Acta Zoologica (Stockholm) **81**: 285–292 (October 2000)

Circulatory organs of abdominal appendages in primitive insects (Hexapoda: Archaeognatha, Zygentoma and Ephemeroptera)

Barbara-Amina Gereben-Krenn and Günther Pass

Department of Evolutionary Biology
Institute of Zoology
University of Vienna
Althanstraße 14
A–1090 Vienna
Austria

Keywords:
morphology, dorsal vessel, heart, cercus, terminal filament, evolution, insects, Hexapoda

Accepted for publication:
11 April 2000

Abstract

Gereben-Krenn, B.-A. and G. Pass 2000. Circulatory organs of abdominal appendages in primitive insects (Hexapoda: Archaeognatha, Zygentoma and Ephemeroptera). — *Acta Zoologica* (Stockholm) **81**: 285–292

The abdominal circulatory organs and the haemolymph supply of the terminal filament and the cerci were investigated in a total of nine species (serial semithin sections, TEM, *in vivo* observations).

In all investigated species, the dorsal vessel features a bidirectional flow. In the 10th abdominal segment, there is an intracardiac valve preventing flow in the anterior direction. The posterior portion of the dorsal vessel differs significantly from the anterior portion in design and wall structure. Pumping actions and frequencies do not correspond with each other in the two portions.

Vessels supply haemolymph to the long terminal appendages. The terminal filament vessel is connected to the dorsal vessel, the two cercal vessels originate from a transverse septum at the base of the terminal filament. All taxa show the same flow pattern through the caudal appendages. Ephemeroptera have a unique spherical body at the posterior end of the dorsal vessel functioning as a backflow valve.

The results indicate that the plesiomorphic state of the circulatory organs in the abdomen of primitive insects differ distinctly from that of higher insects which serve as bases for generalized schemes common to entomological textbooks.

Barbara-Amina Gereben-Krenn, Department of Evolutionary Biology, Institute of Zoology, University of Vienna, Althanstraße 14, A–1090 Vienna, Austria. E-mail: Barbara-Amina.Gereben@univie.ac.at

Introduction

In the open circulatory system of insects, the dorsal vessel handles the bulk of haemolymph transport within the general body cavity. However, in elongate body appendages, haemolymph exchange relies on accessory circulatory organs. In hexapods, these structures feature an amazing diversity in functional morphology: they may be plain guiding structures, such as vessels, diaphragms or elastic tracheae and at a more complex level accesssory pulsatile organs acting as autonomous hearts and functioning independently from the dorsal vessel (reviews: Jones 1977; Pass 1998; Wasserthal 1998).

As yet, the haemolymph supply of long abdominal appendages, such as the terminal filament and the cerci of primitive Hexapoda, has received little attention. In the dipluran *Campodea*, one vessel per cercus has been found linked to the dorsal vessel (Gereben-Krenn and Pass 1999). In Archaeognatha (Bär 1912; Barth 1963), Zygentoma (Rousset 1974) and Ephemeroptera (Meyer 1931), each cercus and the terminal filament house a vessel. According to these descriptions, the linkage mode of the vessels to the dorsal vessel is different among the various taxa. However, since some of these papers are not very detailed and contain inconsistencies, a thorough reinvestigation is indispensable.

The present study is a comparative investigation of the abdominal circulatory organs and the haemolymph supply of the long terminal appendages by serial semithin sections and TEM on representative species of Archaeognatha, Zygentoma and Ephemeroptera. *In vivo* observations further substantiate the anatomical data. This study is part of a series of papers dedicated to the reconstruction of circulatory organ evolution in arthropods (Krenn and Pass 1994, 1995; Gereben-Krenn and Pass 1999; Pass 2000).

Materials and Methods

Animals

Specimens in different developmental stages of

Archaeognatha: *Machilis hrábei* Kratochvil, 1945

Zygentoma: *Lepisma saccharina* Linné, 1758, *Thermobia domestica* (Packard, 1873)

Ephemeroptera: *Epeorus sylvicola* (Pictet, 1865); *Ephemera danica* Müller, 1764; *Cloeon* sp.; *Rhithrogena* sp.; Heptageniidae sp.

All specimens were collected in the surroundings of Vienna, Austria.

Light microscopy

Specimens were fixed in Dubosq-Brasil mixture and stored in 70% ethanol. The isolated body parts were dehydrated with ethanol, using acetone or propylene oxide as an intermedium. They were then embedded in ERL-4206 epoxy resin (Spurr medium) under vacuum impregnation. Serial semithin sections (1 μm) were made with glass or diamond knives (Diatome) and stained in a mixture of 1% azure II and 1% methylene blue in an aqueous 1% borax solution for approximately 30 s at 80 °C.

Transmission electron microscopy (TEM)

Specimens were dissected and fixed in a 2% paraformaldehyde/2.5% glutaraldehyde mixture and, subsequently, in 1% osmium tetroxide. Both fixatives were buffered in 0.1 M sodium cacodylate (pH 7.4). Dehydration and embedding was performed as described for semithin sections. Ultrathin sections were stained in uranyl acetate (1 h, 40 °C) and lead citrate (80 s, 20 °C) in a LKB 2169 Ultrastainer and examined with a Zeiss EM 902.

