



New species of Dactylogyridae Bychowsky, 1933 infecting the gills of *Myloplus schomburgkii* (Jardine) and *Colossoma macropomum* (Cuvier) in the Peruvian Amazon

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Abstract Four new dactylogyrid species are described, three species (*Anacanthorus camposbacae* n. sp., *Anacanthorus carmenrosae* n. sp. and *Notozothecium nanayensis* n. sp.) from the gills of *Myloplus schomburgkii* (Jardine), captured in the River Nanay, Iquitos, Peru, and one new species (*Mymarothecium iiapensis* n. sp.) from the gills of *Colossoma macropomum* (Cuvier) in a fish pond in Iquitos, Peru. *Anacanthorus camposbacae* n. sp. differs from its congeners by possessing the accessory piece with a sub-medial branch having tentacle-like extensions. *Anacanthorus carmenrosae* n. sp. is characterised by possessing a Y-shaped accessory piece.

Notozothecium nanayensis n. sp. differs from its congeners by the shape of both anchors and the presence of a short anteromedial triangular process in the ventral bar and by possessing an accessory piece with spoon-shaped proximal end. *Mymarothecium iiapensis* n. sp. is characterised by presenting a wrench-tool-shaped accessory piece and by possessing both anchors with conspicuous fold on the superficial roots.

Introduction

Myloplus schomburgkii (Jardine), popularly known in Peru as “banda negra” is a species of Serrasalminae (Characiformes) widespread in the middle and lower Amazon River basin, Nanay River and upper Orinoco River basin in South America (Jégu, 2003). This fish is considered as a potential species for the diversification of aquaculture production in the Peruvian Amazon (García et al., 2012). *Colossoma macropomum* (Cuvier), popularly known in Peru as “gamitana” is a serrasalminid characiform fish species native to tropical South America. It is widely distributed in the basins of the Orinoco and Amazon rivers, and is one of the species of greatest importance for aquaculture in the Amazon (Ribeiro-Neto et al. 2016).

Several studies involving monogeneans from cultured fish have been conducted in the Peruvian Amazon region (Mathews et al., 2012, 2013a, b, 2014a, b; Soberon et al., 2014; Gonzales et al., 2016)

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but to date, no new species have been discovered from fish cultivated in fish farms in the Peruvian Amazon. The present study reports two new species of *Anacanthorus* Mizelle & Price, 1965 and one new species of *Notozothecium* Boeger & Kritsky, 1988 from *M. schomburgkii* collected in the River Nanay and one new species of *Mymarothecium* Kritsky, Boeger & Jégu, 1996 from *C. macropomum* cultivated in fish ponds. Herein, the new species are described.

Materials and methods

Ten specimens of *M. schomburgkii* (standard length 21.2 ± 5.3 cm) and 20 *C. macropomum* (standard length 30.2 ± 2.1 cm) were collected in July 2018. Specimens of *M. schomburgkii* were collected in the River Nanay and specimens of *C. macropomum* were collected in a fish pond located in the “Centro de Investigaciones Fernando Alcántara Bocanegra” (CIFAB) of the “Instituto de Investigaciones de la Amazonía Peruana” (IIAP), in Iquitos, Peru. Fish were sacrificed and gill arches were removed and placed in vials containing heated water (68°C). Each vial was shaken vigorously, and 96% ethanol was added.

In the laboratory, the content of each vial was examined using a dissecting microscope, and monogeneans were removed from the gills or sediment using dissection needles. Some specimens were cleared with Hoyer's medium for identification based on the morphology of the sclerotised structures and others were stained with Gomori's trichrome (Humason, 1979; Boeger & Viana, 2006) and mounted in Dammar gum to determine internal soft structures.

Sclerotised structures of the new species were photographed with a camera (Axiocam RC 5s) connected to a Zeiss Primo Star microscope, and the images were used to obtain the measurements of the body, male copulatory organ (MCO) and haptoral sclerites: anchors, bars, hooks and 4A hooks (4A's). Measurements were made following the procedures of Mizelle & Klucka (1953). Measurements are in micrometers and represent straight-line distances between extreme points of the structures measured, and are expressed as the range followed by the mean and number (n) of structures measured in parentheses. Illustrations were prepared with the aid of a microprojector.

Type-specimens were deposited in the Helminthological Collection of the Museum of Natural History at

the San Marcos University (MUSM) Lima, Peru, and in the Zoological Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA) Manaus, Brazil.

