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Walteriella n. g. (Monogenoidea: Dactylogyridae) from the gills of pimelodid catfishes (Siluriformes: Pimelodidae) from the Peruvian Amazonia based on morphological and molecular data

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Abstract On the basis of morphological and molecular data, *Walteriella* n. g. is proposed to accommodate the dactylogyrid parasites *Walteriella conica* n. sp. (type-species) from the gills of *Platynematichthys notatus* (Jardine) (type-host) and *Brachyplatystoma juruense* (Boulenger), and *Walteriella ophiocirrus* n. sp. from the gills of *Platystomatichthys sturio* (Kner) (type-host), all fish host belonging to the Pimelodidae collected in the Peruvian Amazonia. Species of the new genus are distinguished from other dactylogyrids infecting Neotropical catfishes by the presence of a folded germarium, a male copulatory organ basally articulated to the accessory piece, a cone-shaped seminal receptacle, and ventral and dorsal bars with projections directed anteriorly. Both species of the new genus form a strongly supported lineage closely related to Dactylogyridae gen. sp. 13, a parasite of *Hypophthalmus edentatus* Spix & Agassiz (Pimelodidae) in an analysis of partial sequences of the 28S rRNA gene. The clade formed by species of *Walteriella* n. g. and Dactylogyridae gen. sp. 13 is closely related to other dactylogyrids infecting pimelodid catfishes. Based on the morphology of its haptoral elements and copulatory complex, the species Dactylogyridae gen. sp. 13 is morphologically different from species of *Walteriella* n. g., but its generic assignment is still unclear.

Introduction

In the Peruvian Amazonia, northeastern Peru, several species of pimelodid catfishes (Siluriformes: Pimelodidae) are greatly appreciated particularly for local consumption, including *Platynematichthys notatus* (Jardine), commonly known as ‘lince’. This catfish is relatively rare and it is distributed across the lowlands of the Orinoco and the Amazon basins (Lundberg

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et al., 2011). Other catfishes are captured for aquarium trade and exported worldwide because of their striking beauty, such as *Brachyplatystoma juruense* (Boulenger) and *Platystomatichthys sturio* (Kner), locally known as 'alianza' and 'toa zorro', respectively. These three catfish species share their distribution range in the Amazon River basin.

Despite their economic importance, little is known about the helminth parasites of these catfishes. Only the proteocephalid tapeworm *Brayela karuatayi* (Woodland, 1934), the dactylogyrid *Unibarra paranoplatensis* Suriano & Incorvaia, 1995, and two undescribed dactylogyrids characterised molecularly (Dactylogyridae gen. sp. 9 and Dactylogyridae gen. sp. 26 in Mendoza-Palmero et al. 2015), have been reported from *Pln. notatus* (see Mendoza-Palmero et al., 2012, 2015; de Chambrier et al., 2014), *Demidospermus mortenthaleri* Mendoza-Palmero, Scholz, Mendoza-Franco & Kuchta, 2012 was described from *B. juruense* (see Mendoza-Palmero et al. 2012), and there are no records of helminth parasites for *Pls. sturio*.

Recently, Mendoza-Palmero et al. (2015) assessed the interrelationships of dactylogyrids parasitising Neotropical catfishes using partial sequences of the 28S rRNA gene. Some of these dactylogyrids were not identified to the genus and species level and may represent undescribed taxa. In the present study, *Walteriella conica* n. g., n. sp. (syn. Dactylogyridae gen. sp. 10 of Mendoza-Palmero et al. 2015) from the gills of *Pln. notatus* (type-host) and *B. juruense*, and *W. ophiocirrus* n. g., n. sp. from *Pls. sturio* (type-host) are described based on morphological evaluation of specimens collected from Iquitos, Peruvian Amazonia, and on new molecular data.

Materials and methods

Morphological studies

Specimens of *Platynematichthys notatus*, *Brachyplatystoma juruense* and *Platystomaticthys sturio* were provided by local fishermen and aquarium fish traders from the surroundings of Iquitos, Region of Loreto, northeastern Peru, in October 2011 and April 2018. To avoid confusion between generic names of fish hosts, we used herein the abbreviations "*Pln.*" for *Platynematichthys* and "*Pls.*" for *Platystomatichthys.* Fish were euthanised *via* spinal cord severance and immediately examined for ectoparasites. Entire gill arches were removed from the carcasses, placed in Petri dishes with tap water and examined individually under a stereomicroscope. Parasites were removed individually from the gills and fixed in GAP (a mixture of glycerine and ammonium picrate) to study their sclerotised structures (Malmberg, 1957). After their morphological study, these specimens were remounted in Canada balsam following the procedure of Ergens (1969). Other specimens were fixed with hot water (*c.*80°C) and kept in vials with 96% ethanol for morphological and molecular evaluation. To study their internal and external anatomy, some of these specimens were stained with Gomori's trichrome and mounted in Canada balsam.

Illustrations were made with an optical microscope (Olympus BX51, Tokyo, Japan) equipped with a drawing tube. Measurements are presented in micrometres and represent straight-line distances between extreme points and are expressed as the range followed by the mean and number (n) of specimens measured in parentheses. The direction of the coil of the male copulatory organ (MCO) (i.e. clockwise *vs* counterclockwise) was determined following Kritsky et al. (1985). Numbering and distribution of hook pairs followed Mizelle (1936) (see also Mizelle & Price, 1963). Type- and voucher specimens were deposited in the helminthological collection of the Institute of Parasitology, Biology Centre of the Czech Academy of Science, České Budějovice, Czech Republic (IPCAS), and in the Colección Nacional de Helmintos (CNHE), UNAM, Mexico as indicated in the species description. Host names follow Froese & Pauly (2018).

