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## Amphilinidea

# Amphilinidea Poche, 1922 (Order)

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Phylum Platyhelminthes

Class Cestoda

Subclass Cestodaria

Order Amphilinidea

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### Amphilinidea Poche, 1922 (Order)

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#### Introduction

The cestodes (tapeworms) are a large group of endoparasitic worms infecting various vertebrates. Most species are included in the Eucestoda (true tapeworms), characterized (with few exceptions) by a number of segments (proglottids). Examples are *Taenia* (the pig and cattle tapeworms, of which the adults live in humans) and *Diphyllobothrium* (the broad fish tapeworm) infecting humans. Two groups of cestodes, the Gyrocotylidea and Amphilinidea, do not possess proglottids. The Amphilinidea are discussed here. Only 8 species included in 3 genera are known. They have little economic significance, although 1 species was shown to adversely affect sturgeon, the producers of caviar. Amphilinids are of considerable interest to biologists because they may cast light on the phylogeny of tapeworms and of related forms.

They are large (several cm-long), dorsoventrally flattened worms infecting the body cavity of freshwater and marine teleost (bony) fishes and freshwater turtles. Larvae are **ciliated** and possess 10 posterior **hooks**, which are retained in the adult. A well-known species is *Austramphilina* (= *Gigantolina*) *elongata* from Australia, with freshwater crustaceans as intermediate hosts and freshwater turtles as final (definitive) hosts.

A considerable number of studies deal with its morphology, electron microscopy, and life cycle (Rohde and Georgi, 1983; Rohde and Garlick, 1985a; 1985b; 1985c; 1985d; Rohde, 1986; 1987; 1994; Rohde et al., 1986; Rohde and Watson, 1986; 1987; 1988; 1989; 1990a; 1990b). Brief overviews of the Amphilinidea are by Rohde (2005) and Read (2007). The Tree of Life webpage by Rohde (1998) (available at http://tolweb.org/Amphilinidea) contains an account of all aspects of Amphilinidea and an extensive bibliography. Older references can be found in Dubinina (1982). Important papers on some aspects of *Amphilina foliacea* are by Bisserova et al. (2000) and Dudicheva and Bisserova (2000). *Austramphilina elongata* is also discussed in greater detail.

#### Structure of the Adult Austramphilina elongata

The adult worm reaches a length of about 150 or more mm, with a width of about 14 or more mm (Figure 1). As in all amphilinids, the **uterus** forms 3 loops in the body; it extends from the posteriorly located **ovary** to the anterior end, turns back and forward again, opening through a **uterine pore** at the anterior end. The **vagina** opens at the posterior



Figure 1. *Austramphilina elongata*. Several worms in the body cavity of the freshwater turtle *Chelodina longicollis*. Source: K. Rohde. License: CC BY-NC-SA 4.0.



Figure 2. *Austramphilina elongata*, whole mount. X = bodies of unknown function. Source: K. Rohde. License: CC BY-NC-SA 4.0.



Figure 3. *Austramphilina elongata*, receptors of adult. Source: Adapted from Rohde and Watson, 1990b. License: CC BY-NC-SA 4.0.

end. Vitellaria extend in the lateral parts of the body from the anterior to the posterior ends of the body. **Testes** are scattered throughout the body and the male **gonopore** is located near the female one at the posterior end (Figure 2). Electron microscope studies have shown several types of **sensory receptors** (Figure 3).

#### Structure of Larval Austramphilina elongata

The larvae are **ciliated** and possess 10 posterior **hooks** of 3 different kinds. Two pairs are serrate, the others are sickleshaped (Figures 4 and 5). Ducts of clusters of **gland cells** open near the anterior end. The **protonephridial** (excretory/ osmoregulatory) system consists of 3 **flame cells** or **bulbs** on each side of the body, with paired **excretory pores** located near the posterior end (Figure 4). A large number of transverse **muscle bands** extend below the **tegument** (surface layer) of the larva. There are several clusters of **sensilla** (**sensory receptors**) (Figures 5 and 6).

The larvae possess a ciliated **epidermis** located on an underlying tegument which becomes the surface layer (**neo-dermis**) once the epidermis is shed by the invading larva (Figure 7).

The larva possesses a considerable number of sensory receptor types differing with respect to the presence or absence of cilia, the number and shape of the cilia, and the shape of the basal bodies/ciliary **rootles** (Figure 8).



Figure 4. *Austramphilina elongata* larva. Note the bundles of secretory glands opening near the anterior end, the protonephridial system with 3 flame bulbs on each side opening near the posterior end, and the 10 posterior hooks. Source: Adapted from K. Rohde, 1986. License: CC BY-NC-SA 4.0.