Family Dactylogyridae Bychowsky, 1933 Genus *Anacanthorus* Mizelle & Price, 1965

Anacanthorus camposbaeae n. sp.

Type-host: *Myloplus schomburgkii* Jardine (Characiformes: Serrasalminidae)

Type-locality: River Nanay (3°50'42"S, 73°23'12"W), Community of Ninarumi, Iquitos, Peru.

Type-material: The holotype (MUSM 3881) and 3 paratypes (MUSM 3881a-c) were submitted to the Helminthological Collection of the Museum of Natural History at the San Marcos University (MUSM) Lima, Peru, and 6 paratypes (INPA 796 a-f) were submitted to the Zoological Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA) Manaus, Brazil.

Site in host: Gill filaments.

Infection parameters: Prevalence: 40% of 15 hosts examined; mean intensity: 5.25 parasites per infected host; mean abundance: 2.1 parasites per host.

Etymology: The specific name is in honor of Dr Luis Campos Baca, Instituto de Investigaciones de la Amazonía Peruana, in recognition of his valuable work in the Peruvian Amazon.

Description (Fig. 1)

[Based on 10 specimens: 4 stained, 6 cleared.] Body elongate, 773–804 (787; n = 10) long, greatest width 150–240 (180; n = 10). Cephalic lobes 4 (2 terminal and 2 bilateral), well developed. Head organs 3 pairs, lying in cephalic lobes; cephalic glands not observed. Eyes 2 pairs, anterior pair smaller than posterior pair; granules elongate ovate, small. Pharynx muscular, subspherical, 39–48 (42; n = 10) wide; oesophagus short; intestinal caeca 2, confluent posteriorly, lacking diverticula. Peduncle long, narrow; haptor short, bilobate 139–161 (149; n = 10) long, 91–144 (121) wide. Hooks 7 pairs (4 ventral and 3 dorsal), similar in shape, each with slightly depressed thumb, shank expanded proximally; slightly curved shaft; hook pairs 1, 4, 7: 19–31 (25; n = 10 per hook pair) long; hook pairs 2, 3, 5, 6: 17–28 (22; n = 10 per hook pair) long; filamentous hooklet (FH) loop approximately 3/4 of

