Stage 2.

Figs. 81, 83. Cross sections of egg of *Bo. westwoodi* late in Stage 2 (81) and early in Stage 3 (83).

Figs. 82, 84-89. Embryos of *Bo. westwoodi* early in Stage 3 (82), late in Stage 3 (84), in Stage 4 (85), in Stage 5 (86, 87) and in Stage 6 (88, 89).

Scale (A) =50μm (Figs. 81, 83). Scale (B) =100μm (Figs. 80, 82, 84-89).

Table 2. Middle and late development of *Panorpa pryeri*, *Panorpodes paradoxus*, *Bittacus laevipes* and *Boreus westwoodi*.

P.	*Pd.*	*B.*	*Bo.*	State of development
(1) 3 hr	(1)	(1)	(1)	Pronuclei fuse.
22 hr	28 hr	3 days		Blastoderm is completed.
(2) 28 hr	(2) 32 hr	(2) 3.5 days	(2)	Embryonic area appears.
46 hr	3 days	4 days		Mesoderm differentiates.
(3) 51 hr	(3) 4 days	(3) 9 days	(3)	Embryo is covered with amnion.
(4) 54 hr	(4) 15 days	(4) 21 days	(4)	Gnathal segments appear.
(5) 60 hr	(5) 19 days	25 days	(5)	Ten abdominal segments appear.
70 hr	21 days	(5) 55 days		Gnathal segments move forward.
(6) 80 hr	(6) 22 days	(6) 250 days	(6)	Spiracles appear from t2 to a8.
(7) 100 hr	23 days	260 days		Spiracles appear on t1.
(8) 116 hr	(7) 26 days	(7) 263 days		Embryo rotates.
120 hr	27 days	264 days		Dorsal closure is completed.
(9) 130 hr	(8) 31 days	(8) 268 days		Egg tooth appears.
(10) 150 hr	(9) 33 days	(9) 270 days		Larva hatches.

The number in parentheses indicates the number of stage. *P.*, *P. pryeri*; *Pd.*, *Pd. paradoxus*; *B.*, *B. laevipes*; *Bo.*, *Bo. westwoodi*; t1, t2, first and second thoracic segments; a8, eighth abdominal segment.

Discussions with respect to the external features of embryos are given in Chapter VII, 1, i, c 'Tracheae' in Observations, and Chapter I 'Homology in larval abdominal prolegs and thoracic legs' and Chapter II 'Interpretation of metamerism in terminal region of abdomen' in General Discussion and Conclusion.

V. Change in egg size and hatching

Panorpa pryeri, *Panorpodes paradoxus* and *Bittacus laevipes*

To obtain measurements of eggs, fixed eggs were always used.

The change in egg size from oviposition to just before hatching is shown in Fig. 90. The volume of the mature egg increased by approximately 2.5 times that of the newly laid one in *P. pryeri*, about 2 times in *Pd. paradoxus*, and 3.4 times in *B. laevipes*.

The time of the hatching was roughly the same for all eggs in a single cluster in *P. pryeri* and *Pd. paradoxus*, though the time varied within a cluster and the difference sometimes was as much as a month in *B. laevipes*. The larva cut the chorion with the egg tooth to escape from egg in *P. pryeri*. Newly hatched larvae often eat the chorion, and then they were fed

chopped insects, in the rearing of *P. pryeri* and *B. laevipes*. However, newly hatched larvae of *Pd. paradoxus* never ate the chorion or chopped insects, so that they seem not to be carnivorous.

Discussion of change in egg size and hatching

There are not many reports on change of egg size during the embryogenesis of mecopteran insects. YIE (1951) studied in detail the change of egg size in *P. falsa*. According to him, the egg volume just before hatching is about 2.2 times that of the newly laid egg.

The present data on the volume of a mature egg compared to a newly laid one agrees with approximately the data obtained in other mecopteran insects, for example, 2.6 times in *P. nuptialis* (BYERS, 1963), ca. 4 times in *B. stigmaterus* (SETTY, 1931) and about 3.5 times in *Bo. notoperates* (COOPER, 1974).

GASSNER (1963) observed that the larva of *P. nuptialis* cut the chorion and came out of the egg as observed in *P. pryeri*. The first-instar larva of *Bo. notoperates* has no egg tooth and cut the chorion with the mandibles, according to COOPER (1974).

The fact that newly hatched larvae eat the chorion is already known in *Panorpa* (YIE, 1951 ; BYERS, 1963) and in *Bittacus* (SETTY, 1940).

VI. Formation of mesoderm and embryonic envelopes

Panorpa pryeri

In Stage 2, the future middle plate and lateral ones differentiate in the embryonic area, and the future amnion becomes discernible in the anterior part of the embryonic area (Fig. 91). Midway through Stage 2, the amniotic folds appear in the posterior and anterior margins of the embryonic area. At the same time, cells of the embryonic area thicken a little near the posterior pole of the egg.

As development proceeds, the primitive groove arises from near the posterior pole of the egg, and reaches to the posterior part of the protocephalon (Fig. 26). The amnion covers a large part of the ventral surface of the embryo. The manner of formation of the amniotic fold is shown diagrammatically in Fig. 92. The extraembryonic area becomes the serosa, and the amnion nearly covers the embryo. At the same time, several cells of the middle plate begin to invaginate at the preoral region (Fig. 93).

Late in Stage 2, the middle plate has formed multicellular layers in the preoral region. At the postoral region, however, the mesoderm (inner layer) has already begun to form in the manner of the invagination of the tubular middle plate (Fig. 94). Soon it begins to get out of tubular shape.

At the beginning of Stage 3, the ventral side of the embryo is completely covered with the amnion, and the primitive groove reaches to the anterior margin of the protocephalon. In the preoral region, the multicellular layered middle plate invaginates as a tube. The primitive groove near the caudal end of the embryo begins to close. In the middle of Stage 3, the

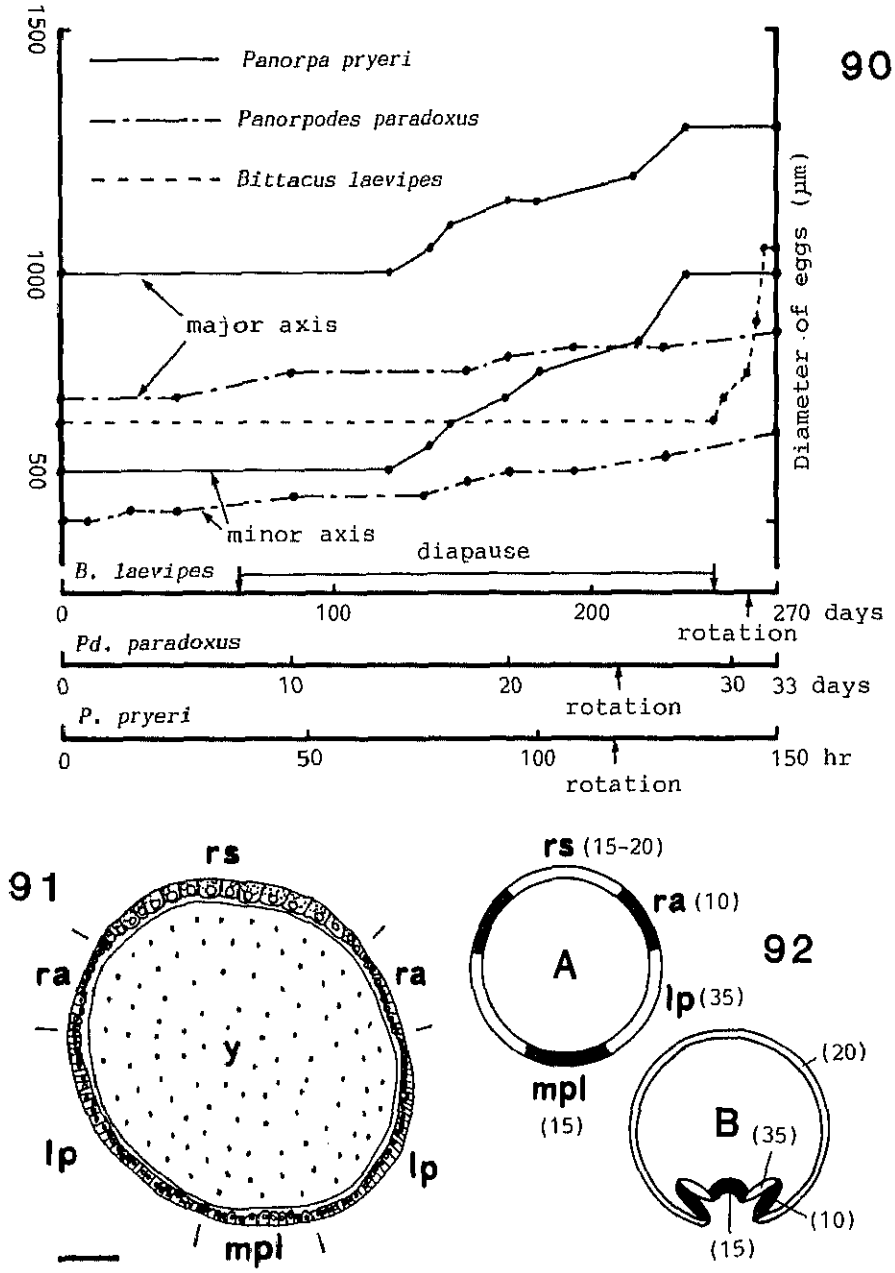


Fig. 90. Change of egg size of *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.
Fig. 91. Cross section through middle part of egg of *P. pryeri* in Stage 2.
Fig. 92. Diagrams of cross sections of egg of *P. pryeri* in the middle of Stage 2 (A) and late in Stage 2 (B) showing embryonic envelope formation. The number in parentheses indicates the number of cells.
Scale=100μm.

primitive groove has disappeared along much of the length of the embryo.

Panorpodes paradoxus

In the middle of Stage 2, the germ rudiment begins to invaginate into the yolk mass. Nuclei of the developing serosa at this time can be heavily stained with hematoxylin, as mentioned before (Fig. 47).

Late in Stage 2, the primitive groove and mesoderm occur on the median line of the sac-like germ rudiment (Fig. 95). The serosa becomes thin (about 15μm), though the amnion keeps its original thickness.

In the middle of Stage 3, the serosa becomes still thinner (about 10μm), and the yolk spherules in cells disappear in the embryonic and extraembryonic areas.

Late in Stage 3, the serosa cells become much thinner (5μm) and their nuclei become flat. The developing amnion becomes membranous, and the mesoderm at the protocephalon spreads to the sides, though near the caudal end it retains its tubular form.

Bittacus laevipes

Late in Stage 2, it is observed that cells near the median line of the embryo proliferate inward (Fig. 96, arrows) to become the mesoderm.

As development proceeds, the mesodermal cells increase in number and are found along the median line of the embryo from the anterior end to the posterior one, at the beginning of Stage 3 (Fig. 97).

By the same time, the amnion covers the ventral surface of the embryo.

Boreus westwoodi

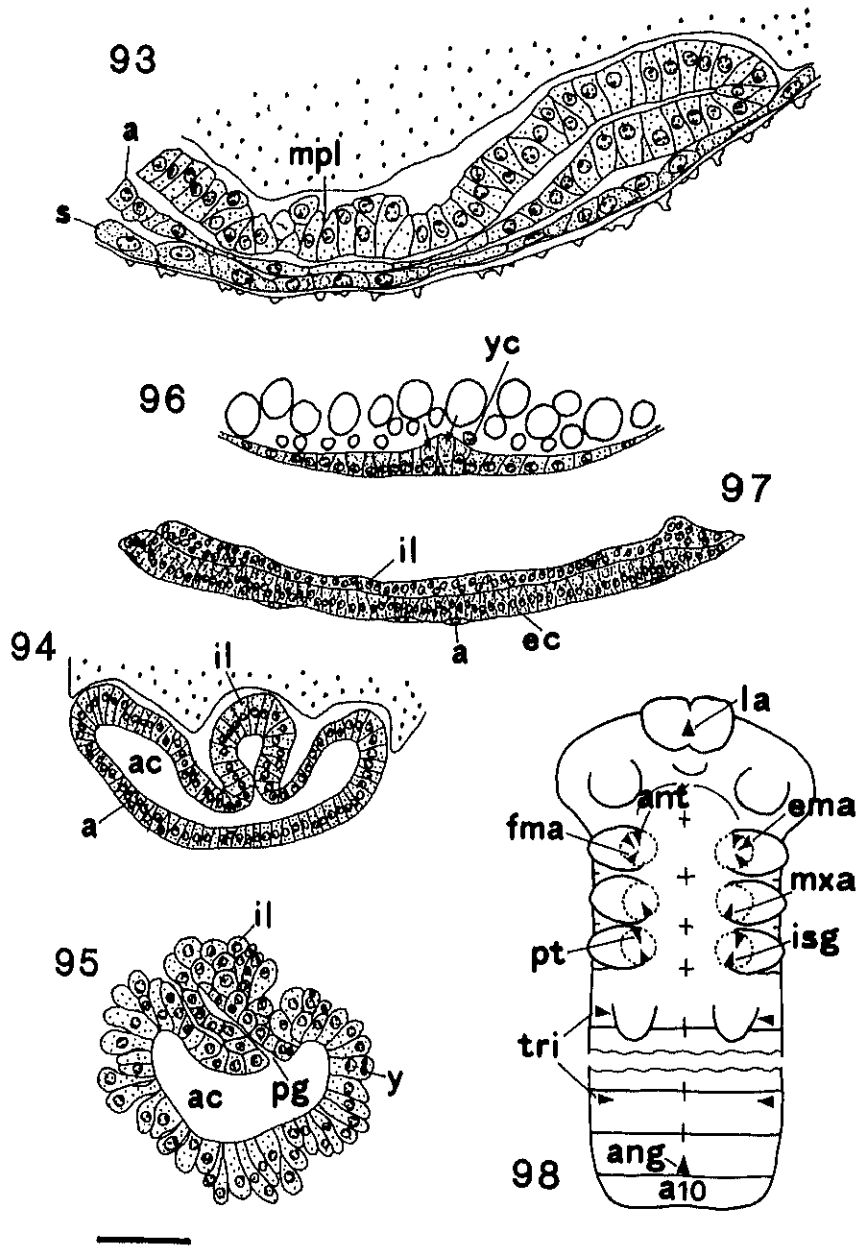
In Stage 3, the middle plate, which differentiated in Stage 2, becomes tubular and invaginates on the median line of embryo (Fig. 83).

At the same time, the ventral surface of the embryo is covered with the vacuolated amnion. As development proceeds, the tubular form of the median plate breaks down at the anterior part of the embryo, though it persists in the posterior part, finally disappearing in Stage 4.

Discussion of formation of mesoderm and embryonic envelopes

JOHANNSEN and BUTT (1941) classified the mesoderm formation of pterygotes into the following three types. In the first type, the mesoderm is formed by a sinking middle plate which will be converted into a sunken tube by the approach and fusion of the lateral plates. In the second type, the mesoderm is formed by a sinking middle plate which is cut off from the lateral plates. In the third type, the mesoderm is formed by the inward proliferation of cells along the median line of the embryo without differentiation of the middle plate.

The first type is observed in many holometabolan insects, for example, megalopteran *Sialis mitsuhashii* (SUZUKI *et al.*, 1981), neuropteran *Chrysopa perla* (BOCK, 1939) and *Ascalaphus ramburi* (KAMIYA and ANDO, 1985), trichopteran *Stenopsyche griseipennis* (MIYAKAWA, 1974a),



Figs. 93, 94. Cross sections of embryo of *P. pryeri* in the middle of Stage 2 (93) and late in Stage 2 (94).
Fig. 95. Cross section of embryo of *Pd. paradoxus* late in Stage 2.
Fig. 96. Cross section of embryo of *B. laevipes* late in Stage 2.
Fig. 97. Sagittal section of embryo of *B. laevipes* in Stage 3.
Fig. 98. Diagram showing positions of ectodermal invaginations.
Scale=50μm (Figs. 93, 95-97) and 100μm (Fig. 94).

coleopteran *Tenebrio molitor* (ULLMANN, 1964), dipteran *Dacus tryoni* (ANDERSON, 1962), etc., and some hemimetabolans such as hemipteran *Pyrilla perpusilla* (SANDER, 1956) and *Gerris paludum* (MORI, 1969).

The second type is observed in isopteran *Kalotermes flavicollis* (STRIEBEL, 1960), hemipteran *Oncopeltus fasciatus* (BUTT, 1949), and hymenopteran *Apis mellifera* (NELSON, 1915).

The third type is reported in some hemimetabolan insects, *e.g.*, some Odonata (ANDO, 1962), blattarian *Blattella germanica* (WHEELER, 1889), and orthopteran *Locusta migratoria* (ROONWAL, 1936), and holometabolan Symphyta of the Hymenoptera, *Pteronidea ribessii* (SHAFIQ, 1954). It is generally thought that the third type is more primitive than two former types.

In the some Lepidoptera, *e.g.*, *Pieris rapae* (EASTHAM, 1927), *Heliothis zea* (PRESSER and RUTSCHKY, 1957) and *Chilo suppressalis* (OKADA, 1960), the type of the inner layer formation differs from one part of the embryo to another, so that several embryologists doubt the phylogenetic significance of the inner layer formation. However, it is also possible to regard the partial difference as a significant charater for those species.

In *P. pryeri*, *Pd. paradoxus* and *Bo. westwoodi*, the mesoderm is formed by the first type, as in many holometabolan insects. On the other hand, in *B. laevipes* it is formed by the third type. Considering the rarity of mesoderm formation of the third type in the Holometabola, it seems that *B. laevipes* is more primitive than *P. pryeri*, *Pd. paradoxus* and *Bo. westwoodi* in mesoderm formation.

In the psocopteran *Liposcelis divergens* (GOSS, 1953) and lepidopterans *Pieris rapae* (EASTHAM, 1927) and *Diacrisia virginica* (JOHANNSEN, 1929), the developing amnion grows by mitosis and covers the ventral surface of the embryo. In *P. pryeri* and *Pd. paradoxus*, however, few mitotic formations were observed in the developing amnion.

VII. Organogenesis

1. Ectodermal derivatives

i) Invaginations

a) Endoskeleton, salivary and anal glands

Panorpa pryeri

Six pairs of ectodermal invaginations were observed in the cephalognathal region in this species (Fig. 98). These are the anterior and posterior tentoria, extensor and flexor mandibular apodemes, maxillary apodemes and salivary glands. Moreover, the labral apodeme and anal gland appear singly on the median line.

