

Female morphology and systematic status of *Philometroides cyprini* (Nematoda: Philometridae), a parasite of carp

František Moravec^{1,*}, Stanislav Červinka^{2,3}

¹Institute of Parasitology, Academy of Sciences of the Czech Republic, Branišovská 31, 370 05 České Budějovice, Czech Republic

²Fishery Technical School, Zátíší 480/2, 389 19 Vodňany, Czech Republic

³Fishery Educational Establishment, Táboritská 941/II, 379 01 Třeboň, Czech Republic

ABSTRACT: Gravid females of *Philometroides* were found under the scales of stock carp *Cyprinus carpio* L. imported from Lithuania to the Czech Republic in the spring of 1997 and kept in a quarantine pond. These specimens and those obtained from pond-cultured carp in Russia (Moscow district) were used for detailed morphological studies, including scanning electron microscopy. The results confirm that the valid name of this parasite is *Philometroides cyprini* (Ishii, 1931), whereas *Philometra lusii* Vismanis, 1962, *Philometra schikhobalovae* Belous, 1965, *Philometra lusiana* Vismanis, 1966 and *Philometroides lusiana* (Vismanis, 1966) Ivashkin et al., 1971 are its junior synonyms. *P. cyprini*, the agent of a serious disease of carp, seems to have been originally distributed in east Asia, from where it was introduced into Europe in the early 1960s.

KEY WORDS: Parasitic nematode · *Philometra* · *Philometroides cyprini* · *Philometroides lusiana* · Morphology · *Cyprinus carpio* · Species introduction · Czech Republic

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INTRODUCTION

Vismanis (1962) described a new philometrid species, *Philometra lusii*, from gravid females found under the scales of common carp *Cyprinus carpio* L. in Lithuania. However, because of some mistakes in the original description, he later redescribed and reestablished this species and renamed it *P. lusiana* Vismanis, 1996 (see Vismanis 1966). Ivashkin et al. (1971) transferred the species to the genus *Philometroides* Yamaguti, 1935. This pathogenic nematode has subsequently been reported, mainly from pond-cultured carps, from many areas of Russia (e.g. Kaliningrad [Königsberg] district, Moscow and Smolensk regions), Belarus and Ukraine (Vasilkov 1967, 1968a, Avdeeva & Evdokimova 2004).

The parasite causes philometroidosis disease of carp, which manifests itself especially in heavily infected fish and may even lead to the death of young carp (Vasilkov 1967, 1983, Kudentsova et al. 1975). Various species of copepods (Copepoda) function as the inter-

mediate host, and the life cycle of the parasite in European conditions is annual. Maturation is strictly seasonal; gravid females bearing larvae in the uterus occur only from the end of May until the beginning of June (Vismanis 1964, 1970, Vasilkov 1967, 1968a,b).

In 1976, Nakajima redescribed *Philometroides cyprini* (Ishii, 1931) from female specimens collected from *Cyprinus carpio* in Japan. He considered *P. cyprini* to be synonymous with *Philometroides lusiana* (Vismanis, 1966) Ivashkin et al., 1971 and remarked that the former species had long been known as a common parasite of cultured carp in Japan.

However, Vismanis et al. (1987) did not agree and regarded *Philometroides cyprini* and *Philometroides lusiana* as 2 independent species. According to them, females of *P. lusiana* possess 4 cephalic papillae and 4 caudal projections, whereas those of *P. cyprini* have no cephalic papillae and only 2 caudal projections. They considered the specimens studied by Nakajima (1976) to belong to *P. lusiana* and those originally described

*Email: moravec@paru.cas.cz

by Ishii (1931) to *P. cyprini*. As a junior synonym of the latter Vismanis et al. (1987) designated *Philometra shikhobalovae* Belous, 1965, a species established by Belous (1965) from specimens collected from *Cyprinus carpio* of Lake Khanka (Amur River basin), Russian Far East. Vismanis et al. (1987) argue that Ishii (1931) did not mention any cephalic papillae in the original description of *P. cyprini* and Belous (1965) did not observe cephalic papillae in her specimens, which had 2 caudal projections. It is necessary to remark, however, that the descriptions provided by these 2 authors are inadequate.