In vivo *observations*

The observations on Ephemeroptera larvae were conducted with a stereomicroscope using heat-protecting filters. Animals were placed in an immersed chamber with continuously sustained water current. Specimens of *Thermobia domestica* were put on microscopical slides and, again, shielded by heat-protecting filters.

Results

The first chapter of this section gives an outline of the common characteristics of the terminal circulatory organs. The subsequent chapters offer comparisons and detailed descriptions in Archaeognatha, Zygentoma and Ephemeroptera.

General morphology

No fundamental differences exist between the circulatory system of immatures and imagines. The morphological characters of the terminal circulatory organs of the investigated taxa can be attributed to three complexes: (i) design of the posterior portion of the dorsal vessel. The posterior most ostia pair is located in the 9th abdominal segment – the haemolymph entering the dorsal vessel here is propelled towards the front and the rear. In the 10th abdominal segment, there is an intracardiac valve preventing flow in the anterior direction. Thus, the dorsal vessels of all investigated taxa feature bidirectional haemolymph flow. The structure of the posterior portion of the dorsal vessel in the 10th segment differs in the investigated taxa; (ii) separation of the abdominal appendage haemocoels from the body cavity. Transverse septa of connective tissue divide the cerci and the terminal filament from the body cavity in all investigated species. These septa are reinforced by muscle cells in certain taxa which are designated in the following as musculo-connective septa. The terminal filament septum completely seals off the haemocoel from the body cavity. In contrast, the cercal septa contain a small opening allowing haemolymph flow into the abdominal body cavity; (iii) circulatory structures within the terminal appendages. The cerci and terminal filament house one vessel each in all investigated species. The vessel wall does not contain any muscle cells and is hence not contractile. The terminal filament vessel is connected to the dorsal vessel. The cercal vessels, in contrast, are not linked with the dorsal vessel but originate at the transverse septum of the terminal filament. They pass through the transverse septa of the cerci and run to the tip of these appendages.

From the morphological data and *in vivo* observation of haemocyte movements, we conclude that the haemolymph flow route through the terminal appendages is uniform in all investigated taxa: from the dorsal vessel into the terminal filament vessel, from the terminal filament haemocoel into the cercal vessels and then through the cercal haemocoels back into the abdominal body cavity (Fig. 1A–D).

Archaeognatha

In *Machilis hrábei*, the posterior portion of the dorsal vessel differs greatly from the anterior portion: its outer diameter amounts to only one fifth the size of the anterior portion, whereas the muscle layer is about four times as strong. Muscle cells form distinct protuberances protruding deeply into the lumen and probably obliterate it. This posterior portion of *Machilis* is designated in the following as 'caudal tube'.

Transverse septa isolate the haemocoels of the terminal appendages from the body cavity. These septa consist of connective tissue only (thickness about 5–10 μm) and are not muscular. The septum of the terminal filament extends