Late in Stage 5, the paired invaginations of the anterior tentorium appear at the posterior edges of the intercalary segment or anterolateral edges of the bases of the mandibular appendages. Paired invaginations of salivary glands arise posteromedially near the bases of the labial appendages (Fig. 99).

In the middle of Stage 6, the paired invaginations of mandibular flexor apodemes arise

posterolaterally at the bases of the mandibular appendages, and the paired invaginations of the posterior tentorial arms appear at the anterolateral bases of the labial appendages (Figs. 100, 101). At the same time, the tips of the invaginations of the salivary glands become sac-shaped in the second thoracic segment.

In Stage 7, maxillary apodemes begin to invaginate at the posteromedial bases of the maxillary appendages. The distal ends of the posterior tentorial arms fuse with each other and form a central body, and then distal ends of the anterior tentorial arms fuse with the central body (Fig. 102). The developing flexor mandibular apodemes become L-shaped in transverse section at their mid-length. The rudiments of extensor mandibular apodemes arise at the anterior mandibular bases. The openings of the rudimental salivary glands fuse with each other, forming a common duct. From there, the forked ducts extend to the third thoracic segment, and the sac-shaped distal ends occupy the first and second abdominal segments.

At the same time, a shallow invagination, which is the rudiment of the median labral apodeme, is formed at the dorsocentral point of the clypeolabral suture. Another shallow invagination makes its appearance at the intersegmental zone between the ninth and tenth abdominal segments on the medioventral line.

By the middle of Stage 7, the median labral apodeme becomes deeper (Fig. 103), and from its distal end muscles extend posteriorly.

In Stage 8, the developing anal gland becomes deeper and surrounding mesodermal cells begin to differentiate into muscles and fat bodies. The depth of the mediam labral apodeme is the same as in the prior stage.

Figure 104 shows the endoskeletal ingrowths of the first-instar larva diagrammatically. Only the tentorium is directly continuous with the cuticle of the cranium. The maxillary apodemes indirectly connect with the labium through a membranous zone, or conjunctiva. The mandibular apodemes connect only with the conjunctiva located between the mandible and cranium.

The anal gland of the first-instar larva has a lumen, and is often surrounded by fat bodies.

Panorpodes paradoxus and *Bittacus laevipes*

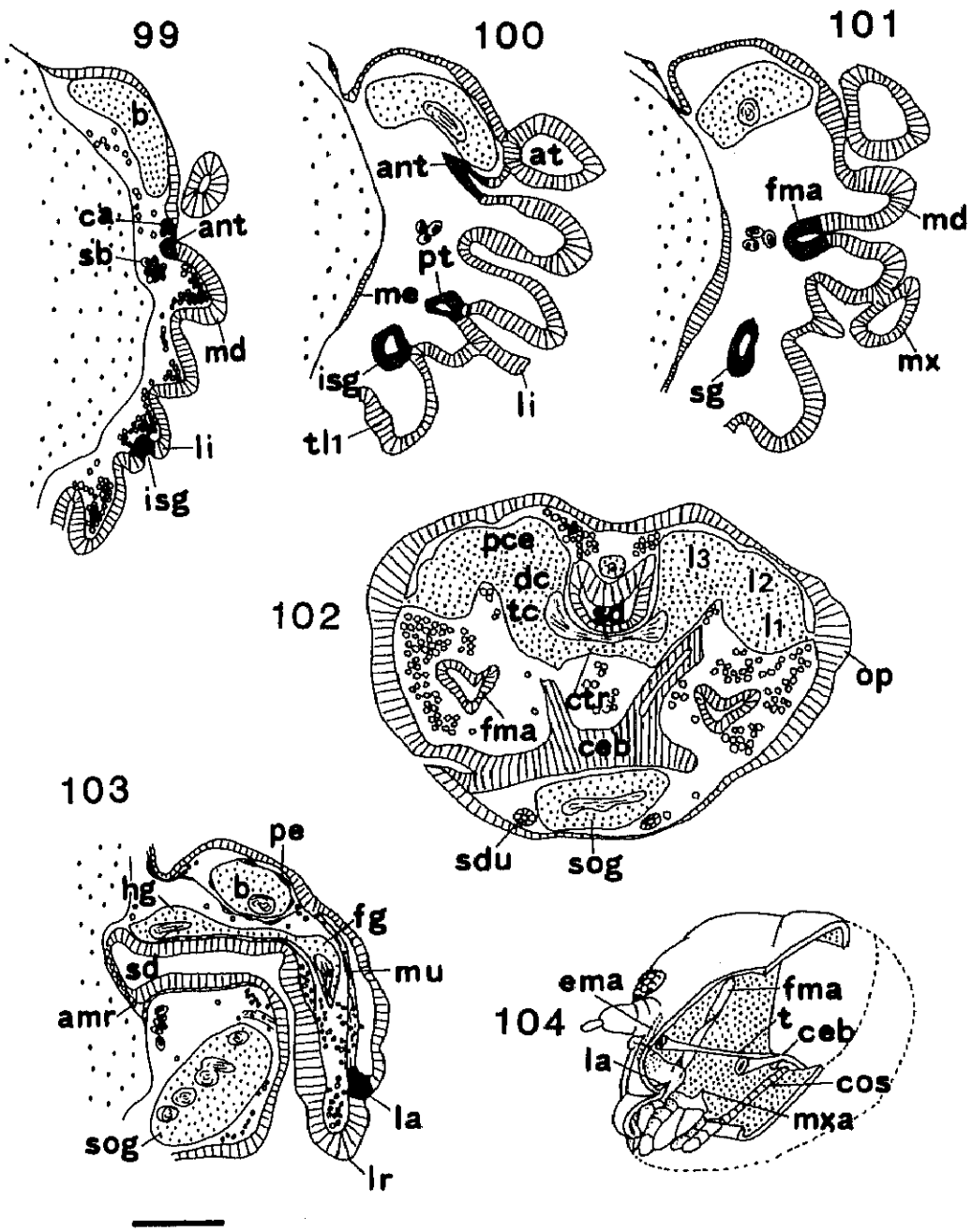
In *Pd. paradoxus* and *B. laevipes*, the number of ectodermal invaginations and their positions in the embryo are the same as described for *P. pryeri*.

In *Pd. paradoxus*, ectodermal invaginations arise at the end of Stage 4, except for those of the labral apodeme and anal gland, which are formed in Stage 6.

In *B. laevipes*, ectodermal invaginations begin to appear early in Stage 5, again except for the labral apodeme and anal gland, which arise late in Stage 6. The anal gland of the first-instar larva has a lumen as in *P. pryeri*.

In the larva of *Bo. westwoodi*, no anal gland was found.

Discussion of ingrowths for endoskeleton and salivary and anal glands



Figs. 99-101, 103. Longitudinal sections of embryo of *P. pryeri* late in Stage 5 (99), in the middle of Stage 6 (100, 101) and in Stage 7 (103).

Fig. 102. Cross section through head of embryo of *P. pryeri* in Stage 7.

Fig. 104. Diagram showing endoskeletons of head of first-instar larva of *P. pryeri*.

Scale=100μm.

At the cephalognathal region, four pairs of ectodermal invaginations appear in many insects, *viz.*, odonatan *Epiophlebia superstes* (ANDO, 1962), trichopteran *Stenopsyche griseipennis* (MIYAKAWA, 1974b), lepidopteran *Pieris rapae* (EASTHAM, 1930), hymenopteran *Apis mellifera* (NELSON, 1915), etc. The paired anterior tentorial arms invaginate from the posterior part of the intercalary segment or the anterior margin of the mandibular segment. The mandibular apodemes appear at the posterior part of the mandibular segment. In the coleopteran *Lytta viridana* (REMPEL and CHURCH, 1971), *Tenebrio molitor* (ULLMANN, 1967), and lepidopteran *Chilo suppressalis* (OKADA, 1960), as well as in *P. pryeri*, two pairs of mandibular apodemes appear, *i.e.*, flexor and extensor apodemes.

EASTHAM (1930) considered that the arrangement of cephalic apodemes follows a metameric sequence, and this view is generally accepted. There are, however, some disputes concerning the association of some apodemes with particular segments, especially the anterior tentorium.

EASTHAM (1930) and ULLMANN (1967) maintained that the anterior tentorial arms are derived from the antennal segment, because they observed that their invaginations appeared at the posterior part of the antennal base. OKADA (1960), and REMPEL and CHURCH (1971) asociated the tentorial arms with the intercalary segment, and I agrees with them because in *P. pryeri*, *Pd. paradoxus* and *B. laevipes* the anterior tentorial invaginations never arise at the bases of the antennae.

REMPEL and CHURCH (1971) found the paired labral apodemes in *Lytta* and suggested the possibility of the appendicular nature of the labrum. The labral apodeme of *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, however, develops singly from its first appearance, so that this apodeme seems to have a different nature from the other apodemes mentioned above.

The central body of the tentorium is derived from the posterior tentorial invaginations in the odonatan *Tanyopteryx pryeri* (ANDO, 1962), orthopteran *Locusta migratoria* (ROONWAL, 1937), coleopteran *Tenebrio molitor* (ULLMANN, 1967) and *Lytta viridana* (REMPEL and CHURCH, 1971), or from the anterior tentorial invaginations in the coleopteran *Calandra oryzae* (TIEGS and MURRAY, 1938). The formation of the central body is the same as in the former group in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

REMPEL and CHURCH (1971) associated the extensor apodeme of the mandible with the antennal segment in *Lytta*. I could not determine the association of the extensor apodeme because the invagination of the apodeme appears between the anterior edge of the mandibular base and the posterior part of the antennal base. Considering, however, that only one pair of ectodermal invaginations is basically associated with one segment in the cephalognathal region (EASTHAM, 1930; MATSUDA, 1965; ULLMANN, 1967), the extensor mandibular apodemes should be considered derived from the antennal segment. This is because the flexor apodeme is associated with the mandibular segment, and the anterior tentorial arms are associated with the intercalary segment in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

The anal gland is unknown in insects other than *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

The "anal gland" of some coleopteran insects appears in the epidermis of the rectum (SNODGRASS, 1935) and is a different organ from that of the mecopteran insects. The anal gland of *P. pryeri*, *Pd. paradoxus* and *B. laevipes* is a similar characteristic to the labial apodeme, because it arises as a single invagination. The gland has few glandular features, at least in the first-instar larva.

b) Corpora allata and prothoracic glands

Bittacus laevipes

In Stage 6, there appears a pair of cell clusters in the ectoderm just in front of the invaginating anterior tentorial arms (Fig. 105). Each of which cluster consists of about a dozen cells, the cytoplasm stains faintly with eosin, and this is the rudimental corpus allatum.

Late in Stage 6, the anlage of the corpus allatum moves inward, retaining a commection with the body wall, and is situated between the anterior tentorial arms, mandibular flexor apodeme and suboesophageal ganglion (Fig. 106).

In Stage 7, the rudimental corpus allatum loses its connection with the body wall, and takes a position on the side of the stomodaeum at the posterior margin of the cranium of the embryo, which is the same as that of the first-instar larva.

A shallow ectodermal invagination becomes visible at the anterior margin of the first thoracic segment, on the medioventral line, late in Stage 6. The invagination deepens and passes between the developing interganglionic connectives. The distal part of the invagination grows bilaterally, and the cytoplasm of the diverging parts can be stained faintly with eosin. These distal parts probably will be future prothoracic glands.

I failed to observe the further development of these rudiments. The prothoracic glands are found on connectives in front of the first thoracic ganglion in the first-instar larva.

Panorpa pryeri and *Panorpodes paradoxus*

In *P. pryeri*, a pair of cell clusters appears at the posterior part of the intercalary segment or in front of the invaginations of the developing anterior tentorial arms (Fig. 99). Each cell cluster is composed of four to five cells which have cytoplasm poorly stained with hematoxylin, and this cell mass to be a future corpus allatum.

I could not follow the further development of the corpora allata.

The corpora allata of the first-instar larva of these species are located just behind the corpora cardiaca (Fig. 121).

Discussion of corpora allata and prothoracic glands

The origin of the corpora allata is generally considered ectodermal (NELSON, 1915; EASTHAM, 1930; ROONWAL, 1937; ANDO, 1962, etc.), though TIEGS and MURRAY (1938) suggested that they are derived from the antennal coelomic sac, showing their mesodermal origin, in *Calandra*.

The original segment of the corpora allata, however, seems to be not specified. They are

associated with the intercalary segment in *P. pryeri* and *B. laevipes*; with the mandibular segment in dragonflies (ANDO, 1962), *Pieris* (EASTHAM, 1930), *Chilo* (OKADA, 1960), Papilionidae (TANAKA, 1987); or with the maxillary segment in *Apis* (NELSON, 1915), *Oncopeltus* (DORN, 1972), *Lytta* (REMPEL *et al.*, 1977), *Stenopsyche* (MIYAKAWA, 1974b).

The developing corpora allata are surrounded with a cellular sheath derived from the antennal mesoderm in *Locusta* (ROONWAL, 1937), but this condition was never observed in mecopteran species.

The corpora allata of *P. pryeri* and *Pd. paradoxus* finally come to lie close to the corpora cardiaca, as has also been observed in *Chilo* (OKADA, 1960), *Oncopeltus* (DORN, 1972) and *Lytta* (REMPEL *et al.*, 1977).

The prothoracic glands are generally derived from a pair of invaginations formed at the posterior margin of the labial segment or the anterior margin of the first thoracic segment (OKADA, 1960; ANDO, 1962; MIYAKAWA, 1974b; KOBAYASHI and ANDO, 1983; TANAKA, 1985b). In contrast, the prothoracic glands of *B. laevipes* seem to be derived from a single invagination, but there are some doubts about their formation. REMPEL *et al.* (1977), however, reported that the prothoracic glands of *Lytta* are derived from a single invagination at the labral-prothoracic intersegmental region on the medioventral line, as in *B. laevipes*.

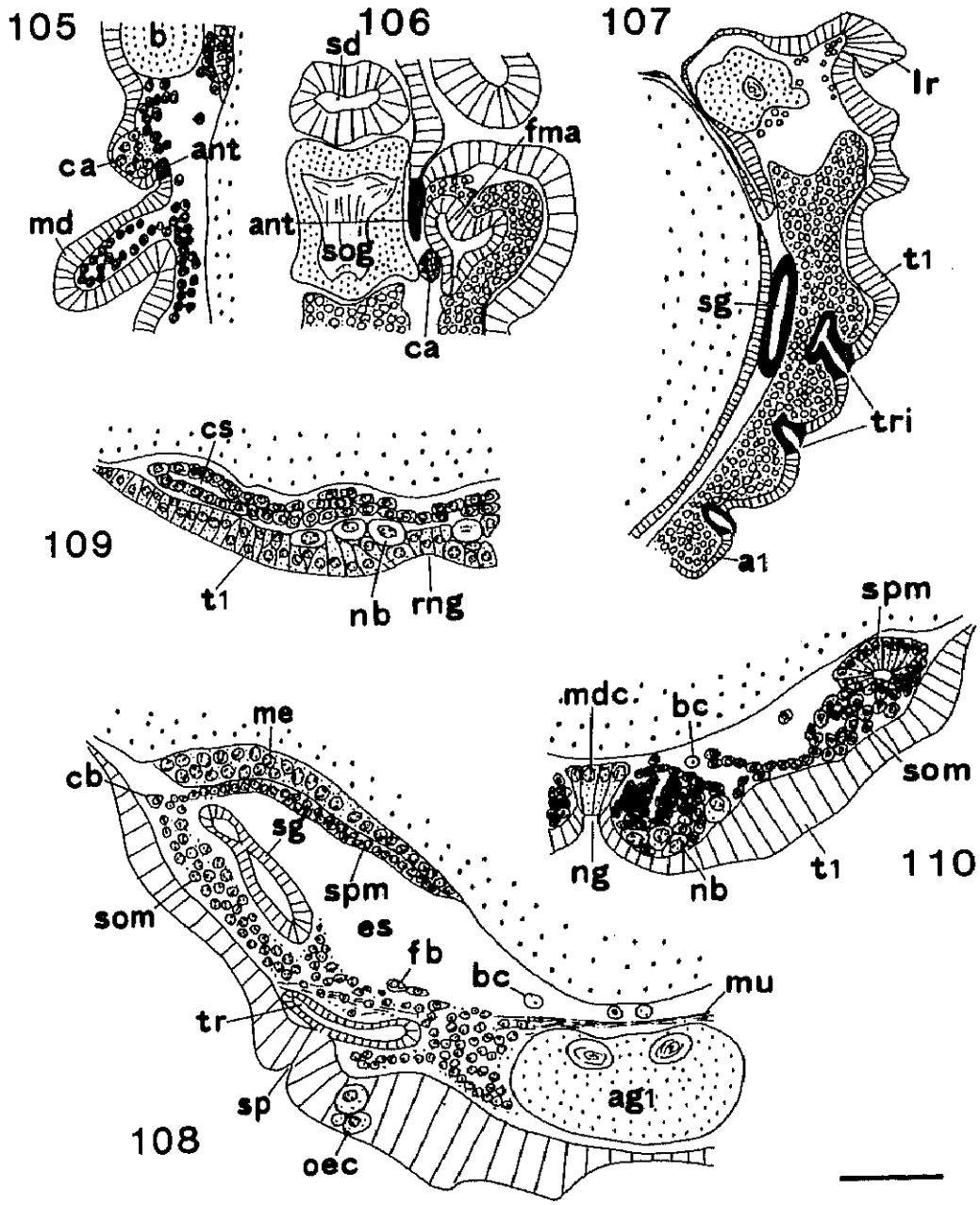
c) Tracheae

Panorpa pryeri

Late in Stage 5, paired tracheal invaginations become evident on the second thoracic segment to the eighth abdominal segment. They are located at the anterolateral bases of the developing second thoracic appendages and it comparable regions of each of the following nine segments. These invaginations are shallow, though those of the second thoracic segment are deeper than the others and grow anteriorly. In the middle of Stage 6, the tracheal rudiments in the mesothorax begin to fork (Fig. 107), while the others possess their original depth.