For comparative purposes, the following type- and voucher specimens were studied: *Demidospermus mortenthaleri* Mendoza-Palmero, Scholz, Mendoza-

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Franco & Kuchta, 2012 (paratypes IPCAS M-522); *Cosmetocleithrum bulbocirrus* Kritsky, Thatcher & Boeger, 1986 (voucher IPCAS M-525); *Aphanoblastella aurorae* Mendoza-Palmero, Scholz, Mendoza-Franco & Kuchta, 2012 (paratype IPCAS M-524), *Paracosmetocleithrum trachydorasi* Acosta, Scholz, Blasco-Costa, Alves & Silva, 2018 (paratype IPCAS M-675); *Vancleaveus janauacaensis* Kritsky, Thatcher & Boeger, 1986 (voucher IPCAS M-527); *Nanayella aculeatrium* Acosta, Mendoza-Palmero, Silva & Scholz, 2019 (holotype and paratypes IPCAS M-694); and *Unilatus unilatus* Mizelle & Kritsky, 1967 (voucher IPCAS M-530). Field codes (PI = Peru, Iquitos) of each fish species examined are provided in the species taxonomic summary; photographs of hosts examined are available from authors upon request.

Molecular characterisation and phylogenetic analyses

A partial sequence of the 28S rRNA gene of Dactylogyridae gen. sp. 10 from *Pln. notatus* of Mendoza-Palmero et al. (2015), which corresponds to *Walteriella conica* n. g., n. sp., was retrieved from GenBank (KP056226), whereas sequences of *W. conica* n. g., n. sp. of two isolates from *B. juruense* and three isolates of *W. ophiocirrus* n. g., n. sp. from *Pls. sturio*, were newly generated in the present study.

Specimens used for molecular characterisation were cut in half with fine needles, either the haptor or the anterior part of the body were mounted in GAP for morphological identification, remounted in Canada balsam following the procedure of Ergens (1969), and deposited as molecular vouchers (hologenophores of *W. conica* n. sp. IPCAS M-699, and those of *W. ophiocirrus* n. sp. IPCAS M-700) (see Pleijel et al., 2008 for terminology). The rest of the worm was placed in a sterilised Eppendorf tube. DNA extraction and gene amplification followed the procedure described in Mendoza-Palmero et al. (2015). Previously published sequences of the 28S rRNA fragment (720–1,575 bp long) of a total of 41 dactylogyrids from siluriforms (see Mendoza-Palmero et al., 2015; Acosta et al., 2017, 2018; Franceschini et al., 2018; Yamada et al., 2018), together with those newly generated in the present study, were used for the assessment of phylogenetic relationships of the new genus. Sequences of three species of the Diplectanidae were used as outgroups.

Sequences were aligned using default parameters of MAFFT implemented in Geneious v.11.1.4 (Kearse et al., 2012). The extremes of the alignment were trimmed, resulting in 819 nucleotide positions for analysis. Phylogenetic analyses were run under Maximum Likelihood (ML) and Bayesian Inference (BI) criteria, applying the model of nucleotide evolution GTR+Γ+I, estimated using jModelTest 2.1.1 (Darriba et al., 2012). ML analyses were performed with RAxML v.8 (Guindon & Gascuel, 2003), model parameters and bootstrap support values (1,000 replicates) were estimated with RAxML. BI trees were generated using MrBayes v.3.2 (Ronquist et al., 2012) running two independent MCMC runs of four chains for 10^7 generations and sampling tree topologies every 10^3 generations. 'Burn-in' was set to the first 25,000 generations. MrBayes and RAxML analyses were carried out on the computational resource CIPRES (Miller et al., 2010). Genetic divergences were calculated using uncorrected p-distances model in MEGA v.6 (Kimura, 1980; Tamura et al., 2001), and are presented in Supplementary Table S1. Phylogenetic trees were edited in FigTree v.1.3.1 (Raumbaut, 2009).

Subclass Polyonchoinea Bychowsky, 1937
Order Dactylogyridea Bychowsky, 1937
Family Dactylogyridae Bychowsky, 1933

Walteriella n. g.

Diagnosis

Body fusiform, consisting of cephalic region, trunk, peduncle and haptor. Tegument smooth. Two terminal and 2 bilateral cephalic lobes; 2 pairs of bilateral head organs. Cephalic glands unicellular, anterior or posterolateral to pharynx. Subspherical chromatic granules clustered together. Mouth subterminal, midventral, opening into wide buccal tube immediately on anterior margin of pharynx; pharynx muscular; oesophagus short; intestinal caeca 2, confluent posterior to gonads, lacking diverticula. Common genital pore midventral, near level of intestinal bifurcation. Gonads in tandem, intercaecal. Testis postgermarial; vas deferens looping left intestinal caeca; seminal vesicle a dilation of vas deferens; prostatic reservoir single. Copulatory complex composed of basally articulated male copulatory organ (MCO) and accessory piece. MCO tubular, coiled with basal flange; coil with clockwise direction; accessory piece rod-shaped, serving as guide of terminal portion of MCO. Germarium folded in ventral/dorsal direction; oviduct connected to germarium leading to uterus; uterus runs anteriorly on midline leading to genital pore. Vaginal aperture sinistral, emptying to a cone-shaped seminal receptacle. Vitelline follicles dense, coextensive with intestinal caeca. Haptor bilobed on lateral margin, armed with 2 anchor-bar complexes and 7 pairs of hooks. Ventral and dorsal anchors similar. Ventral and dorsal bars robust, both with projections directed anteriorly. Hooks with undilated shanks and depressed thumbs with ancyrocephaline distribution (Mizelle, 1936). Parasites on the gills of Neotropical catfishes (Pimelodidae).