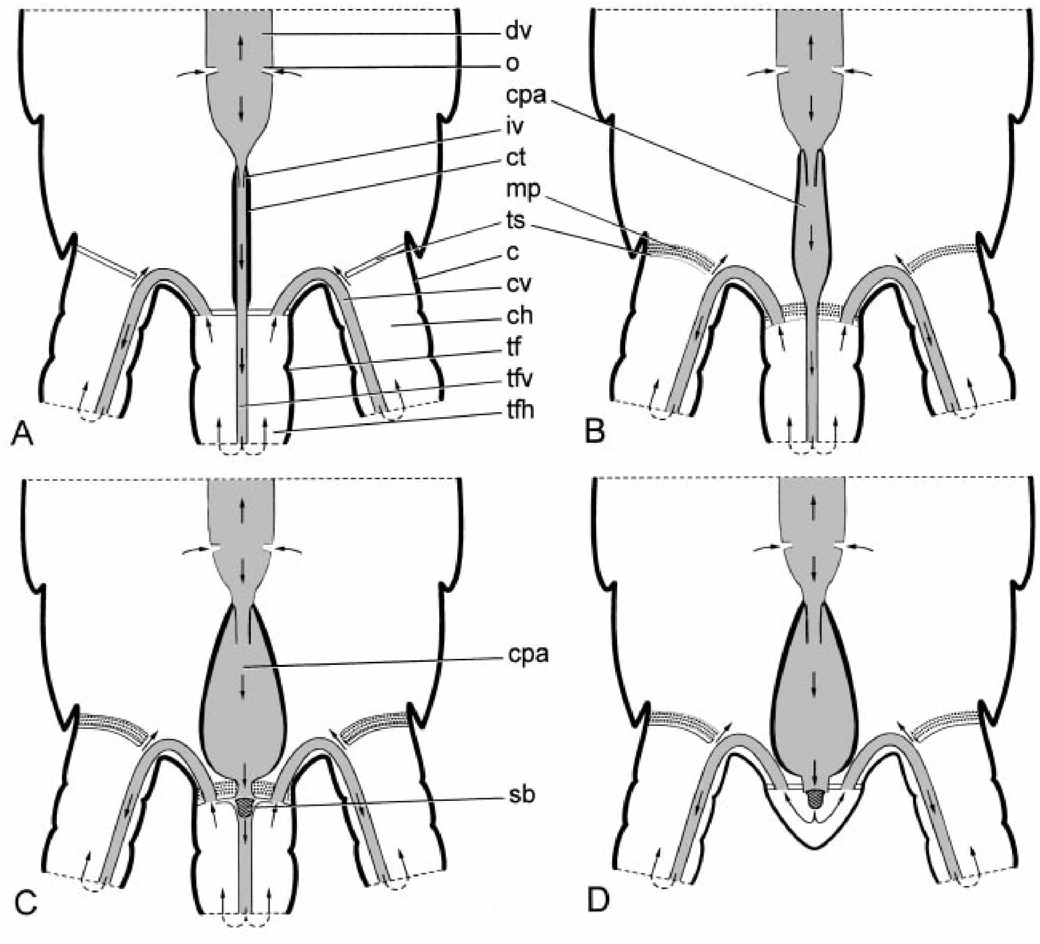

Fig. 1—Design of the abdominal circulatory organs in Archaeognatha (**A**), Zygentoma (**B**) and Ephemeroptera with developed terminal filament (**C**) and reduced terminal filament (**D**). Arrows indicate the haemolymph flow directions in the posterior portion of the dorsal vessel and through the terminal filament and the cerci. c, cercus, ch, cercus haemocoel; cpa, caudal pulsatile ampulla; ct, caudal tube; cv, cercal vessel; dv, dorsal vessel; iv, intracardiac valve; mp, musculo-connective septa; o, ostium; sb, spherical body; tf, terminal filament; tfh, terminal filament haemocoel; tfv, terminal filament vessel; ts, transverse septum.

along the entire appendage base (Fig. 2A); the septum of each cercus permits haemolymph exchange between cercal haemocoel and body cavity via a small lateral opening.

The terminal filament vessel connects to the dorsal vessel and opens into the haemocoel just short of the tip. The origin of the cercal vessels is ventro-lateral at the transverse septum of the terminal filament (Fig. 2B).

Zygentoma

In contrast to Archaeognatha, the posterior portion of the dorsal vessel in *Lepisma saccharina* and *Thermobia domestica* is slightly widened and the wall barely thicker than in the anterior portion. This structure is designated in the following as 'caudal pulsatile ampulla'.

The haemocoels of the terminal body appendages are separated from the body cavity by musculo-connective septa (Fig. 2C). The muscle cells attach at the appendage bases. Considering the location of the attachment sites and the arched course of these musculo-connective septa, it is difficult to fathom their function; in any case, they cannot play a role in appendage movement.

The dorsal vessel is continuous with the vessel of the terminal filament. The latter is somewhat dilated at its base. The cercal vessels originate at the transverse septum of the terminal filament and run into the cerci (Fig. 2D).

Unique to Zygentoma are numerous long flaps on the epidermis occurring in the terminal filament and cerci. They probably constitute valves preventing haemolymph backflow towards the rear.

In *Thermobia domestica*, haemolymph flow into the dorsal vessel through ostia of the 9th abdominal segment could be clearly observed, as well as the bidirectional haemolymph flow. There are bursts of activity of the caudal pulsatile