Based on literature data and partly on personal observations of specimens, Moravec (1994) followed the conception of Nakajima (1976) and took *Philometroides lusiana* to be a junior synonym of *P. cyprini*. However, the name *P. lusiana* has been commonly used for this species in the recent Russian parasitological literature (e.g. Avdeeva & Evdokimova 2004) and in web electronic databases. Moreover Cakić et al. (2002) reported *P. lusiana* and *P. cyprini* as 2 distinct species from the abdominal cavity of the Mediterranean barbel *Barbus meridionalis* Risso, from Yugoslavia.

MATERIALS AND METHODS

In the spring of 1997, live stock carp *Cyprinus carpio* were imported from Lithuania to the Czech Republic by the State Fishery Hluboká nad Vltavou (South Bohemia) and were placed in a quarantine pond (Haklovský pond). During the 30 d quarantine period, 15 fish were examined to check their health and condition. All fish were infected by gravid *Philometroides* females located under the scales. Subsequently, the entire fish stock was killed, and the pond was dried out and disinfected in order to eliminate the parasite.

A few of the above mentioned *Philometroides* specimens and those previously provided by Dr. G. V. Vasilkov (collected from pond-cultured carp at a fish farm in Gzhelka, Moscow district, Russia, in May 1963) were first studied by light microscopy (LM) and scanning electron microscopy (SEM).

The nematodes were fixed in hot 4% formaldehyde solution in physiological saline and then kept in 4% formaldehyde solution in water. For LM examination, they were cleared with glycerine. Drawings were made with the aid of a Zeiss drawing attachment. Specimens used for SEM were postfixed in 1% osmium tetroxide, dehydrated through a graded ethanol series, critical point dried and sputter-coated with gold; they were examined using a JEOL JSM-6300 scanning electron microscope at an accelerating voltage of 15 kV.

RESULTS

The gravid female nematodes were 7.02 to 10.9 cm long and 843 to 1339 μ m in maximum width. Live specimens were red; those fixed in 4% formaldehyde solution in physiological saline were brownish. The body surface was covered with numerous irregularly distributed cuticular bosses (Figs. 1A,D,E & 2D,F). At the cephalic end there were 4 submedian slightly elevated lobes, each with a pair of small cephalic papillae of the external circle (Figs. 1B,C & 2A,C); 4 small single submedian and 2 lateral papillae of the internal circle surrounded the small oral aperture (Figs. 1C & 2B). The oesophagus was 2.24 to 2.78 mm long and 122 to 174 μ m in maximum width, with a moderately developed anterior bulb (Fig. 1B); the oesophageal gland containing a large cell nucleus was well developed. The nerve ring was 218 to 278 μ m from the anterior extremity. The anus and