In Stage 7, the mesothoracic tracheal rudiments elongate into the gnathal segments, and then their openings, or spiracles, disappear and a pair of new spiracles is found on the posterior part of the prothorax (Fig. 39). These new spiracles seem to be anteriorly shifted spiracles of the mesothorax, because tracheal invaginations are never found on the pro- and mesothoracic segments at once. The tracheal invaginations on the posterior prothorax already have long branched tracheal trunks from their earliest appearance. The openings of the tracheal invaginations on the metathorax become vestigial. Each tracheal invagination connects with those of the next anterior and posterior segments by Stage 8. Midway through Stage 8, the proximal parts of the tracheal invaginations commence to form rudimental atria. The openings of the tracheal invaginations on the metathorax finally close, though vestigial spiracular tracheae contact the inner side of the body wall. Such connections are observed also in the first-instar larvae.

In Stage 9, the developing atria become chitinous, and those of the first-instar larva have



Figs. 105, 107. Parasagittal sections of embryo of *P. pryeri* in Stage 6 (105) and in the middle of Stage 6 (107).

Fig. 106. Horizontal section through head of embryo of *P. pryeri* late in Stage 6.

Figs. 108-110. Cross sections of embryo of *P. pryeri* in Stage 7 (108), late in Stage 4 or in Stage 5 (109), and in Stage 6 (110).

Scale=50μm (Figs. 105, 106, 108-110) and 100μm (Fig. 107).

chitinous compartments around the openings.

Panorpodes paradoxus and *Bittacus laevipes*

Tracheal formation in *Pd. paradoxus* and *B. laevipes* is similar to that of *P. pryeri*. In both *Pd. paradoxus* and *B. laevipes*, in Stage 6, paired tracheal invaginations appear on the mesothoracic segment to the eighth abdominal segment. A pair of tracheal invaginations shifts from the mesothoracic segment to the prothoracic one late in Stage 6 in *Pd. paradoxus* and *B. laevipes*, while the openings of the tracheal invaginations on the metathoracic segment disappear. The vestigial tracheae of the metathoracic segment, however, retain contact with the inner side of the metathoracic body wall in the first-instar larvae of *Pd. paradoxus* and *B. laevipes*, as in *P. pryeri*.

Discussion of tracheal development

The embryonic development of the tracheal system has been investigated in detail by ANDO (1962), and REMPEL and CHURCH (1972).

Paired tracheal invaginations at first arise on the mesothoracic segment to the seventh or eighth abdominal segment of the embryos in many insects, for example, in the Odonata (ANDO, 1962), orthopteran *Locusta migratoria* (ROONWAL, 1937), siphonapteran *Ctenocephalides felis* (KESSEL, 1939), coleopteran *Tenebrio molitor* (ULLMANN, 1967), and in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

In *Locusta* (ROONWAL, 1937), the spiracles of the meso- and metathoracic segments migrate forward as development proceeds, and finally the former is located at the intersegmental zone between prothorax and mesothorax, and the latter at the posterior margin of the mesothoracic pleuron.

In holometabolous embryos, the migration of the mesothoracic spiracles to the prothoracic segment is known in *Ctenocephalides* (KESSEL, 1939), *Lytta* (REMPEL and CHURCH, 1972), as well as in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*. This fact supports MATSUDA's view (1970) that in the Holometabola, "the anterior thoracic spiracle has shifted its position anteriorly from the primary position on the mesothorax."

SNODGRASS (1935) suggests that in the adult of *Heterojapyx gallardi* (Japygidae) the spiracles of the thorax do not correspond in their position to those of the abdomen, so that the thoracic spiracles are not serially homologous with the abdominal spiracles.

The thoracic spiracles of *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, however, are apparently arranged in a row with the abdominal spiracles during the embryonic stage. Therefore, I believe that the thoracic spiracles are serially homologous with the abdominal ones.

In the hymenopteran *Apis* (NELSON, 1915) and *Athalia* (FAROOQI, 1963), a pair of tracheal invaginations arises in the maxillary segment in the embryonic stage, and in *Apis*, tracheae in the head are derived from these invaginations. In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, however, such invaginations were never observed.

During embryogenesis, the spiracles of the metathoracic segment disappear, and only

vestigial tracheae connect with the inner side of the body wall in the first-instar larvae in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*. The same fact is reported by REMPEL and CHURCH (1972) in *Lytta*.

In the mecopteran *Microchorista philpotti* (Nannochoristidae), there are nonfunctional, vestigial spiracles on the metathoracic segment in the larval stage (PILGRIM, 1972), which suggests the primitive nature of this species, to judge from the results of the present study.

ii) Oenocytes and trichogen cells

Panorpa pryeri

In Stage 7, paired cell clusters in the ectoderm are found posteromedially from the spiracles of the first eight abdominal segments. These cell clusters, which stain poorly with hematoxylin, consist of four to five large cells (about 20μm) with large nuclei (ca. 10μm in diameter, Fig. 108). These cell clusters are the rudimental oenocytes. There are also paired cell clusters in the ectoderm at the medial bases of the developing dorsal processes on the first nine abdominal segments. These consist of ten to twelve cells each and have characters similar to those seen in the rudimental oenocytes. Moreover, there arise paired cell clusters in the ectoderm laterally from the bases of the developing abdominal prolegs of the first eight segments. These clusters contain five to six cells each and resemble the rudimental oenocytes.

Late in Stage 9, all of these cell clusters stain much lighter with hematoxylin, and they are situated at almost their original positions against the inner side of the body wall in first-instar larvae. During embryogenesis, cell divisions of the oenocytes were never seen.

Also observed are some cells which have larger nuclei (ca. 15μm in diameter) and cytoplasmic processes toward the body surface. These are the rudimental trichogen cells.

Panorpodes paradoxus and *Bittacus laevipes*

In *Pd. paradoxus*, the rudimental oenocytes appear as paired cell clusters in the ectoderm, just posteromedially from the spiracles of the first eight abdominal segments, in Stage 5, and in Stage 6 in *B. laevipes*. In *Pd. paradoxus*, this cell cluster consists of seven to eight cells, having nuclei 5 to 6μm in diameter, slightly larger than those of the surrounding ectodermal cells.

In *B. laevipes*, this cell cluster contains about ten cells, which stain lightly with hematoxylin and have large nuclei (ca. 7μm in diameter). At the same time, rudimental trichogen cells become apparent in the ectoderm of the body wall, and oenocytes are still found in the first-instar larvae in *Pd. paradoxus* and *B. laevipes* as in *P. pryeri*.

The oenocyte-like cell cluster found on both sides of the rudimental oenocytes in *P. pryeri* are never found throughout the embryonic stage in *Pd. paradoxus* and *B. laevipes*.

Discussion of oenocytes and trichogen cells

Oenocytes appear in the ectoderm posteromedially from the spiracles on the first seven or

eight abdominal segments in many insects, *i.e.*, the orthopteran *Locusta migratoria* (ROONWAL, 1937), neuropteran *Chrysopa perla* (BOCK, 1939), lepidopteran *Diacrisia virginica* (JOHANNSEN, 1929), coleopteran *Euryope terminalis* (PATERSON, 1932), dipteran *Aedes aegypti* (RAMINANI and CUPP, 1978), etc. In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, the manner of oenocyte formation is the same in the above mentioned species. The existence of the oenocyte-like cell masses found on both sides of the oenocytes in *P. pryeri* may be unknown in other insect embryos.

In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, it seems that the oenocytes do not increase in number during the embryonic stage, but they increase in number in *Euryope* (PATERSON, 1932).

Rudimental trichogen cells first appear at the stage when oenocytes are formed, and the nuclei are larger than those of the oenocytes in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*. These characters of the trichogen cells have also been observed in *Chryosopa* (BOCK, 1939) and *Diacrisia* (JOHANNSEN, 1929).

iii) Nervous system

a) Ventral nerve cord

Panorpa pryeri

Late in Stage 4 or at the beginning of Stage 5, neuroblasts are discernible in the ectoderm of the gnathal and following segments on both sides of the medioventral line of the embryo (Fig. 109). These cells are easily distinguished from the surrounding ectodermal cells, since they stain only lightly with hematoxylin, are large in size (about 20μm), and have large nuclei (10 to 12μm in diameter). There are about ten neuroblasts on each side of the segment in the thoracic region. At the same time, there appears a shallow furrow along the medioventral line and it is the rudimental neural groove.

Late in Stage 5, the number of neuroblasts increases to fifteen to twenty on each side of a thoracic segment. In Stage 6, the neuroblasts repeat mitoses and pile up inward two rows of daughter cells which become the future ganglion cells. The peripheral daughter cells become slightly flat and cover the developing ganglia (Fig. 110).

The neural groove deepens and the ectodermal cells of the medioventral line become situated between the developing ganglia. They are the rudimental median cord. As development of the median cord proceeds, there occur three to four cells which stain lightly with hematoxylin and have large nuclei (about 10μm in diameter, Fig. 111). They are similar to the neuroblasts in the characters mentioned above and are found in each of the segments.

In the middle of Stage 6, the paired developing ganglia begin to connect with each other by two commissures, though by only one commissure in the mandibular segment. The neuroblast-like cells of the median cord seem to participate mainly in the formation of the transverse commissures of the ganglion (Fig. 112). At the same time, the peripheral daughter cells mentioned before begin to become membranous, and seem to be the developing perilemma (perineurium, in the strict sense).

Each ganglion in which the neuropiles have not yet differentiated is connected with

adjacent ganglia by two connectives. The median cord almost loses its connection with the ventral epidermis. In the interganglionic regions, median cord cells do not participate in ganglion formation and disintegrate during late embryogenesis.

Late in Stage 6, the ganglia of the mandibular, maxillary and labial segments, and of the eighth to tenth abdominal segments begin to fuse. In Stage 7, these ganglionic concentrations consolidate further, the former becoming the large suboesophageal ganglion at the posterior part of the head, and the latter forming a large synganglion in the eighth abdominal segment.

Panorpodes paradoxus, *Biltacus laevipes* and *Boreus westwoodi*

The formation of the ventral nerve cord in *Pd. paradoxus* and *B. laevipes* is basically the same as that of *P. pryeri*. In *Pd. paradoxus* and *B. laevipes*, late in Stage 4, neuroblasts are distinguishable in the ectoderm of the gnathal and following segments on both sides of the medioventral line. In each of the thoracic and abdominal segments, there are about a dozen neuroblasts on each side. Several large cells of the median cord seem to take part in the formation of the transverse commissure in *Pd. paradoxus* and *B. laevipes*, as in *P. pryeri*.

In *Pd. paradoxus*, in Stage 6, the gnathal ganglia begin to consolidate, and the same tendency is observed in the eighth to the tenth abdominal ganglia in *B. laevipes* in Stage 5.

In *Bo. westwoodi*, neuroblasts are discernible in the gnathal and following segments at the end of Stage 4, and about ten neuroblasts occur on each side of the thoracic segments.

b) Brain

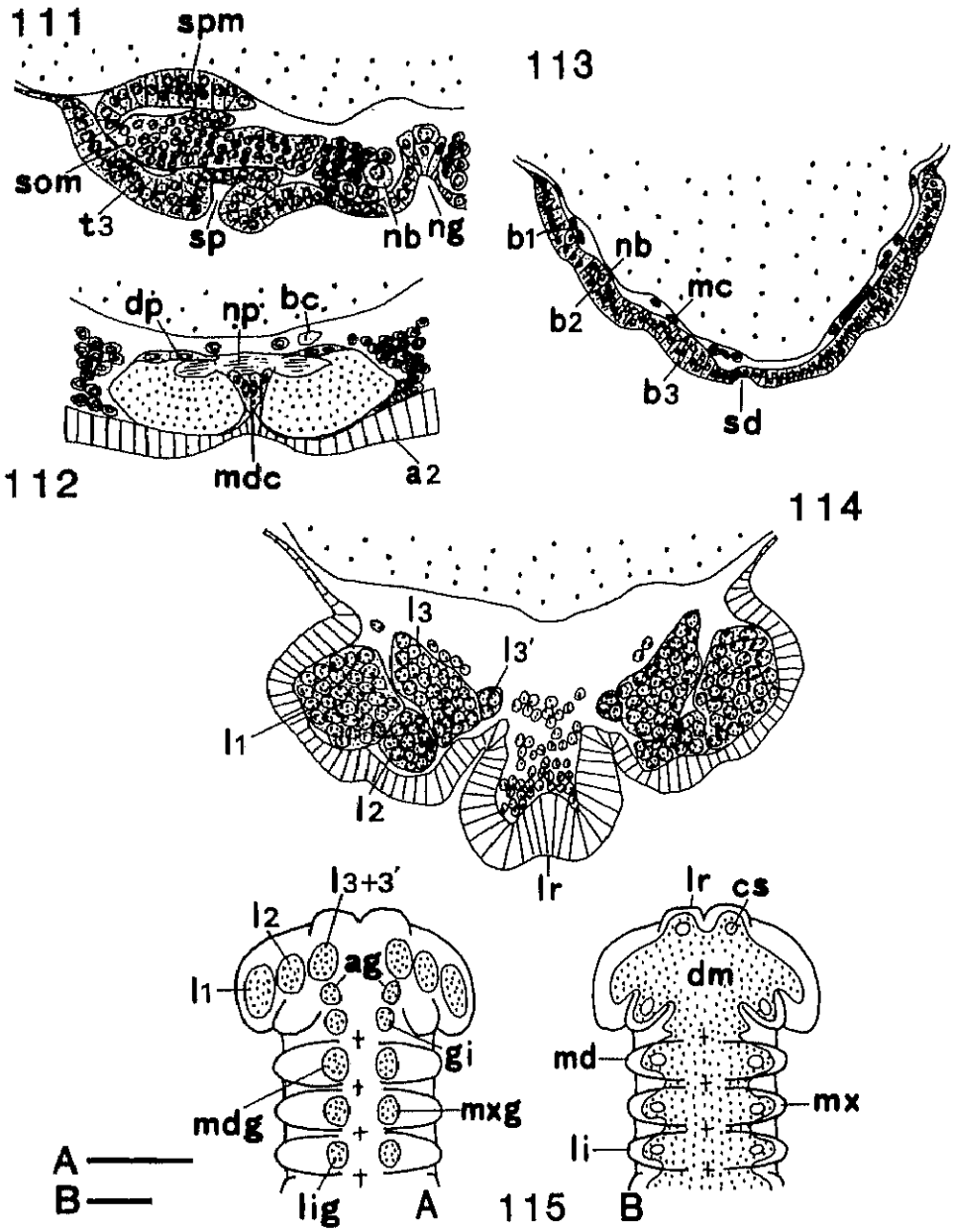
Panorpa pryeri

In Stage 4, there arise three pairs of protocephalic ectodermal bulges (Fig. 113) in which there are some round cells, namely neuroblasts.

Midway through Stage 5, the neuroblasts increase in number and stain lightly with hematoxylin. These neuroblasts begin to form the tissues of lobus 2 and lobus 3 of the future protocerebrum. At the same time, paired neuroblast masses which become the deutocerebrum are found in the ectoderm at both sides of the developing stomdaeum. Another pair of neuroblast masses appears in the intercalary segment, and these develop into the tritocerebrum. Figure 115 shows diagrammatically the distributions of the ganglia and mesodermal cells of the cephalognathal region in this stage.

In the middle of Stage 6, the neuroblasts of lobus 2 and lobus 3 produce a large number of daughter cells by mitoses and these lobi begin to separate from the protocephalic body wall. There is also a pair of small neuroblast clusters connecting with lobi 3, and these clusters seem to be lobi 3' (Fig. 114). The lobi 1, 2, 3 and 3' fuse with each other to form the future protocerbrum, and at this time, the developing proto-, deuto- and tritocerebrums become connected with each other. The optic plate differentiates in the protocephalic body wall connecting with lobus 1 (Fig. 116).

In Stage 7, the paired lobi 3 are connected by a commissure, and the connective also appears between the proto- and deutocerebrums, and neuropiles are formed in the developing



Figs. 111, 112. Cross sections of embryo of *P. pryeri* in Stage 6 (111) and in the middle of Stage 6 (112).

Figs. 113, 114. Horizontal sections of embryo of *P. pryeri* in Stage 4 (113) and in the middle of Stage 6 (114).

Fig. 115. Diagram showing distribution of ganglia (A) and mesodermal cells (B) in the cephalognathal region of embryo of *P. pryeri*.

Scale (A) =50μm (Figs. 111, 112, 114). Scale (B) =100μm (Fig. 113).

brain. The lobus 1 differentiates into proximal and distal parts (Fig. 117); the former is the future medulla, and the latter is the future lamina ganglionaris. The commissure of the tritocerebrum forms beneath the developing stomodaeum (Fig. 102).

Late in Stage 8, or in Stage 9, the brain of the embryo reaches completion. In the upper part of lobus 2, there arise two to three round cells which have large nuclei (about 20μm in diameter). These nuclei are only slightly stained with hematoxylin and contain a few particles stained with eosin. Because of their position, they are considered as globuli cells of the corpora pedunculata. At the same time, innervations are found from the deutocerebrum to the antennae and from the tritocerebrum to the frontal ganglion.

In lobus 1, about a dozen cells which stain slightly deeper with hematoxylin than surrounding cells are discernible at the distal margin of the medulla. Their cytoplasm stains darkly with eosin in the first-instar larva.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

In *Pd. paradoxus*, the neuroblasts in the protocephalic ectoderm arise late in Stage 4, and lobus 1 of the protocerebrum seems to be formed by invagination of the ectoderm.

In *B. laevipes*, there are three pairs of protocephalic bulges in Stage 4. Late in the stage, neuroblasts appear in the protocephalic ectoderm. The later embryonic development of the brain, including that of *Pd. paradoxus*, is basically the same as described for *P. pryeri*.