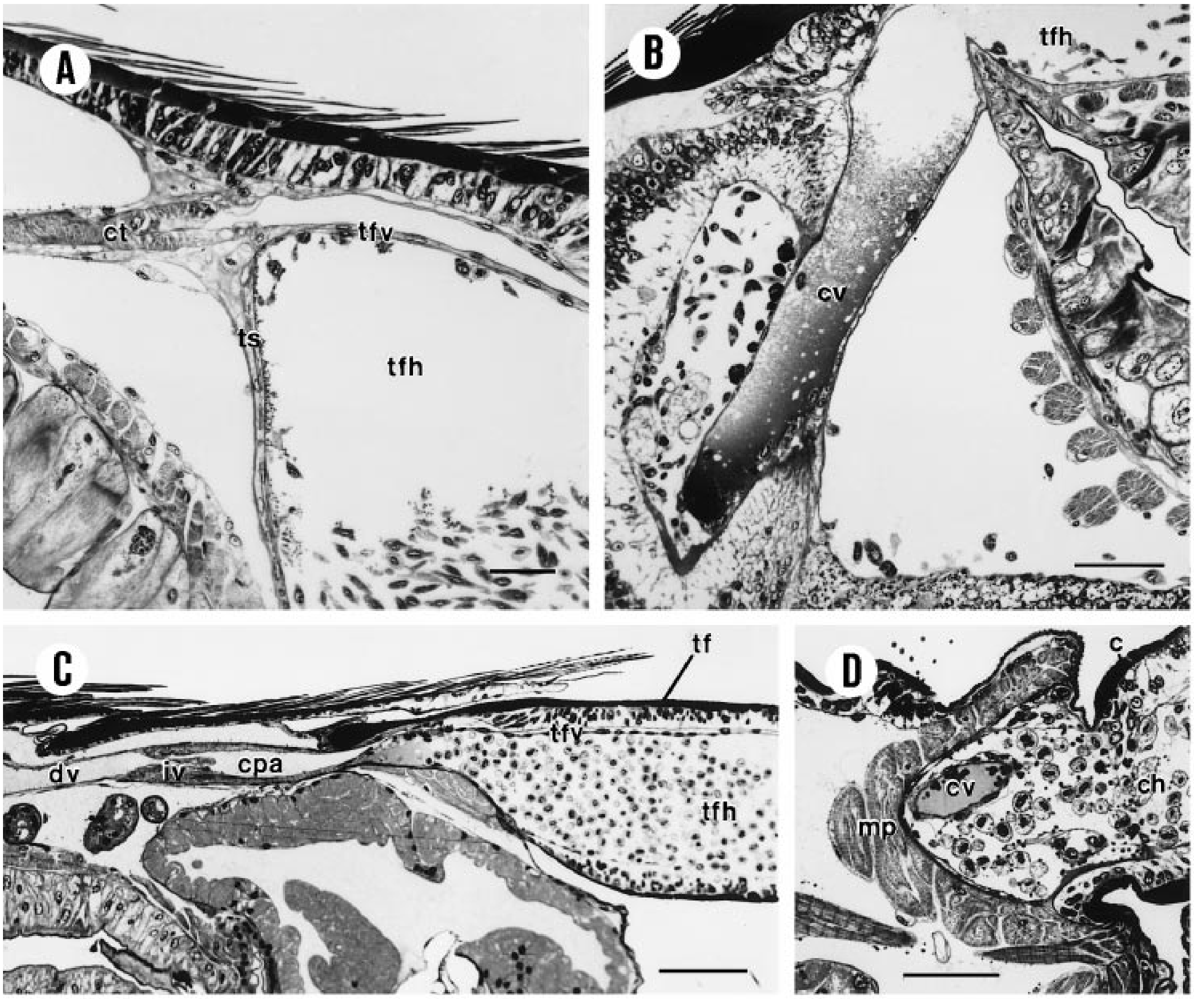

Fig. 2—**A**, *Machilis hrábei*, sagittal section through the proximal region of the terminal filament. The transverse septum (ts) separates the haemocoels of the abdomen and terminal filament (tfh). The caudal tube (ct) of the dorsal vessel continues into the terminal filament vessel (tfv). Scale bar: 50 µm. —**B**, *Machilis hrábei*, cross section through distal region of left side of the last abdominal segment. The haemocoel of the terminal filament (tfh) is connected to the cercal vessel (cv). Scale bar: 50 µm. —**C**, *Thermobia domestica*, sagittal section through last abdominal segment and base of terminal filament (tf). The dorsal vessel (dv) continues into the caudal pulsatile ampulla (cpa) which is connected to the terminal filament vessel (tfv). The caudal pulsatile ampulla is characterized by a thick muscular wall and at its anterior end by the intracardiac valve (iv). The transverse musculo-connective septum separates the haemocoels of abdomen and terminal filament (tfh). Scale bar: 100 µm. —**D**, *Thermobia domestica*, horizontal section through last abdominal segment and the base of cercus (c). The transverse musculo-connective septum (mp) separates the haemocoels of the abdomen and the cercus (ch). A section of the cercal vessel (cv) is visible in the cercus. Scale bar: 50 µm.

ampulla and long periods of inactivity in-between. Furthermore, we were able to follow the haemolymph flow through the caudal appendages as described above.

Ephemeroptera

In all investigated larvae and imagines, the posterior portion of the dorsal vessel is distinctly different from the anterior one. Compared with Zygentoma, the modification of this portion of the dorsal vessel, the caudal pulsatile ampulla, is much more noteworthy. The pear-shaped ampulla has a conspicuous wall which is about 6–10 times thicker than the wall of the anterior portion of the dorsal vessel (Fig. 3A,B). Furthermore, the wall of the pulsatile ampulla consists of two layers. The inner two thirds of the wall are composed of circular muscle, the outer third of connective tissue including collagen fibers in a matrix of fine filaments (Fig. 3C).