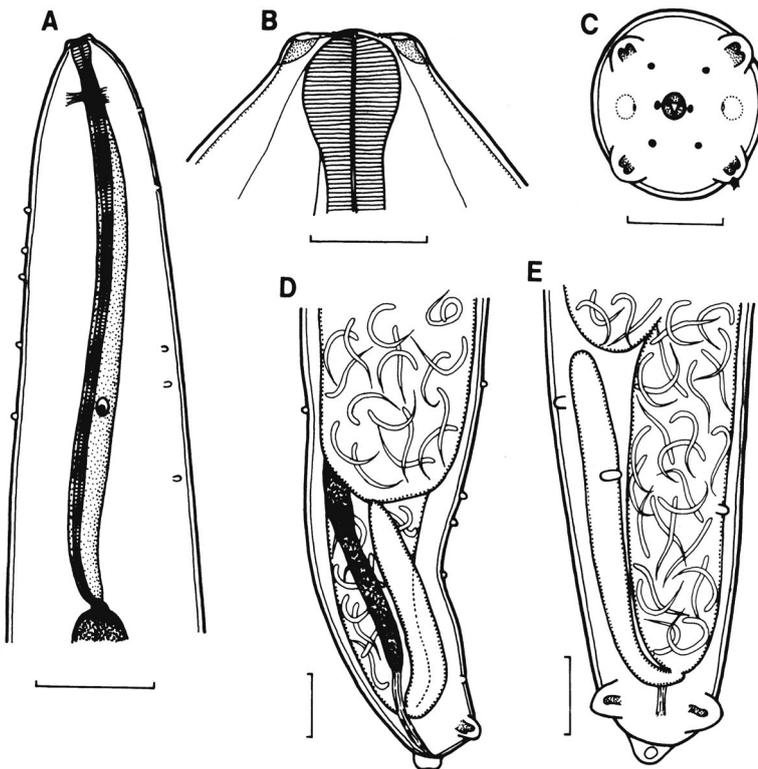


Fig. 1. *Philometroides cyprini*. (A) Anterior part of subgravid female (lateral view). (B,C) Cephalic end (lateral and apical views). (D,E) Posterior part of gravid female (lateral and dorsal views). Scale bars: A = 0.5 mm; B = 0.05 mm; C = 0.1 mm; D,E = 0.2 mm

