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# First Detection of Podocytes in the Circulatory System of Enigmatic Echiurids (Annelida: Thalassematidae)

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**Abstract**—The fine structure of echiurid blood vessels in the proboscis is known in detail, but the circulatory system of the trunk is still understood mainly at the level of general anatomy. The trunk circulatory system was studied in *Bonellia viridis* females, and specialized podocytes were found to form the walls of the ring vessel and the anterior part of the ventral vessel. Podocytes were for the first time described in the echiurid circulatory system. Podocytes of *B. viridis* displayed a typical cell architecture, which is known for other bilaterians. A podocyte consists of a cell body; primary processes; and pedicels, which extend from the primary processes and are interconnected via specialized slit diaphragms. The presence of podocytes indicates that the ventral and ring vessels act as ultrafiltration sites, where the plasma is filtered through the basal lamina into the body cavity.

**Keywords:** podocyte, morphology, circulatory system, ultrafiltration, metanephridia, anal sac

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Echiurids are a small group of marine benthic unsegmented worms [1, 2]. The taxonomic position of the group was revised repeatedly, but now echiurids are considered to unquestionably belong to Annelida [3]. In contrast to the majority of other annelids, echiurids lost metameric body segmentation and are completely devoid of dissepiments [1]. Their trunk thus contains a single coelom, which is not divided into compartments [1]. Other systems of organs were transformed with the loss of a metameric organization. For example, metameric metanephridia are absent in echiurids, and unusual anal sacs act as excretory organs. The anal sacs have multiple ciliated funnels and communicate with the posterior gut to form a single cloaca.

Most echiurids have a closed circulatory system, which has been studied at the level of general anatomy [4] and 3D reconstructed from serial sections [2]. An ascending dorsal vessel and a descending ventral vessel are recognized in the circulatory system of echiurids, like in other annelids [5]. However, the dorsal blood vessel occurs only in the anterior part of the echiurid trunk and is absent from the middle and posterior parts, being replaced with a sinus surrounding the

digestive tube [1]. There are three blood vessels in the proboscis. One is axial and is an extension of the dorsal vessel; two lateral vessels fuse together in the trunk to form the ventral blood vessel. The structure of the blood vessels of the proboscis is well understood [6, 7], while the fine structure of the trunk circulatory system remains almost unknown. New data on podocyte location in the circulatory system and the detailed podocyte structure will shed light on the function of the metanephridial system in echiurids and expand our knowledge of the physiology of these unusual annelids.

