

## NOTE

## Role of *Leptodora kindti* (Cladocera: Leptodoridae) in the life cycle of *Raphidascaris biwakoensis* (Nematoda: Anisakidae), a fish parasite in Lake Biwa, Japan

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**ABSTRACT:** Advanced third-stage larvae (body length 2.92 to 5.49 mm) of the fish nematode *Raphidascaris biwakoensis* Fujita, 1928 were found as frequent parasites of the leptodoriid cladoceran *Leptodora kindti* (Focke) in Lake Biwa, southern Honshu, Japan, in July 1995. This is the first confirmed record of the L<sub>3</sub> of this nematode species from an invertebrate host, showing that *L. kindti* may serve as a true intermediate host to *R. biwakoensis*, probably in addition to lower aquatic vertebrates. The high degree of prevalence of the nematode larvae in *L. kindti* in Lake Biwa indicates that this plankton crustacean is an important source of infection with *R. biwakoensis* for fish definitive and paratenic hosts.

**KEY WORDS:** Parasitic nematode · *Raphidascaris biwakoensis* · Intermediate host · *Leptodora kindti* · Japan

During recent hydrobiological studies carried out by two of the authors (Y.T. and T.N.) in Lake Biwa, southern Honshu, Japan, a plankton sample containing numerous specimens of the leptodoriid cladoceran *Leptodora kindti* (Focke) was collected from Shiotsu Bay (4 m depth) on 21 July 1995. Many of them proved to be infected by marked larval anisakid nematodes, identified as the third-stage larvae of *Raphidascaris biwakoensis* Fujita, 1928, an intestinal parasite of some piscivorous and zooplanktivorous freshwater fishes in southern Japan. Of 25 infected *L. kindti*, most specimens harboured 1 nematode larva in the haemocoel of the abdomen; only 2 leptodoriids contained 2 larvae each. Since the body of *L. kindti* is translucent, the

presence of large, white-coloured nematodes in them is conspicuous (Fig. 1). Numerous *Daphnia*-like cladocerans, mostly *Daphnia galeata* Sars, present in the sample were uninfected.

The larvae of *Raphidascaris biwakoensis* (Fig. 2) were not encapsulated. Their bodies (n = 5) measured 2.92 to 5.49 mm in length and 124 to 235 µm in maximum width. The cuticle was thin and smooth; cervical alae were absent. The ventral cephalic tooth was well developed and present in all larvae. The lip anlagen were only feebly developed. The oesophagus with somewhat expanded posterior part measured 340 to 560 µm in length and 44 to 80 µm in maximum width, the ventriculus was 28–60 × 48–84 µm; the ventricular appendix was 272 to 580 µm long and 20 to 60 µm wide. The distance of the nerve ring and the excretory pore from the anterior extremity was 132 to 184 µm and 192 to 280 µm, respectively. The brownish intestine was broad, occupying most of the space of the body; it ended in a short colourless rectum surrounded by a few well-developed unicellular rectal glands. Developing sexual glands were tubular, 84 to 1800 µm long, forming a few coils in the region near the mid-body, reaching anteriorly up to the posterior end of the ventricular appendix in the most advanced larvae; neither vagina nor vulva were visible. The tail was slender, sharply pointed, 84 to 132 µm long. No signs of the next larval moult were observed.

Although several *Raphidascaris* species (some established on larval forms) were reported from fishes in Lake Biwa (Fujita 1928, Kataoka & Momma 1932, 1934), Smith (1984a) suggested that *R. biwakoensis* is the only freshwater representative of this genus in