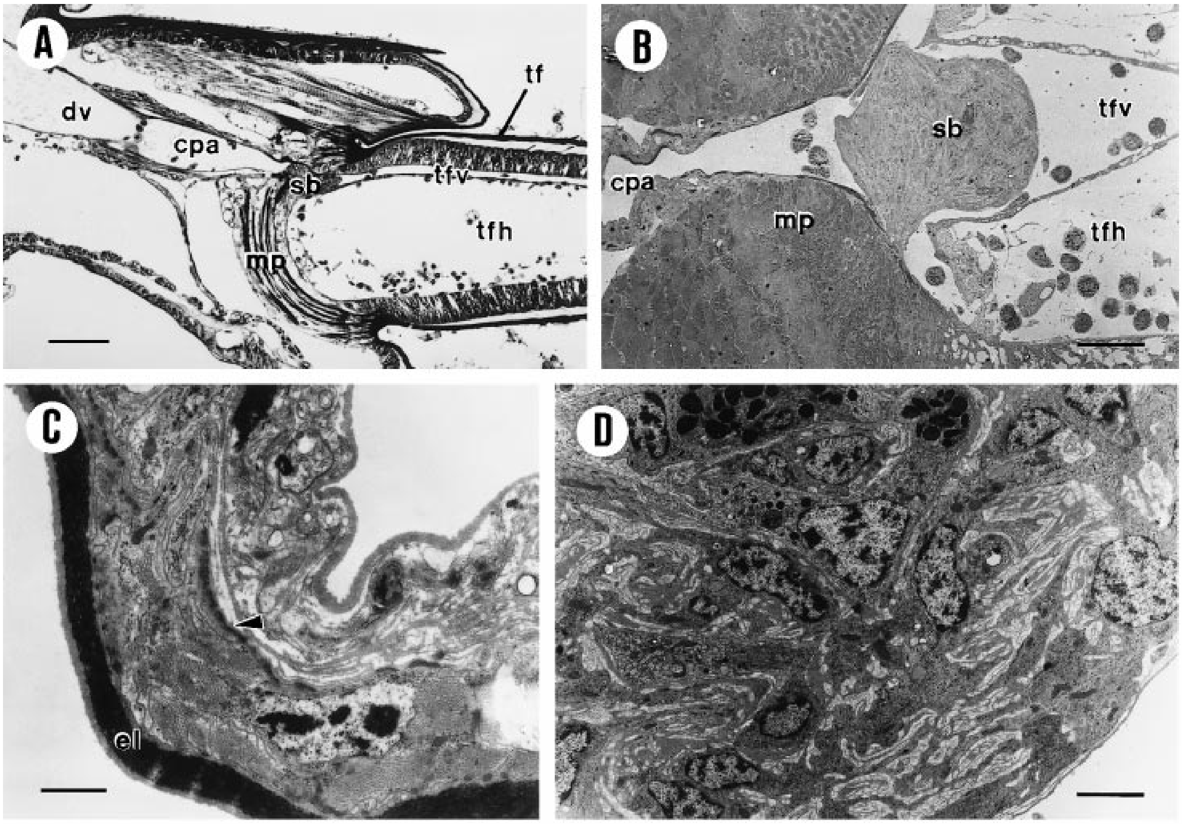

Fig. 3—**A**, *Rhithrogena* sp., sagittal section through last abdominal segment and base of the terminal filament (tf). At the transition zone between the caudal pulsatile ampulla (cpa) and the terminal filament vessel (tfv) is the spherical body (sb). The transverse musculo-connective septum (mp) separate the body cavity from the terminal filaments haemocoel (tfh); note the peculiar arched course of the musculo-connective septum. Scale bar: 50 μm. —**B**, *Ephemera* sp., horizontal TEM section of the transition zone between pulsatile ampulla and terminal filament vessel. The caudal pulsatile ampulla (cpa) opens into the funnel-shaped proximal region of the terminal filament vessel (tfv) where the spherical body (sb) is located. Scale bar: 30 μm. —**C**, *Ephemera* sp., horizontal TEM section through wall of caudal pulsatile ampulla. It consists of an inner muscular layer and an outer layer of connective tissue (boundary between the two layers marked by arrowhead). At the luminal side thick external lamina (el) of elastic filaments. Scale bar: 1,7 μm. —**D**, *Ephemera* sp., TEM section through part of the spherical body mass. At the upper end two cells with large lysosomes. The more translucent areas between the cells consist of extracellar matrix. Scale bar: 2,5 μm.

The separation of the appendage haemocoels resembles that in Zygentoma: transverse musculo-connective septa extend across the entire appendage base. The openings in the cercal septa are formed as valves. The caudal pulsatile ampulla protrudes into the musculo-connective septa of the terminal filament and opens into the funnel-shaped, enlarged terminal filament vessel. Peculiarly, this vessel has openings in both sides of the funnel region. A spherical body is located between the end of the caudal pulsatile ampulla and the base of the terminal filament vessel (Fig. 3A,B); it is attached to the transverse septum of the terminal filament. The spherical body does not constitute compact tissue; there are large intercellular spaces with an amorphic matrix of heterogeneous density (Fig. 3D). Its cells are especially rich in ribosomes which occur freely within the cytoplasm or associated with dilated ER cisternae. There are a few cells in addition to electron-dense bodies which probably are lysosomes.

The larvae of *Epeorus sylvicola* show a modified condition (Fig. 1D). Their terminal filament is largely reduced and the remaining epiproct contains no vessel. The epiproct haemocoel is separated from the body cavity by a thin transverse connective septum. The caudal pulsatile ampulla opens directly into this haemocoel. A spherical body exists also in this species and is located at the ampulla opening.

In vivo observation of larvae of *Rhithrogena* sp. showed that contraction of their caudal pulsatile ampulla is independent of that in the anterior dorsal vessel portion. Furthermore, the ampulla pulses intermittently and the anterior portion continuously. Upon contraction of the caudal pulsatile ampulla, haemolymph is propelled into all three appendage vessels simultaneously. Hemocytes issue out of the vessel tips and subsequently flow within the appendage haemocoels towards the abdomen.