Type-species: Walteriella conica n. sp. (syn. Dactylogyridae gen. sp. 10 of Mendoza-Palmero et al., 2015).

Other species: Walteriella ophiocirrus n. sp.

ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the *International Code of Zoological Nomenclature* (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Walteriella* n. g. is urn: B7550305-8435-4C69-8D2E-8F249194E309.

Etymology: The genus is named after Dr Walter A. Boeger (Brazil) in recognition of his extensive investigation on Neotropical monogenoids. The genus name is appended to the diminutive *-ella* and should be treated as feminine.

Remarks

Species of *Walteriella* n. g. can be distinguished from other species of the Dactylogyridae parasitic on Neotropical freshwater catfishes, i.e. *Ameloblastella* Kritsky, Mendoza-Franco & Scholz, 2000; *Amphocleithrum* Price & González-Romero, 1969; *Aphanoblastella* Kritsky, Mendoza-Franco & Scholz, 2000; *Cosmetocleithum* Kritsky, Thatcher & Boeger, 1986; *Demidospermus* Suriano, 1983; *Heteropriapulus* Kritsky, 2007; *Kritskyia*, Kohn, 1990; *Paracosmetocleithrum* Acosta, Scholz, Blasco-Costa, Alves & Silva, 2018; *Pavanelliella* Kritsky & Boeger, 1998; *Philocorydodas* Suriano, 1986; *Trinigyrus* Hanek, Molnar & Fernando, 1974; *Unibarra* Suriano & Incorvaia, 1995; *Unilatus* Mizelle & Kritsky, 1967 and *Vancleaveus* Kritsky, Thatcher & Boeger, 1986; by the presence of a folded germarium, a cone-shaped seminal receptacle and haptoral bars with projections directed anteriorly.

A cone-shaped seminal receptacle has never been reported in any other dactylogyrids infecting Neotropical fishes (Thatcher, 2006; Cohen et al., 2013). In most dactylogyrids, the seminal receptacle is represented by a subspherical structure located anterior or near the germarium. Species of *Walteriella* n. g. share the presence of the MCO basally articulated to the accessory piece with species of *Ameloblastella*, *Philocorydoras* and *Unibarra*, but species of *Walteriella* n. g. differ from species of these genera in the shape of the germarium, seminal receptacle and haptoral bars. Additionally, species of *Walteriella* n. g. resemble species of the recently erected genus *Nanayella* by the shape of the germarium (folded), but species of *Walteriella* n. g. can be distinguished from species of *Nanayella* by the presence of a coiled MCO, no sclerotised vaginal aperture, ventral and dorsal bars with projections directed anteriorly, and hooks of similar size.

Walteriella conica n. sp.

Syn. Dactylogyridae gen. sp. 10 of Mendoza-Palmero et al. (2015)

Type-host: *Platynematichthys notatus* (Pimelodidae) (field code PI 798).

Other host: *Brachyplatystoma juruense* (Pimelodidae) (PI 1010).

Type-locality: River Nanay in Santa Clara de Nanay near Iquitos (03°41′60″S, 73°16′27″W), Loreto Region, Peru.

Other locality: Amazon River basin, Loreto Region, Iquitos, Peru (*B. juruense*; precise locality of fish host is not known).

Type-material: Holotype (IPCAS M-699) collected on *Pln. notatus* (5.x.2011), 11 and 4 paratypes (IPCAS M-699; CNHE 11161), respectively, and 5 and 10 vouchers (IPCAS M-699; CNHE 11162), respectively.

Site in host: Gills.

Representative DNA sequences: 1,473 bp long partial sequence of the 28S rRNA gene of *W. conica* n. sp. from *Pln. notatus* (GenBank: KP056226; designated as Dactylogyridae gen. sp. 10 by Mendoza-Palmero et al., 2015); 1,380 and 1,460 bp long partial sequences of the 28S rRNA gene of *W. conica* n. sp. from *B. juruense* (GenBank: MK834513-14, respectively).

ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the *International Code of Zoological Nomenclature* (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Walteriella conica* n. sp. is urn: 7B080A40-A5DE-4E79-9E34-45CE9779B761.
Etymology: The specific name is from Latin (*conica* = cone shaped) and refers to the shape of the seminal receptacle.

Description (Fig. 1A–G)

[Based on 16 specimens fixed in GAP and 18 stained with Gomori's trichrome and mounted in Canada balsam, all collected from *Pln. notatus.*] Body elongated, relatively large, 602–825 (727; n = 18) long, with greatest width at level of gonads, 90–155 (116; n = 18) wide (Fig. 1A). Cephalic lobes well developed, 2 terminal and 2 bilateral. Chromatic granules clustered together at level of anterior margin of pharynx. Pharynx subspherical, 35–50 (42; n = 16) wide.