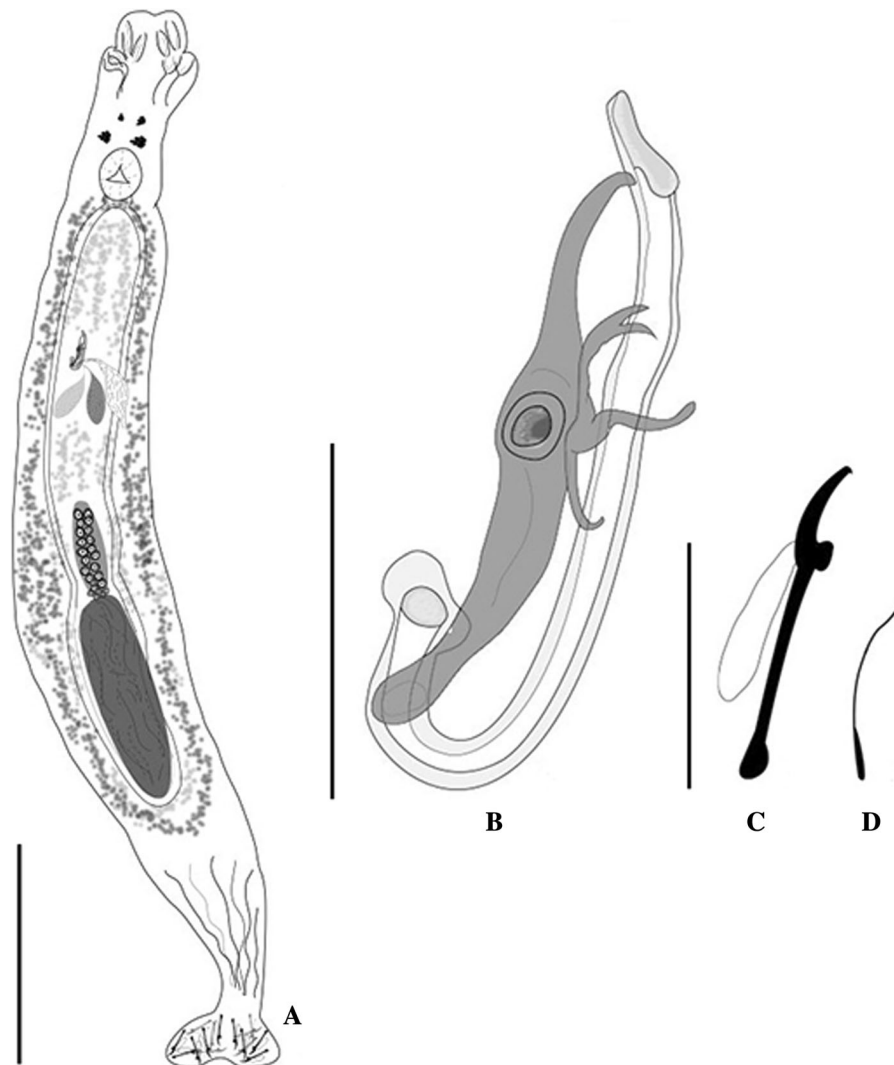


Fig. 1 *Anacanthorus camposbaecae* n. sp. A, Holotype, whole-mount, ventral view; B, Copulatory complex; C, Hook; D, 4A hook. Scale-bars: A, 200 µm; B, 50 µm; C, D, 25 µm

shank. 4A's similar, each 8–12 (10; n = 6 per hook pair) long. Testis elongate, post-germinal; germarium elongate, approximately 1/4 the size of testis; seminal vesicle elongate; prostatic reservoirs 2, pyriform. Copulatory complex comprising male copulatory organ (MCO) and accessory piece. MCO 82–90 (86; n = 10) long, J-shaped, with sub-terminal aperture and slightly sclerotised walls on both sides. Accessory piece 70–82 (78; n = 10) long, non-articulated with MCO, with recurved tip and submedial tentacle-like extensions at distal end. Eggs, vagina, oötype and

uterus not observed. Vitellarium dense, coextensive with caeca.

Remarks

The new species is allocated to *Anacanthorus* Mizelle & Price, 1965 due to the lack of anchors and bars and the possession of 7 pairs of haptor hooks (4 ventral and 3 dorsal). *Anacanthorus camposbaecae* n. sp. resembles *Anacanthorus carmenrosae* n. sp. in the shape of the body and the distribution of the internal organs but can be differentiated by the structure and

shape of the accessory piece, which presents a submedial branch with tentacle-like extensions (vs Y-shaped).

***Anacanthorus carmenrosae* n. sp.**

Type-host: *Myloplus schomburgkii* Jardine (Characiformes: Serrasalminidae).

Type-locality: River Nanay (3°50'42"S, 73°23'12"W), Community of Ninarumi, Iquitos, Peru.

Type-material: Holotype, MUSM 3882; 3 paratypes, MUSM 3882 a-c; 6 paratypes were submitted to the Helminthological Collection of the Museum of Natural History at the San Marcos University (MUSM) Lima, Peru, and 6 paratypes, INPA 800 a-f were submitted to the Zoological Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA) Manaus, Brazil.

Site in host: Gill filaments

Infection parameters: Prevalence: 40% of 15 hosts examined, mean intensity: 3.0 parasites per infected host; mean abundance: 1.2 parasites per host.

Etymology: The specific name is in honor of Dr Carmen Rosa García Dávila, Instituto de Investigaciones de la Amazonía Peruana, in recognition of her valuable work in the Peruvian Amazon.

Description (Fig. 2)

[Based on 10 specimens: 4 stained, 6 cleared.] Body elongate, 812–946 (864; n = 10) long, greatest width 202–280 (234; n = 10). Cephalic lobes 4, well developed, 2 terminal and 2 bilateral. Head organs 3 pairs, lying in cephalic lobes; cephalic glands not observed. Eyes 2 pairs, anterior pair smaller than posterior pair; granules elongate ovate, small. Pharynx muscular, subspherical, 30–33 (32; n = 10) wide; oesophagus short; intestinal caeca 2, confluent posteriorly, lacking diverticula. Peduncle short; haptor short, bi-lobate 146–171 (159; n = 10) long, 168–198 (178; n = 10) wide. Hooks 7 pairs (4 ventral and 3 dorsal), similar in shape, each with slightly depressed thumb, shank expanded proximally; slightly curved shaft; hook pairs 1, 7: 39–44 (43; n = 10 per hook pair) long; hook pairs 2–6: 41–51 (49; n = 10 per hook pair) long; filamentous hooklet (FH) loop approximately 3/4 of shank. 4A's similar, each 18–22 (20; n = 6 per hook pair) long. Testis elongate,

post-germinal; germarium circular; seminal vesicle elongate; prostatic reservoirs 2, pyriform. Copulatory complex comprising MCO and accessory piece. MCO 81–103 (92; n = 10) long, J-shaped, presenting slightly sclerotised walls on both sides, with a kind of rough coat covering the base. Accessory piece 60–85 (75; n = 10) long, non-articulated, Y-shaped, with one extreme longer than the other; longer extreme with a sharp termination. Eggs, vagina, oötype and uterus not observed. Vitellarium dense, coextensive with caeca.

