

HELMINTHOLOGIA, 52, 2: 113 - 117, 2015

***Philometroides khalili* n. sp., a new philometrid nematode (Philometridae) from the operculum of the cyprinid fish *Labeo rosae* in Zimbabwe**

F. MORAVEC¹, A. HALAJIAN², S. TAVAKOL², I. NYAGURA², W. J. LUUS-POWELL²

¹Institute of Parasitology, Biology Centre of the Academy of Sciences of the Czech Republic, Branišovská 31, 370 05 České Budějovice, Czech Republic, *E-mail: moravec@paru.cas.cz; ²Department of Biodiversity, University of Limpopo, Turfloop Campus, Private Bag X1106, Sovenga 0727, South Africa

Article info

Received October 20, 2014
Accepted December 15, 2014

Summary

A new nematode species, *Philometroides khalili* n. sp. (Philometridae), is described from female specimens recovered from the operculum of the freshwater cyprinid fish *Labeo rosae* Steindachner (Cyprinidae, Cypriniformes) caught in the Bubi River, Zimbabwe. Based on light and scanning electron microscopical examination, the new species mainly differs from the only other African congeneric species *P. africanus* Moravec et Van As, 2001 in the body shape (filiform), length of gravid female (56 – 71 mm), in possessing a large oesophageal gland with a conspicuously large cell nucleus and in the shape (transversely oval), size (up to 33 – 36 µm high) and distribution of cuticular bosses. From other congeners it differs by a combination of morphological and biometrical features
Keywords: parasitic nematode; Dracunculoidea; cyprinid fish; river; Africa

Introduction

Data on philometrid nematodes (Philometridae) parasitizing African freshwater fishes are rather scarce. Until now, only six valid nominal species, all described solely from females, have been reported, representing four different genera (Moravec *et al.*, 2009): *Afro-philometra hydrocyoni* (Fahmy, Mansour et El-Naffar, 1976) from the fins of *Hydrocynus forskahlii* (Cuvier) and *H. vittatus* Castelnau in Egypt and Kenya, *Nilonema gymmarchi* Khalil, 1960 from the swimbladder of *Gymnarchus niloticus* Cuvier (Gymnarchidae) in the Sudan, *Philometra bagri* (Khalil, 1965) from the subcutaneous tissue of *Bagrus bajad* (Forsskal) (Bagridae) in the Sudan, Egypt and Kenya, *P. lati* Moravec, Charo-Karisa et Jirků, 2009 and *P. spiriformis* Moravec, Charo-Karisa et Jirků, 2009 from the abdominal cavity and operculum, respectively, of *Lates niloticus* (Linnaeus) in Kenya, and *Philometroides africanus* Moravec et Van As, 2001 from gill arches and operculum of *Hepsetus odoe* (Bloch) (Hepsetidae) in Botswana (Khalil, 1960, 1965, 1969, 1974; Fahmy *et al.*, 1976; El-Naffar *et al.*, 1983; Moravec & Van As, 2001). *Philometra congolensis* Schuurmans Stekhoven, 1937 described from *Clarias* sp. in the then Belgian Congo (= Democratic Republic of Congo, former Zaire) proved to be a misidentification of *Eustrongylides* larvae (Moravec, 2006). Two unidentified species of

Philometroides Yamaguti, 1935 were recorded from *Labeo altivelis* Peters (Cyprinidae) in Zimbabwe (Khalil, 1974) and from *Lates niloticus* (Linnaeus) (Latidae) in Egypt (El-Naffar *et al.*, 1983). Boomker (1994) reported unidentified species of the Philometridae from *Schilbe intermedius* Rüppel (Schilbeidae), *Serranochromis meridianus* Jubb (Cichlidae) and *Synodontis zambezensis* Peters (Mochokidae) in South Africa (see also Khalil & Polling, 1997). During recent studies of fish parasites in Zimbabwe, a few female specimens of *Philometroides* were recovered from the opercula of *Labeo rosae* Steindachner (Cyprinidae) (Fig. 3). A subsequent examination showed that they represent a new species, which was previously reported by Khalil (1974) as *Philometroides* sp. in the congeneric host (see above). The new species is described herein.