The neuroblasts appear in the protocephalic ectoderm in Stage 5 in *Bo. westwoodi*.

c) Stomatogastric nervous system

Panorpa pryeri

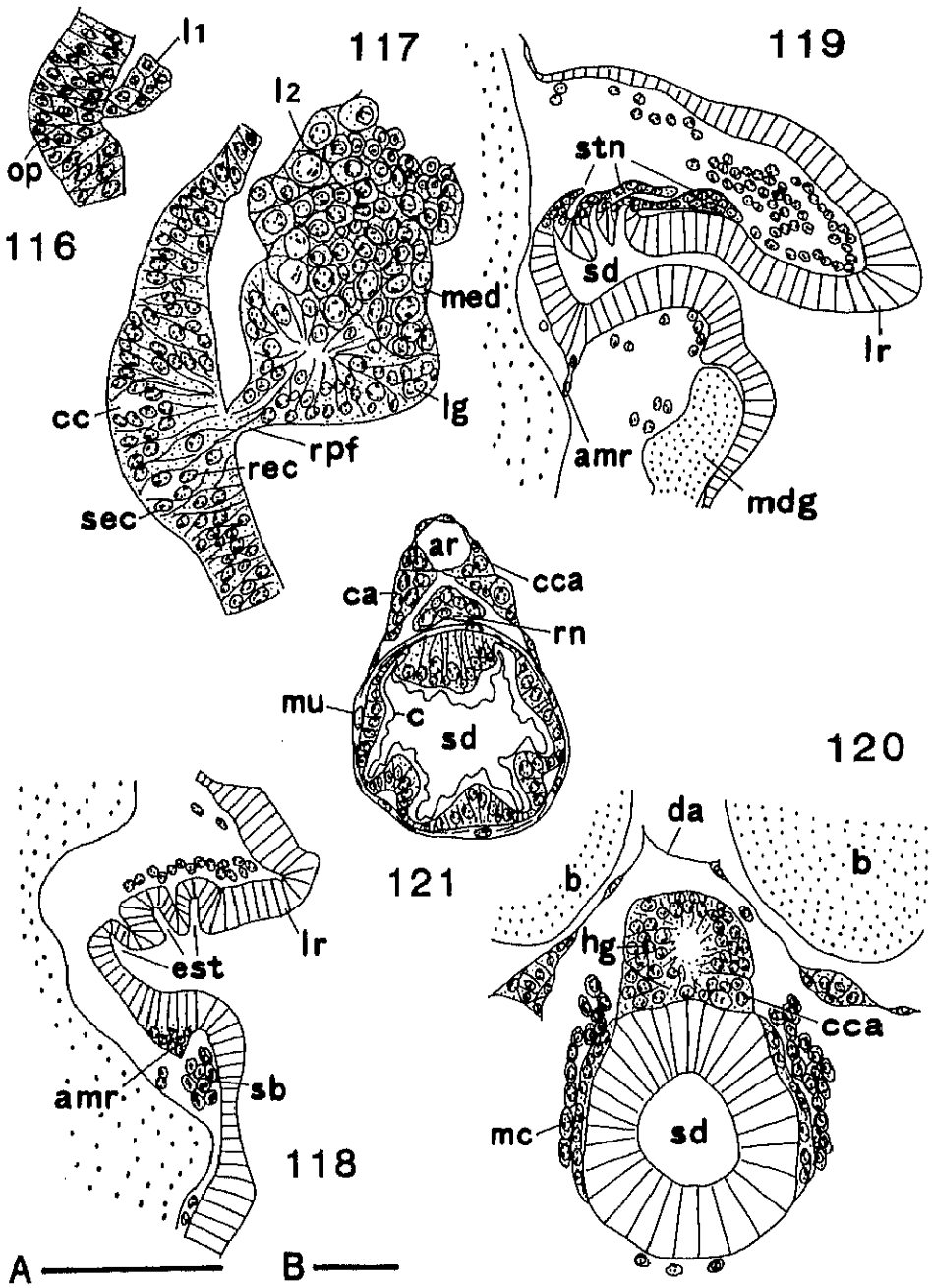
In the middle of Stage 5, three evaginations first become apparent along the mediodorsal line of the developing stomodaeum (Fig. 118). The future stomatogastric nervous system is derived from these evaginations.

Halfway through Stage 6, the evaginations elongate forward along the stomodaeal roof (Fig. 119). The rudiment of the frontal ganglion arises from the anterior evagination; rudiments of the recurrent nerve and hypocerebral ganglion are derived from the median and posterior evaginations, respectively. As development proceeds, the tubular form of the evaginations begins to break down.

In Stage 7, the developing frontal ganglion migrates forward, and nerve fibers are formed within the frontal and hypocerebral ganglia and recurrent nerve. The anlagen of the corpora cardiaca begin to differentiate from the anteroventral part of the developing hypocerebral ganglion, which is situated near the bottom of the developing stomodaeum (Fig. 120).

In the middle of Stage 7, the openings of the anterior two evaginations close, and then that of the posterior one also closes. Late in Stage 7, or in Stage 8, the rudimental corpora cardiaca begin to shift laterally.

In the first-instar larva, the corpora cardiaca are located posterolaterally from the hypocerbral ganglion. A pair of paracardiac nerves, which run forward from the anterior



Figs. 116, 117, 120. Cross sections of embryo of *P. pryeri* in the middle of Stage 6 (116) and in Stage 7 (117, 120).

Figs. 118, 119. Sagittal sections of embryo of *P. pryeri* in the middle of Stage 5 (118) and in Stage 6 (119).

Fig. 121. Cross section of first-instar larva of *P. pryeri*.

Scale (A) =50μm (Figs. 116-118, 120, 121). Scale (B) =50μm (Fig. 119).

ends of the corpora cardiaca, can be seen. The corpora allata are united with the posterior ends of the corpora cardiaca (Fig. 121).

Panorpodes paradoxus and *Bittacus laevipes*

In *Pd. paradoxus* the stomatogastric nervous system is formed from three evaginations from the stomodaeal roof at the beginning of Stage 5 (Fig. 122), and late in Stage 4 in *B. laevipes*, and the formation of the system is fundamentally the same as in *P. pryeri*.

Discussion of development of nervous system

Embryogenesis of the brain in *P. pryeri*, *Pd. paradoxus* and *B. laevipes* is similar to that observed in other insects (ROONWAL, 1937; KESSEL, 1939; ANDO, 1962; MIYAKAWA, 1974b; SUZUKI *et al.*, 1987).

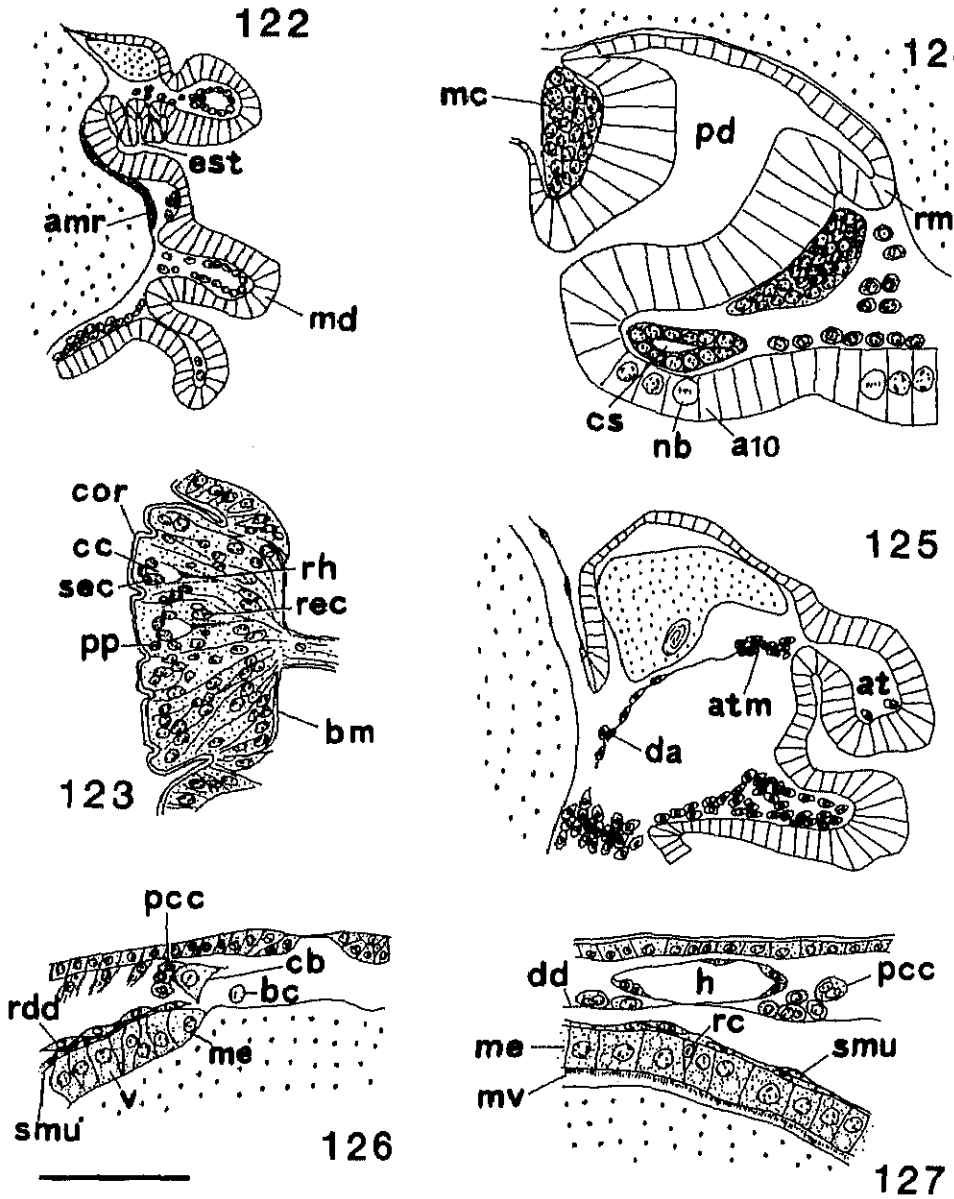
In *Tenebrio* (ULLMANN, 1967), the neuroblasts never appear in lobus 1 or the optic lobe during embryogenesis. The neuroblast-like, large round cells of lobus 1 in *P. pryeri* , *Pd. paradoxus* and *B. laevipes* seem not to divide teloblastically, so they may not be true neuroblasts in the strict sense as defined by ULLMANN (1967) and REMPEL *et al.* (1977). Furthermore, ULLMANN (1967) states that lobus 1 is derived from an ectodermal invagination, in the same manner as described in *Lytta* (REMPEL *et al.*, 1977), and as observed earlier in *Pd. paradoxus* (SUZUKI, 1985).

The stomatogastric nervous system originates from an evagination of the stomodaeal roof in *Epiophlebia* (ANDO, 1962). However, in *Chilo* (OKADA, 1960) and *Aedes* (RAMINANI and CUPP, 1978), no evaginations appear and the system is derived directly from the epipharyngeal roof. In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, the system originates from three evaginations of the stomodaeal roof, as many other insects (ROONWAL, 1937; TIEGS and MURRAY, 1938; ULLMANN, 1967; REMPEL and CHURCH, 1969; MIYAKAWA, 1974b; TANAKA *et al.*, 1985).

Embryogenesis of the ventral nerve cord presents two problems. The first is the participation of the median cord in ganglionic formation, and the second is the origin of the perineurium.

BADEN (1936) suggested that the median cord participates in neither ganglionic formation nor other organic formation, but degenerates. It seems to be the general view, however, that the median cord takes part in ganglionic formation (OKADA, 1960; SPRINGER, 1967; SPRINGER and RUTSCHKY, 1969; MIYAKAWA, 1974b, etc.), and results of the present study agree with this view.

As for the origin of the perineurium, there are three views. The first is the lateral cord origin (NELSON, 1915; EASTHAM, 1930; ROONWAL, 1937; ANDO, 1962; ASHHURST, 1965; REMPEL *et al.*, 1977; SUZUKI *et al.*, 1987), the second is the median cord origin (TIEGS and MURRAY, 1938; OKADA, 1960), and the third is mesodermal origin (BADEN, 1936; LARINK, 1969). In *P. pryeri*, peripheral ganglionic cells derived from the lateral cord seem to form the perineurium.



Figs. 122, 124. Longitudinal sections of embryo of *Pd. paradoxus* in Stage 5 (122) and late in Stage 4 (124).

Figs. 123,125. Longitudinal sections of embryo of *P. pryeri* in Stage 9 (123) and in the middle of Stage 6 (125).

Fig. 126. Cross section through first abdominal segment of embryo of *P. pryeri* in Stage 8.

Fig. 127. Cross section through third abdominal segment of embryo of *P. pryeri* in Stage 9.

Scale=100μm (Figs. 122, 125) and 50μm (Figs. 123, 124, 126, 127).

iv) Sense organs: larval eyes

Panorpa pryeri

Midway through Stage 6, at the lateral body wall of the embryonic head, rudimental optic plates become evident. They are thicker than surrounding ectodermal structures and connected with rudimental postretinal fibers of lobus 1. Simultaneously, the cell arrangement of developing optic plates begins to be irregular (Fig. 166).

In Stage 7, rudiments of the retinular and Semper's cells differentiate in the developing optic plates, and crystalline cones are formed by the Semper's cells (Fig. 117).

In Stage 9, the developing crystalline cones become conspicuous (Fig. 123), because they do not become stained with hematoxylin or eosin. Rudimental rhabdoms are discernible beneath the crystalline cones. On the surface of the developing larval eyes, there is a corneal zone 1 to 1.5μm-thick secreted by the corneagenous cells or primary pigment cells, and a basement membrane is distinguishable. As development proceeds, the cornea thickens to about 6μm at points of maximun thickness, and that of the first-instar larva appears slightly biconvex. Judging from its staining characteristics, the cornea is divided into three layers. The other layer (approximately 4μm-thick) remains transparent, the middle layer (1μm-thick) stains with eosin, and the inner layer stains with hematoxylin. The eyes, located close behind the antennal bases, consists of about thirty ommatidia each. An ommatidium has four Semper's cells and eight retinular cells.

Bittacus laevipes and *Panorpodes paradoxus*

In *B. laevipes*, the rudimental postretinal fiber appears beneath the developing optic plate in Stage 5. Cell differentiation occurs in the optic plate, which begins to thicken in Stage 6, and the developing crystalline cones are discernible in Stage 7. The components in an ommatidium of the first-instar larva are similar to those in *P. pryeri*.

In *Pd. paradoxus*, a rudimental postretinal fiber elongates from lobus 1 of the protocerebrum to the future optic plate at the end of Stage 4 or the beginning of Stage 5. In Stage 6, the rudimental optic plate thickens slightly, though no cell differentiation is observed. Late in this stage, however, the postretinal fiber degenerates, so the optic plate has no connection with lobus 1. Subsequently the optic plate becomes thin again. As a result, the first-instar larva of *Pd. paradoxus* has no eyes.

Discussion of larval eyes

Generally speaking, larvae of holometabolous insects have lateral ocelli, or stemmata (SNODGRASS, 1935; PAULUS, 1979). Embryogenesis of the stemmata seems to be fundamentally of two types. In the first type, the formation of the stemma is accompanied by ectodermal invagination of the developing optic plate, and invaginated ectodermal cells differentiate into retinular cells. This type is reported in the coleopteran *Hydrophilus* (PATTEN, 1887).

In the second type, there is no ectodermal invagination of the optic plate, and as the plate

thickens the elements of the future stemma, including retinular cells, form within the developing optic plate. This type is known in the lepidopteran *Ephestia kuhniella* (BUSSELMANN, 1935) and *Heliothis zea* (PRESSER and RUTSCHKY, 1957), and the mecopteran *P. pryeri* and *B. laevipes*. The compound eye formation of hemimetabolous insects, *e.g.*, *Epiophlebia* (ANDO, 1957) and *Oncopeltus* (BUTT, 1949), also belong to this type. The insects included in the second type are further divided into two groups. The first group has a typical ommatidium which has four Semper's cells and eight retinular cells; *Epiophlebia*, *Oncopeltus* and other hemimetabolous insects belong to this type (SNODGRASS, 1935). The second group includes insects in which the stemmata have three Semper's cells and seven retinular cells (*Ephestia*, BUSSELMANN, 1935; *Heliothis*, PRESSER and RUTSCHKY, 1957; Trichoptera and Lepidoptera, PAULUS and SCHMIDT, 1978).

The ommatidium of *P. communis* (BIERBRODT, 1942), *P. pryeri* and *B. laevipes* (ANDO and SUZUKI, 1977; SUZUKI and NAGASHIMA, 1989) has four Semper's cells and eight retinular cells, so that they are included in the first group, together with hemimetabolous insects. PAULUS (1979) suggested that the stemmata of the second group are modified from the ommatidia of the hemimetabolous insects. Accordingly, the larval eyes of *P. communis* (BIERBRODT, 1942), *P. pryeri* and *B. laevipes* are thought to be more akin to those of the Hemimetabola than those of the coleopterans, in the manner of their development, and than those of the lepidopterans on the degree of the modification of the ommatidia.

In *Pd. paradoxus*, the first-instar larva has no eyes, as observed by ISSIKI (1959), and this seems to be an apomorphic character, judging from its embryogenesis (SUZUKI, 1985).

2. Mesodermal derivatives

i) Segmentation of inner layer, formation of coelomic sac and differentiation of splanchnic and somatic mesoderm

Panorpa pryeri

In Stage 4, mesodermal cells of the preoral region lie scattered (Fig. 113). Those of the gnathal segments begin to lie segmentally and expand laterally before the segmentation of the embryonic ectoderm, while a layer of mesodermal cells exists in the intersegmental regions. In the posterior part, behind the gnathal segments, however, segmentation of the mesoderm does not yet occur in Stage 4, and the primary median mesoderm is still distributed along the medioventral line of the embryo. The mesodermal cells of caudal end of the embryo exist in a large mass (Fig. 130).

In Stage 5, the segmentation of the mesoderm finally reaches to the tenth abdominal segment. Simultaneously, paired coelomic sacs appear in the third abdominal segment (Fig. 109).

In the middle of Stage 5, coelomic sacs of the tenth abdominal segment are finally formed. Anterior to the mandibular segment, coelomic sacs are found only in the labral and antennal regions. Figure 115 shows diagrammatically the distribution of the mesodermal cells and coeloms in the cephalognathal region.

In Stage 6, the ventral wall of the coelom begins to extend and lines the ectoderm of the embryo. As development proceeds, the cells of the coelomic ventral wall scatter and become rudimental somatic mesodermal cells (Fig. 110). At the same time, the mesodermal cells of the dorsal wall persist in their original arrangement and are the future splanchnic mesoderm.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

In *Pd. paradoxus* and *B. laevipes*, mesodermal cells are distributed from the anterior end to the posterior end of the embryo (Fig. 97) by the middle in Stage 3 and late in Stage 2, respectively. Segmentation of the mesoderm begins before the ectodermal segmentation of the embryo.