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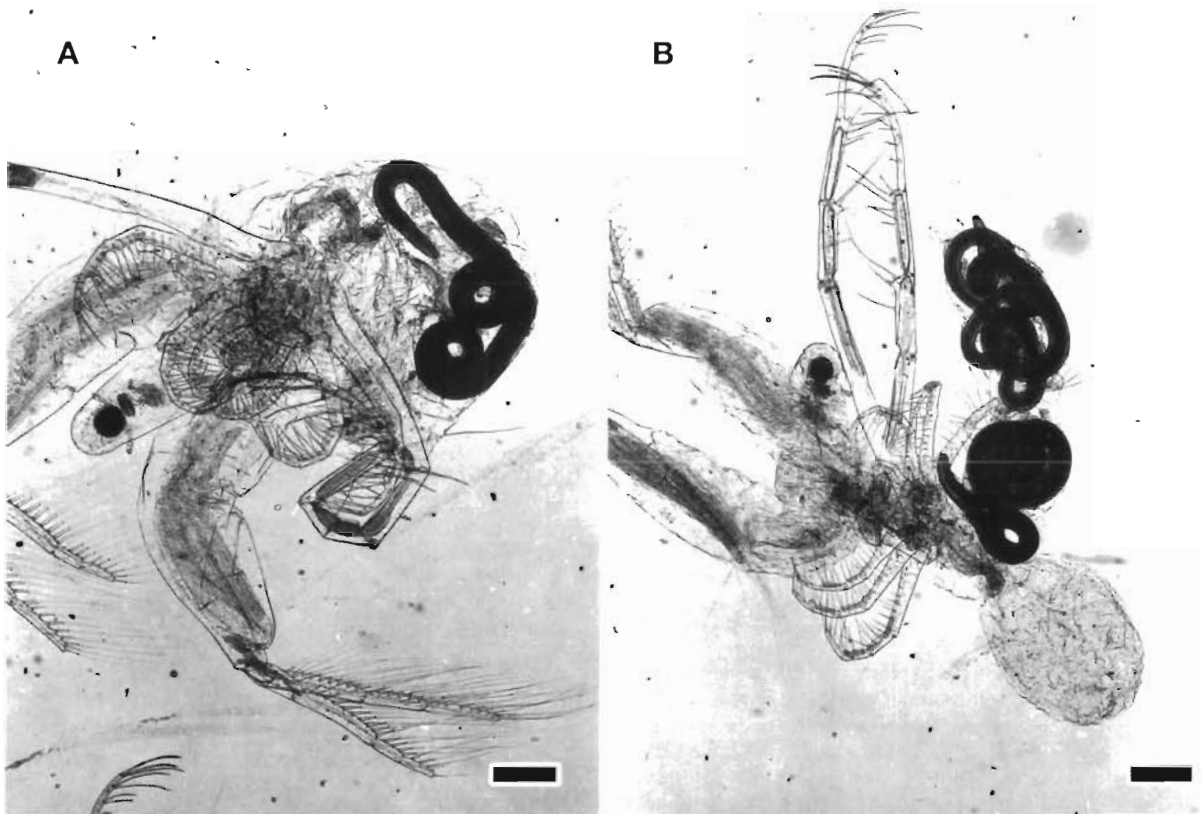


Fig. 1 *Leptodora kindti* infected with *Raphidascaris biwakoensis* third-stage larvae (fresh mounts). (A) With 1 larva, (B) with 2 larvae. Scale bars = 0.5 mm

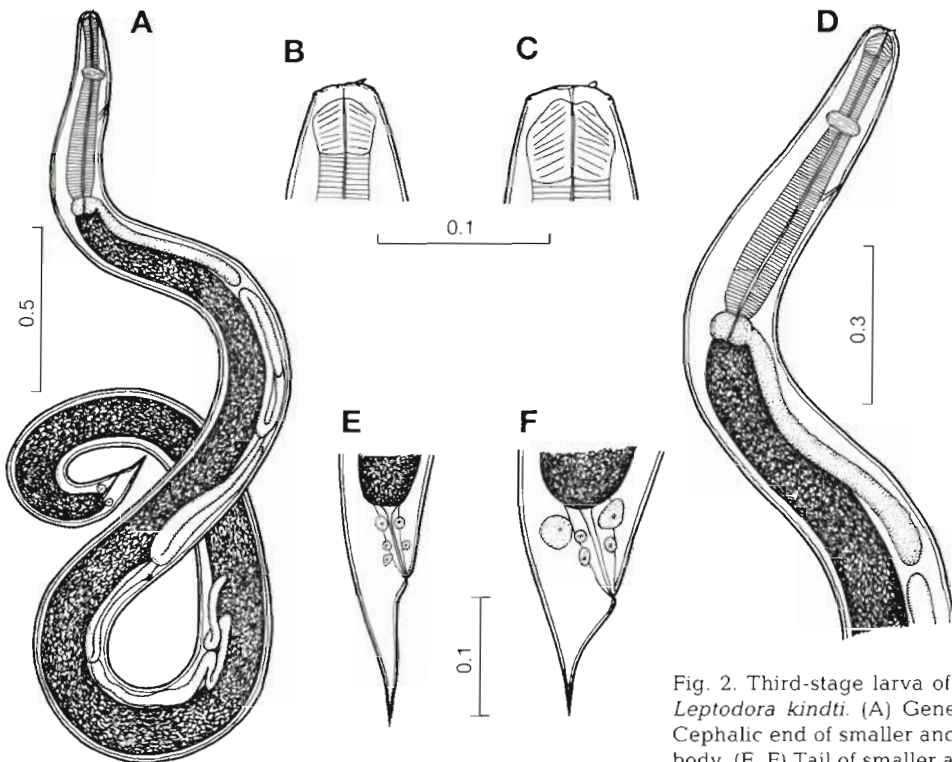


Fig. 2. Third-stage larva of *Raphidascaris biwakoensis* from *Leptodora kindti*. (A) General view of female larva. (B, C) Cephalic end of smaller and larger larva. (D) Anterior part of body. (E, F) Tail of smaller and larger larva. Scale bars in mm