Material and Methods

Fish were caught by gill nets in the Bubi River located in the middle Limpopo River basin, Zimbabwe. The nematodes were washed in physiological saline and then fixed either in 4 % formalin or 96 % ethanol. Only two of the four nematodes were intact and in a proper shape for morphological study. For light microscopical examination, the nematodes were cleared with glycerine. Drawings were made with the aid of a Zeiss drawing attachment. Anterior

and posterior ends of both available specimens were then used for scanning electron microscopy (SEM). They were postfixed in 1 % osmium tetroxide (in phosphate buffer), dehydrated through a graded acetone series, critical-point-dried and sputter-coated with gold; they were examined using a JEOL JSM-7401F scanning electron microscope at an accelerating voltage of 4 kV (GB low mode). The rest of the alcohol-fixed specimen was maintained in 96 % ethanol for a subsequent molecular examination. All measurements are in micrometres unless otherwise indicated. The fish

nomenclature adopted follows FishBase (Froese & Pauly, 2014).

Results

Family Philometridae Baylis et Daubney, 1926

Philometroides khalili n. sp.

Syn. *Philometroides* sp. Khalil, 1974.

(Figs. 1, 2)

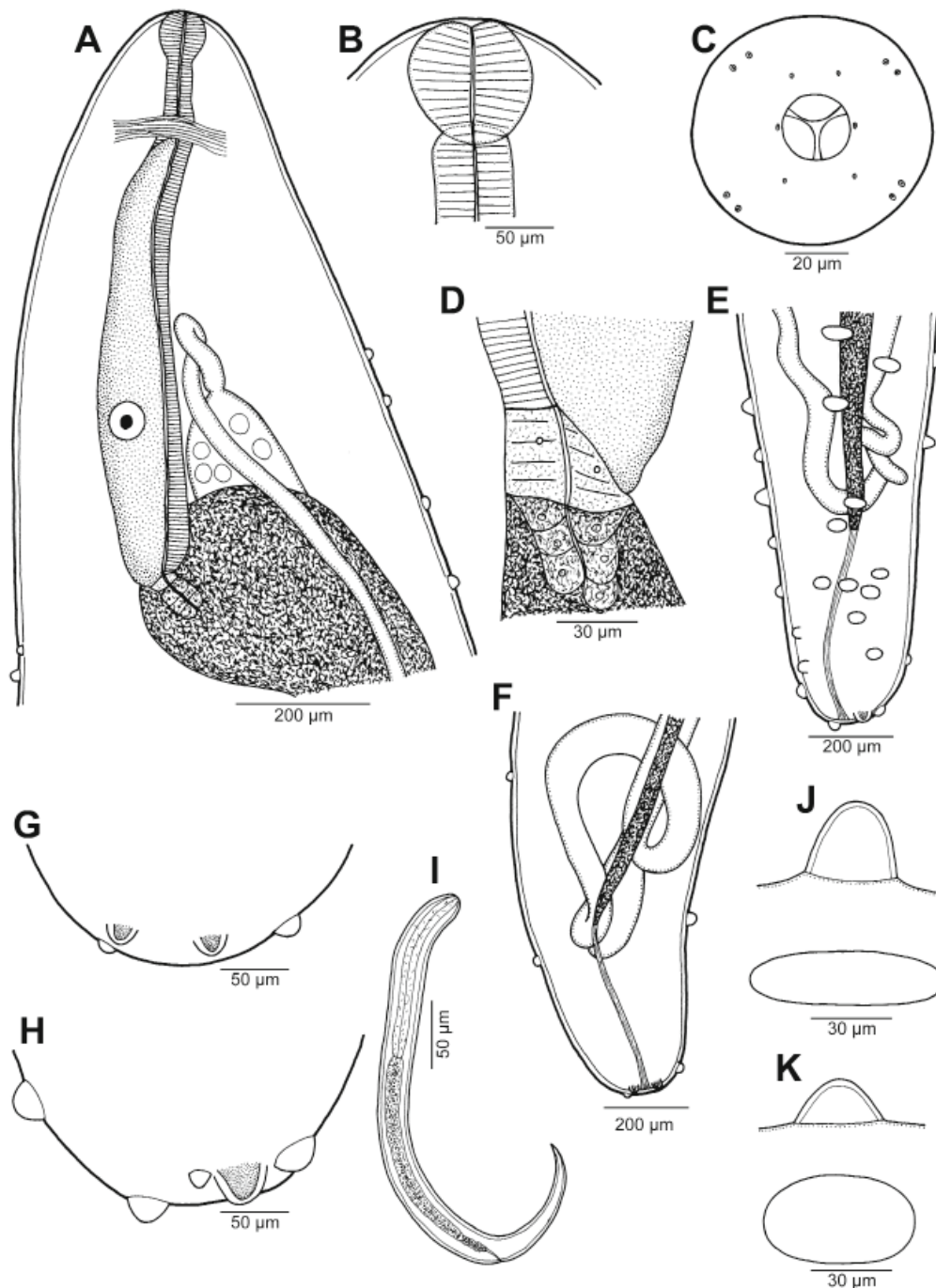


Fig. 1. *Philometroides khalili* n. sp., gravid female. A – anterior end of body (holotype), lateral view; B, C – cephalic end of paratype, lateral and apical views; D – oesophago-intestinal junction, lateral view (paratype); E – posterior end of body, lateral view (paratype); F – posterior end of body, dorsoventral view (holotype); G – caudal end, dorsoventral view (holotype); H – caudal end, lateral view (paratype); I – larva from uterus, lateral view; J, K – two different cuticular bosses, each in lateral and apical views