In *Pd. paradoxus*, late in Stage 4, paired coelomic sacs are formed in each of the gnathal and following segments. The sacs appear somewhat earlier in Stage 4 in *B. laevipes*. In the cephalic region, coelomic sacs of the antennae and labrum are found, and in the abdominal region paired coelomic sacs are discernible in all ten segments in both species (Fig. 124).

The coelomic sacs of the gnathal and thoracic segments commence to differentiate into the developing somatic and splanchnic mesoderm in Stage 5, in *Pd. paradoxus* and *B. laevipes*.

In *Bo. westwoodi*, the primary median mesoderm migrates laterally in Stage 4. As development advances, the segmentation of the mesoderm occurs in the gnathal and thoracic segments before the ectodermal metamerization of the embryo. Late in Stage 4, the coelom begins to form, and its wall differentiates into the somatic and splanchnic mesoderm in Stage 5.

Discussion of coelomic sacs

The segmentation of the mesoderm is generally prior to the metamerization of the ectoderm of the embryo (JOHANNSEN and BUTT, 1941; ANDO, 1962; REMPEL and CHURCH, 1969), and it is also true in *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*.

Splanchnic mesoderm arises from the coelomic dorsal wall, and somatic mesoderm is derived from the coelomic ventral wall, in general (NELSON, 1915; BOCK, 1939; LUGINBILL, 1953; SANDER, 1956, etc.). This differentiation is undergone in the same way in *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*.

In the Hemimetabola, coelomic sacs often appear in the labral, antennal, intercalary and three gnathal segments in the cephalognathal region (MELLANBY, 1936; ROONWAL, 1937; ANDO, 1962), and in the embryonic abdomen they are found to the tenth (ANDO, 1962) or eleventh segment (ROONWAL, 1937).

On the other hand, in several Holometabola, coelomic sacs of the intercalary segment disappear (Trichoptera, MIYAKAWA, 1974c; Lepidoptera, EASTHAM, 1930; Siphonaptera, KESSEL, 1939), and furthermore, coelomic sacs of the mandibular and maxillary segments disappear in the coleopteran *Calandra* (TIEGS and MURRAY, 1938) and hymenopteran *Athalia* (FAROOQI, 1963). In the trichopteran *Stenopsyche* (MIYAKAWA, 1974c) and siphonapteran *Ctenocephalides* (KESSEL, 1939), coelomic sacs of the tenth abdominal segment do not appear during

embryonic development. In *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*, compared with the hemimetabolan insects, only the coelomic sacs of the intercalary and eleventh abdominal segments disappear. In the development of the coelomic sac, mecopteran insects seem to possess more primitive characters than those holometabolous insects mentioned above.

ii) Circulatory system

a) Aorta

Panorpa pryeri

Midway through Stage 6, mesodermal cells located near the bases of the rudimentary antennae become membranous and begin to elongate backward in the embryonic head (Fig. 125). This membranous structure is to be rudimental aorta.

In Stage 7, the developing aorta is found above the stomodaeum, though it is not yet tubular (Fig. 120). In Stage 8, the aorta becomes tubular. As development proceeds, the tube of the rudimental aorta grows backward and joins the developing heart or dorsal blood vessel in the first thoracic segment.

In *Pd. paradoxus* and *B. laevipes*, the formation of the aorta occurs by a process similar to that in *P. pryeri*.

b) Heart, pericardial cells, dorsal diaphragm and blood cells

Panorpa pryeri

In Stage 6, median mesodermal cells situated above the ventral nerve cord migrate laterally, and only a small amount of mesoderm remains above the ganglia. Simultaneously, among the mesodermal cells above the ganglia, there appear a few cells that stain poorly with hematoxylin (Fig. 110). They are rudimental blood cells.

In Stage 7, cells having large nuclei (about 6μm in diameter) and stained only lightly with hematoxylin are discernible at the lateral margin of the somatic mesoderm of the thoracic and following segments. They are the rudimental cardioblasts.

In Stage 8, they become crescent-shaped in transverse section. A few cells, the future pericardial cells, are located at beside the abdominal cardioblast in a transverse section (Fig. 126). Several cells of somatic mesoderm below the developing splanchnic muscles become less stained with hematoxylin, and are arranged in a row (Fig. 126). They are considered to be the future dorsal diaphragm.

Late in Stage 8, or in Stage 9, the tubular heart appears on the mediodorsal line of the embryo, extending from the first thoracic to the ninth abdominal segment. The vacuolated pericardial cells (Fig. 127) are observed especially at the sides of the heart, from the second to the ninth abdominal segment. In the same segments, the dorsal diaphragm is well developed.

Figure 128 shows the heart, pericardial cells and the dorsal diaphragm at the intersegmental region between the second and third abdominal segments in the first-instar larva.

The dorsal diaphragm contacts the body wall only in the intersegmental regions. Pericardial cells often have two nuclei that stain faintly with eosin, and their cytoplasm is stained only slightly with hematoxylin. The blood cells are found especially in the head and heart, and around the heart.

Panorpodes paradoxus and *Bittacus laevipes*

In *Pd. paradoxus*, cardioblasts first become discernible at the lateral margins of the somatic mesoderm of the thoracic and abdominal segments in Stage 6, and in Stage 5 in *B. laevipes*.

In Stage 6, the nuclei of the cardioblasts become slightly enlarged and, in *B. laevipes*, are less stained with hematoxylin.

In *Pd. paradoxus* and *B. laevipes*, blood cells are derived from scattered median mesodermal cells. The formation of the pericardial cells and dorsal diaphragm is essentially the same as in *P. pryeri*.

Discussion of cirulatory system

In *Stenopsyche* (MIYAKAWA, 1974c), the aorta is formed from the labro-antennal coelom, and in *Locusta* (ROONWAL, 1937), *Pyrilla* (SANDER, 1956) and *Epiophlebia* (ANDO, 1962) the aorta is derived from the antennal coelom. The origin in *P. pryeri*, *Pd. paradoxus* and *B. laevipes* is the same as in *Locusta*, *Pyrilla* and *Epiophlebia*. The cardioblasts originate from the lateral edges of the somatic mesoderm, and the dorsal diaphragm originates from the dorsal side of the somatic mesoderm in many insects (EASTHAM, 1930; ROONWAL, 1937; ANDO, 1962; MIYAKAWA, 1974c, etc.) as well as in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

The pericardial cells are derived from somatic mesodermal cells located at the sides of the cardioblasts in *Locusta* (ROONWAL, 1937) and *Melanoplus* (KESSEL, 1961), and they arise from the first thoracic to the eighth abdominal segment. In *P. pryeri*, pericardial cells appear in association with cardioblasts, as in *Locusta*. They, however, do not appear in the thoracic segments.

The blood cells of *P. pryeri*, *Pd. paradoxus* and *B. laevipes* originate from scattered primary mesodermal cells, as in *Pieris rapae* (EASTHAM, 1930), *Chryosopa perla* (BOCK, 1939), *Pimpla turionellae* (BRONSKILL, 1959), *Chilo suppressalis* (OKADA, 1960) and *Stenopsyche griseipennis* (MIYAKAWA, 1974c). In *Epiophlebia* (ANDO, 1962) and *Gerris* (MORI, 1969), in contrast, the blood cells are formed from secondary median mesodermal cells which migrate from the lateral mesoderm.

iii) Suboesophageal bodies

Panorpa pryeri

In the middle of Stage 5, mesodermal cells belonging to the intercalary segment are located beneath the developing midgut rudiment, which elongates backward from the blind end of the stomodaeum. As development advances, these cells are less stained with hematoxylin and have two nuclei each (Fig. 118); they are the rudimental suboesophageal bodies. They

migrate backward accompanying the invagination of the rudimental anterior tentorial arms. At first they are located on the ventral side of the stomodaeum, then they surround it.

In the first-instar larva, the suboesophageal bodies surround the stomodaeum at the level of the posterior end of the cranium. During embryogenesis, mitoses were never found in the suboesophageal bodies.

Panorpodes paradoxus and *Bittacus laevipes*

In *Pd. paradoxus*, the suboesophageal bodies develop from mesodermal cells of the intercalary segment at the beginning of Stage 5, and in Stage 6, in *B. laevipes*. They adjoin the sides and venter of the stomodaeum at the posterior end of the cranium in the first-instar larvae.

Discussion of suboesophageal bodies

The suboesophageal bodies are found in orthopteran, plecopteran, isopteran, mallophagan, coleopteran and lepidopteran insects (JOHANNSEN and BUTT, 1941). Also, they are known in the Trichoptera (PATTEN, 1884; MIYAKAWA, 1974c) and Diptera (OKADA, 1960).

PATTEN (1884) suggested that the suboesophageal bodies are of endodermal origin. Many other embryologists, however, have reported that the bodies are mesodermal in origin. These bodies are derived from mesodermal cells of the mandibular segment in *Locusta migratoria* (ROONWAL, 1937), *Melanoplus differentialis* (KESSEL, 1961) and *Bombyx mori* (WADA, 1955). They are formed from the mesodermal cells of the intercalary segment in *Pieris* (EASTHAM, 1930), *Calandra* (TIEGS and MURRAY, 1938), *Chilo* (OKADA, 1960), and *Stenopsyche* (MIYAKAWA, 1974c).

REMPEL and CHURCH (1969) reported that the suboesophageal bodies are derived from ectodermal proliferation in the intersegmental region between the intercalary and mandibular segments in *Lytta*, and they emphasized the necessity for the re-examination of the origin of suboesophageal bodies.

These bodies, however, originate from mesodermal cells which belong to the intercalary segment in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

In *Bombyx* (TOYAMA, 1902; WADA, 1955), the suboesophageal bodies proliferate the blood cells. The suboesophageal bodies of *P. pryeri*, *Pd. paradoxus* and *B. laevipes* seem not to participate in the formation of blood cells, because I could not find any mitotic figures in those bodies during embryogenesis.

iv) Gonads

Panorpa pryeri

Late in Stage 3, several large cells are observed in the mesodermal cell cluster at the caudal end of the embryo (Fig. 129). They seem to be rudimental germ cells.

At the end of Stage 3 or the beginning of Stage 4, fifteen to twenty germ cells are discernible on the mesodermal cell mass of the tenth abdominal segment (Fig. 130). In Stage

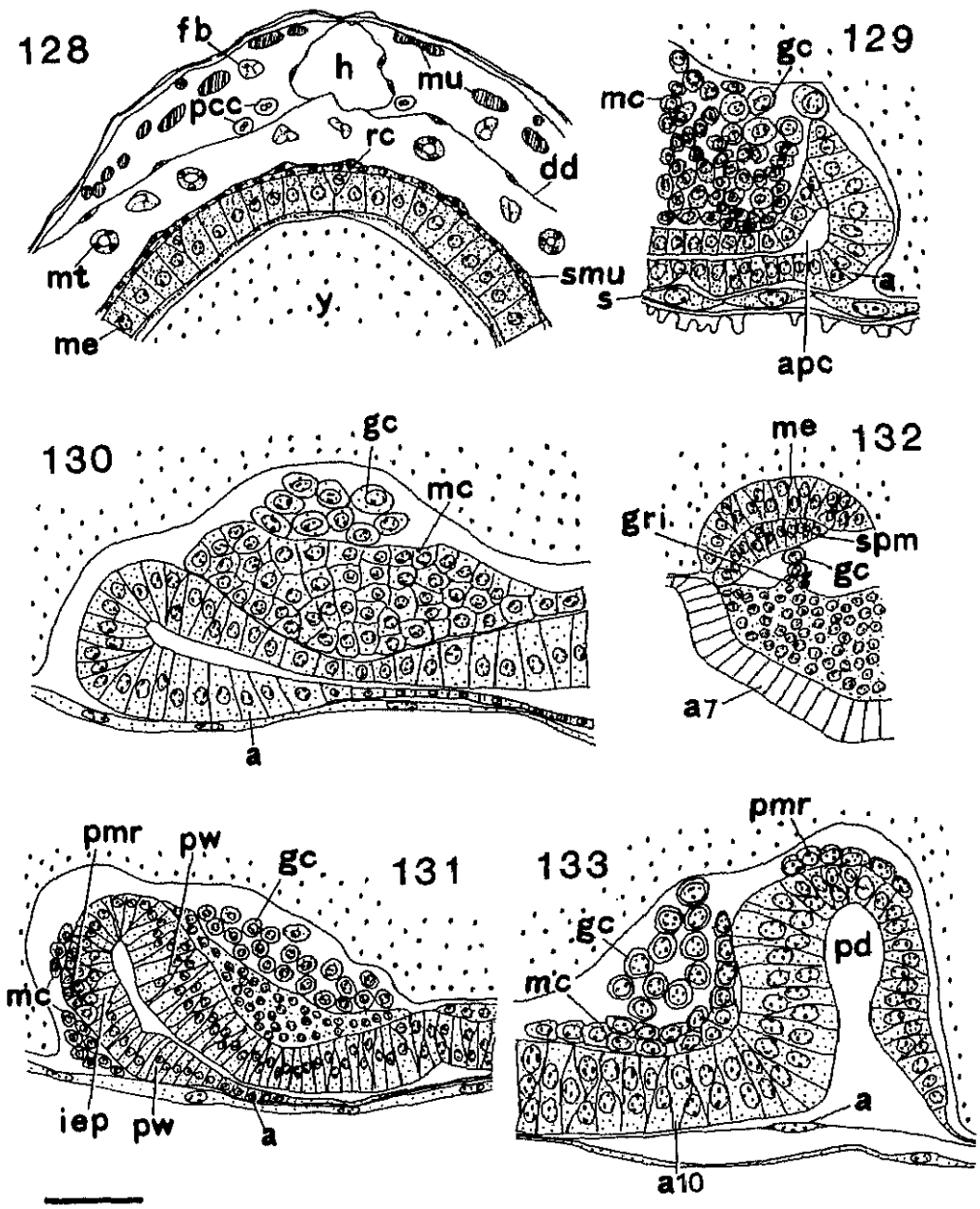


Fig. 128. Cross section between second and third abdominal segments of first-instar larva of *P. pryeri*.

Figs. 129-131. Longitudinal sections through posterior end of abdomen of embryo of *P. pryeri* late in Stage 3 (129), in Stage 4 (130) and in Stage 5 (131).

Fig. 132. Cross section of embryo of *P. pryeri* in the middle of Stage 6.

Fig. 133. Longitudinal section of embryo of *B. laevipes* in Stage 4.

Scale=50μm (Figs. 128-132) and 25μm (Fig. 133).

5, they begin to become rounded, stain more lightly with hematoxylin, and migrate forward with the development of the proctodaeum (Fig. 131). By the middle of Stage 5, they are distributed from the sixth to the tenth abdominal segment.

Midway through Stage 6, these caudal germ cells are scattered, but the center of their distribution is in the seventh abdominal segment. At this stage, one or two germ cells are observed on the ridges of the somatic mesoderm, the rudimental genital ridges, in all abdominal transverse sections (Fig. 132). In Stage 7, the distribution of germ cells is limited to the sixth and seventh abdominal segments. They become a pair of cell masses surrounded by vacuolated mesodermal cells. These vacuolated cells become a cell strand that elongates backward from the germ cell mass. In Stage 8, the germ cell masses become round and take their position in the sixth abdominal segment. They are almost the same shape as the gonads of the first-instar larva.

In the first-instar larva, each gonad contains about 120 germ cells and stains deeply with hematoxylin and eosin. The gonads and the ducts which run from the gonads to the eighth abdominal segment are associated with fat bodies.

Panorpodes paradoxus, Bittacus laevipes and *Boreus westwoodi*

In *Pd. paradoxus* and *B. laevipes*, gonad formation is similar to that in *P. pryeri*.

The rudimental germ cells become recognizable in the middle of Stage 4 as a cluster which contains about a dozen cells slightly larger than the surrounding mesodermal cells of the tenth abdominal segment, in *Pd. paradoxus*, and late in Stage 4 in *B. laevipes* (Fig. 133).

In the first-instar larva, the gonads are near the posterior margin of the sixth abdominal segment in *Pd. paradoxus*, and of the fifth abdominal segment in *B. laevipes*, and have short ducts at their posterior ends. Each gonad is composed of seven to eight germ cells in *Pd. paradoxus*, and their cytoplasm stains only faintly with eosin.

In *Bo. westwoodi*, about fifteen rudimental germ cells are found on the mesodermal cell mass of the tenth abdominal segment, at the end of Stage 4.

Discussion of origin of gonads

In many insects, the germ cells appear after gastrulation of the embryo. Their position is in the anterior part of the abdomen in *Bombyx* (MIYA, 1958), center of the abdomen in *Ctenocephalides* (KESSEL, 1939), and posterior part of the abdomen in *Epiophlebia* (ANDO, 1962), *Lytta* (CHURCH and REMPEL, 1971) and *Stenopsyche* (MIYAKAWA, 1974c). They are posterior in *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*.

As development advances, the germ cells become distributed, from the sixth to the eighth abdominal segment in *Bombyx* (MIYA, 1958), from the fifth to the sixth in *Epiophlebia* (ANDO, 1962), and from the second to the seventh in *Stenopsyche* (MIYAKAWA, 1974c), but in *Ctenocephalides* (KESSEL, 1939) the germ cells do not migrate from their original position.

The germ cell strands at the sides of the embryonic abdomen metamerize temporarily, take their position on each genital ridge, then they gather into a pair of cell masses, *i.e.*,

rudimental gonads (MIYA, 1958 ; ANDO, 1962, etc.). However, I failed to find the temporary segmentation of the germ cell strand in *P. pryeri*, *Pd. paradoxus* and *B. laevipes*.

v) Differentiation of muscles and fat bodies

Panorpa pryeri

In Stage 6, the coelomic sacs of each segment begin to break down and differentiate into the somatic and splanchnic mesoderm. In Stage 7, in the somatic mesoderm, there are cells which have a somewhat fibrous shape and cytoplasm that stains lightly with eosin (Fig. 108). They are developing muscle cells. Other cells of the somatic mesoderm are rudimental fat bodies. As development proceeds, the rudimental fat bodies become vacuolated and stain slightly with eosin.

In the first-instar larva, fat bodies are associated mainly with the gonads, heart and midgut but are also observed in the thoracic legs and abdominal prolegs.

Panorpodes paradoxus and *Bittacus laevipes*

In *Pd. paradoxus* and *B. laevipes*, differentiation of the rudiments of muscles and fat bodies from somatic mesoderm occurs in Stage 6.