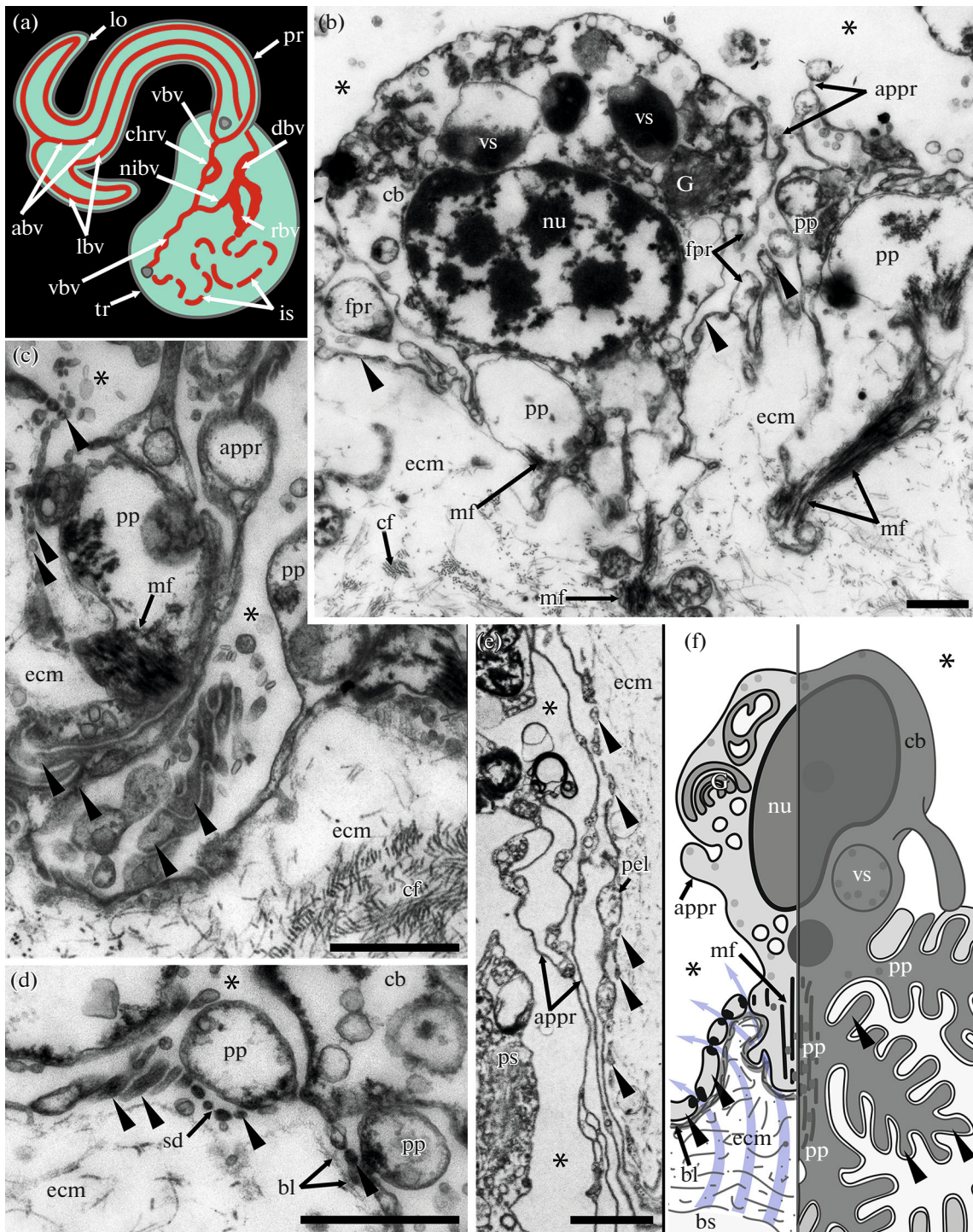
The objective of this work was to study the fine structure of podocytes in the circulatory system of *Bonellia viridis* females.

Adult females representing *B. viridis* Rolando, 1822 were collected near the shore of Marseille (Provence, France; 43°16'38" N, 5°18'24" E). Their morphology was studied using a Leica M165C stereo microscope equipped with a Leica DFC420 digital camera (Leica Microsystems). After dissection, separate parts of the circulatory system were fixed with 2.5% glutaraldehyde in 0.05 M cacodylate buffer supplemented with NaCl at 4°C for 8 h. The specimens were washed with the cacodylate buffer three times for 4 h and postfixed with 1% osmium tetroxide in the same buffer. The samples were dehydrated with increasing ethanol concentrations and embedded in Embed-812 (Electron Microscopy Science). Semithin and thin sections were obtained using a Leica UC7 ultramicrotome (Leica