Remarks

The new species is allocated to *Anacanthorus* due to the lack of anchors and bars and the possession of 7 pairs of haptor hooks (4 ventral and 3 dorsal). *Anacanthorus carmenrosae* n. sp. resembles *Anacanthorus acuminatus* Kritsky, Boeger & Van Every, 1992 and *Anacanthorus chaunophallus* Kritsky, Boeger & Van Every, 1992 in having distal branches in the accessory piece, giving it the shape of the letter "Y". The new species differs from the mentioned species in having only two branches in the accessory piece while *A. acuminatus* and *A. chaunophallus* present three branches.

Genus *Notozothecium* Boeger & Kritsky, 1988

***Notozothecium nanayensis* n. sp.**

Syn. *Notozothecium bethae* Boeger & Kritsky, 1988 of Gonzáles et al. (2015)

Type-host: *Myloplus schomburgkii* Jardine (Characiformes: Serrasalminidae).

Type-locality: River Nanay (3°50'42"S, 73°23'12"W), Community of Ninarumi, Iquitos, Peru.

Type-material: Holotype, MUSM 3883; 3 paratypes, MUSM 3883a-c; 6 paratypes were submitted to the Helminthological Collection of the Museum of Natural History at the San Marcos University (MUSM) Lima, Peru, and six paratypes, INPA 799 a-f were submitted to the Zoological Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA) Manaus, Brazil.

Site in host: Gill filaments

Infection parameters: Prevalence: 47% of 15 hosts examined, mean intensity: 6.0 parasites per infected host; mean abundance: 3.0 parasites per host.

