A new species of Tereancistrum (Monogenea: Dactylogyridae), parasite of Prochilodus lineatus (Characiformes: Prochilodontidae) from southeast Brazil

Uma nova espécie de Tereancistrum (Monogenea: Dactylogyridae), parasito de Prochilodus lineatus (Characiformes: Prochilodontidae) do sudeste do Brasil

Lucas Aparecido Rosa Leite; Larissa Sbeghen Pelegrini*; Rodney Kozlowiski de Azevedo; Vanessa Doro Abdallah

1 Instituto de Biociências de Botucatu, Universidade Estadual Paulista – UNESP, Botucatu, SP, Brasil
2 Programa de Pós-graduação em Análise de Sistemas Ambientais, Centro Universitário CESMAC, Maceió, AL, Brasil

How to cite: Leite LAR, Pelegrini LS, Azevedo RK, Abdallah VD. A new species of Tereancistrum (Monogenea: Dactylogyridae), parasite of Prochilodus lineatus (Characiformes: Prochilodontidae) from southeast Brazil. Braz J Vet Parasitol 2020; 29(2): e017019. https://doi.org/10.1590/S1984-29612020024

Abstract
A new species of Tereancistrum Kritsky, Thatcher & Kayton, 1980 collected of Prochilodus lineatus gills from the Batalha River, Tietê-Batalha basin, São Paulo State, Brazil is described. The new species can be distinguished from its congener mainly by the configuration of the ventral bar, which has an anvil-shaped characteristic with corrugated anterior projection. Tereancistrum takemotoi n. sp. is morphologically similar to T. toksonum Lizama, Takemoto & Pavanelli, 2004 in terms of their dorsal bars (Y-shaped), their dorsal anchors with divergent roots (superficial and deep) wherein their deep root rather elongated, and by the fact that they both have the male copulatory organ counterclockwise. However, only T. takemotoi n. sp. presents the male copulatory organ with 2½ rings and shows undulations in the anterior margin of the dorsal bar. These undulations are absent in T. toksonum (which only has 1¼ rings). This is the fourth Tereancistrum species described for P. lineatus and the first described for the region from the Tietê-Batalha basin.

Keywords: Freshwater fish, gills parasite, taxonomy, Dactylogyridea, Platyhelminthes, Batalha river.

Resumo
É descrita uma nova espécie de Tereancistrum Kritsky, Thatcher & Kayton, 1980, coletada das brânquias de Prochilodus lineatus do rio Batalha, bacia do Tietê-Batalha, estado de São Paulo, Brasil. A nova espécie pode ser diferenciada de seus congêneres, principalmente pela configuração da barra ventral, que tem o formato de bigorna e apresenta uma projeção anterior com ondulações. Tereancistrum takemotoi n. sp. é morfologicamente semelhante a T. toksonum Lizama, Takemoto & Pavanelli, 2004, em termos de suas barras dorsais (em forma de Y), suas âncoras dorsais com raízes divergentes (superficiais e profundas) sendo que a raiz profunda é bastante alongada, e pelo fato de ambos terem o órgão copulatório masculino no sentido anti-horário. No entanto, apenas T. takemotoi n. sp. apresenta o órgão copulatório masculino com 2¾ anéis e também exibe ondulações na margem anterior da barra dorsal, diferentemente do T. toksonum (que possui apenas 1¾ anel em seu órgão copulatório masculino). Essa é a quarta espécie de Tereancistrum descrita para P. lineatus e a primeira descrita para a região da bacia do Tietê-Batalha.

Palavras-chave: Peixes de água doce, parasitos branquiais, taxonomia, Dactylogyridea, Platyhelminthes, rio Batalha.
Introduction

Prochilodus lineatus Valenciennes, 1837, known in Brazil as curimba, curimbata, or curimatã, is one of the most abundant native fish species in the floodplains of the upper Paraná River, although it is distributed across the country. It has substantial commercial appeal and is considered a medium to large species that engages in extensive migrations for food and reproduction (Agostinho et al., 1997). Due to anthropogenic interferences in aquatic environments (such as dams), its natural stocks have been declining, a process that has attracted several ecological, parasitological, and molecular studies on the species (Lizama et al., 2005, 2006; Rosa & Lima, 2008; Oyakawa et al., 2009).

The parasitic fauna of *P. lineatus* is relatively well known in the upper Paraná basin, and many studies have considered taxonomic and ecological aspects of this host. Monogenean ectoparasites are particularly common, with several species having been recorded (Lizama et al., 2004; Leite et al., 2018). The species of the family Dactylogyridae Bychowsky, 1933 are the most know gill parasites in the Neotropical region and are generally not highly pathogenic (Boeger & Vianna, 2006). This family of parasites includes *Tereancistrum* Kritsky, Thatcher & Kayton, 1980 whose main morphological characteristic is the presence of sclerites associated with ventral anchors (Kritsky et al., 1980).

In this study, a new species of *Tereancistrum* was found in the gills of *P. lineatus* from the Tietê-Batalha River basin, São Paulo State, Brazil during an investigation of the parasitic fauna of this fish. We provide a morphological description and illustrations pertinent to its identification.

Material and Methods

Fifty specimens of *P. lineatus* were collected at two sites on the Batalha River, part of the Tietê-Batalha basin, in São Paulo State, Brazil. This river is located in the municipalities of Reginópolis (21°53'17"S, 49°13'31"W) and Piratininga (22°24'46"S, 49°05'05"W). Fish samplings were performed between June 2015 and June 2016. The hosts presented mean standard length and mean weight of 28.45 ± 6.24 cm and 671.48 ± 542.74 g, respectively.