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**Fig. 1.** Podocytes in the circulatory system of *B. viridis* females. (a) A schematic organization of the circulatory system. (b) A podocyte in a cross section of the ventral blood vessel, TEM. (c) Primary processes and pedicels of podocytes of the ventral blood vessel, TEM. (d) Pedicels in the ring blood vessel, TEM. (e) A schematic structure of a podocyte with pedicels. Arrows show the direction of fluid movement from the circulatory system into the trunk coelom. Designations: abv, axial blood vessel; appr, apical process; bl, basal lamina; bs, blood space; cb, cell body; cf, collagen filament; chrv, chaetal ring vessel; dbv, dorsal blood vessel; ecm, extracellular matrix; G, Golgi apparatus; is, intestinal sinus; lbv, lateral blood vessel; lo, lobe of the proboscis; mf, myofilament; nibv, neuro-intestinal blood vessel; nu, nucleus; pp, primary process of a podocyte; pr, proboscis; rbv, ring blood vessel; tr, trunk; sd, slit diaphragm; vbv, ventral blood vessel; vs, vesicle. Asterisks (\*) indicate the coelomic cavity; arrow tips indicate the pedicels of the podocyte. Scale bar, 1  $\mu$ m.

Microsystems). Ultrathin sections were stained with 0.5% uranyl acetate and 0.4% lead citrate and examined under a JEM-1011 electron microscope (JEOL).

The circulatory system of the echiurid trunk includes a descending part, which consists of the ventral blood vessel; an ascending part, which consists of the perienteric sinus and the dorsal blood vessel; and transversal blood vessels, which connect the two parts. A neurointestinal vessel extends from the ventral vessel in the anterior one-third of the trunk and ends in a ring vessel (Fig. 1a). Walls of the ventral blood vessel in its anterior one-third and the ring vessel are built of podocytes. Podocytes display common structural features and minor differences, depending on their localization. A podocyte consists of a broadened cell body, primary processes, and pedicels at the level of cell architecture (Fig. 1b). The cell body includes a large nucleus with a distinct nucleolus, a Golgi apparatus, numerous phagosomes, and multivesicular bodies (Fig. 1b). Many apical protrusions extend from the cell body and primary processes into the body cavity. The primary processes are 0.5–3 µm in diameter and are flattened on the surface of a basal lamina. Thin myofilament bundles pass through the primary process cytoplasm (Figs. 1b, 1c). The primary processes give origin to multiple thin pedicels, which branch and lie between pedicels of the adjacent podocytes, together producing a characteristic maze-like pattern (Figs. 1b, 1c). The pedicles reach 0.1–0.25 µm in width on average (Fig. 1d). Contacts are detectable between pedicles of neighbor podocytes and can be described as slit diaphragms, thin plates of electron-dense matter. A distance between pedicles and, accordingly, the width of a slit diaphragm does not exceed 20 nm. The pedicels lie on the dense basal lamina, which is underlain by a thick extracellular matrix layer penetrated by collagen fibers (Figs. 1c, 1d). Along with common structural features, there are considerable differences between podocytes of the ventral blood vessel and those of the ring vessel. The pedicels of ring-vessel podocytes (Fig. 1e) are far larger in diameter and form a simpler, less branched maze; the basal lamina is not as pronounced; and the apical protrusions are appreciably longer and more abundant.

Podocytes are thought to occur in certain vascular regions where ultrafiltration sites form [8]. Pedicels interconnected via specialized slit diaphragms are considered to be a main feature that distinguishes podocytes from all other cells. Fluid ultrafiltration from the vascular lumen into the coelomic cavity occurs through the basal lamina of a vessel and slit diaphragms between podocyte pedicels [9]. Primary urine forms in the coelomic cavity as a result of ultrafiltration, is passed into an excretory channel by the function of ciliated funnels of metanephridia, and undergoes secondary absorption there [10]. Podocytes found in *B. viridis* (Fig. 1f) are cytologically similar to podocytes described in other Bilateria. This makes it possible to assume that ultrafiltration sites are in the

anterior part of the ventral vessel and the ring vessel in *B. viridis* and, possibly, other echiurids. That is, the sites are in the anterior part of the trunk, while specialized metanephridia (anal sacs) are in its posterior part [11].