Figure 5. Posterior end of a larval *Austramphilina elongata*. Note the cluster of sensilla, transverse muscle bands, ciliated epidermis, and 5 pairs of hooks of 3 types. Source: K. Rohde. License: CC BY-NC-SA 4.0.

Transverse muscle fibres Sensilla

Figure 6. Larva of *Austramphilina elongata* impregnated with silver. Note the transverse muscle bands and receptors (sensilla). Source: K. Rohde. License: CC BY-NC-SA 4.0.



Figure 7. Larval *Austramphilina elongata*, diagram of electronmicroscopic structure of surface layers. Note larval syncytial and ciliated epidermis at the surface, based on the tegument (neodermis) that has insunk (below the surface) nuclei (only the process leading to 1 nucleus is illustrated). Source: K. Rohde. License: CC BY-NC-SA 4.0.

#### Life Cycle of Austramphilina elongata

The eggs of *Austramphilina elongata* have to get into freshwater for further development (Figure 9). The escape route from the host is unknown. Larvae hatch in freshwater. They swim around in water until they get into contact with a crayfish (phylum Arthropoda: class Crustacea: order Decapoda). On the crayfish, the larva bends in such a way that both the anterior and posterior ends are located close together on the cuticle of the host. The sickle-shaped hooks pierce into the cuticle, the serrate ones perform sawing movements, cutting through the cuticle. The 3 types of anterior



Figure 8. *Austramphilina elongata*, diagrams of larval receptors as seen under the transmission electron microscope. Source: Adapted from Rohde et al., 1986a. License: CC BY-NC-SA 4.0.



Figure 9. Life cycle of *Austramphilina elongata*. Note: Escape route of egg from turtle is unknown. Source: K. Rohde. License: CC BY-NC-SA 4.0.



Figure 10. Section though the esophageal wall of a turtle, *Chelodina longicollis*, showing a penetrating *Austramphilina* juvenile (arrow). Source: K. Rohde. License: CC BY-NC-SA 4.0.



Figure 11. Two juvenile *Austramphilina* specimens (arrows) migrating along the trachea towards the body cavity of a turtle. Source: K. Rohde. License: CC BY-NC-SA 4.0.

glands apparently produce a secretion (which, however, has not been identified) dissolving the surface layer. The larva penetrates into the host's tissue, shedding the ciliated epidermis in the process. Penetration is observed to occur through the gills, and through the thin junctions between the crayfish's segments within 30 minutes after first contact. Larvae infective to turtles are several mm long and may be observed in the abdomen of crayfish. Turtles become infected by eating crayfish. Juvenile worms penetrate through the wall of the esophagus (Figure 10), migrate along the trachea (Figure 11), and through the septum into the body cavity where they mature. Adult worms are seen mainly in the body cavity, but occasionally also in the lungs. This suggests that eggs may



Figure 12. *Amphilina foliacea*, adult. Source: Adapted from Dubinina, 1982. License: CC BY-NC-SA 4.0.

leave the host via the trachea and mouth cavity from where they are spit out into water. Once, an adult was also seen in the urinary bladder, and once in the oviduct of a turtle, suggesting that eggs may be shed through the cloaca. Freshwater shrimps could also be infected experimentally, but larvae did not reach a size infective to turtles in them.

#### **Other Species**

Only 1 other species has been studied in detail, namely, *Amphilina foliacea*. It differs from *Austramphilina* in a number of morphological features (Figure 12). Its protonephridial system forms a network of canals, differing from that of other species, for example, *Gephyrolina paragonopora* (Figure 13).

Amphilina foliacea uses freshwater amphipods (class Crustacea: order Amphipoda) as intermediate hosts and Accipenser (sturgeon) as final hosts. It inhabits the body cavity of the final host and eggs escape through the coelomic pore which connects the body cavity to the outside (it is not present in turtles!). Eggs containing infective larvae are ingested by the amphipods, whose mouthparts break the eggshell allowing the larva to escape and penetrate into the host.

Adult *Nesolecithus africanus* infect African freshwater fish. Juveniles have been recovered from freshwater prawns (class Crustacea: order Decapoda).



Figure 13. Protonephridial canal system of *Amphilina foliacea* (A) and of *Gephyrolina paragonopora* (B). Adapted from Dubinina, 1982. License: CC BY-NC-SA 4.0.