Discussion

Functional morphology

Modifications of the posterior portion of the dorsal vessel. The modifications in the posterior portion of the dorsal vessel in the investigated taxa have been previously described to some extent, however, a lack of accuracy and detail in these studies has led to conflicting conclusions regarding the functioning (Bär 1912; Meyer 1931; Barth 1963; Rousset 1974). All these abdominal circulatory structures are to be seen in the context of ensuring haemolymph supply to the terminal body appendages and enabling the bidirectional flow in the dorsal vessel. This specific mode of flow could be demonstrated in all investigated taxa of Archaeognatha, Zygentoma and Ephemeroptera. It is enhanced by the intracardiac valve in the dorsal vessel which shunts haemolymph flow towards the rear and prevents flow towards the front.

Posterior to the valve region, there is the caudal muscular portion of the dorsal vessel which is characterized by an especially strong wall indicating increased contraction power. This portion differs in detail among the investigated taxa. In Archaeognatha, it forms a caudal tube. Its wall has specific formations on the luminal side which were noted by Barth (1963) and described as septa and protuberances. Barth's functional interpretation of these structures as a haemocyte filter seems far-fetched, although we cannot offer a more plausible hypothesis. In Zygentoma and Ephemeroptera, the caudal muscular portion forms a pulsatile ampulla. In the latter, it is an especially voluminous structure and was therefore called 'Schwanzkammer' by Meyer (1931). *In vivo* observations confirmed that this portion contracts intensively and with a frequency strikingly different from that of the anterior portion of the dorsal vessel. Moreover, there is no synchronization of the pumping actions of the two portions. In the mayfly *Cloeon dipterum*, Meyer (1931) recorded contraction frequencies of 60–70 beats.min^{-1} in the anterior portion of the dorsal vessel and, simultaneously, 15 beats per min in the caudal pulsatile ampulla.

Therefore, we can state that the caudal muscular portion is a histologically distinct and largely autonomous structure with a specific function. This holds especially true for the caudal pulsatile ampulla of Ephemeroptera which was interpreted as an accessory pulsatile organ (Jones 1977).

Hemocoel separation between abdomen and appendages. A common feature of all investigated species is the separation of the cerci and terminal filament haemocoels from the abdominal body cavity. In an unspecified machilid species, Barth (1963) described a single expansive transverse septum separating a narrow terminal sinus and resulting in the confluence of the cercal and terminal filament haemocoels (cf. his anatomical reconstruction Fig. 4). However, our investigation of *Machilis hrábei* identifies one transverse septum for each caudal appendage partitioning the haemocoels from each other. Consequently, these two incongruous descriptions lead to conflicting conclusions on the flow patterns in cerci and terminal filament (see below).

Musculo-connective septa are present at the appendage bases in Zygentoma and Ephemeroptera. Although, it is not possible to unambiguously compare the anatomical descriptions of previous investigators with our results, we assume that these musculo-connective septa were already noted (Zygentoma: Birket-Smith 1971, 1974; Rousset 1973; Ephemeroptera: Birket-Smith 1971). None of these authors, however, offer an explanation for the functional significance of these septa. The attachment sites of the muscle cells of these septa are located at the base of the terminal filament and the cerci, respectively. Therefore, they cannot contribute to movement of these appendages. Since they are peculiarly arched and not stretched as muscles usually are, it is likely that contraction of these musculo-connective septa causes haemolymph volume fluctuations at the appendage bases. Then, these structures must be regarded as accessory pumping structures forcing haemolymph out of the appendage haemocoels back into the abdominal body cavity. Unfortunately, the anatomical situation at the caudal appendage base is rather congested which also prevents *in vivo* observation of septum contraction and determining possible effect on haemolymph flow.

The mayfly *Epeorus sylvicola* has a diminutive terminal filament. Remarkably, the transverse septum at the filament base consists of connective tissue only. Obviously, the combination of the caudal pulsatile ampulla and the musculo-connective cercal septa suffices for transporting haemolymph through the diminutive terminal filament and the cerci.

Circulatory organs of the terminal filament and the cerci. All investigated species possess one vessel each in the terminal filament and the cerci. However, the anatomical origin of these vessels has been accounted for in very different ways in the literature which has led to various conclusions as to the circulation routes through the terminal appendages.

In machilids, Barth (1963) described the two cercal vessels as not linked with the dorsal vessel instead they issue into the body cavity through a valve-like opening at the transverse septum. As a consequence of these findings and the analysis of the anatomy of the transverse septum, Barth derived the following circulation pattern (cf. his Fig. 4, p. 376): haemolymph from the dorsal vessel flows up the terminal filament vessel to its tip, back within the terminal filament haemocoel and, eventually, directly into the cercus haemocoel. He supposed that haemolymph flows into the cercal vessel at the cercus tip and back into the abdominal body cavity in an afferent flow. Results of our investigations on *Machilis hrábei* contradict this scheme: the cercal vessels are, in fact, separate from the dorsal vessel but originate at the transverse septum of the terminal filament. Moreover, linkage with the abdominal body cavity is not effected via the cercal vessel but via a small opening in the transverse septa of the cerci. Based on these results, we obtain the following scheme of haemolymph circulation in the caudal appendages of machilids: haemolymph issues out of the dorsal vessel into the terminal filament vessel, flows back to the appendage base within the terminal filament haemocoel and enters the cercal vessels at the base. There is an efferent flow within the cercal vessels; haemolymph returns via the cercal haemocoel into the body cavity through a small opening in the transverse septum.