Numerous phagosomes and lysosome derivatives observed in *B. viridis* podocytes suggest highly intense endocytosis, which is similarly characteristic of podocytes of other annelids [12]. Myofilaments found at the bases of primary processes (Fig. 1f) point to incomplete separation of functions between podocytes and myoepithelial vascular cells. Myofilaments occurring in the basal regions of podocytes have been described in many annelids and other bilaterians, for example, Phoronida [13, 14]. Numerous apical protrusions are an outstanding feature of *B. viridis* podocytes. Similar protrusions have previously been observed in coelomic epithelial cells of the lateral channels and myoepithelial cells of the axial and lateral vessels of the proboscis in *B. viridis* females [7]. Thus, several cytological features preserved in podocytes suggest their common origin with the vascular myoepithelium and coelothelium.

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#### ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Invertebrates of the phylum Annelida were used in this work. In accordance with Directive 2010/63/EU of 22 September, 2010 on the protection of animals used for scientific purposes, Chapter 1, Paragraph 3, the requirements of bioethics do not apply to invertebrate animals with the exception of the class Cephalopoda.

#### CONFLICT OF INTEREST

The authors of this work declare that they have no conflicts of interest.

#### REFERENCES

1. Pilger, J.F., Echiura, in *Microscopic Anatomy of Invertebrates*, New York: Wiley-Liss, 1993, vol. 12, pp. 186–236.
2. Maiorova, A.S. and Adrianov, A.V., Biodiversity of echiurans (Echiura) of the Kuril-Kamchatka Trench area, *Prog. Oceanogr.*, 2020, vol. 180, p. 102216.
3. Goto, R., Monnington, J., Sciberras, M., et al., Phylogeny of Echiura updated, with a revised taxonomy to reflect their placement in Annelida as sister group to Capitellidae, *Invertebr. Syst.*, 2020, vol. 34, no. 1, p. 101.
4. Amor, A., Modelos de sistema vascular en Echiura, *Physis*, 1973, vol. 32, no. 84, pp. 115–120.

5. Schmidt-Rhaesa, A., *The Evolution of Organ Systems*, Oxford: Oxford Univ. Press, 2007.
6. Bosch, C., Les vaisseaux sanguins de la trompe chez *Bonellia viridis* (Echmrien) et leur musculature a fonction multiple, *Ann. Sci. Nat., Zool. Biol. Anim.*, 1984, vol. 6, pp. 3–32.
7. Kuznetsov, P. and Temereva, E., Ultrastructure of proboscis blood vessels in females of *Bonellia viridis* (Annelida: Bonellinae): New reconstructions, *J. Morphol.*, 2022, vol. 283, no. 6, pp. 771–782.
8. Peters, W., Possible site of ultrafiltration in *Tubifex tubifex* (Annelida, Oligochaeta), *Cell Tissue Res.*, 1977, vol. 179, pp. 367–375.
9. Bunke, D., Ultrastructure of the metanephridial system in *Aeolosoma bengalense* (Annelida), *Zoomorphology*, 1994, vol. 114, pp. 247–258
10. Smith, P.R., Polychaeta: Excretory system, in *Microscopic Anatomy of Invertebrates*, New York: Wiley-Liss, 1992, vol. 7, pp. 71–108.
11. Lehrke, J. and Bartolomaeus, T., Ultrastructure of the anal sacs in *Thalassema thalasseum* (Annelida, Echiura), *Zoomorphology*, 2011, vol. 130, pp. 39–49.
12. Bunke, D., Ultrastructure of the metanephridial system in *Aeolosoma bengalense* (Annelida), *Zoomorphology*, 1994, vol. 114, pp. 247–258.
13. Storch, V. and Hermann, K., Podocytes in the blood vessel linings of *Phoronis muelleri* (Phoronida, Tentaculata), *Cell Tissue Res.*, 1978, vol. 190, pp. 553–556.
14. Temereva, E.N. and Malakhov, V.V., Ultrastructure of the blood system in phoronid *Phoronopsis harmeri* Pixell, 1912: 1. Capillaries, *Russ. J. Mar. Biol.*, 2004, vol. 30, no. 1, pp. 28–36.

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