Haptor subhexagonal, 50–95 (76; n = 16) long, 70–110 (93; n = 15) wide (Fig. 1A). Ventral anchor with elongate and rounded superficial root, short deep root, short and robust shaft, and perpendicular straight and elongate point, slightly exceeding base width, 42–47 (45; n = 14) long; base width 21–28 (23; n = 14) (Fig. 1B). Dorsal anchor with elongate superficial root, short and reduced deep root, wide and robust shaft, elongated and straight point, slightly exceeding base width; 41–45 (43; n = 14) long; base width 20–25 (23; n = 14) (Fig. 1C). Ventral bar 51–58 (55; n = 13) long, rod-shaped with conspicuous anterior medial groove (Fig. 1D). Dorsal bar 44–51 (47; n = 13) long, open U-shaped, with conspicuous ornamentations in its medial region and on posterior ends (Fig. 1E).

Hooks similar in shape and size; each with slender shank, slightly curved proximally, depressed thumb, straight shaft and short and curved point; filamentous hooklet (FH) loop about 90% of shank length; hooks 14–15 (n = 16) long (Fig. 1F). MCO 60–85 (68; n = 15) long, a coiled tube of about 2 ½ clockwise rings, distally acute (Fig. 1G), proximal ring diameter 29–35 (32; n = 15). Accessory piece 24–34 (30; n = 10) long, delicate, appearing as sigmoid-shaped; articulation piece represented by a fine tube extending from base of MCO to medial region of accessory piece (Fig. 1G). Vaginal aperture not sclerotised. Vaginal canal sinuous, folded in its proximal portion, narrowing at its middle into a delicate sclerotised channel and leading to a cone-shaped seminal receptacle lying sinistral of germarium (Fig. 1A). Germarium 80–130 (97; n = 15) long, 36–77 (52; n = 16) wide (Fig. 1A); testis elongate-oval, 105–170 (142; n = 16) long, 50–68 (58; n = 15) wide. Seminal vesicle lying sinistral of midline and continuing into ascending twisted duct connected to MCO (Fig. 1A). Prostatic reservoir single, pyriform. Oviduct and uterus conspicuous (Fig. 1A).

Remarks

Walteriella conica n. sp. is typified by the characteristics used to define the new genus.

The new species was found on two different hosts (*Pln. notatus* and *B. juruense*). Because of mixed infection of parasites on both hosts (up to six species of dactylogyrids on the same individual), it was not possible to estimate parameters of infection for any of the species of *Walteriella* n. g.

Walteriella ophiocirrus n. sp.

Type-host: *Platystomatichthys sturio* (Pimelodidae) (PI 1042).
Type-locality: Iquitos, Loreto Region, Peru (precise locality of fish host is not known).
Type-material: Holotype (IPCAS M-700) collected on *Pls. sturio* (19.iv.2018), 10 and 7 paratypes (IPCAS M-700; CNHE 11163), respectively, and 4 and 5 vouchers (IPCAS M-700; CNHE 11164), respectively.
Site in host: Gills.
Representative DNA sequence: 1,480 (two isolates) and 1,477 bp long partial sequences of the 28S rRNA gene of *W. ophiocirrus* n. sp. from *Pls. sturio* (GenBank: MK834511-12, 15).
ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the *International Code of Zoological Nomenclature* (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Walteriella ophiocirrus* n. sp. is urn: B212ABB3-A6E8-48E4-9F1A-FC80A664D18E.
Etymology: The specific name is from Greek (*ophios* = snake) and refers to the shape of the male copulatory organ.