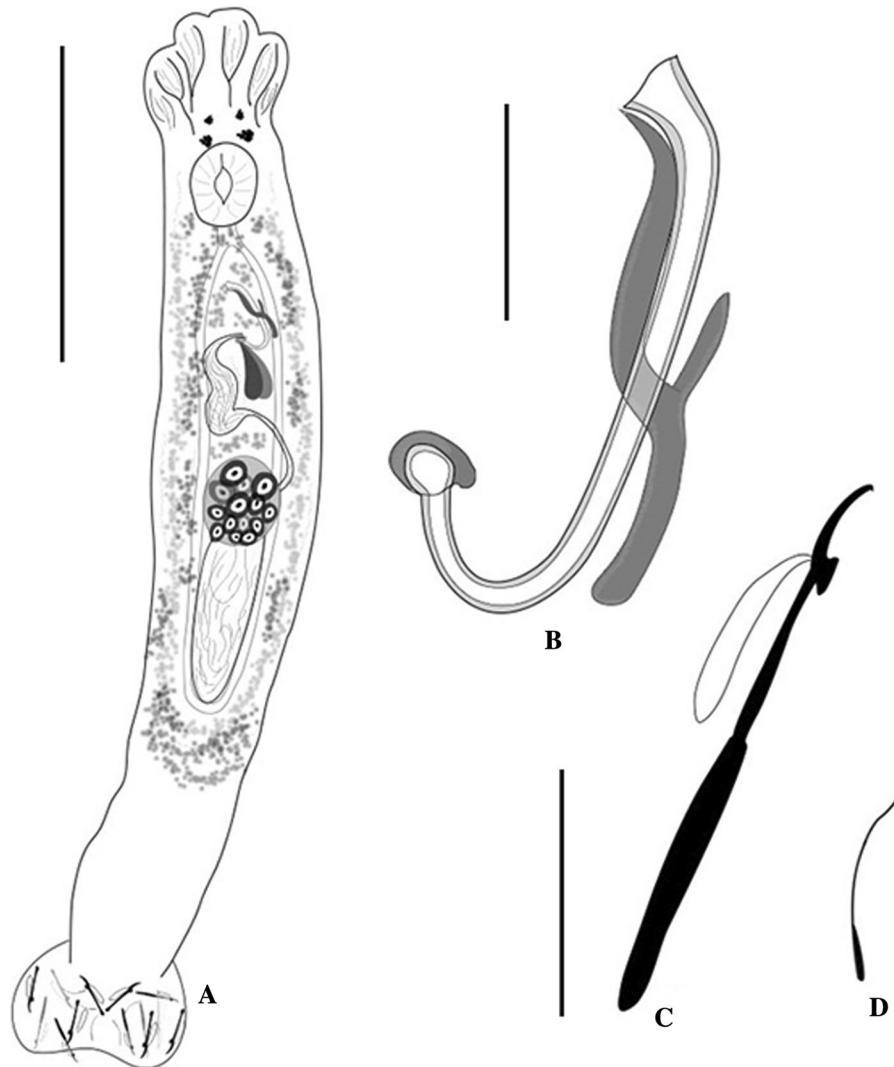


Fig. 2 *Anacanthorus carmenrosae* n. sp. A, Holotype whole-mount, ventral view; B, Copulatory complex; C, Hook; D, 4A. Scale-bars: A, 200 μ m; B, 30 μ m; C, D, 25 μ m

Etymology: The specific name is derived from the type-locality, River Nanay, where hosts were collected.

Description (Fig. 3)

[Based on 10 specimens: 3 stained and 7 cleared.] Body elongated, 381–645 (523; $n = 10$) long, greatest width 108–230 (152; $n = 10$). Cephalic lobes well developed, lateral lobes prominent. Head organs 4 pairs, lying in cephalic lobes; cephalic glands not observed. Eyes 2 pairs, anterior pair smaller than

posterior pair; accessory granules usually in cephalic region, granules elongate ovate, small. Pharynx muscular, subspherical, 20–35 (29; $n = 10$) wide; intestinal caeca 2, confluent posteriorly, lacking diverticula. Peduncle short and broad, haptor short subhexagonal 81–115 (94; $n = 10$) long, 100–196 (140; $n = 10$) wide. Ventral anchor 40–56 (50; $n = 10$) long, 27–36 (30; $n = 10$) wide, with prominent superficial and deep root, with distinct small hump on superficial root, curved shaft with moderately long point. Dorsal anchor 33–45 (41; $n = 10$) long, 20–27 (23; $n = 10$) wide, with elongate superficial root, short deep root, with distinct