Description: Gravid female (1 complete holotype and 1 incomplete paratype specimens; measurements of latter in parentheses): Body of fixed specimens yellowish, filiform, 71 (not established) mm long and 1,224 (898) in maximum width; maximum width/length ratio of body in holotype 1: 61. Width of cephalic end 231 (340), that of caudal end 272 (272). Cuticle of nearly entire body bearing numerous, irregularly scattered bosses 15 – 36 (15 – 27) high and transversely oval to almost circular in apical view; bosses absent from short anterior part of body at oesophageal region (Figs. 1A,E – H,J,K, 2D – G). Cephalic end rounded, cephalic papillae indistinct in lateral view (Fig. 1A,B). Oral aperture circular, relatively small. Bottom of mouth formed by three flat oesophageal lobes. Cephalic papillae very small, arranged in two circles:

external circle formed by four pairs of submedian papillae, internal circle consisting of six papillae (four submedian and two lateral) surrounding oral aperture. Lateral amphids indistinct (Figs. 1C, 2A – C). Oesophagus muscular, inflated at anterior end to form distinct bulb, 1.17 (1.32) mm long, representing 1.6 % (-) of body length; maximum width of oesophagus including oesophageal gland 171 (136). Oesophageal bulb spherical, 87 (90) long and 102 (81) wide. Dorsal oesophageal gland large, starting at level of nerve ring and extending posteriorly to end of oesophagus; markedly large nucleus of oesophageal gland situated 830 (993) from anterior end of body (Fig. 1A). Small ventriculus 24 (24) long and 45 (45) wide, opening into intestine through valve (Fig. 1D). Nerve ring 218 (299) from anterior extremity. Intestine light-