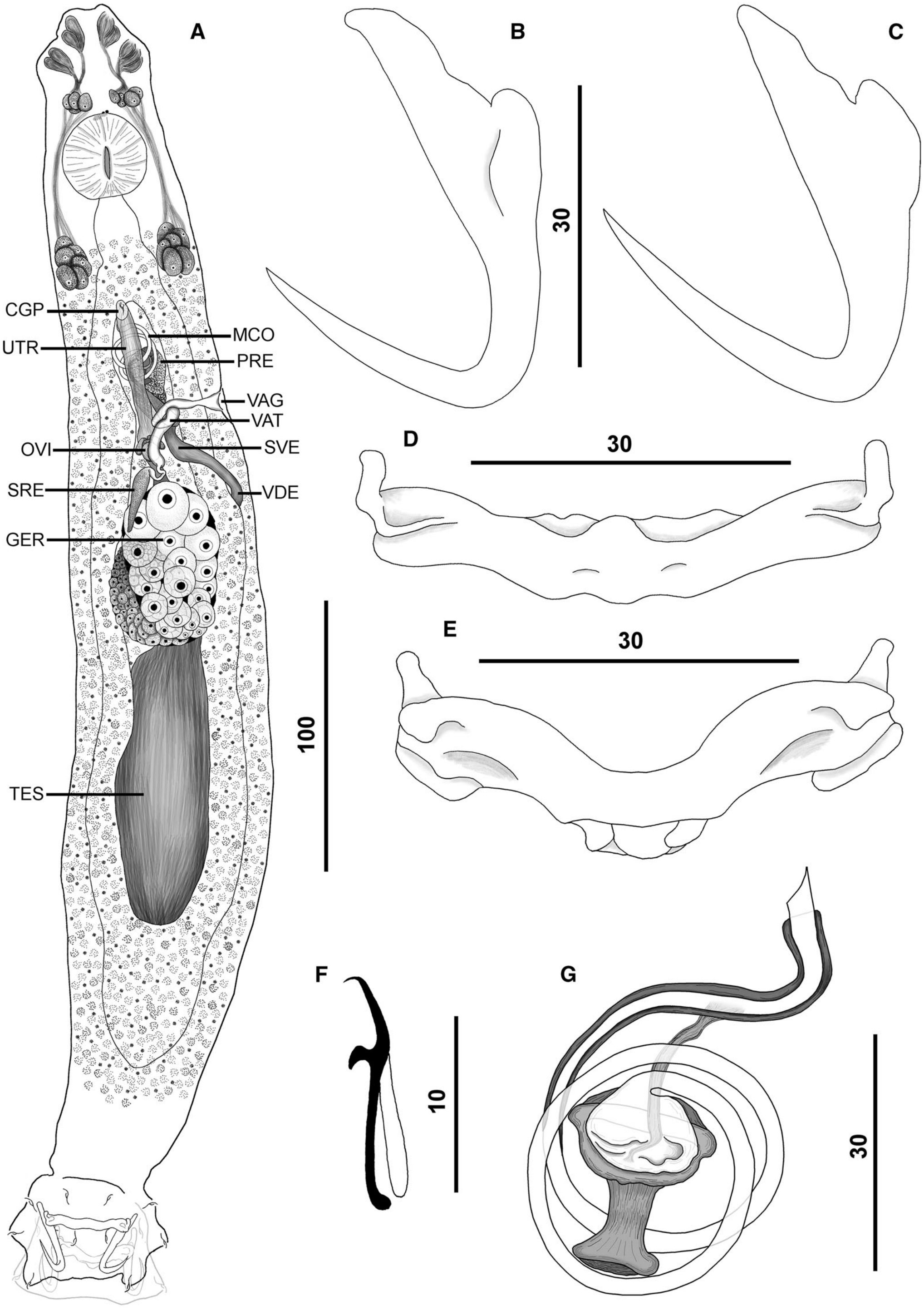

Fig. 1 *Walteriella conica* n. g., n. sp. from *Platynematichthys notatus*. A, Whole specimen in ventral view (composite); B, Ventral anchor; C, Dorsal anchor; D, Ventral bar; E, Dorsal bar; F, Hook; G, Copulatory complex (ventral). *Scale-bars:* A, 100 µm; B–E, G, 30 µm; F, 10 µm. *Abbreviations*: CGP, common genital pore; UTE, uterus; MCO, male copulatory organ; PRE, prostatic reservoir; VAG, vagina; VAT, vaginal tube; OVI, oviduct; SVE, seminal vesicle; VDE, vas deferens; SRE, seminal receptacle; GER, germarium

Description (Fig. 2A–G)

[Based on 16 specimens fixed in GAP and 18 stained with Gomori's trichrome and mounted in Canada balsam, all collected from *Pls. sturio*.] Body elongated, small and slender, 245–336 (276; n = 17) long; greatest width at level of gonads, 33–50 (43; n = 17) (Fig. 2A). Cephalic lobes moderately developed, 2 lobes located anteriorly and 2 bilateral. Spherical chromatic granules clustered together. Pharynx subspherical, 15–20 (18; n = 17) wide.

Haptor subrectangular, 37–51 (43; n = 15) long, 41–60 (49; n = 14) wide (Fig. 2A). Ventral anchor with elongated and pointed superficial root, short deep root, straight shaft, elongated and delicate point, perpendicular to shaft, 33 (32–35; n = 16) long; base width 15–20 (17; n = 16) (Fig. 2B). Dorsal anchor with well-developed and pointed superficial root, inconspicuous deep root, short shaft, elongated and delicate point, perpendicular to shaft, 28–31 (29; n = 16) long; base width 15–21 (18; n = 16) (Fig. 2C). Ventral bar 31–40 (34; n = 15) long, rod-shaped (Fig. 2F). Dorsal bar 25–31 (28; n = 15) long, open U-shaped, with conspicuous ornamentations in its medial region and on posterior ends (Fig. 1E).

Hooks similar; each with slender shank, slightly curved proximally, depressed thumb, evenly curved shaft and short point; filamentous hooklet (FH) loop about 90% of shank length; hooks 13–15 (14; n = 15) long (Fig. 2E). MCO 50–64 (57; n = 14) long, a robust, thick-walled coiled tube of about 2 ½ clockwise rings, distal region blunt and expanded (Fig. 2G), proximal ring diameter 20–23 (22; n = 14). Accessory piece 31–42 (35; n = 14) long, sheath-like, distally expanded, accompanying distal portion of MCO; articulation piece represented by fine tube extending from base of MCO and connected to first third region of accessory piece (Fig. 2D). Vagina not sclerotised, leading to vaginal tube folded in its first third, narrowing in its middle portion into a delicate channel (apparently sclerotised), emptying to a short cone-shaped seminal receptacle lying antero-sinistral to germarium. Germarium, 23–35 (28; n = 12) long, 13–30 (18; n = 12) wide; testis elongate-oval, 28–40 (33; n = 10) long, 12–19 (16; n = 10) wide (Fig. 2A). Seminal vesicle and prostatic reservoir not observed. Oviduct connected to germarium anteriorly, uterus not observed.

Remarks

Walteriella ophiocirrus n. sp. closely resembles the type-species, *W. conica* n. sp., but can be distinguished by its smaller body size (245–336 *vs* 602–825 µm), the shape of the copulatory complex (thick-walled and distally rounded MCO, with a robust accessory piece in *W. ophiocirrus* n. sp. *vs* a slender and distally acute MCO, and a delicate accessory piece in *W. conica* n. sp.), and hooks with an evenly curved shaft *vs* hook with a straight shaft in *W. conica* n. sp.