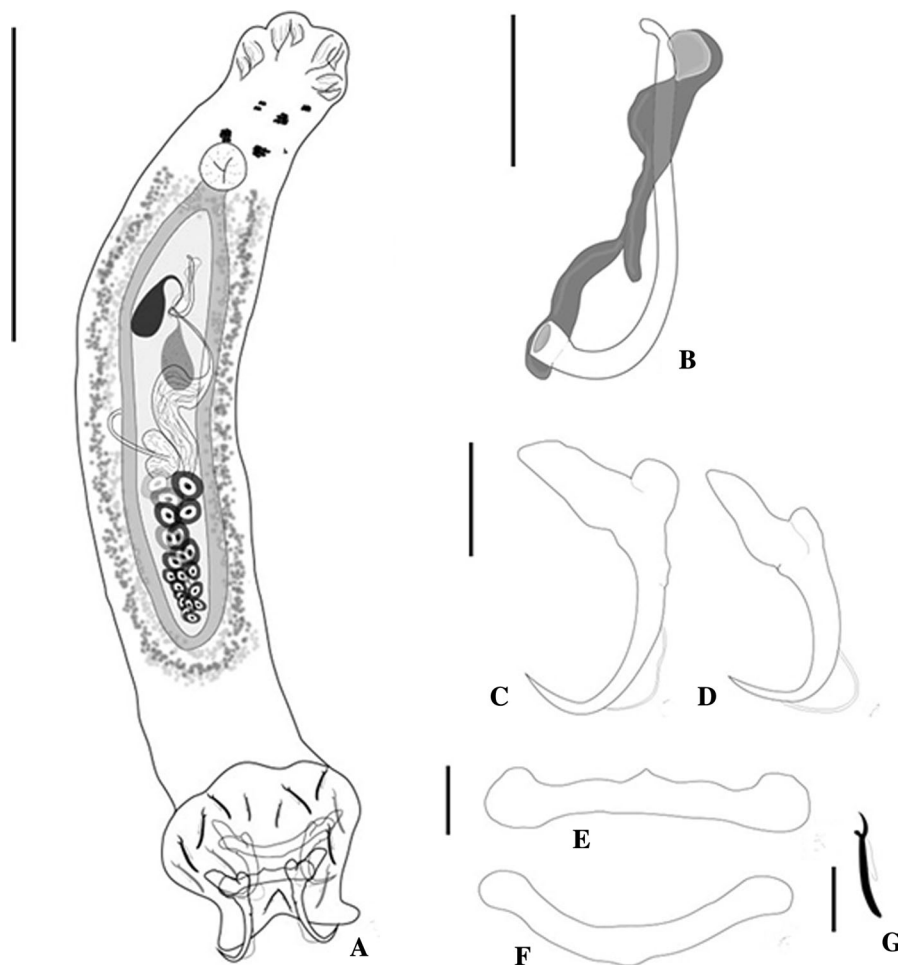


Fig. 3 *Notozothecium nanayensis* n. sp. A, Holotype whole-mount, ventral view; B, Copulatory complex; C, Ventral anchor; D, Dorsal anchor; E, Ventral bar; F, Dorsal bar; G, Hook. Scale-bars: A, 200 μm ; B–D, 20 μm ; E–F, 10 μm ; G, 15 μm

small hump on superficial surface of base, curved shaft with moderately long point. Ventral bar 47–71 (58; $n = 10$) long, 6–10 (8; $n = 10$) wide, yoke-shaped with enlarged terminations, short triangular anteromedial process. Dorsal bar 42–68 (55; $n = 10$) long, 7–10 (8; $n = 10$) wide, U-shaped, with slightly enlarged ends. Hooks 7 pairs (5 ventral and 2 dorsal), similar in shape, each with delicate point, protruding thumb, slightly curved shaft; hook pairs 1–7: 19–30 (25; $n = 10$ per hook pair) long; filamentous hooklet (FH) loop approximately half of shank. Testis ovate, dorsal to germarium, germarium conical; seminal vesicle elongate; prostatic reservoirs 2, pyriform. Copulatory complex comprising MCO and accessory piece. MCO 31–50 (39; $n = 10$) long, J-shaped. Accessory

piece 19–29 (41; $n = 10$) long, articulated with MCO by an elongate articulation process, distal rod long, straight, with spoon-shaped proximal end, with a prominent hump near its mid-length. Vagina delicate, flared into cone before entering kidney-shaped seminal receptacle. Eggs, oötype and uterus not observed. Vitellarium limited in trunk, absent in regions of reproductive organs.

Remarks

The new species is allocated to *Notozothecium* Boeger & Kritsky, 1988, based on the morphology of the male copulatory organ that is an elongate tube, accessory piece with proximal articulation process and distal rod,

ventral bar with anteromedial projection. The new species resembles *Notozothecium robustum* Kritsky, Boeger & Jégu, 1996 in having a J-shaped MCO and an accessory piece articulated with MCO by an elongate articulation process. However, *N. nanayensis* n. sp. differs from *N. robustum* by the shape of both anchors and mainly by the presence of a short triangular anteromedial process of the MCO, while in *N. robustum*, it is rod-shaped. *Notozothecium nanyensis* n. sp. also resembles *N. bethae* in the morphology of both bars but differs in the shape of the anchors and also in the shape of the MCO (a J-shaped tube vs a delicate coiled tube, comprising about 1¼ ring in *N. bethae*).

Genus *Mymarothecium* Kritsky, Boeger & Jégu, 1996

Mymarothecium iiapensis n. sp.

Type-host: *Colossoma macropomum* (Cuvier) (Characiformes: Serrasalminidae).

Type-locality: Fish pond, located in CIFAB, IIAP, Iquitos, Perú (3°49.1'11"S, 73°19.20'31"W)

Type-material: Holotype, MUSM 3884; 2 paratypes, MUSM 3883a-b were submitted to the Helminthological Collection of the Museum of Natural History at the San Marcos University (MUSM) Lima, Peru, and 4 paratypes, INPA 798 a-d, were submitted to the Zoological Collection of the Instituto Nacional de Pesquisas da Amazônia (INPA) Manaus, Brazil.

Site in host: Gill filaments

Infection parameters: Prevalence: 10% of 20 hosts examined, mean intensity: 21.5 parasites per infected host; mean abundance: 2.15 parasites per host.

Etymology: The specific name is derived from the type-locality, Instituto de Investigaciones de la Amazonía Peruana (IIAP) where the parasite was found.