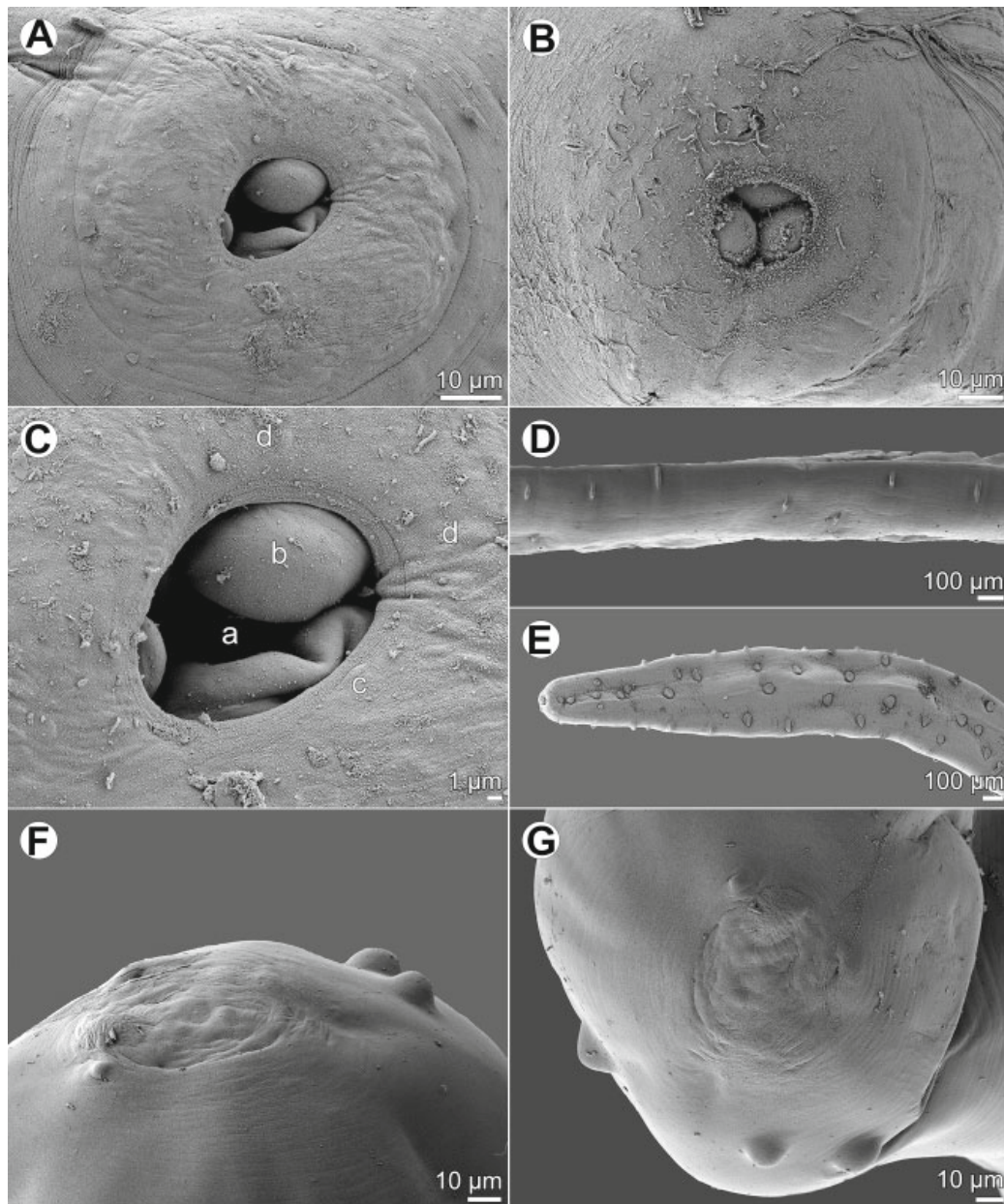


Fig. 2. *Philometroides khalili* n. sp., scanning electron micrographs of gravid female. A, B – cephalic end of holotype and paratype, respectively, apical views, C – region of oral aperture, apical view, D, E – middle and posterior parts of body, respectively, lateral views, F, G – caudal end of holotype specimen, subapical and apical views, respectively.
Abbreviations: a – oral aperture, b – dorsal anterior lobe of oesophagus, c – lateral cephalic papilla of internal circle, d – submedian cephalic papilla of external circle