In *Thermobia domestica,* Rousset's (1974) description of circulation is in line with our own observations on this species and identical to our results in *Machilis hrábei* given above.

In Ephemeroptera, Meyer (1931) described both the cercal and the terminal filament vessels as emanating from the caudal pulsatile ampulla (cf. his schematic drawings fig. 2, p. 4 and fig. 4, p. 8). However, we did not find any linkage of the caudal pulsatile ampulla with the cercal vessels. The cercal vessels likewise originate at the transverse septum as described above for Archaeognatha and Zygentoma. We therefore surmise that Meyer erred in his otherwise very elaborate fig. 16 (p. 22) that the cercal vessels terminate at the cercus base and that they are not linked with the dorsal vessel: this may betray his own doubts on this subject.

All previous investigators have overlooked the spherical body at the funnel-shaped base of the terminal filament vessel. It is attached to the transverse musculo-connective septum and may function as a valve hindering haemolymph backflow from the terminal filament vessel. The cells of this spherical body contain remarkably high numbers of ribosomes indicating strong synthetic activity. This feature may suggest an additional endocrine function of this structure. Remarkably, a similar spherical cell mass was found in the heads of moths associated with their antennal circulatory organs (Selvatico 1887; Schneider and Kaissling 1959; Vondran and Wasserthal 1998). This structure, too, is regarded as a backflow valve with a possible endocrine function.

Evolutionary analysis

Obviously, the design of the posterior portion of the dorsal vessel of the basic hexapods must be seen in context with the respective types of abdominal appendages. Primitive groups (Diplura, Archaeognatha, Zygentoma, Ephemeroptera) with long filiform appendages consistently have a dorsal vessel with bidirectional haemolymph flow. Appendages are supplied with haemolymph via vessels which can be linked with the dorsal vessel in different ways. In Diplura, there is a pair of cerci whose vessels are connected with the dorsal vessel via a caudal chamber (Gereben-Krenn and Pass 1999). A long-terminal filament is present in Archaeognatha, Zygentoma and Ephemeroptera in addition to the cerci pair. As a consequence in these animals, haemolymph flows from the dorsal vessel into the terminal filament vessel, then it passes into the terminal filament haemocoel and subsequently enters the cercal vessels. An autapomorphy of Ephemeroptera is the spherical body located between the end of the caudal pulsatile ampulla and the funnel-shaped enlarged base of the terminal filament vessel.

In Archaeognatha, Zygentoma and Ephemeroptera, the caudal muscular portion within the 10th abdominal segment effects the haemolymph supply of the abdominal appendages. Whether this structure must be interpreted as part of the dorsal vessel or whether it represents an accessory pulsatile organ of its own cannot be ascertained at present; ontogenetic studies may shed light on this issue.

A plesiomorphic character of primitive Hexapoda is the presence of septa made up of connective tissue separating the abdominal appendages from the body cavity. In Zygentoma and Ephemeroptera, the musculo-connective septa are obviously capable of reinforcing the propulsion of the haemolymph stream. This character state can be considered as a synapomorphy of Dicondylia. In Zygentoma, we have indications for the evolutionary origin of the transverse septum muscles of the terminal filament: according Rousset (1973) it derives from a distinct skeletal muscle (M68).

In Odonata and Plecoptera, the dorsal vessel ends in the 9th abdominal segment and its rear end is closed (Odonata: Gereben-Krenn and Pass unpublished; Zawarzin 1911; Plecoptera: Schwermer 1914). In both groups, the dorsal vessel is not directly involved in the haemolymph supply of the caudal appendages. In Plecoptera, the cerci house one vessel each which are neither anatomically nor functionally connected to the dorsal vessel. The driving force for circulation in the cerci stems from an autonomous pulsatile organ located in each paraproct (Pass 1987). The evolutionary impetus for the appearance of these cercus-hearts may have been the reduction of the terminal filament. Coincidental to the reduction, the rear end of the dorsal vessel has probably become closed. In consequence the dorsal vessel was decoupled from the haemolymph supply of the cerci.

Cercal vessels are not known in any other pterygote insects, although, information on this point is sketchy. In Odonata (Pass unpublished) and Blattodea (Murray 1967), a connective tissue diaphragm exists within the cerci which partitions the haemocoel into a dorsal and a ventral sinus featuring countercurrent haemolymph streams.