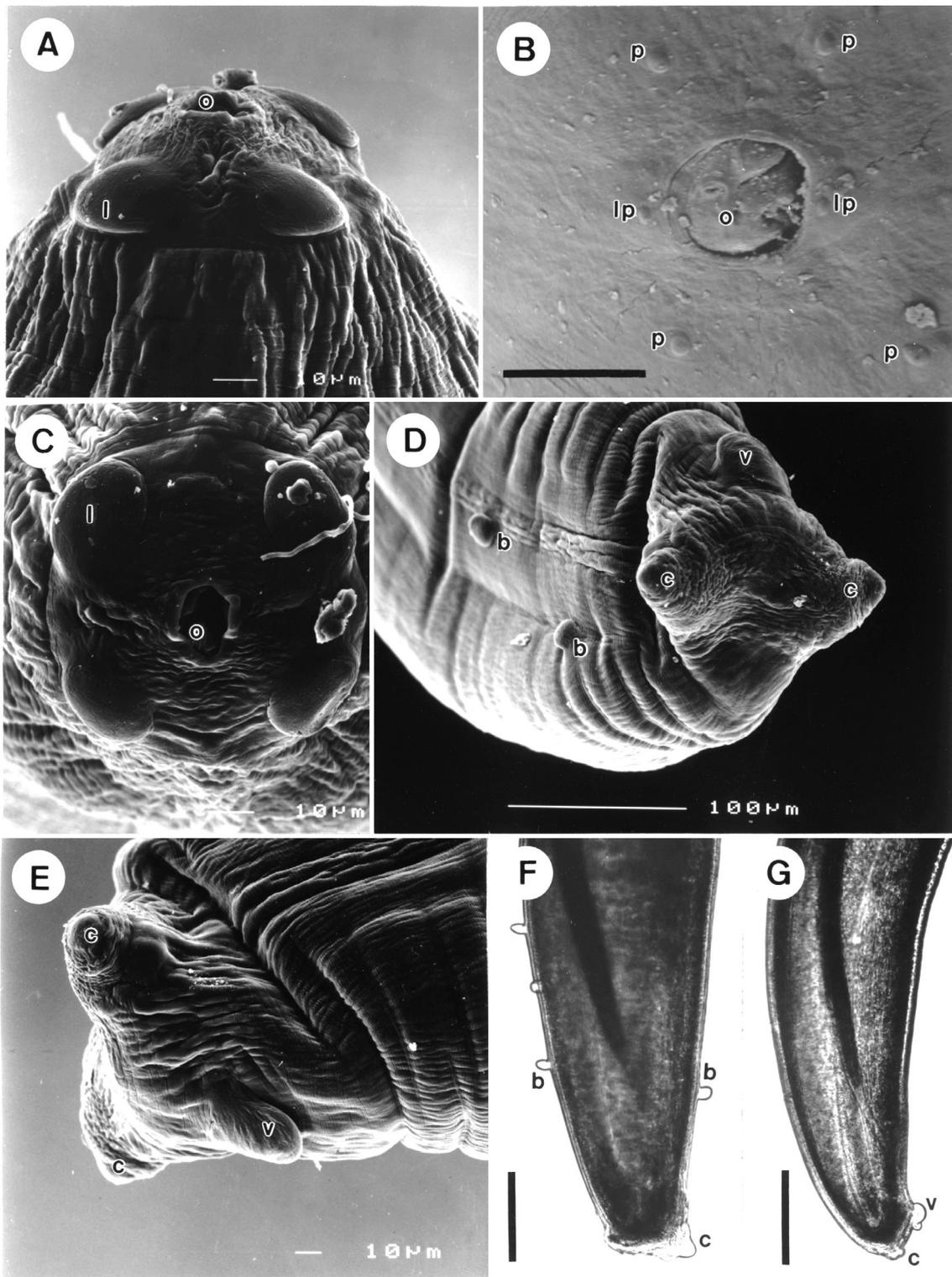


Fig. 2. *Philometroides cyprini*. (A–E) Scanning electron micrographs of gravid female showing (A,C) cephalic end (lateral and apical views), (B) region of oral aperture (apical view) and (D,E) caudal end (subapical and subventral views). (F,G) Subdorsal and lateral views of the posterior part of the nematode (light microscope). Abbreviations: b = cuticular boss; c = dorsalolateral caudal projection; l = submedian cephalic lobe containing pair of outer papillae; lp = lateral cephalic papilla of inner circle; o = oral aperture; p = submedian cephalic papilla of inner circle; v = ventral caudal cuticular lobe. Scale bars: A,C,E = 10 µm; B = 30 µm; D = 100 µm; F,G = 500 µm

vulva were atrophied. The posterior end bore 2 large dorsolateral caudal projections 54 µm long, sometimes appearing to be doubled inside; a large cuticular lobe was present on the ventral side (Figs. 1D,E & 2D–G). The uterus was filled with first-stage larvae 383 to 560 µm long and 14 to 16 µm wide.

DISCUSSION

Because the cephalic papillae of most philometrids are difficult to observe by LM, these nematodes have sometimes been described as having no such papillae; therefore, the use of SEM is the only reliable means of studying the papillae (Moravec 2004). However, the *Philometroides* females parasitizing the skin of carp have not been previously studied by SEM, and for the most part their cephalic ends have not been observed by LM in an apical view. In fact, the 4 'papillae' reported in these nematodes by Vismanis (1966) and Nakajima (1976) are the submedian cephalic lobe-like elevations, each with a pair of small, submerged papillae of the external circle. As confirmed in this study, these elevations may be only poorly developed in some specimens and, consequently, difficult to observe. The papillae of the internal circle have not been observed at all by previous authors. The number and arrangement of cephalic papillae as found in this study is typical of many other philometrid species (Moravec 2004).

As confirmed by SEM, the posterior end of these female nematodes bears 2 caudal projections, as in the majority of other philometrid species in which these are present. According to Vismanis (1966), there are 2 smaller lateral projections of cuticular character present between the large caudal projections. However, in a later study (Vismanis 1967) he illustrated the caudal end bearing, in addition to the 2 large simple lateral caudal projections, a large ventral and a smaller dorsal projection. The present study has revealed the double inner structure of the dorsolateral caudal projection and the presence of a large ventral cuticular projection (see also Moravec 1994).

We have shown that the differences in descriptions of *Philometroides cyprini* and *P. lusiana* arose due to insufficient knowledge of the morphology of these nematodes and that both species are identical. The valid name is *Philometroides cyprini*, whereas *Philometra lusii*, *Philometra schikhobalovae*, *Philometra lusiana* and *Philometroides lusiana* are its junior synonyms.

It can be assumed that *Philometroides cyprini* was originally distributed in east Asia — with reports of the species occurrence in Japan (Ishii 1916, 1931), China (Chen 1973, Pan et al. 1990) and in the Russian Far East (Belous 1965) — and that it was introduced from there into Europe in the 1960s.

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