coloured, straight, displaced laterally by uterus; posterior end of intestine atrophied, forming short ligament 105 (99) long attached to posterior extremity. Posterior end of body rounded, with two small, subterminal papilla-like caudal projections (Figs. 1E – H, 2F,G). Ovaries long, narrow, situated near anterior and posterior body ends (Fig. 1A,E,F). Uterus occupying major part of body, filled with numerous larvae and eggs. Larvae (n = 5) in holotype 384 – 429 long, maximum width 21 – 24; oesophagus 123 – 141 long, representing 33 – 38 % of body length; length of tail 87 – 96, representing 21 – 23 % (Fig. 1I).

Male: Unknown.

Taxonomic summary

Type host: Rednose laboe *Labeo rosae* Steindachner (Cyprinidae, Cypriniformes), standard body length 22.2 – 26.5 cm.

Site of infection: Operculum (outer side).

Type locality: Bubi River, Drummond Ranch (20°40'52.40" S, 28°22'48.60" E), Zimbabwe (collected 15 July 2014).

Prevalence and intensity: 14 % (3 fish infected/21 fish examined); 1 – 2 nematode specimens.

Type specimens: Body ends of holotype and paratype mounted on SEM stubs and the middle part of holotype body preserved in ethanol in the Helminthological Collection of the Institute of Parasitology, Biology Centre, Academy of Sciences of the Czech Republic, in České Budějovice (Cat. No. N-1067).

Etymology: This species has been named in honour of the well-known helminthologist Dr. Lotfi F. Khalil, who was the first to discover this *Philometroides* species.

Discussion

In having many irregularly scattered cuticular bosses on the body surface, the present specimens belong to the genus *Philometroides* Yamaguti, 1935 (see Rasheed, 1963; Moravec, 2006). At present this genus includes 31 valid species (Moravec & Manoharan, 2013), which are tissue-dwelling parasites of marine, brackish-water and freshwater fishes. Most of them were de-

scribed only from females, whereas conspecific males are known for eight species. As other philometrids, *Philometroides* spp. are known to exhibit a relatively high degree of host specificity, and individual species are characterized, in addition to morphological features and molecular sequence data, by the location of gravid females in the host (Moravec, 2006; Moravec & de Buron, 2013).

To date, the only valid species of *Philometroides* reported from a freshwater fish in Africa is *P. africanus* Moravec et Van As, 2001, described solely from females found in the gill arches and operculum of the African pike *Hepsetus odoe* (Bloch) (Hepsetidae, Characiformes) in Botswana (Duba Lagoon in the Okavango River delta) (Moravec & Van As, 2001). *Philometroides hydrocyoni* Fahmy, Mansour et El-Naffar, 1976 described from the fins of *Hydrocynus forskahlii* (Cuvier) and *H. vittatus* Castelnau (syn. *H. lineatus*) in Egypt (Fahmy et al., 1976) was subsequently transferred to the genus *Afrophilometra* Moravec, Charo-Karisa et Jirků, 2009 (see Moravec et al., 2009). Two unidentified species of *Philometroides* were recorded from *Labeo altivelis* (Cyprinidae) in Zimbabwe (Khalil, 1974) and from *Lates niloticus* (Latidae) in Egypt (El-Naffar et al., 1983) (see above).

In contrast to *Philometroides* sp. from *Lates niloticus* reported by El-Naffar et al. (1983), Khalil (1974) gave a short description and illustration of the only available gravid *Philometroides* sp. female with a damaged caudal end, collected from the operculum of *Labeo altivelis* in the Banyati River, Zimbabwe. According to his data, the specimen was filiform, 55.9 mm long and 750 µm in the maximum width, noted for the presence of a large oesophageal gland. Its general morphology was practically the same as that of specimens of the present material from *L. rosae*, except for the length of first-stage larvae from uterus; while Khalil (1974) reported the larval body length to be 890-960 µm, the larvae of one (holotype) of the present specimens were only 384-429 µm long. Unfortunately, Khalil's specimen is no longer available for a re-examination, but it is apparent that the reported length of larvae was given erroneously, because none of the known philometrid species has such big larvae (nearly 1 mm long!) (see Moravec 2006), which could be hardly swallowed up by presumed copepod intermediate hosts.