Description (Fig. 4)

[Based on 10 specimens: 4 stained, 6 cleared.] Body short, robust, 215–311 (264; n = 15) long, greatest width 100–74 (89; n = 15). Cephalic lobes well developed, lateral lobes prominent. Head organs 4 pairs, lying in cephalic lobes; cephalic glands not observed. Eyes 2 pairs, anterior pair smaller than posterior pair; accessory granules usually in cephalic

region, granules elongate, ovate, small. Pharynx muscular, subspherical, 23–16 (18; n = 15) wide; intestinal caeca 2, confluent posteriorly, lacking diverticula. Peduncle short and broad, haptor short, subhexagonal 56–39 (47; n = 15) long, 96–50 (72; n = 15) wide. Anchors similar, each with well-differentiated robust superficial root, with conspicuous fold on superficial roots, short deep root, curved shaft, elongated point. Ventral anchor 22–26 (24; n = 10) long, 11–15 (13; n = 10) wide. Dorsal anchor 20–25 (23; n = 10) long, 14–12 (13; n = 10) wide. Ventral bar 41–37 (39; n = 10) long, 3.1–3.9 (3.5; n = 10) wide, arcuate, with expanded ends, short cylindrical posteromedial process present. Dorsal bar 36–38 (37; n = 10) long, 3.1–4.0 (3.7; n = 10) wide, slightly arched. Hooks 7 pairs (5 ventral and 2 dorsal), similar in shape, each with delicate point, protruding thumb, shank expanded proximally; slightly curved shaft; hook pairs 1, 5: 9–14 (12; n = 10 per hook pair) long; hook pairs 2, 6: 16–20 (19; n = 10 per hook pair) long; hook pairs 3, 4, 7: 21–29 (26; n = 10 per hook pair) long; filamentous hooklet (FH) loop approximately half of shank. Testis subovate, dorsal to germarium; germarium elongate; seminal vesicle elongate; prostatic reservoirs 2, pyriform. Copulatory complex comprising MCO and accessory piece. MCO 24–34 (29; n = 10) long, J-shaped. MCO presenting slightly sclerotised walls on both sides. Accessory piece 37–47 (42; n = 10) articulated with MCO by long copulatory ligament, wrench-tool-shaped. Eggs, vagina, oötype and uterus not observed. Vitellarium dense in 2 lateral fields.

Remarks

The new species is allocated to *Mymarothecium* Kritsky, Boeger & Jégu, 1996 based on the presence of an articulated accessory piece, a tubular non-coiled male copulatory organ, and a medial projection in the ventral bar. The new species resembles *Mymarothecium whittingtoni* Kritsky, Boeger & Jégu, 1996 and *Mymarothecium ianwhittingtoni* Leão, São Clemente & Cohen, 2015, in having a J-shaped MCO and a hook-shaped distal end of the accessory piece. However, the new species differs from *M. whittingtoni*, *M. ianwhittingtoni* and other congeners by presenting a wrench-tool-shaped accessory piece and by possessing both anchors with conspicuous fold on the superficial roots.