In summary, we conclude that a posteriorly, open dorsal vessel with bidirectional haemolymph flow represents the plesiomorphic state in the circulatory organs of Hexapoda (Gereben-Krenn and Pass 1999). This character state is functionally associated with haemolymph supply to the long-terminal body appendages. The condition of a posteriorly closed dorsal vessel with unidirectional haemolymph flow typical of higher pterygotes must then be regarded a derived character state. It is probably a consequence of the loss of the terminal filament and the reduction or loss of the cerci.

Acknowledgements

We are very grateful to G. Rothe and T. Gatschnegg for their excellent technical assistance, T. Gatschnegg for his help with the illustrations, E. Christian and J. Rusek for determination of specimens, T. Gatschnegg, H.W. Krenn and N. Szucsich for critically reading the manuscript, and T. Micholitsch and J. Plant for linguistic assistance. This study was supported by the Austrian Science Foundation (Project P 10631-BIO).

References

Barth, R. 1963. Über das Zirkulationssystem einer Machilide (Thysanura). – *Memorias Do Instituto Oswaldo Cruz Rio de Janeiro* **61**: 371–399.

Bär, H. 1912. Beiträge zur Kenntnis der Thysanuren. – *Jenaische Zeitschrift für Naturwissenschaften* **48**: 1–92.

Birket-Smith, J. R. 1971. The abdominal morphology of *Povilla adusta* Navas (Polymitarcidae) and of Ephemeroptera in general. – *Entomologica Scandinavica* **2**: 139–160.

Birket-Smith, J. R. 1974. On the abdominal morphology of Thysanura (Archaeognatha and Thysanura s. str.). – *Entomologica Scandinavica Supplementum* **6**: 5–67.

Gereben-Krenn, B.-A. and Pass, G. 1999. Circulatory organs of Diplura (Hexapoda): the basic design in Hexapoda? – *International Journal of Insect Morphology and Embryology* **28**: 71–79.

Jones, J. C. 1977. *The Circulatory System of Insects*. Thomas, Springfield.

Krenn, H. W. and Pass, G. 1994. Morphological diversity and phylogenetic analysis of wing circulatory organs in insects, part I: Non-Holometabola. – *Zoology* **98**: 7–22.

Krenn, H. W. and Pass, G. 1995. Morphological diversity and phylogenetic analysis of wing circulatory organs in insects, part II: Holometabola. – *Zoology* **98**: 147–164.

Meyer, E. 1931. Über den Blutkreislauf der Ephemeriden. – *Zeitschrift für Morphologie und Ökologie der Tiere* **22**: 1–48.

Murray, J. A. 1967. Morphology of the cercus in *Blattella germanica* (Blattaria: Pseudomopinae). – *Annals of the Entomological Society of America* **60**: 10–16.

Pass, G. 1987. 'Cercus heart' in stoneflies – a new type of accessory circulatory organ in insects. – *Naturwissenschaften* **74**: 440–441.

Pass, G. 1998. Accessory pulsatile organs. In: F. Harrison and M. Locke (Eds): *Microscopic Anatomy of Invertebrates,* Vol. 11B *Insecta*, pp. 621–640. Wiley-Liss, New York.

Pass, G. 2000. Accessory pulsatile organs: Evolutionary innovations in insects. – *Annual Review of Entomology* **45**: 495–518.

Rousset, A. 1973. Squelette et musculature des régions génitales et postgénitales de *Thermobia domestica* (Packard). Comparaison avec la région génitale de *Nicoletia* sp. (Insecta: apterygota: Lepismatida). – *International Journal of Insect Morphology and Embryology* **2**: 55–80.

Rousset, A. 1974. Les différenciations postérieures du vaisseau dorsal de *Thermobia domestica* (Packard) (Insecta, Lepismatida). Anatomie et innervation. – *Comptes Rendus Hebdomadaires des Séances de l'Académie des Sciences* **278** (Série D): 2449–2452.

Schneider, D. and Kaissling, K.-E. 1959. Der Bau der Antenne des Seidenspinners *Bombyx mori* L. III. Das Bindegewebe und das Blutgefäß. – *Zoologisches Jahrbuch für Anatomie und Ontogenie der Tiere* 77: 111–132.

Schwermer, W. 1914. Beiträge zur Biologie und Anatomie von *Perla marginata* Scopoli. – *Zoologisches Jahrbuch für Anatomie und Ontogenie der Tiere* **37**: 267–312.

Selvatico, S. 1887. Die Aorta im Brustkasten und im Kopf von *Bombyx mori*. – *Zoologischer Anzeiger* **10**: 562–563.

Vondran, T. and Wasserthal, L. T. 1998. The cephalic arterial system in Lepidoptera. – *Proceedings of the* VI$^{\text{th}}$ *European Congress of Entomology, Ceske Budejovice*, 69.

Wasserthal, L. T. 1998. The open hemolymph system of Holometabola and its relation to the tracheal space. In: F. Harrison and M. Locke (Eds): *Microscopic Anatomy of Invertebrates,* Vol. 11B *Insecta*, pp. 583–620. Wiley-Liss, New York.

Zawarzin, A. 1911. Histologische Studien über Insekten. I. Das Herz der Aeschnalarve. – *Zeitschrift für Wissenschaftliche Zoologie* **97**: 481–510.