In *P. pryeri*, *Pd. paradoxus* and *B. laevipes*, the differentiation and formation of the muscles and fat bodies is similar to that of many other insects (NELSON, 1915 ; ROONWAL, 1937 ; BOCK, 1939 ; ANDO, 1962 ; MORI, 1969, etc.). Therefore, there is no special discussion of this subject.

vi) Musculature of thoracic and abdominal segments of the first-instar larva

Panorpa pryeri

In the first-instar larva, the main thoracic musculature is essentially the same in the three thoracic segments. The main abdominal musculature is also the same in each of the abdominal segments except the caudal one. Accordingly, Fig. 134 shows the main musculatures of the metathorax and first abdominal segment for the convenience of comparison.

It is noteworthy that in the first eight abdominal segments, there are lateral muscles connecting to the inside of small processes mentioned before (Fig. 37), and they seem to be homologous with the lateral thoracic muscles connecting with the bases of thoracic legs (Fig. 134, arrows). None of the muscles of the anal legs are likely to be homologous with those of the thoracic and abdominal segments.

3. Alimentary canal formation

i) Stomodaeum or foregut

Panorpa pryeri

At the end of Stage 4, a shallow invagination of the stomodaeum arises in the center of the protocephalon (Fig. 32). As development proceeds, the anlage of stomodaeum elongates backward, and its inner end becomes flat, in the middle of Stage 6 (Fig. 119). By the middle

of Stage 7, the developing stomodaeum elongates and begins to slender, and the inner end slightly evaginates toward the midgut (Fig. 103). The closing membrane breaks and disappears late in Stage 9, and the cardiac valve is formed at the distal end of the stomodaeum.

The first-instar larva has a long oesophagus following the pharynx, and there is no evident proventriculus.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

In *Pd. paradoxus* and *Bo. westwoodi*, a shallow invagination of the rudimental stomodaeum arises in the center of the cephalic lobes late in Stage 4 (Figs. 53, 85), and in Stage 4 in *B. laevipes* (Fig. 64).

The further development of the stomodaeum of *Pd. paradoxus* and *B. laevipes* is essentially the same as that of *P. pryeri*. The proventriculus is not differentiated in these three species, either.

ii) Proctodaeum, or hindgut, and Malpinghian tubules

Panorpa pryeri

In Stage 4, a thick amnion, which includes the future dorsal wall and the blind end of the proctodaeum, continues for a length of approximately 100μm from the caudal end of the embryo (Fig. 130).

In Stage 5, the posterior end of the embryo starts to become immersed in the yolk. At this time, the mesodermal cells which later form the muscles surrounding the rudimental proctodaeum begin to cover the sides of the proctodaeum, and then its dorsal surface (Fig. 131).

Late in Stage 5, the inner end of the proctodaeum becomes thin (Fig. 135), and a thin epithelial rudiment of the midgut is found on it. At this stage, three paired anlagen of the Malpighian tubules evaginate from the anterior end of the proctodaeal wall (Fig. 135).

Midway in Stage 6, the developing proctodaeum elongates forward and has a loop near its mid-length. The looped part occupies the seventh abdominal segment, where the blind end of the proctodaeum is located, and the eighth abdominal segment. The Malpighian tubules extend forward to the second abdominal segment and turn back to the eighth. As development advances, the end of proctodaeum and the midgut rudiment on it fuse with each other, transforming into a thick closing membrane by Stage 8. Late in Stage 9, the pyloric valve is formed just behind the attachments of the Malpighian tubules, and the duodenal valve follows the pylorus. The closing membrane breaks down just before hatching. Figure 136 shows the alimentary canal of the first-instar larva.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

The formation of the proctodaeum and Malpinghian tubules of *Pd. paradoxus* and *B. laevipes* occur basically in the same manner as that observed in *P. pryeri*.

The sinking of the developing proctodaeum into the yolk begins in the middle of Stage 4 in *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*. The three pairs of rudimental Malpighian tubules grow outward from the blind end of the developing proctodaeum late in Stage 4 in *Pd. paradoxus*, and in Stage 5 in *B. laevipes*. The proctodaeum develops a loop late in Stage 5 in *Pd. paradoxus*, and in Stage 6 in *B. laevipes*.

iii) Midgut

Panorpa pryeri

Anterior midgut rudiment

At the end of Stage 4, the anterior midgut rudiment arises from a few proliferating cells in the inner end of the invaginating stomodaeum (Fig. 137).

In Stage 5, the anterior midgut rudiment elongates backward between the yolk and mesodermal cells and it begins to divide at its distal end about the middle of Stage 5.

As development proceeds, the distal ends of the rudiments, which assume the form of a pair of ribbons, extend forward to the first thoracic segment.

Posterior midgut rudiment

In Stage 5, a few cells of the posterior midgut rudiment appear at the blind end of the future proctodaeum (Fig. 131) and subsequently increase in number.

Late in Stage 5, as the developing proctodaeum invaginates, the blind end becomes flat. A pair of ribbons of the posterior midgut rudiment (Fig. 135) elongates as in the anterior midgut rudiment, and reaches anteriorly to the eighth abdominal segment.

Further development of midgut rudiments

The ribbon-like rudiments elongate along the dorsal surfaces of the coelomic sacs.

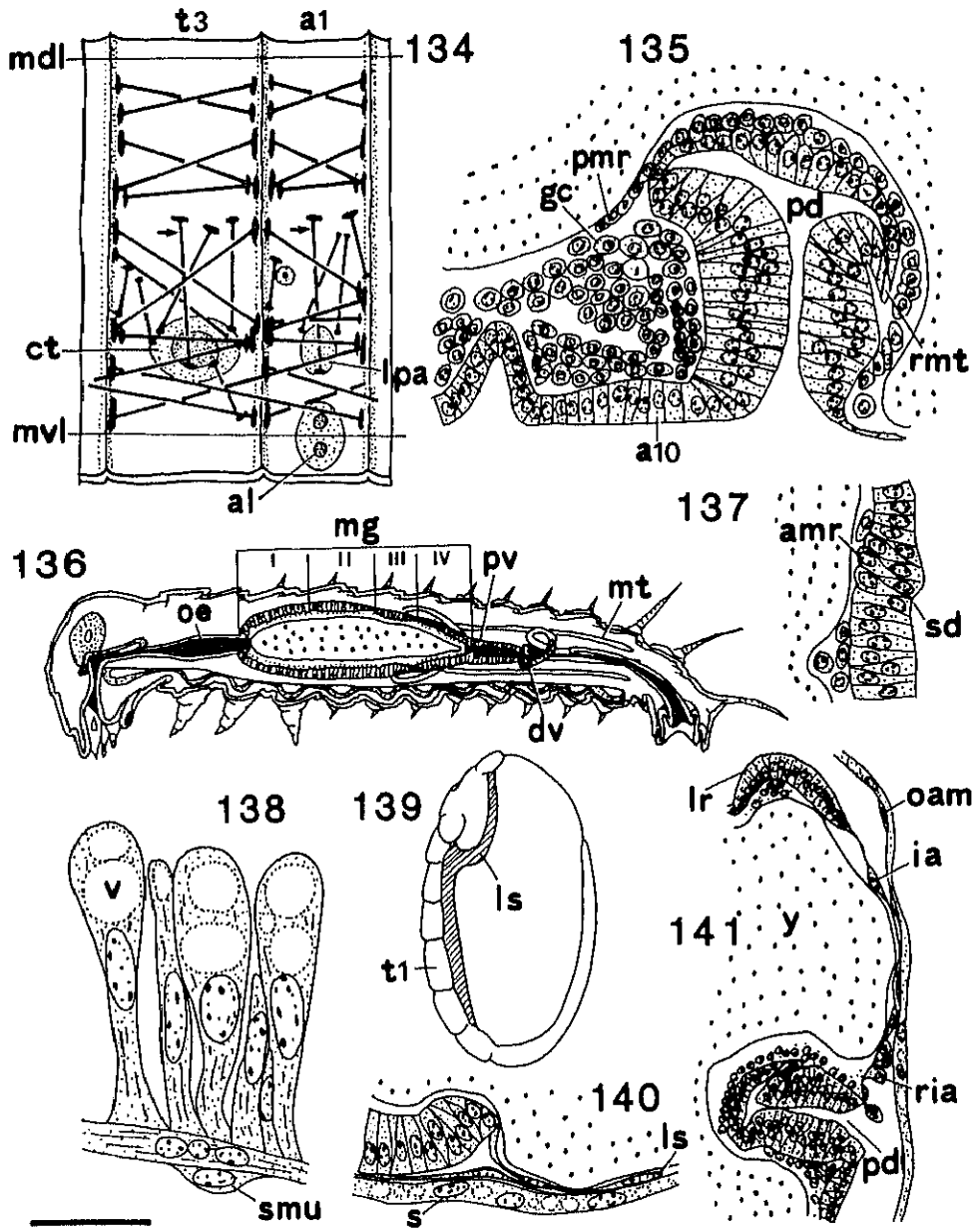
Late in Stage 6, the ribbons of the both midgut rudiments fuse in the third or fourth abdominal segment, and the rudiments continue between the inner ends of the stomodaeum and proctodaeum. The ribbons widen, cover the yolk ventrally, and the splanchnic mesoderm follows and coats them (Fig. 132).

In Stage 7, the ribbons become wider and almost cover the ventral side of the yolk except above the ventral nerve cord (Fig. 108). The splanchnic mesoderm commences to thin and form a double layer. At the same time the epineural sinus is considerably enlarged above the ventral nerve cord, and a remarkable consumption of yolk occurs.

In Stage 8, the developing midgut epithelium completely covers the ventral side of the yolk. It then covers the dorsal side of the yolk, so that the developing midgut assumes a tubular form.

In Stage 9, the midgut epithelium is thicker on the dorsal side than on the ventral, and has many cytoplasmic processes that later differentiate into microvilli (Fig. 127). Regenerative cells are found more commonly in the dorsal epithelium than in the ventral.

The epithelium of a newly hatched larva has many vacuoles, and morphologically



Figs. 134, 136. Diagrams showing alimentary canal (136) and main musculature of thorax and abdomen (134) of first-instar larva.

Figs. 135, 137, 141. Longitudinal sections of embryo of *P. pryeri* late in Stage 5 (135), at the end of Stage 4 (137) and in Stage 5 (141).

Fig. 138. Midgut epithelial cells of first-instar larva of *P. pryeri*.

Fig. 139. Diagram showing lateral spread of amnion of *P. pryeri* in Stage 4.

Fig. 140. Cross section through head of embryo of *P. pryeri* in Stage 5.

Scale=50μm (Figs. 135, 137, 140), 10μm (Fig. 138) and 100μm (Fig. 141).

differentiates in four regions of the midgut, *i.e.*, Regions I-IV (Fig. 136). In Region III, the epithelial cells show a characteristic shape (Fig. 138). These cells seem to correspond to those of 'Type II' in *P. communis* (GRELL, 1938).

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

The midgut formation of *Pd. paradoxus* and *B. laevipes* is bipolar and proceeds principally in the same manner as that of *P. pryeri* as just mentioned.

The anterior midgut rudiment is derived from the posterior end of the invaginating stomodaeum in the middle of Stage 4, in *Pd. paradoxus* and *B. laevipes*, and in Stage 4 in *Bo. westwoodi*.

The posterior midgut rudiment originates from the anterior end of the developing proctodaeum in the middle of Stage 4 in *Pd. paradoxus*, and at the end of Stage 4 in *B. laevipes*, but in *Bo. westwoodi* it does not yet appear in that stage.

In the first-instar larva of *B. laevipes*, the midgut epithelial cells in the anterior part of the posterior half of the midgut seem to be comparable to the cells of Region III in *P. pryeri*, though their distal ends are not rounded.

In *Pd. paradoxus*, there are no epithelial cells that seem to be comparable to the cells of Region III of *P. pryeri*.

Discussion of the alimentary canal

Formation of the alimentary canal in *P. pryeri* was studied in detail by SUZUKI and ANDO (1981), and the results of the present study agree with them. Formation of the stomodaeum in Mecoptera is the same as in many other insects (JOHANNSEN and BUTT, 1941; ANDERSON, 1972; HAGET, 1977). In the Mecoptera, the thick amnion of the embryonic caudal end participates in the formation of the proctodaeum, as in the Odonata (ANDO, 1962), in the hemipterans *Oncopeltus* (BUTT, 1949) and *Pyrilla* (SANDER, 1956), siphonapteran *Ctenocephalides* (KESSEL, 1939), trichopteran *Stenopsyche* (MIYAKAWA, 1975) and lepidopterans *Endoclita* (KOBAYASHI *et al.*, 1981) and *Neomicropteryx* (KOBAYASHI and ANDO, 1983). However, proctodaeal formation in the Mecoptera, in which the future blind end of the proctodaeum wholly originates from part of the thickened amnion, differs from that in the above species.

In the Mecoptera, it is also interesting that the pyloric valve is situated just behind the openings of the Malpighian tubules. In the larva of *P. communis*, there is a duodenal valve at the posterior end of the pylorus (GRELL, 1938), and the same is true for *Panorpodes* and *Bittacus*. A similar structure is seen in the cockroach *Blatta orientalis* (HENSON, 1944).

As for the midgut formation, in some Hemimetabola, the midgut epithelium is formed by cells or outgrowths of both the stomodaeum and proctodaeum, that is, by bipolar formation. Examples in the Hemiptera are *Oncopeltus* (BUTT, 1949) and *Pyrilla* (SANDER, 1956). The yolk cells participate together with the ectodermal bipolar formation of the midgut epithelium in the psocopteran *Liposcelis divergens* (GOSS, 1953) and embiopteran *Haploembia solieri*

(STEFANI, 1961). The participation of yolk cells in the midgut epithelium formation is also reported for the hemipteran *Gerris* (MORI, 1976). According to KISHIMOTO and ANDO (1986), the midgut epithelium of the plecopteran *Kamimuria tibialis* arises only from the end of the proctodaeum, *i.e.*, by unipolar formation, and they regard the unipolar condition as derived from the bipolar one.

In the Holometabola, as reviewed by HAGET (1977) and MORI (1983), formation of the midgut epithelium is usually bipolar (Neuroptera, BOCK, 1939; Coleoptera, LUGINBILL, 1953; REMPEL and CHURCH, 1969; Hymenoptera, BRONSKILL, 1964; Trichoptera, MIYAKAWA, 1975; Lepidoptera, MIYA, 1976; KOBAYASHI *et al.*, 1981; KOBAYASHI and ANDO, 1983; and Mecoptera). In the Mecoptera, the formation begins as soon as the stomodaeum and proctodaeum invaginate. However, the formation starts in an earlier stage in the siphonapteran *Ctenocephalides* (KESSEL, 1939) and dipteran *Dacus* (ANDERSON, 1962). In the Holometabola, the midgut rudiment is ectodermal in origin and is formed in the bipolar manner, in general, though there are some differences in the time of appearance of the midgut rudiments.

Cells of the mesoderm partially participate in the formation of the midgut rudiments in the megalopteran *Sialis lutaria* (STRINDBERG, 1915), coleopteran *Donacia crassipes* (HIRSCHLER, 1909), and lepidopteran *Catocala nupta* (HIRSCHLER, 1928). The participation of the mesoderm in the midgut formation, however, seems to be exceptional in the Holometabola, and these three investigations should be re-examined.

4. Other structures and phenomena

i) Fate of embryonic envelopes

Panorpa pryeri

In Stage 4, the original amnion covering the ventral surface of the embryo begins to spread laterally over the yolk mass from the cephalic to the thoracic region (Fig. 139). In Stage 5, the lateral spread of the original amnion (Fig. 140) extends to the seventh abdominal segment. As development proceeds, the amniotic spread almost covers the entire egg surface except the region near the caudal end of the embryo. In the middle of Stage 5, the embryo and yolk mass are covered completely by the amnion. Consequently, at this time the yolk mass is covered with the inner or dorsal amnion, and the outer amnion is situated just beneath the serosa. A few round cells of the inner amnion separate from the caudal end of the embryo and enter the space between the outer and inner envelopes (Fig. 141).

In Stage 7, the inner amniotic cells on the dorsal side of the egg become vacuolated and thick, and they thicken further late in Stage 7. This thickening part of the inner amnion becomes indistinguishable at the stage of the embryonic rotation. Late in Stage 8, the inner amnion disappears by the completion of the dorsal closure of the embryo, but the outer amnion persists at hatching.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

In *Pd. paradoxus* and *B. laevipes*, the formation and fate of the inner and outer amnions are the same as in *P. pryeri*.

The yolk mass is completely covered by the inner amnion by the middle of Stage 4 in *Pd. paradoxus*, by late in Stage 4 in *B. laevipes*, and during Stage 5 in *Bo. westwoodi*. In *Bo. westwoodi*, a thickening of the serosa at the anterodorsal part of the egg was observed late in Stage 4 (Fig. 142).

Discussion of the embryonic envelopes

As far as I am aware, there are few papers in which the differentiation of the inner and outer amnions is described. In some species of coleopteran insects, *e.g.*, *Brachyrhinus ligustici* (BUTT, 1936), *Apion apricans* and *Polydrosus sericeus* (KRZYSZTOFOWICZ, 1960) both amnions are found. The existence of inner and outer amnions seems to be a characteristic of the Mecoptera, considering that *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi* all have both the inner and outer ones.

The related phenomenon of the amnion covering the entire embryo and persisting until just before hatching is observed generally in the Lepidoptera (EASTHAM, 1930; PRESSER and RUTSCHKY, 1957; OKADA, 1960; ANDERSON and WOOD, 1968; KOBAYASHI *et al.*, 1981, etc.). In these lepidopteran insects, the amnion is comparable to the outer amnion of the Mecoptera mentioned above. In the Lepidoptera the continuous amnion is formed as a result of completion of the dorsal closure, and the yolk remains in the space between the amnion and serosa. The amnion formation in the Lepidoptera and Mecoptera seems to be a significant character when considering the relationship of these orders.

The partial thickening of the serosa in *Bo. westwoodi* resembles morphologically the hydropyle cells or columnar serosa of the heteropteran (COBBEN, 1968; MORI, 1969, 1970; MADHAVAN, 1974), plecopteran (KISHIMOTO and ANDO, 1985) and lepidopteran insects (KOBAYASHI and ANDO, 1983), and it seems to correlate with the moist or aquatic condition of the site where the eggs are deposited.

ii) Formation of abdominal prolegs

Panorpa pryeri

In Stage 7, small paired processes, which consist of ectodermal cell masses (Fig. 143), first become evident below each ganglion of the first eight abdominal segments; these are anlagen of abdominal prolegs. In Stage 8, the paired cell masses commence to protrude cytoplasmic processes on the ventral surface of the embryo.