#### **Taxonomy and Phylogeny**

Gibson (1994) has provided a key to the species (see also Schmidt, 1986) and Dubinina (1982), in a detailed monograph of the Amphilinidea, has discussed the position of the group in the phylum Platyhelminthes (see also Galkin, 1999). Eight species have been described:

- 1) Amphilina foliacea synonyms Monostomum foliaceum, A. neritina
- 2) *Am. japonica* synonyms *Am. bipunctata*, *A. foliacea*
- Gephyrolina paragonopora synonyms Am. paragonopora, Hunteroides mystel, Schizochoerus paragonopora
- 4) Schizochoerus liguloideus synonyms M. liguloideum, Am. liguloidea
- 5) Nesolecithus janickii synonyms Am. liguloidea, M. liguloideum, S. janickii
- 6) *N. africanus* synonym *S. africanus*
- 7) Austramphilina elongata synonyms Kosterina Kuiperi, Gigantolina elongata
- 8) Gigantolina magna synonyms Am. magna, Gyrometra albotaenia, Gy. kunduchi

The Gyrocotylidea have often been considered to be the sister group of the amphilinids, both comprising the Cestodaria (non-segmented tapeworms) (Bandoni and Brooks, 1987). However, later studies do not support a monophyletic group, Cestodaria. Instead, gyrocotylids appear to be the earliest divergent lineage within the cestodes followed by the amphilinids and then the eucestodes (true cestodes) (Waeschenbach et al., 2012; Littlewood et al., 2015; Waeschenbach and Littlewood, 2017). The Cestoda must be considered to be the sister group of the Trematoda (see, for example, Park et al., 2007) and all the large groups of parasitic flatworms Polyopisthocotylea and Monopisthocotylea (= "Monogenea"), Trematoda, and Cestoda (including the Eucestoda, Amphilinidea, and Gyrocotylidea) are monophyletic comprising the Neodermata, as first proposed by Ehlers (1985) and later confirmed by numerous electron microscope and DNA studies (for example, Egger et al., 2015). Various hypotheses of these relationships are currently being tested using deep sequencing of DNA at the genome level.

#### Acknowledgement

- Based on the author Rohde's online articles available at https://krohde.wordpress.com/2009/08/03/the-amphilinidea-a-small-group-of-xk923bc3gp4-21/ and https://krohde.wordpress.com/2009/08/03/die-amphilini
  - dea-eine-kleine-gruppe-xk923bc3gp4-22/

#### Literature Cited

- Bandoni, S. M., and D. R. Brooks. 1987. Revision and phylogenetic analysis of the Amphilinidea Poche, 1922 (Platyhelminthes: Cercomeria: Cercomeromorpha). Canadian Journal of Zoology 65: 1,110–1,128. doi: 10.1139/z87-175
- Biserova, N. M., V. A. Dudicheva, N. B. Terenina, M. Reuter, et al. 2000. The nervous system of *Amphilina foliacea* (Platyhelminthes, Amphilinidea): An immunocytochemical, ultrastructural and spectrofluorometrical study. Parasitology 121: 441–453. doi: 10.1017/s0031182099006411
- Dubinina, M. N. 1982. [Parasitic worms of the class Amphilinida (Platyhelminthes)]. Trudy Zoologicheskogo Institut, Akademiia Nauk SSSR 100: 1–143. [In Russian.]
- Dudicheva, V. A., and N. M. Biserova. 2000. [Distribution of sensory organs on surface of adult *Amphilina foliacea* (Plathelminthes, Amphilinida).] Zoologicheskiĭ Zhurnal 79: 1,139–1,146. [In Russian.]
- Galkin, A. K. 1999. [Position of Amphilinidea in the system of Cercomeromorphae.] Parazitologiya 33: 497–506. [In Russian.]
- Gibson, D. I. 1994. Order Amphilinidea Poche 1922. In L. F. Khalil, A. Jones, and R. A. Bray, eds. Keys to the Cestode Parasites of Vertebrates. CAB International, Wallingford, United Kingdom, p. 3–10.