Japan. According to his revisional paper (Smith 1984a), there are only 2 valid *Raphidascaris* species parasitizing freshwater fishes, *R. acus* (Bloch, 1779) in continental palaeartic Eurasia and North America and *R. biwakoensis* Fujita, 1928 in Japan (3 other congeneric species were later described from freshwater fishes in South America). Both of these species are easily distinguishable, because the former has broad cervical alae (in adults as well as in third- and fourth-stage larvae—see Moravec 1970) which are missing in the latter; the absence of cervical alae in *R. biwakoensis* fourth-stage larvae was confirmed by Moravec & Nagasawa (1989). Recently, Bruce (1990) transferred *R. biwakoensis* to the newly re-erected genus *Ichthyascaris* Wu, 1949 (synonymized with *Raphidascaris* Raillet et Henry, 1915 by Hartwich 1957). However, since the morphologies of both the type species of *Ichthyascaris*, *I. lophii* Wu, 1949, and *R. biwakoensis* are insufficiently known (Bruce et al. 1994), we consider it is reasonable to retain *R. biwakoensis* in the genus *Raphidascaris*.

The general morphology of *Raphidascaris* larvae from *Leptodora kindti* shows that they are advanced third-stage larvae of *R. biwakoensis*. Larvae with the same morphology, but distinctly smaller (1.74 to 2.38 mm long), were recorded from 3 specimens of *L. kindti* in Lake Biwa as early as in 1932 by Kataoka & Momma, who identified them as immature forms of *Raphidascaris* sp.; there is no doubt that they were *R. biwakoensis* too.

The hosts of adult *Raphidascaris biwakoensis* are some piscivorous and zooplanktivorous fishes [*Pseudobagrus (Pelteobagrus) nudiceps* (Sauvage), *Oncorhynchus mykiss* (Walbaum), *Plecoglossus altivelis* Temminck et Schlegel] (Fujita 1928, Kataoka & Momma 1934, Yamaguti 1935), whereas various species of forage fishes apparently serve as intermediate or paratenic hosts, as for *Raphidascaris acus* (Bloch, 1779) (see Moravec 1994).

The life cycle of this parasite is not known. That of the congeneric species *Raphidascaris acus* involves obligate intermediate hosts, various species of fish and cyclostomes, and, less often, amphibians, whereas various invertebrates (oligochaetes, snails, plankton and benthic crustaceans and aquatic insects) have generally been considered as pre-intermediate paratenic hosts of second-stage larvae (Moravec 1970, 1994, Smith 1984b, Torres & Alvarez-Pellitero 1988). Only recently has Moravec (1996) proved that an invertebrate (*Gammarus*) may serve, in addition to some lower aquatic vertebrates, as a true intermediate host of *R. acus*.

The striking features of *Raphidascaris biwakoensis* larvae from *Leptodora kindti* are the markedly large size of the body and a highly advanced stage of genital

primordia. Whereas the largest third-stage larva of *R. acus* recorded from an invertebrate (*Gammarus fos-sarum*) was only 1.77 mm long, with only a small, oval-shaped genital primordium (Moravec 1996), some larvae of *R. biwakoensis* from *L. kindti* attained a body length of more than 5 mm and their genital primordia were highly advanced, even in the smallest larvae (1.74 to 2.38 mm long) reported by Kataoka & Momma (1932). Such advanced genital primordia are usually typical of fourth-stage larvae, but the presence of the ventral cephalic tooth and the absence of a vulva in female larvae as well as their poorly developed lip anlagen clearly show that they are third-stage larvae. The fourth-stage larvae of *R. biwakoensis* described from the fish *Cottus reini* Hilgendorf from the same locality (Lake Biwa) by Moravec & Nagasawa (1989) were larger (5.71 to 7.71 mm long) and the cephalic tooth was missing; from the same fish they also described a third-stage larva (4.2 mm long) morphologically identical with those from *L. kindti*.

The presence of large-sized third-stage larvae of *Raphidascaris biwakoensis* with advanced genital primordia in *Leptodora kindti* indicates that this plankton crustacean plays the role of a true intermediate host to this nematode parasite. Since *L. kindti* is predatory, preying on other aquatic invertebrates, it may acquire *R. biwakoensis* infection by eating either the free-living second-stage larvae of this nematode or some invertebrates (copepods, oligochaetes?) serving as its possible pre-intermediate paratenic hosts. It is highly probable that, similar to the case of *R. acus*, a variety of fish species can serve as the intermediate hosts of *R. biwakoensis* in addition to some invertebrates (*L. kindti*). However, a frequent occurrence of infected *L. kindti* in Lake Biwa suggests that this plankton crustacean is an important source of infection with *R. biwakoensis* for fishes serving both as the definitive or the paratenic hosts of this nematode. Since *R. biwakoensis* larvae and adults are frequent parasites of a variety of fish species in Lake Biwa, including those of economic importance, further studies on the life cycle of this parasite are highly desirable.

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