Late in Stage 8, or in Stage 9, the paired cytoplasmic processes increase in size, and several nuclei migrate from the proximal part to the middle of the developing abdominal prolegs (Fig. 144). As these nuclei move to the peripheral zone of the abdominal proleg, a cavity appears within the proleg.

Panorpodes paradoxus and *Bittacus laevipes*

In *B. laevipes*, paired small cell masses, the rudimental abdominal prolegs, become recognizable beneath the first eight abdominal ganglia by the middle of Stage 6. The further development of the abdominal prolegs is essentially the same as in *P. pryeri*.

In *Pd. paradoxus*, no abdominal prolegs can be observed during embryonic development. However, a small process is found on the medioventral line at the center of each of the first eight abdominal segments in the first-instar larva. This small process is formed as follows :

Late in Stage 6, a small cell mass in the ectoderm arises at the position mentioned above. In the middle of Stage 7, these cell masses, except those of the first and second abdominal segments, begin to protrude cytoplasmic processes on the ventral surface of the embryo (Fig. 145).

As development proceeds, they elongate, although their nuclei remain in the proximal portion of each process. The processes have several nuclei at their base, and their cytoplasm stains lightly with eosin. The processes of the first and second abdominal segment become small.

Discussion of abdominal prolegs

In the Holometabola, larval abdominal prolegs or pseudopods are known, for example, in the Lepidoptera, Tenthredidae of the Hymenoptera and Blephariceridae of the Diptera (SNODGRASS, 1935 ; MATSUDA, 1976).

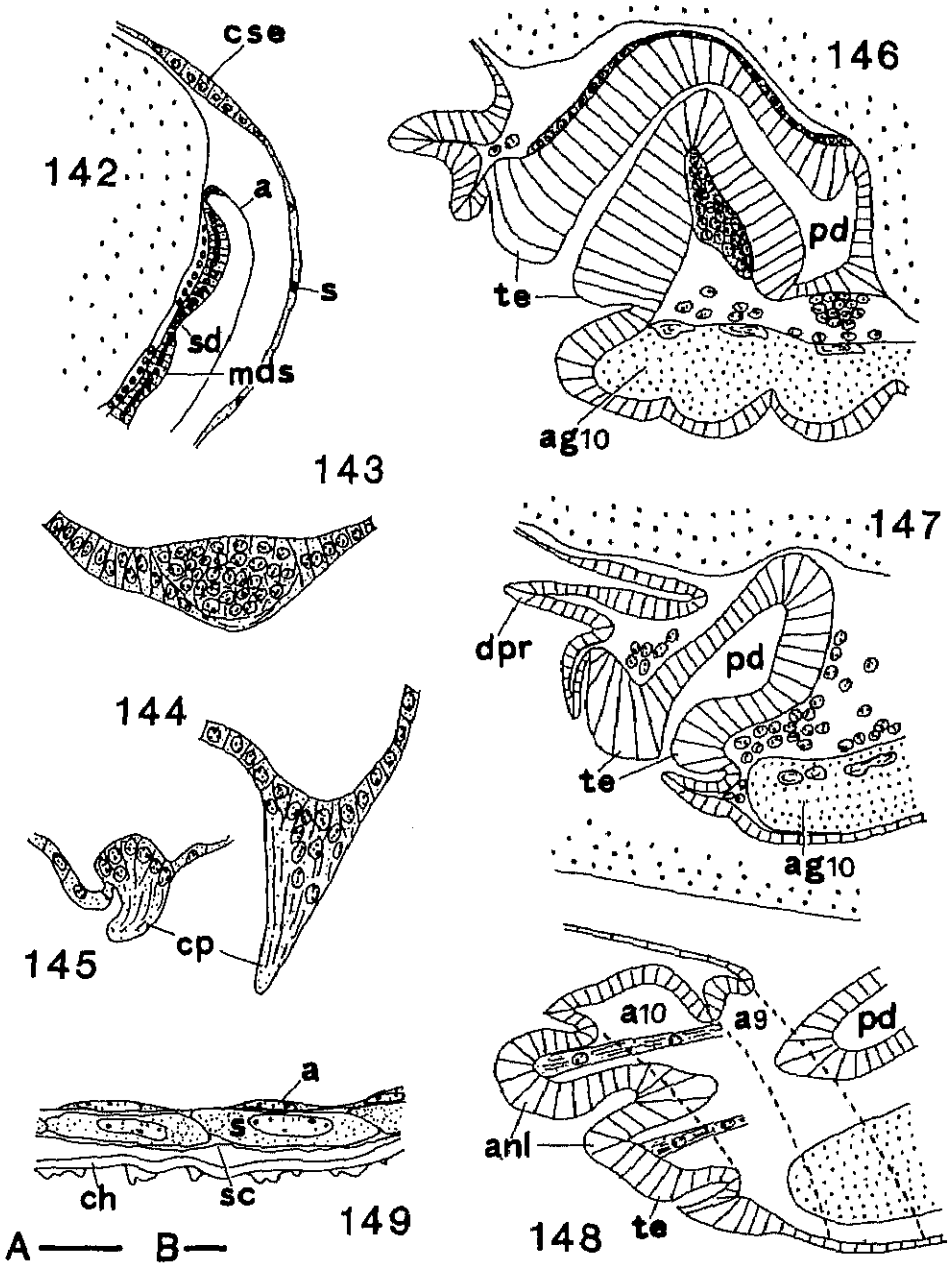
Thoracic leg formation in the Mecoptera starts as a slight thickening of the ectoderm, which is transformed into the evagination which is filled with subsomitic mesodermal cells. As development advances, lining mesodermal cells differentiate into muscles and fat bodies. Abdominal proleg formation proceeds in the same manner in the lepidopteran *Diacrisia virginica* (JOHANNSEN, 1929), hymenopteran *Athalia proxima* (FAROOQI, 1963) and dipteran *Neocurupira chiltoni* (CRAIG, 1967).

The abdominal proleg formation in *P. pryeri* and *B. laevipes* is conspicuously different from thoracic leg formation, and is rather similar to the development of trichogen cells. This suggests that the abdominal prolegs of *P. pryeri* and *B. laevipes* are different from the thoracic legs. ANDO and HAGA (1974) observed that no pleuropodia occur in the embryos of *P. pryeri* and *B. mastrillii*, and a pair of styliform appendages or abdominal prolegs are formed in each of the first to the eighth abdominal segments along the medioventral line of mature embryos. They, however, did not discuss homology of the thoracic and abdominal appendages. In *Pd. paradoxus*, small processes on the medioventral line of the larval abdomen are similar to abdominal prolegs, in the manner of their formation.

iii) Formation of anal legs and telson

Panorpa pryeri

Late in Stage 5, the lateral wall of the developing proctodaeum is very thick (Fig. 135), and this thickness persists till Stage 7. In Stage 7, the boundary between the posterior end of the tenth abdominal segment and the proctodaeum becomes distinct (Fig. 146). As development



Figs. 142, 145. Longitudinal sections of embryo of *Bo. westwoodi* late in Stage 4 (142) and in the middle of Stage 7 (145).

Figs. 143, 144, 146-148. Longitudinal sections of embryo of *P. pryeri* in Stage 7 (143), 146, 147), in Stage 9 (144) and in the middle of Stage 7 (148).

Fig. 149. Serosal cuticle of *P. pryeri* in Stage 5.

Scale (A) =50μm (Figs. 142, 146-148). Scale (B) =10μm (Figs. 143-145, 149).

advances, the lateral wall of the proctodaeum becomes thin, except for the posterior end of the abdomen, or telson (Fig. 147). At the same time, as the ganglion of the tenth abdominal segment moves forward, the posterior margin of the segment becomes a fold and surrounds the telson (Fig. 147).

In the middle of Stage 7, the ectoderm of the telson begins to grow thin, and several mesodermal cells which line the telson differentiate into muscles connecting with the distal end of the telson (Fig. 148). Four points with which these muscles connect are the lateral and dorsolateral sides of the proctodaeal opening, and these points subsequently become ectodermal protrusions. They are the developing anal legs. After turning of the embryo, these protrusions invaginate into the body cavity . For discussion of this subject, see Chapter III, 'Interpretation of metamerism in terminal region of abdomen' in General Discussion and Conclusion.

iv) Serosal cuticle

Panorpa pryeri

In Stage 5, there is a very thin envelope that stains only slightly with hematoxylin between the serosa and the chorion (Fig. 149). It is the serosal cuticle, secreted by the serosa. Midway through this stage, it thickens slightly and has two or three layers stained faintly with hematoxylin.

In Stage 6, the cuticle composed of multilayers, becomes about 10μm-thick. The serosal nuclei stain poorly with hematoxylin. In Stage 7, the cuticle is ca. 15μm-thick, and becomes 5 to 10μm-thick in Stage 9. The serosal nuclei indicate degenerating figures, though the amniotic nuclei are relatively intact in Stage 9. The cytoplasm of the serosal cells becomes granular and stains faintly with eosin. The serosal cuticle persists until just before hatching.

Panorpodes paradoxus, *Bittacus laevipes* and *Boreus westwoodi*

The serosal cuticle begins to be secreted late in Stage 2 in *Pd. paradoxus*, in Stage 3 in *B. laevipes*, and in Stage 4 in *Bo. westwoodi*. In *Pd. paradoxus*, the serosal cuticle reaches maximum thickness in Stage 6 (15μm), and in *B. laevipes* late in Stage 4 (about 7μm).

Discussion of serosal cuticle

Serosal cuticle has been observed in many insects, *e.g.*, in the collembolan *Tomocerus ishibashii* (UEMIYA and ANDO, 1987a, b), thysanuran *Pedetontus unimaculatus* (MACHIDA, 1981a ; MACHIDA and ANDO, 1985), Odonata (ANDO, 1962), orthopteran *Melanoplus differentialis* (SLIFER, 1937), plecopteran *Pteronarcys proteus* (MILLER, 1940), hemipteran *Pyrilla perpusilla* (SANDER, 1956), and the coleopteran *Lytta viridana* (CHURCH and REMPEL, 1971), etc.

As a rule, it is thought that the serosal cuticle together with the chorion protects the egg. In some Odonata (ANDO, 1962), *Tomocerus* (UEMIYA and ANDO, 1987a, b) and *Pedetontus* (MACHIDA, 1981a, b ; MACHIDA and ANDO, 1985), the serosal cuticle becomes a protective membrane because the chorion in these insects ruptures during embryonic development. The

chorion in *P. pryeri* and *Pd. paradoxus* does not rupture, but is so thin and frail that the serosal cuticle in these species seems to have an important role in the protection of the egg contents.

In *B. laevipes*, however, the chorion is very thick (25μm in thickness) and the serosal cuticle is only a half as thick as in *P. pryeri* and *Pd. paradoxus*.

VIII. Other parnorpid species observed

I have in addition observed the embryogenesis of three panorpid species other than *Panorpa pryeri*: *P. japonica*, *P. nipponensis* and *P. helena*. The general features of the egg and embryogenesis of these panorpids closely resemble those of *P. pryeri*, and there is no need to repeat the details. We may accept the embryogenesis of *P. pryeri* as being representative of the Panorpidae.

General Discussion and Conclusion

The results obtained in the present study have been discussed for each of the structures or systems. Here, the subjects considered signgficant in the comparative embyrology or morphology of insects will be examined further.

In this section, the term Mecoptera will be used to represent *P. pryeri*, *Pd. paradoxus*, *B. laevipes* and *Bo. westwoodi*, and these species will be mentioned by the names of the families to which they belong for convenience of description when no confusion will result.

I. Homology in larval abdominal prolegs and thoracic legs

Accoring to BERLESE's theory, the first-instar larvae of hemimetabolous insects have only thoracic legs (oligopod), but when the holometabolous larvae hatch they have legs on the thoracic and abdominal segments (polypod). HINTNON (1958) considered that the larval abdominal prolegs of the panorpoid insects (Mecoptera, Trichoptera, Lepidoptera, Siphonaptera and Diptera) are secondary, adaptive structures and not homologous with the thoracic legs. MATSUDA (1976) gave attention to the position of abdominal prolegs in insect embryos and suggested that the larval abdominal prolegs are serially homologous with the thoracic legs.

I shall attempt to homologize the larval abdominal prolegs and the thoracic ones in the Mecoptera by comparison with other holometabolous insects which have larval abdominal prolegs. I will recognize abdominal prolegs as homologous with thoracic legs when the following two criteria are satisfied: The first is that in each segment paired abdominal prolegs must be arranged in a row with the thoracic legs. The second is that the abdominal legs must develop histologically in the same manner as the thoracic legs, at least in their early and middle development.

Abdominal prolegs or appendages are found in larvae of some holometabolous insects, *e.*

g., the Megaloptera, Lepidoptera, Tenthredinidae of Hymenoptera and Blephariceridae of Diptera.

In the larvae of the lepidopteran *Diacrisia* (JOHANNSEN, 1929), hymenopteran *Athalia* (FAROOQI, 1963) and dipteran *Neocurupira* (CRAIG, 1967), the abdominal prolegs are arranged in a row with the thoracic legs, and the embryological formation of abdominal prolegs is similar to that of the thoracic legs. Since the larval abdominal prolegs of these insects satisfy the above two criteria, I recognize them homologous with the thoracic legs.

In the coleopteran *Lytta* (CHURCH and REMPEL, 1971) and neuropteran *Chrysopa* (BOCK, 1939), abdominal appendages occur only on the first abdominal segment, but disappear later during embryonic stage. These appendages, *i.e.*, pleuropodia, are thought to be homologous to the thoracic legs. KOBAYASHI and ANDO (1990) concluded that the abdominal appendage-like swellings of the embryos are homologous with thoracic legs in the trichopteran *Nemotaulius admorsus*. In his study of the megalopteran *Protohermes grandis*, MIYAKAWA (1979) said of the abdominal appendages of the embryo: "The abdominal segments have two pairs of swellings. The median ones immediately lateral to the ventral nerve are small and homotopous with the pleuropodia. These median swellings later disappear. The lateral swellings are conspicuous structures located between the median swellings and tracheal pits, and they later give rise to the abdominal filaments or tracheal gills." In this species, median swellings seem to be homologous with thoracic legs judging from his statement that the median swellings are homotopous with the pleuropodia, because the pleuropodia are generally thought to be homologous with thoracic legs (WHEELER, 1890; HUSSEY, 1926; MACHIDA, 1981a). Consequently, the lateral swellings or the abdominal filaments of *Protohermes* are not considered to be homologous with the thoracic legs, and the same is true of the megalopteran *Sialis* (ANDO *et al.*, 1985).

In the mecopterans *P. pryeri* and *B. laevipes*, on the ventral surface of the embryonic abdomen there appear two pairs of processes, *i.e.*, median and lateral processes, on the first eight abdominal segments (Figs. 40, 72). The lateral processes are located in the positions comparable to those of thoracic legs, and persist in the first-instar larva. The median processes are located below each of the abdominal ganglia, and become abdominal prolegs. The formation of abdominal prolegs is considerably different from that of the thoracic legs (see Chapter VII, 4, ii, 'Formation of abdominal prolegs' in Observations).

Furthermore, in the first-instar larva of *P. pryeri*, there are lateral abdominal muscles connecting with the lateral processes, and they seem to be homologous with the lateral muscles associated with thoracic legs (Fig. 134, arrows). From these observations, I conclude that the median processes (median larval abdominal legs) of *P. pryeri* and *B. laevipes* are not homologous with the thoracic legs.

In conclusion, in many cases, the larval abdominal prolegs of holometabolous insects are thought to be homologous with the thoracic legs, as mentioned by MATSUDA (1976). In some cases, *e.g.*, in *Protohermes* (MIYAKAWA, 1979), *Panorpa* and *Bittacus*, however, the median larval abdominal legs or filaments seem to be not homologous with the thoracic legs.

II. Interpretation of metamerism in terminal region of abdomen

Concerning the metamerism in terminal region of the insect abdomen, HEYMONS (1895, 1896), in his embryological works on hemimetabolous insects, regarded the telson as a twelfth abdominal segment that consists of the subanal and supraanal lobes, despite the lack of segmental ganglia, coelomic sacs and appendages. On the other hand, SNODGRASS (1935) argued that "In most insects no trace of a twelfth segment is to be found, and the periproct (=telson) must be supposed to be represented, it at all, by a circumanal membrane at the end of the eleventh segment." MATSUDA (1976), however, from detailed examination of the studies concerned with segmentation of the insect abdomen, concluded that, supporting HEYMONS' view, the abdomen consists of eleven segments and a telson (=the twelfth abdominal segment) in the lower insects. MACHIDA (1981a, b), from his embryological study of the thysanuran *Pedetontus*, supported HEYMONS' (1895, 1896) and MATSUDA's (1976) views.

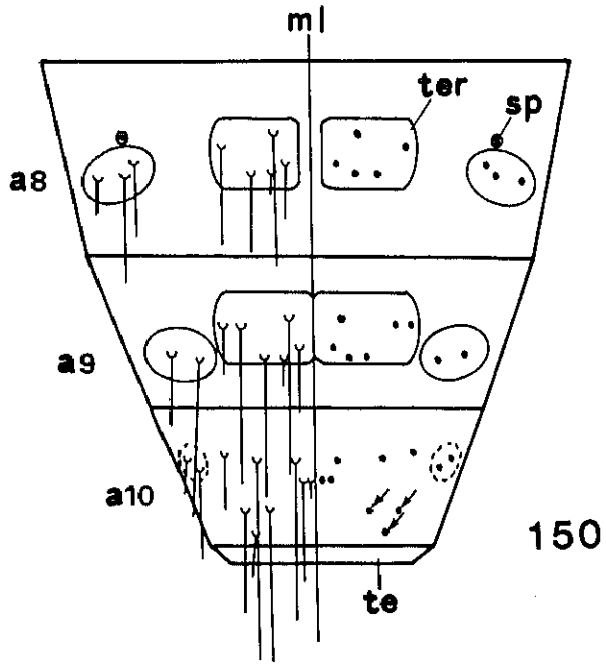
As mentioned above, it seems to be generally accepted that the telson is not derived from the eleventh abdominal segment in the Thysanura and Hemimetabola. The telson of *P. pryeri* also seems to be formed from the posterior end of the proctodaeum (see Chapter VII, 4, iii, 'Formation of anal legs and telson' in Observations), not from the tenth abdominal segment.