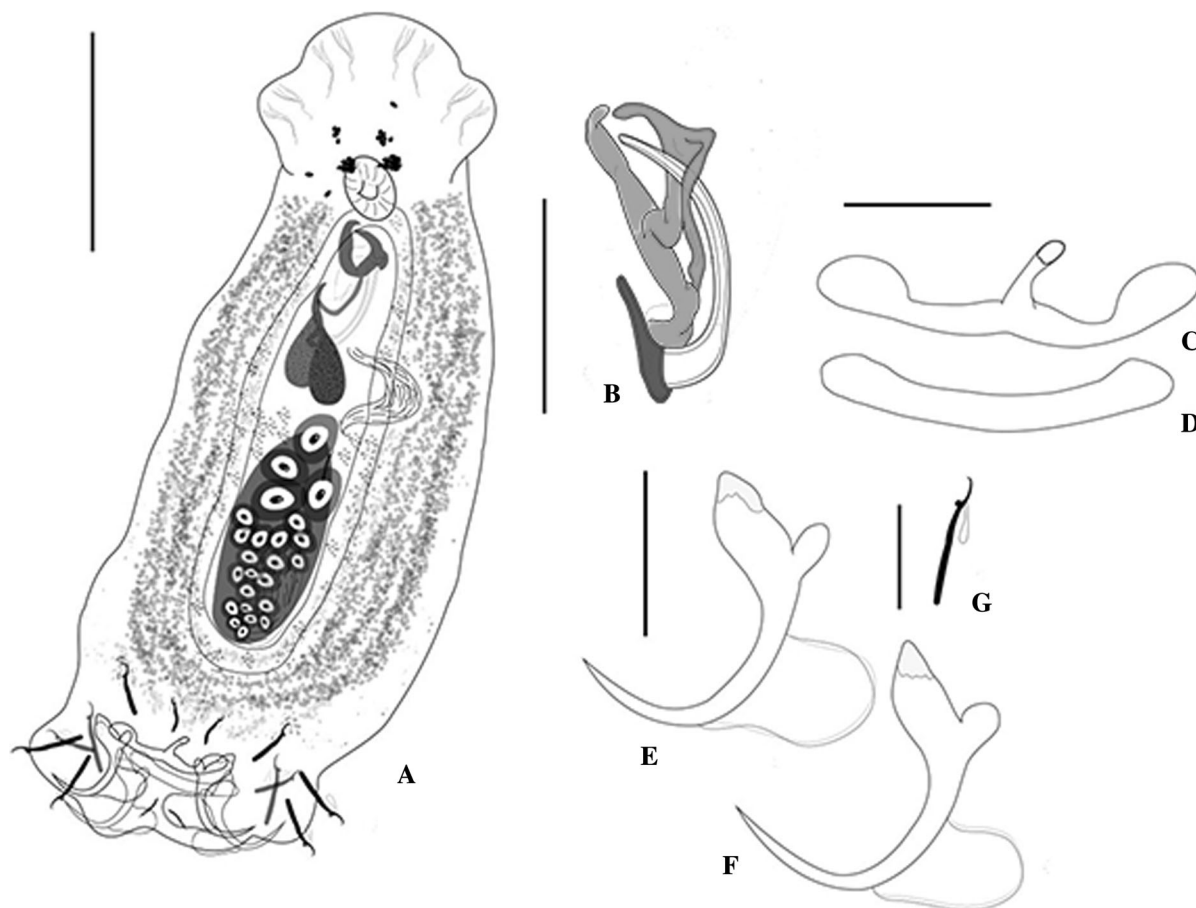


Fig. 4 *Mymarothecium iiapensis* n. sp. A, Holotype whole-mount, ventral view; B, Copulatory complex; C, Ventral bar; D, Dorsal bar; E, Ventral anchor; F, Dorsal anchor; G, Hook. Scale-bars: A, 100 µm; B–F, 15 µm; G, 20 µm

Discussion

For species of *Myloplus* (Jardine), the following monogeneans have been described and reported: *Notozothecium bethae* Kritsky, Boeger & Jégu, 1996 in *Myloplus pacu* (Jardine) and *M. rhomboidalis* (Cuvier); *Anacanthorus hoplophallus* Kritsky, Boeger & Van Every, 1992, *A. pedanophallus* Kritsky, Boeger & Van Every, 1992, *A. spinatus* Kritsky, Boeger & Van Every, 1992, *A. stagmophallus* Kritsky, Boeger & Van Every, 1992 and *Notothecioides llewellyni* Kritsky, Boeger & Jégu, 1997 in *Myloplus rubripinnis* (Müller & Troschel) and *N. llewellyni* in *Myloplus torquatus* (Kner) (see Cohen et al., 2013). For *M. schomburgkii*, *N. bethae* was reported parasitising cultured fishes from the Peruvian Amazon (Gonzales

et al., 2016). Comparing the figures of the sclerotised parts of the parasite presented by Gonzales et al. (2016) with the original drawings and description made by Kritsky et al. (1996), we conclude that the material of Gonzales et al. (2016) was misidentified and in fact represents *Notozothecium nanayensis* n. sp. described herein. Additionally, the two new species of *Anacanthorus* described here from the gills of *M. schomburgkii*, represent the first monogeneans reported for this fish species.

For *C. macropomum*, five species of monogeneans have been described and reported: *Anacanthorus spathulatus* Kritsky, Thatcher & Kayton, 1979, *Linguadactyloides brinkmanni* Thatcher & Kritsky, 1983, *Mymarothecium boegeri* Cohen & Kohn, 2005, *Notozothecium euzeti* Kritsky, Boeger & Jégu, 1996 and

Notozothecium janauachensis Belmont-Jégu, Domingues & Martins, 2004 (see Cohen et al., 2013). The new species parasitising the gills of *C. macropomum* described here, increases the number of known parasites for this fish species to six.

Recently, studies focused on monogenean diversity in the Peruvian Amazon River basin have reported a high diversity with more than 60 undescribed species (Mendoza-Franco & Scholz, 2009; Mendoza-Palmero et al., 2012; Mendoza-Franco et al., 2016). Clearly, the number of known taxa is still low and discoveries of new species will continue.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable institutional, national and international guidelines for the care and use of animals were followed.

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