Phylogenetic position of the new genus

Phylogenetic analyses built on ML and BI criteria using 44 sequences of dactylogyrid parasites along with those used as the outgroup, yielded similar topologies (Fig. 3). Sequences for *Walteriella conica* n. sp. from *P. notatus* retrieved from GenBank (as Dactylogyridae gen. sp. 10 of Mendoza-Palmero et al., 2015) and those newly generated in the present study (two isolates from *B. juruense*) were identical; therefore, only one sequence for *W. conica* n. sp. from *Pln. notatus* and one from *B. juruense* were included in Fig. 3.

In the present study, in which only parasites of siluriforms were analysed, *W. conica* n. sp. and *W. ophiocirrus* n. sp. formed a strongly supported clade (in both ML and BI analyses). Both species of *Walteriella* n. g. are most closely related to Dactylogyridae gen. sp. 13 from the pimelodid *Hypophthalmus edentatus*, with a nucleotide divergence of 15–16% (120 bp for *W. conica* n. sp. and 117 bp for *W. ophiocirrus* n. sp., in comparison to Dactylogyridae gen. sp. 13, respectively). Species of *Walteriella* n. g. and Dactylogyridae gen. sp. 13 clustered together with the sister clade (only supported by BI analysis) composed of species of *Nanayella*, *N. aculeatrium*, *N. fluctuatrium* Acosta, Mendoza-Palmero, Silva & Scholz, 2019, *N. amplofalcis* Acosta, Mendoza-Palmero, Silva & Scholz, 2019 and *N. processusclavis* Acosta, Mendoza-Palmero, Silva & Scholz, 2019 (see Acosta et al., 2019).

Dactylogyrid parasites of pimelodids appeared in two separated clades (labelled 1 and 2 in Fig. 3). Clade 1 is formed by three species of *Ameloblastella* (*Ameloblastella* sp. 23, *A. edentensis* Mendoza-Franco, Mendoza-Palmero & Scholz, 2016 and *A. unapioides* Mendoza-Franco, Mendoza-Palmero &