In *P. pryeri*, the problem is whether or not the rudimental eleventh abdominal segment exists in the posterior part of the tenth abdominal segment. In this species, an eleventh abdominal ganglion and coelomic sacs never appear during embryonic stage (see Chapter VII, 1, iii, a, 'Ventral nerve cord' and 2, i, 'Segmentation of inner layer, formation of coelomic sacs, and differentiation of splanchnic mesoderm' in Observations). I, however, can not deny the possibility that the rudimental eleventh abdominal segment exists in the posterior end of the tenth abdominal segment for the following reasons:

In the first-instar larva of *P. pryeri*, the caetotaxy of the first nine abdominal segments is fundamentally the same. That of the tenth abdominal segment is also similar to the other abdominal segments, except for an additional three pairs of spines which occur on the posterior margin of the tergum (Fig. 12, arrows), as in *P. japonica* (MIYAKE, 1912), *P. nuptialis* (BYERS, 1963), *P. falsa* and ten species of *Panorpa* and *Neopanorpa* (YIE, 1951). This group of spines is never found on the first nine abdominal segments, and seems to be regarded as belonging to the eleventh abdominal segment. Furthermore, in *P. communis* (ROTTMAR, 1966) the imaginal disk of the pupal eleventh abdominal segment is formed in the posterior part of the prepupal tenth abdominal segment.

Judging from these data, the tenth abdominal segment of *P. pryeri* is thought to contain the rudimental eleventh abdominal segment in its posterior part. Consequently, I consider that the abdomen of *P. pryeri* consists of ten segments and a telson, and that the rudimental eleventh abdominal segment is hidden in the tenth abdominal segment.

In *Pd. paradoxus*, ISSIKI (1959) reported that the abdomen of the first-instar larva apparently consists of eleven segments, and on the eleventh abdominal tergum there are three pairs of



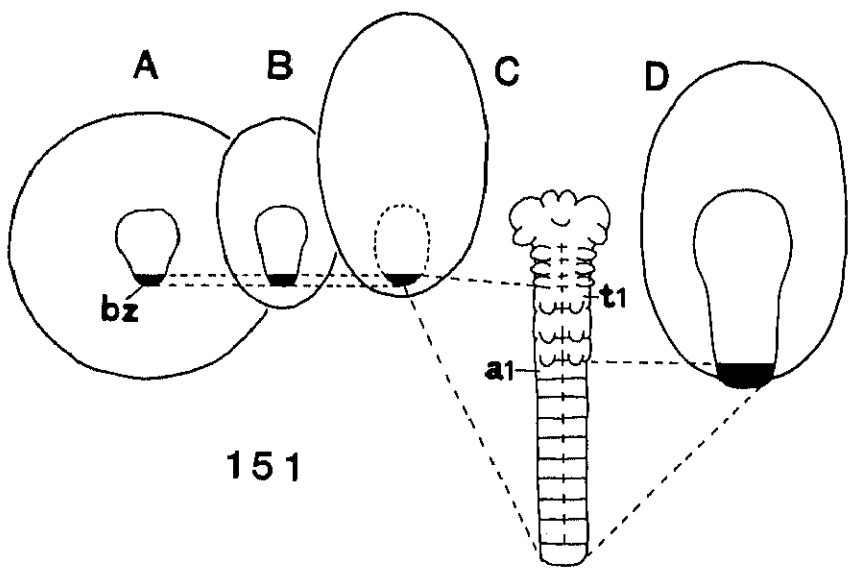


Fig. 150. Diagram showing main caetotaxy of eighth to tenth abdominal segments of first-instar larva of *Pd. paradoxus*. Arrow, spine belonging to vestigial abdominal segment.

Fig. 151. Diagrams showing type of germ band of *B. laevipes* (A), *Bo. westwoodi* (B) *Pd. paradoxus* (C) and *P. pryeri* (D).

spines. I, however, failed to find any distinguishable intersegmental suture at the posterior part of the tenth abdominal segment of the first-instar larvae. Moreover, in the embryo of this species I also could not find any ganglion or coelomic sacs of an eleventh abdominal segment (Fig. 124).

The abdomen of the first-instar larva of *Pd. paradoxus* is composed of ten segments and a telson, and the vestigial eleventh abdominal segment is possibly involved in the posterior part of the tenth abdominal segment, as in *P. pryeri*, since there are three pairs of spines on the posterior margin of the tenth abdominal tergum (Fig. 150, arrows).

MATSUDA (1976) regarded the larval anal legs of *Panorpa* as apomorphically modified cerci of the eleventh abdominal segment. The anal legs of *P. pryeri* are, however, derived directly from the telson (see Chapter VII, 4, iii, 'Formation of anal legs and telson' in Observations); hence, I believe that in *P. pryeri* these structures are not cerci and not derived from the eleventh abdominal segment.

III. Phylogenetic consideration of mecopteran families, from embryological aspects

Heretofore, several embryologists have attempted to examine the phylogenetic relationships of insects by means of comparative embryology. According to JOHANNSEN and BUTT (1941), in the hemimetabolous and holometabolous insects, the types of formation of the germ band and the relation of its length to the form of the egg have no significance for phylogeny. SHAROV (1966) explained the phylogenetic relationship between the thysanuran and pterygote insects in the manner in which the embryonic envelope forms. KRZYSZTOFOWICZ (1966) studied embryogenesis in three families and 28 species of Coleoptera and pointed out the significance of the formation of the embryonic envelope and the presence of liquefied yolk between the amnion and serosa in the comparison of these families.

ANDO (1962) investigated the embryology of seven families of Odonata and divided them into two groups by the type of germ band in relation to the yolk, *i.e.*, the partially invaginated type (Cordulegasteridae and Libellulidae) and the invaginated type (the remaining families). He also showed that grouping the seven families on the basis of comparative embryological data agreed with the taxonomic groups of the Odonata. COBBEN (1968, 1978) studied the eggs, architecture of the egg-shell, gross embryology and eclosion of the Heteroptera in detail, and used the types of blastokinesis of embryos as a character for phylogenetic studies of ten heteropteran superfamilies. HAGA (1985) investigated the oögenesis and embryogenesis of the thysanopteran *Bactrothrips brevitubus* and compared them with those of other paraneopteran species in discussing the phylogeny of the paraneopteran orders.

Recently, the relationship of the Trichoptera and lower Lepidoptera was examined with regard to the formation of the germ rudiment, and it was suggested that there is a close affinity between these orders (ANDO and TANAKA, 1976, 1980; ANDO and KOBAYASHI, 1978; KOBAYASHI and ANDO, 1981, 1982, 1987, 1988; AKAIKE *et al.*, 1982).

From the results quoted above, it may be seen that there are cases in which the data from

comparative embryology provided important criteria for study of insect phylogeny, especially when one investigates the embryology of insects situated at a point of phylogenetic divergence, and compares the results obtained from the early and middle stages of embryonic development.

Therefore, I attempt to examine the relationships of the mecopteran families by means of comparative embryology.

Character 1: Formation of mesoderm

As discussed in Chapter VI, in lower hemimetabolous insects, the mesoderm (inner layer) formation is by cell proliferation along the median line of the embryo. On the other hand, there are only a few holometabolous insects in which the mesoderm is formed in this manner, *i.e.*, the proliferating type. In almost all Holometabola, the mesoderm is formed by the invagination of the middle plate along the median line, *i.e.*, the invaginating type.

In the Mecoptera, formation of the mesoderm in the Bittacidae is of the proliferating type, and that of the Boreidae, Panorpodidae and Panorpidae is of the invaginating type. So the Bittacidae are likely to be more primitive than the other three families in this respect. This character is a synapomorphy of the Boreiae, Panorpodidae and Panorpidae.

Character 2: Oöplasm

In general there is a tendency for eggs of the lower Pteryogota, such as in the Hemimetabola, to be poor in oöplasm (=periplasm and reticuloplasm) as compared with the amount of yolk, and, on the contrary, for the oöplasm to be rich in the eggs of higher pterygotes or Holometabola (KRAUSE, 1939, 1961; ANDERSON, 1972; ANDO, 1981).

In the Mecoptera, the eggs of Bittacidae are poor in oöplasm compared with the amount of yolk. One may think that this characteristic of the eggs of the Bittacidae is an effect of egg hibernation. ANDO (1973), however, denied this supposition, citing the case of hibernatant and nonhibernatant eggs in the Odonata, which exhibit almost the same characteristics concerning the relative amount of oöplasm and yolk. Consequently, the nature of the eggs of Bittacidae seems to be not apomorphic, but plesiomorphic, as suggested by ANDO (1973). The nature of bittacid eggs is likely to be more primitive than that of eggs of the Boreidae, Panorpodidae and Panorpidae.

Character 3: Aggregation of primary yolk cell

In the Panorpidae and Panorpodidae, primary yolk cells form an aggregation deeply in the yolk (Chapter III, 'Early embryonic development' in Observations). This peculiar phenomenon may be recognized as a synapomorphic character of these two families.

The additional increase in volume of periplasm is also characteristic of these two families (see Chapter I, 'Oviposition and organization of newly laid eggs' in Observations).

Character 4: Presence of polar granule

The apparent polar granules are only found in the eggs of some holometabolan orders, that is, Diptera, Coleoptera and Hymenoptera as shown by ANDO (1973). Therefore the presence of the polar granule is regarded as a highly specialized or apomorphic character in the Panorpidae.

Character 5 : Type of germ band

KRAUSE (1939) classified the germ bands of insects into three types. The first type of germ band includes only the future protocephalic and gnathal regions, at first, and the future thoracic and abdominal regions develop from the posterior end of the germ band as it elongates. He termed it the short germ type. The germ band that includes all of the future protocephalic, gnathal, thoracic and abdominal regions from the beginning he termed the long germ type. The germ band that includes the future protocephalic, gnathal and thoracic regions at first, is termed the semilong germ type. He found that the short germ type is often found in hemimetabolans, *e.g.*, Orthoptera, and the long germ type is found in higher Holometabola, *e.g.*, Diptera and Hymenoptera. The semilong germ type is found in some Coleoptera.

In the Mecoptera, the germ band of the Panorpidae is regarded as of the semilong germ type, and those of the other three families are regarded as the short germ type (Fig. 151), hence the germ band of the Panorpidae is thought to be more derived than that of the other families.

Character 6 : Formation of germ band or germ rudiment

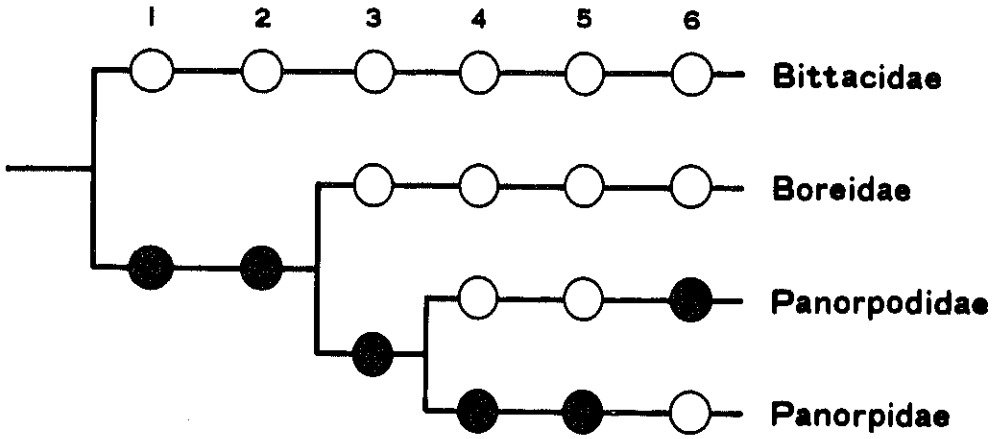
In several species of the coleopteran Chrysomelidae, ZAKHVATKIN (1968) observed that the eggs of *Leptinotarsa decemlineata* and *Phyllodecta vitellinae* are rich in yolk and poor in oöplasm, and the germ bands are formed on the surface of the eggs, that is, they have a superficial type of germ band. On the contrary, the eggs of *Galerucella lineola* and *Chalcoides aurata* are rich in oöplasm and not so rich in yolk, and their germ bands sink into the yolk at first, that is, they are of the invaginated type. Therefore, he concluded that the relative amount of yolk and oöklasm determines the type of early embryogenesis.

In the Mecoptera, the germ band formation in the Bittacidae, Boreidae and Panorpidae is of the superficial type, and the germ band or rudiment formation in the Panorpodidae is of the immersed type, in which the invaginated germ band leaves the egg periphery and sinks completely into the yolk. Because of the considerable difference between the Panorpodidae and Panorpidae in formation of the germ band, although they have similar oöplasm-rich eggs, I think that the affinity of these two families is not very close.

Phylogenetic tree of the Mecoptera based on embryological characters

Finally, the foregoing conclusions from the present study are summed up into a phylogenetic tree in the next figure.

This phylogenetic tree of the four mecopteran families based on embryological characters



Phylogenetic tree showing relationships among the mecopteran families. Arabic numerals refer to character enumeration in text. Open circle indicates plesiomorphic states, and filled circles indicate apomorphic character states.

does not agree with that concluded by PENNY (1975). He considered that the Boreidae and Panorpodidae branched after their common ancestor branched from the ancestor of the Panorpidae, because the Boreidae and Panorpodidae are the only mecopteran families that have scarabaeiform larvae. According to the embryological data, also I can not faver HINTON's proposal (1958) that the Boreidae should be separated from the mecopteran families and established as a new order, Neomecoptera, because of the morphological difference in the larval cranium, maxilla and labium.

The phylogenetic tree proposed by me agrees with that by MICKOLEIT (1978) based on comparative morphology of exoskeletal characters of the female genitalia, and it also agrees with that by WILLMANN (1981, 1987) based on characters of the male genitalia, fossil record, etc.

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* indirect citation

Abbreviations Used in the Figures

a, amnion
a1-10, first to tenth abdominal segments
ac, amniotic cavity
af, amniotic fold
ag, antennal ganglion
ag1-10, first to tenth abdominal ganglia
al, abdominal proleg
amr, anterior midgut rudiment
an, anus
ang, anal gland
anl, anal leg
ant, anterior tentorial arm
apc, amnioproctodaeal cavity
ar, aorta
at, antenna
atm, antennal mesoderm
b, brain
b1-3, protocephalic bulges of 1 to 3
bc, blood cell
bdc, blastoderm cell
bm, basement membrane
bz, budding zone
c, cuticle
ca, corpus allatum
cb, cardioblast
cc, crystalline cone
cca, corpus cardiacum
ceb, central body of tentorium
ch, chorion
cor, cornea
cos, common duct of salivary gland
cox, coxopodite
cp, cytoplasmic process
cr, cytoplasmic reticulum or reticuloplasm
cs, coelomic sac
cse, columnar serosa
ct, coxa of thoracic leg
ctr, commissure of tritocerebrum
da, developing aorta
dc, deutocerebrum
dd, dorsal diaphragm
dm, distribution of mesodermal cells
dp, developing perineurium
dpr, dorsal process
dv, duodenal valve
e, eye
ea, embryonic area
ec, ectoderm
eea, extraembryonic area
ema, mandibular extensor apodeme
en, endochorion
es, epineural sinus
est, evagination of stomatogastric nervous system
et, egg tooth
fb, fat body
fg, frontal ganglion
fm, first maturation division of female pronucleus
fma, mandibular flexor apodeme
gc, germ cell
gd, germ disk
gi, ganglion of intercalary segment
gr, germ rudiment
gri, genital ridge
gs, granular substance
h, heart
hg, hypocerebral ganglion
ia, inner amnion
ic, intercalary segment
iep, inner (or anterior) end of proctodaeum
il, inner layer or mesoderm
ip, inner periplasm
isg, invagination of salivary gland
l1-3, lobi 1 to 3 of protocerebrum
l3', lobus 3' of protocerebrum

la, labral apodeme
lg, lamina ganglionaris
li, labium
lig, labial ganglion
lis, labial segment
lp, lateral plate
lpa, lateral process of abdominal segment
lr, labrum
ls, lateral spread of amnion
mc, mesodermal cell
md, mandible
mdc, median cord
mdg, mandibular ganglion
mdl, mediodorsal line
mds, mandibular segment
me, midgut epithelium
med, medulla
ml, median line
mpl, middle plate
mt, Malpighian tubule
mu, muscle
mv, microvilli
mvl, medioventral line
mx, maxilla
mxa, maxillary apodeme
mxp, maxillary palp
mxg, maxillary ganglion
mxs, maxillary segment
nb, neuroblast
ng, neural groove
np, neuropile
oa, opening of amniotic fold
oam, outer amnion
oe, oesophagus
oec, oenocyte
op, optic plate
pb, polar body
pc, protocorm
pcc, pericardial cell
pce, protocerebrum
pcl, protocephalon or protocephalic lobe
pd, proctodaeum
pe, perineurium
pg, primitive groove
pl, periplasm
pmr, posterior midgut rudiment
pog, polar granule
pp, primary pigment cell
pt, posterior tentorial arm
pv, pyloric valve
pw, proctodaeal wall
ra, rudimental amnion
rc, regenerative cell
rd, rudimental dorsal process
rdd, rudimental dorsal diaphragm
re, rudimental eye
rec, retinular cell
rh, rhabdom
ria, round cell of inner amnion
rmt, rudimental Malpighian tubule
rn, recurrent nerve
rng, rudimental neural groove
rpf, rudimental postretinal fiber
rs, rudimental serosa
s, serosa
sb, suboesophageal body
sc, serosal cuticle
sd, stomodaeum
sec, Semper's cell
sg, salivary gland
sm, second maturation division of female pronucleus
smu, splanchnic muscle
sog, suboesophageal ganglion
som, somatic mesoderm
sp, spiracle
spf, small process on frons
spm, splanchnic mesoderm
stn, stomatogastric nervous system
sz, spermatozoon

t, tentorium
t1-3, first to third thoracic segments
tc, tritocerebrum
te, telson
ter, tergum
tl1-3, first to third thoracic legs
tp, telopodite
tr, trachea
tri, tracheal invagination
v, vacuole
y, yolk or yolk granule
yc, yolk cell (=yolk nucleus)