Taking into account the morphological and biometrical similarity of the present specimens with those of *Philometroides* sp. of Khalil (1974) and since both these forms had the same location in the host (operculum) and were collected from the congeneric hosts in nearby regions, we consider them to belong to the same new species, *P. khalili* n. sp. This new species is morphologically very different from the only other African *Philometroides* species, *P. africanus* (see above). Whereas the body of the gravid female of *P. khalili* n. sp. is filiform, cylindrical, about 56 – 71 mm long, that of *P. africanus* is fusiform, plump, only about 8–9 mm long. The oesophageal gland of the former species is large, extending from the level of the nerve ring to the end of oesophagus, whereas that of the latter is small, poorly demarcated, just below middle of the cylindrical portion of the oesophagus. Conspicuous interspecific differences are also in the embossment of the body: bosses of *P. khalili* are large, up to 33 – 36 µm high, mostly transversely oval in apical view, whereas those of *P. africanus* are small, only 6 – 12 µm high, circular in apical view and they may be even



Fig. 3. Encapsulated *Philometroides khalili* female on the outer surface of operculum of *Labeo rosae*

absent in smaller gravid specimens. It should be also mentioned that the hosts of these two species belong to different fish orders (Cypriniformes vs. Characiformes).

By the body length of gravid female, the number and small size of cephalic papillae, the shape and arrangement of cuticular bosses, the shape of the oesophagus, the presence of markedly large oesophageal gland and cell nucleus or by the combination of these features, *P. khalili* n. sp. also differs from all other congeneric species (see Moravec, 2006; Moravec *et al.*, 2008, 2012a,b; Moravec & de Buron, 2009; Moravec & Manoharan, 2013).

Acknowledgements

We thank Ignace Nyagura, Hendrik E. Hattingh and Willem J. Smit for their assistance during the field work, Paul S. O. Fouché (University of Venda) for verifying the fish identity, and Biodiversity Research Chair (University of Limpopo) for funding the field survey. The authors' thanks are also due to the staff of the Laboratory of Electron Microscopy, Institute of Parasitology, Biology Centre of the AS CR, České Budějovice, for their technical assistance, and to Blanka Škoriková of the same Institute for help with illustrations. This study was partly supported by the Czech Science Foundation (Grant. No. P505/12G112) and by institutional support (RVO:60077344, Institute of Parasitology, BC AS CR). We are grateful to Di Drummond and Keith Sparks for permission to work at Drummond Ranching and their keen interest in fish research.