- Littlewood, D. T. J., R. A. Bray, and A. Waeschenbach. 2015.
  Phylogenetic patterns of diversity in cestodes and trematodes. *In* S. Morand, B. R. Krasnov, and D. T. J. Littlewood, eds.
  Parasite Diversity and Diversification. Cambridge University
  Press, Cambridge, United Kingdom, p. 305–319.
- Park, J.-K., K.-H. Kim, S. Kang, W. Kim, et al. 2007. A common origin of complex life cycles in parasitic flatworms: Evidence from the complete mitochondrial genome of *Microcotyle sebastis* (Monogenea: Platyhelminthes). BMC Evolutionary Biology 7: 11. doi: 10.1186/1471-2148-7-11
- Read, C. P. 2007. Amphilinidea. *In* McGraw-Hill Encyclopedia of Science and Technology, Volume 1. McGraw-Hill, New York, New York, United States.
- Rohde, K. 2005. Amphilinidea. *In* K. Rohde, ed. Marine Parasitology. CSIRO Publishing, Melbourne, and CAB International Publishing, Wallingford, United Kingdom, p. 87–89, 461.
- Rohde, K. 1998. The Amphilinidea. Tree of Life. http://tolweb. org/Amphilinidea
- Rohde, K. 1987. The formation of glandular secretion in larval *Austramphilina elongata* (Amphilinidea). International Journal for Parasitology 17: 821–828. doi: 10.1016/0020-7519(87)90064-6
- Rohde, K. 1994. The minor groups of parasitic Platyhelminthes. Advances in Parasitology 33: 145–234. doi: 10.1016/ s0065-308x(08)60413-3
- Rohde, K. 1986. Ultrastructural studies of Austramphilina elongata Johnston, 1931 (Cestoda, Amphilinidea). Zoomorphology 106: 91–102. doi: 10.1007/BF00312111
- Rohde, K., and P. R. Garlick. 1985a. A muticiliate 'starcell' in the parenchyma of the larva of *Austramphilina elongata* Johnston, 1931 (Amphilinidea). International Journal for Parasitology 15: 403–407. doi: 10.1016/0020-7519(85)90025-6
- Rohde, K., and P. R. Garlick. 1985b. Subsurface sense receptors in the larva of *Austramphilina elongata* Johnston, 1931 (Amphilinidea). Zoomorphology 105: 34–38. doi: 10.1007/ BF00312071
- Rohde, K., and P. R. Garlick. 1985c. Two ciliate sense receptors in the larva of *Austramphilina elongata* Johnston, 1931 (Amphilinidea). Zoomorphology 105: 30–33. doi: 10.1007/ BF00312070
- Rohde, K., and P. R. Garlick. 1985d. Ultrastructure of the posterior sense receptor of larval *Austramphilina elongata* Johnston, 1931 (Amphilinidea). International Journal for Parasitology 15: 339–402. doi: 10.1016/0020-7519(85)90024-4
- Rohde, K., and M. Georgi. 1983. Structure and development of *Austramphilina elongata* Johnston, 1931 (Cestodaria, Amphilinidea). International Journal for Parasitology 13: 273–287. doi: 10.1016/0020-7519(83)90039-5

- Rohde, K., and N. Watson. 1988. Development of the protonephridia of *Austramphilina elongata*. Parasitology Research 74: 255–261. doi: 10.1007/BF00539574
- Rohde, K., and N. Watson. 1990a. Ultrastructural studies of juvenile Austramphilina elongata: Scanning and transmission electron microscopy of the tegument. International Journal for Parasitology 20: 271–277. doi: 10.1016/0020-7519(90)90140-I
- Rohde, K., and N. Watson. 1990b. Ultrastructural studies of juvenile *Austramphilina elongata*: Transmission electron microscopy of sensory receptors. Parasitology Research 76: 336–342. doi: 10.1016/0020-7519(90)90140-I
- Rohde, K., and N. Watson. 1989. Ultrastructural studies of larval and juvenile *Austramphilina elongata* (Platyhelminthes, Amphilinidea); penetration into, and early development in the intermediate host, *Cherax destructor*. International Journal for Parasitology 19: 529–538. doi: 10.1016/0020-7519(89)90083-0
- Rohde, K., and N. Watson. 1987. Ultrastructure of the protonephridial system of larval *Austramphilina elongata* (Platyhelminthes, Amphilinidea). Journal of Sub-Microscopic Cytology 19: 113–118.
- Rohde, K., and N. Watson. 1986. Ultrastructure of the sperm ducts of *Austramphilina elongata* (Platyhelminthes, Amphilinidea). Zoologischer Anzeiger 217: 23–30. doi: 10.1016/0020-7519(89)90083-0
- Rohde, K., N. Watson, and P. R. Garlick. 1986.
  Ultrastructure of three types of sense receptors of larval *Austramphilina elongata* (Amphilinidea).
  International Journal for Parasitology 16: 245–251. doi: 10.1016/0020-7519(86)90051-2
- Waeschenbach, A., and D. T. J. Littlewood. 2017. A molecular framework for the Cestoda. *In J. N. Caira and K. Jensen*, eds. Planetary Biodiversity Inventory (2008–2017): Tapeworms from Vertebrate Bowels of the Earth. University of Kansas, Natural History Museum, Special Publication Number 25. Lawrence, Kansas, United States, p. 431–451.
- Waeschenbach, A., B. L. Webster, and D. T. J. Littlewood. 2012. Adding resolution to ordinal level relationships of tapeworms (Platyhelminthes: Cestoda) with large fragments of mtDNA. Molecular Phylogenetics and Evolution 63: 834–847. doi: 10.1016/j.ympev.2012.02.020

#### **Supplemental Reading**

Schmidt, G. D. 1986. Handbook of Tapeworm Identification. CRC Press, Boca Raton, Florida, United States, 675 p.