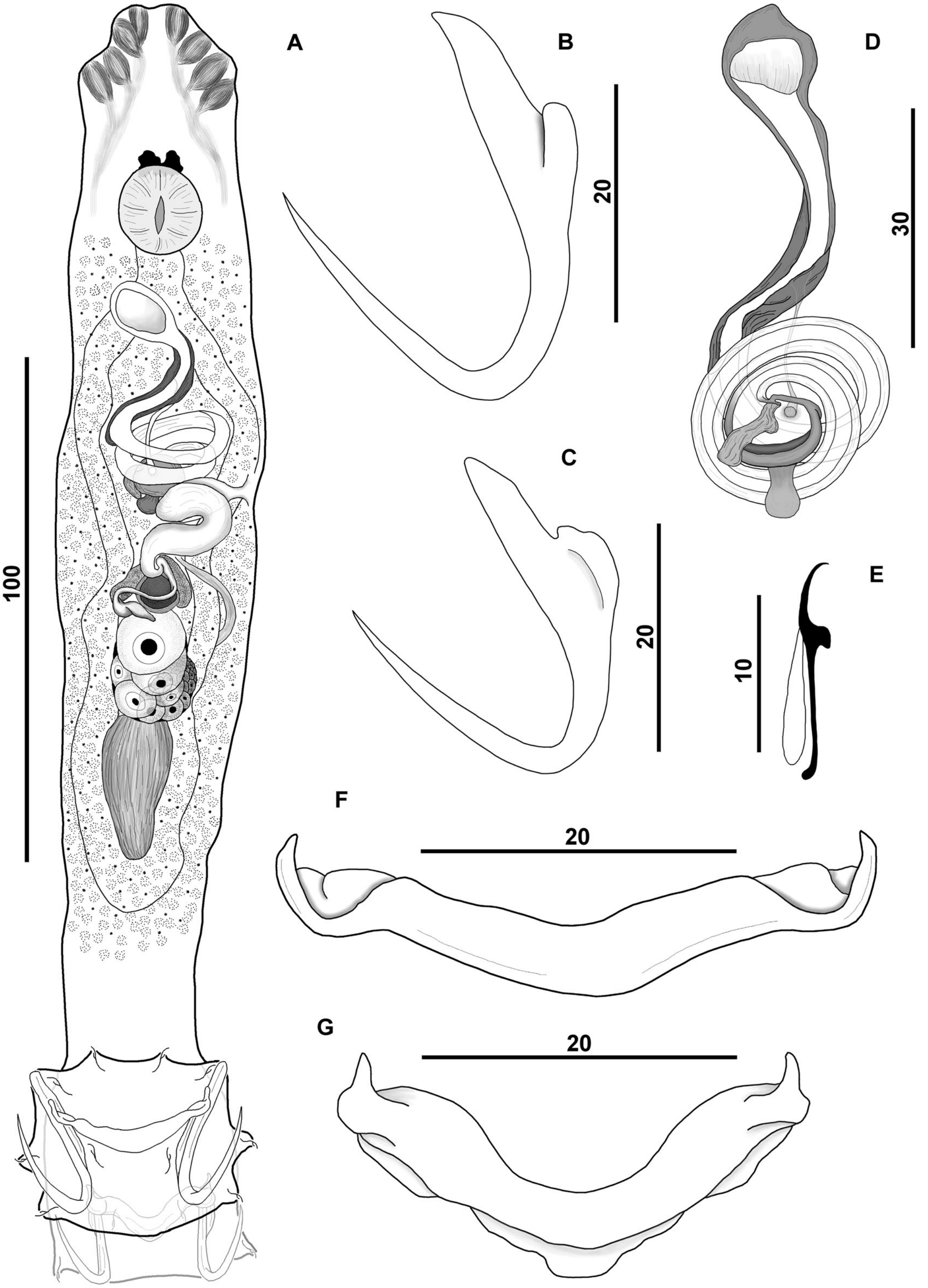

Fig. 2 *Walteriella ophiocirrus* n. g., n. sp. from *Platystomatichthys sturio*. A, Whole specimen in ventral view (composite); B, Ventral anchor; C, Dorsal anchor; D, Copulatory complex (ventral); E, Hook; F, Ventral bar; G, Dorsal var. *Scale-bars:* A, 100 µm; B, C, F, G, 20 µm; D, 30 µm; E, 10 µm

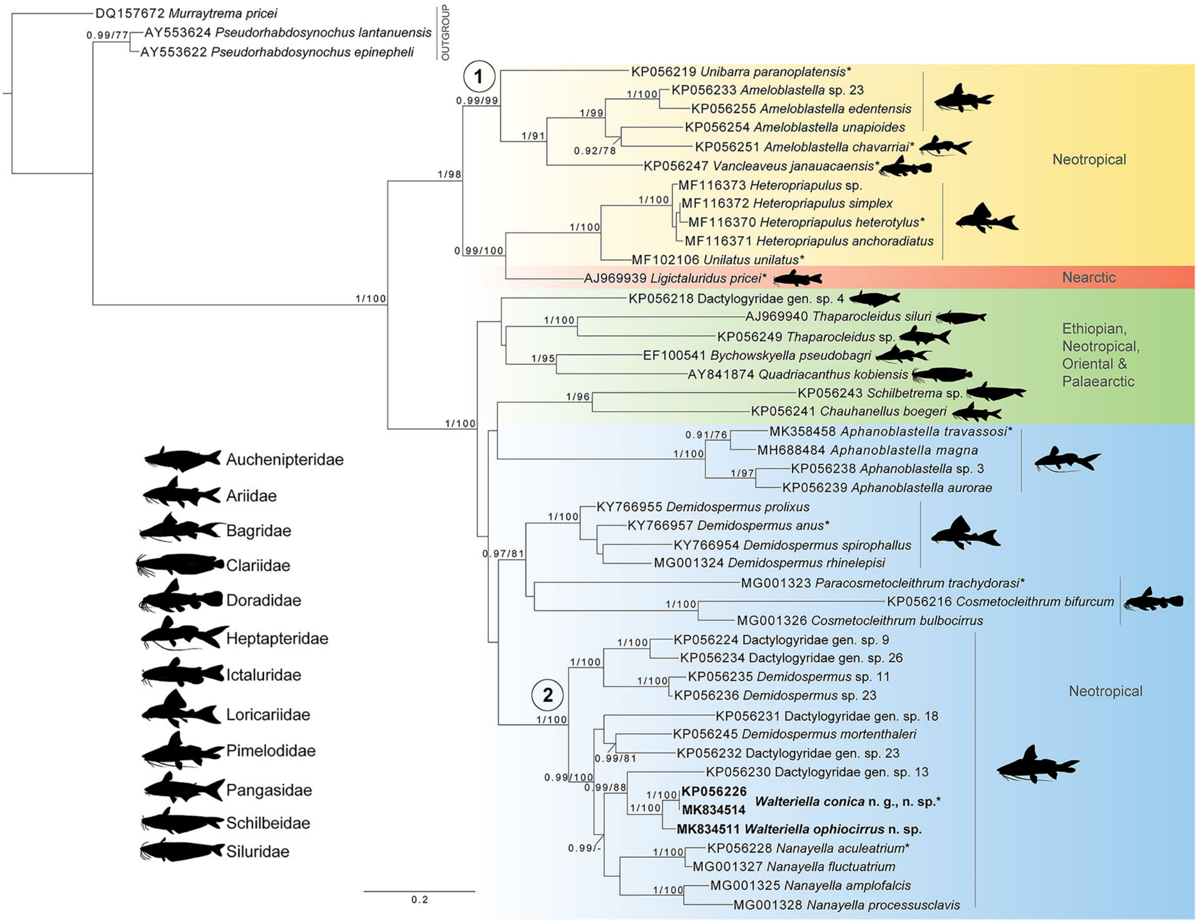

Fig. 3 Molecular phylogeny of monogenoidean parasites of catfishes estimated by Maximum Likelihood (ML) criterion using partial sequences of the 28S rRNA gene showing the position of *Walteriella conica* n. g., n. sp. and *W. ophiocirrus* n. g., n. sp. Species of the Diplectanidae were used as the outgroup. GenBank sequence ID's follow species names. Species newly sequenced in the present study are in bold. Type-species of individual genera are indicated with asterisks. Posterior probabilities from Bayesian inference analysis and bootstrap values (ML) are given above the nodes (posterior probabilities < 0.90 and bootstrap values < 60 are not shown)

Scholz, 2016), and *Unibarra paranoplatensis* (all parasites of pimelodids), including *A. chavarriai* (Price, 1938) (type-species of the genus) and *Vancleavus janauacaensis* Kritsky, Thatcher & Boeger, 1986 from heptapterid and doradid catfishes, respectively.