References

- BOOMKER, J. (1994): Parasites of South African freshwater fishes. VI. Nematode parasites of some fish species in the Kruger National Park. *Onderstepoort J. Vet. Res.*, 61: 35 – 43
- EL-NAFFAR, M. K., SAOUD, M. F., HASSAN, I. M. (1983): A general survey of helminth parasites of some fishes of Lake Nasser at Aswan, A. R. Egypt. *Assiut Vet. Med. J.*, 11: 141 – 183
- FAHMY, M. A. M., MANDOUR, A. M., EL-NAFFAR, M. K. (1976): On some nematodes parasites from the freshwater fishes in Assiut Province, Egypt. *Vet. Med. J., Egypt*, 24: 263 – 276
- FROESE, R., PAULY, D. (Eds) (2014): *FishBase*. World Wide Web electronic publication. <http://www.fishbase.org>, version 10/2014
- KHALIL, L. F. (1960): On a new nematode, *Nilonema gymnarchi* gen. et sp. nov., (Dracunculidae), from a freshwater fish in the Sudan. *J. Helminthol.*, 34: 55 – 58. DOI: 10.1017/S0022149X00020344
- KHALIL, L. F. (1965): On a new philometrid nematode, *Thwaitia bagri* sp. nov., from a freshwater fish in the Sudan. *J. Helminthol.*, 39: 309 – 312. DOI: 10.1017/S0022149X00020733
- KHALIL, L. F. (1969): Studies on the helminth parasites of freshwater fishes of the Sudan. *J. Zool. (Lond.)*, 58: 143 – 170. DOI: 10.1111/j.1469-7998.1969.tb02132.x
- KHALIL, L. F. (1974): Some nematodes from the freshwater fishes of Rhodesia with the description of a new species *Cithariniella petterae* n. sp. *Ann. Parasitol. Hum. Comp.*, 48: 811 – 818
- KHALIL, L. F., POLLING, L. (1997): *Check list of the helminth parasites of African freshwater fishes. Second edition*. University of the North, Pietersburg, 185 pp
- MORAVEC, F. (2006): *Dracunculoid and anguillicoloid nematodes parasitic in vertebrates*. Academia, Prague, 634 pp
- MORAVEC, F., CHARO-KARISA, H., JIRKŮ, M. (2009): Philometrids (Nematoda: Philometridae) from fishes of Lake Turkana, Kenya, including two new species of *Philometra* and erection of *Afrophilometra* gen. n. *Folia Parasitol.*, 56: 41 – 54. DOI: 10.14411/fp.2009.008
- MORAVEC, F., CROSBY, M. D., DE BURON, I., GONZÁLEZ-SOLÍS, D., ROUMILLAT, W. A. (2008): Three new species of philometrids (Nematoda: Philometridae) from centrarchid fishes in the USA. *J. Parasitol.*, 94: 1103 – 1113. DOI: 10.1645/GE-1523.1
- MORAVEC, F., DE BURON, I. (2009): Two new species of philometrids (Nematoda: Philometridae) from marine fishes off South Carolina. *J. Parasitol.*, 95: 722 – 727. DOI: 10.1645/GE-1866.1
- MORAVEC, F., DE BURON, I. (2013): A synthesis of our current knowledge of philometrid nematodes, a group of increasingly important fish parasites. *Folia Parasitol.*, 60: 81 – 101. DOI: 10.14411/fp.2013.010
- MORAVEC, F., JASSIM, A.A.R., AL-SALIM, N.K. (2012a): *Philometroides acanthopagri* sp. nov., a new philometrid (Nematoda, Philometridae) from the musculature of *Acanthopagrus latus* (Sparidae) from marine waters of Iraq. *Acta Parasitol.*, 57: 372 – 377. DOI: 10.2478/s11686-012-0044-3
- MORAVEC, F., MANOHARAN, J. (2013): Gonad-infecting philometrids (Nematoda: Philometridae) including four new species from marine fishes off the eastern coast of India. *Folia Parasitol.*, 60: 105 – 122. DOI: 10.14411/fp.2013.012
- MORAVEC, F., NAGASAWA, K., NOHARA, K. (2012b): Two species of philometrid nematodes (Philometridae) from marine fishes off Japan, including *Philometroides branchiostegi* sp. n. from *Branchiostegus japonicus* (Malacanthidae). *Folia Parasitol.*, 59: 71 – 78. DOI: 10.14411/fp.2012.011
- MORAVEC, F., VAN AS, J. G. (2001): *Philometroides africanus* sp. n. (Nematoda: Philometridae), a new tissue parasite of the African pike *Hepsetus odoe* (Pisces) in Botswana. *Folia Parasitol.*, 48: 127 – 131. DOI: 10.14411/fp.2001.019
- RASHEED, S. (1963): A revision of the genus *Philometra* Costa, 1845. *J. Helminthol.*, 37: 89 – 130. DOI: 10.1017/S0022149X00019672