Clade 2 is composed of two subclades. The first subclade is formed by Dactylogyridae gen. sp. 9 and Dactylogyridae gen. sp. 26, both parasites of *Pln. notatus*, and *Demidospermus* spp. 11 and 23, both parasites of *B. vaillanti* (Valenciennes). The second subclade comprises Dactylogyridae gen. sp. 18, a parasite of *Pseudoplatystoma fasciatum* (L.), *D. mortenthaleri* Mendoza-Palmero, Scholz, Mendoza-Franco & Kuchta, 2012 ex *B. juruense*, Dactylogyridae gen. sp. 23 ex *Platysilurus mucosus* (Vaillant), species of *Walteriella* n. g. which clustered together with Dactylogyridae gen. sp. 13 ex *H. edentatus*, and *Nanayella aculeatrium* and *N. fluctuatrium*, both parasites of *Sorubim lima* (Bloch & Schneider), and *N. amplofalcis* and *N. processusclavis*, both ex *Hemisorubim platyrhynchos* (Valenciennes).

Discussion

In the present study, *Walteriella* n. g. is erected to accommodate *W. conica* n. sp. (type-species) from the gills of *Pln. notatus* (type-host) and *B. juruense*, and

W. ophiocirrus n. sp. from *Pls. sturio* (type-host) from the Peruvian Amazonia. Combination of morphological and molecular evidence enabled us to propose this new genus, which is morphologically characterised by the unusual shape of the germarium (folded), seminal receptacle and bars with projections directed anteriorly.

Specimens of *W. conica* n. sp. are relatively large worms (602–825 µm), from which internal details such as the oviduct, uterus and genital pore could be observed (see Fig. 1), which is often difficult in smaller species of dactylogyrids. It is worth mentioning that the morphological resemblance of anchors and bars between *W. conica* n. sp. and *W. ophiocirrus* n. sp. is remarkable (see Figs. 1 and 2), despite the fact that *W. conica* n. sp. is almost three times larger than *W. ophiocirrus* n. sp.

The occurrence of species of *Walteriella* n. g. on different host species of the same family, i.e. *W. conica* n. sp. on *Pln. notatus* and *B. juruense*, and *W. ophiocirrus* n. sp. on *Pls. sturio*, may appear unexpected, but it has been shown that these three pimelodid species are phylogenetically closely related based on morphological and molecular evidence (Lundberg & Akama, 2005; Lundberg et al., 2011).

A preliminary morphological study of specimens of Dactylogyridae gen. sp. 13 revealed distinct morphology of their haptoral elements and copulatory complex compared to that of both species of *Walteriella* n. g. Therefore, generic assignment of Dactylogyridae gen. sp. 13 is still unclear. A detailed study of the internal morphology of specimens of Dactylogyridae gen. sp. 13 will help us to determine its generic placement. Previous phylogenetic studies have shown that dactylogyrids infecting pimelodids do not form a monophyletic group (Mendoza-Palmero et al., 2015; Franceschini et al., 2018; Yamada et al., 2018). In the present study, a similar topology was recovered for the relationship of parasites of pimelodids within the Dactylogyridae.

The knowledge of the dactylogyrid parasite fauna of Neotropical catfishes has remarkably increased in the last five years, when morphology-based taxonomy and molecular approaches were combined. As a result, new dactylogyrids (19 species) have been described in *Ameloblastella*, *Aphanoblastella*, *Cosmetocleithrum*, *Demidospermus*, *Heteropriapulus*, *Paracosmetocleithrum* and *Pavanelliella* (see Mendoza-Palmero et al., 2015; Mendoza-Franco et al. 2016; Aguiar et al., 2017; Acosta et al., 2017, 2018; Franceschini et al., 2018; Yamada et al., 2018).

The monogenoidean fauna of freshwater fishes in the Neotropical region, especially that of the Dactylogyridae, still represents an overwhelming challenge for taxonomists due to several reasons: (i) extraordinary richness of these dactylogyrids in this region; (ii) morphological complexity of these parasites; (iii) high number of undescribed species *vs* low number of taxonomists actively working on the group; (iv) only partly known diversity of fish hosts and the problematic taxonomy in some groups; and (v) scarcity of phylogenetic studies of dactylogyrids based on molecular data (Mendoza-Palmero et al. 2015; Acosta et al., 2017, 2018, 2019; Aguiar et al., 2017; Franceschini et al., 2018, Yamada et al., 2018). Nevertheless, there is a considerable progress in unravelling the diversity and phylogenetic relationships within the Dactylogyridae. For instance, the monophyly of selected groups such as *Ameloblastella*, *Heteropriapulus*, *Pavanelliella* (all Dactylogyridae) and *Scleroductus* Jara & Cone, 1989 (Gyrodactylidae) has been assessed, and some controversial groups that have become 'catch-all' taxa such as *Demidospermus* Suriano, 1983 have been revised (Kritsky et al., 2013; Mendoza-Palmero et al., 2015; Aguiar et al., 2017; Acosta et al., 2017, 2018; Franceschini et al., 2018). Therefore, in future studies on Neotropical monogenoids, species characterisation based on morphology and molecular data should be considered for the adequate species/-genus level placement of these parasites.

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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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