Hosts were collected using nylon monofilament gillnets with different mesh sizes (with a distance of 2 to 10 cm between nodes) placed at varying heights. At each of the sampling points, the nets were installed perpendicularly and in a half-moon shape at sunset (around 5:00 p.m.), and the fish were collected at sunrise (around 5:00 a.m.). After being collected, fish that were still alive were anesthetized with a eugenol-based solution (clove oil) (65 mg/L) and euthanized through the physical method of medullary collapse. Collected fish were individually packed in plastic bags and frozen for two days until necropsy.

During the necropsy, the gills were removed, and the gill arches were separated and placed in a glass jar with water. It was shaken so that the parasites detached from the filaments. Glass content was filtered through at 53 μm mesh sieve, and the contents retained in the sieve were analyzed under a stereomicroscope. The parasites were collected and stored in 70% ethanol solution. For identification, the parasites were cleared and mounted in Gray & Wess’s (1950) medium, or stained with Gomori’s trichrome and mounted on Canada balsam for the analysis of the internal organs (Gomori, 1950).

The illustrations were prepared with the aid of a camera lucida attached to a Leica DMLS microscope. The identifications and morphological analysis were performed using the Trinocular Nikon E200 microscope and the Motic computerized image analysis system (Moticam 5.0MP). Measurements (in micrometers) were expressed as the mean followed by the standard deviation (SD) and the range in parentheses.

The quantitative descriptors were obtained based on Bush et al. (1997). The terminology related to the sclerites followed the recommendations of Kritsky & Mizelle (1968) and the coils description of the male copulatory organs follows that of Kritsky et al. (1985).

Voucher specimens from the analyzed hosts were deposited in the Fish Collection of the Fish Biology and Genetics Laboratory of the Department of Biosciences of Botucatu at São Paulo State University, Botucatu (UNESP Botucatu) in Botucatu, São Paulo, Brazil (reference number IBB 22918). Parasite holotypes and paratypes were deposited in the Zoology Collection of the National Institute of Amazonian Research (INPA) (holotype nº INPA 809, paratype nº 810) in Manaus, Amazonas, Brazil and in the Helminthological Collection of the Department of Biosciences at UNESP (paratypes nº 578L, 579L, 580L) in Botucatu.

All the procedures followed the guidelines and standards of the Brazilian Biodiversity Information and Authorization System (SISBIO) (authorization nº: 40998-2); in addition, the anesthesia and euthanasia methodologies used on the fish followed the guidelines of the National Council for the Control of Animal Experimentation (CONCEA),
and the research project was submitted to the Research Ethics Committee for Animal Experimentation of the Universidade do Sagrado Coração (authorization nº 3295230615) in the municipality of Bauru, São Paulo State, before it could be carried out.

Results

Monogenea Carus, 1863
Monopisthocotylea Odhner, 1912
Dactylogyridae Bychowsky, 1933
Tereancistrum Kritsy, Thatcher and Kayton, 1980

Tereancistrum takemotoi n. sp.

DESCRIPTION (based on 17 specimens) (Figure 1): Body elongated and fusiform, 539.2 ± 69.5 (359.7-684.1) long; greatest width 130.1 ± 27.6 (95.1-200) at the level of the gonads. Tegument smooth. Cephalic lobes developed; head organs well developed. Eyespots 2 pairs; component granules subspherical. Pharynx ovate, 31.3 ± 3.5 (27.5-36.5) wide, 43.3 ± 5.8 (33.9-52.5) long; esophagus long; intestinal ceca confluent posterior to testis. Peduncle elongate, moderate in width; haptor globose to subhexagonal, 94.5 ± 10.8 (78-109) wide, 62.4 ± 6.2 (54-77.2) long. Hooks 12 ± 1.6 (10.2-14.6) long, with erect thumb, slightly curved shaft and point, and small proximal enlargement of shank; FH loop approximately one half of shank length. Ventral anchor robust, 32.3 ± 2.1 (27.1-35.1) long, with deep root well-developed and superficial root with uneven protuberance, short and straight shaft and recurved point; base 13.7 ± 2.4 (9.5-16.7) wide. Accessory anchor sclerite 30.7 ± 2.1 (28.3-34.7) long, robust, with spatulate

![Figure 1. Tereancistrum takemotoi n. sp. (ventral view). (A) Holotype (ventral view); (B) Vagina; (C) Hook; (D) Copulatory complex; (E) Ventral bar; (F) Dorsal bar; (G) Egg; (H) Accessory anchor sclerite; (I) Dorsal anchor; (J) Ventral anchor.](image-url)
end. Dorsal anchor 31.7 ± 1.6 (27.9-33.4) long, with widely divergent roots, short shaft and straight point; base 23.1 ± 2.6 (17.4-26.3) wide. Ventral bar 50.4 ± 5.7 (42.7-58.7) long, anvil-shaped and with corrugated anterior projection. Dorsal bar 38.9 ± 3.9 (30.845.3) long, Y-shaped, with internal groove and ends corrugated. Gonads overlapping. Male copulatory organ tapered, tubular, coiled, with 2 1/4 counterclockwise rings; ring diameter 19.4 ± 2.2 (14.8-21.6). Accessory piece 23.2 ± 2.4 (20-27.1) long, variable, not articulated with the base of male copulatory organ. Vagina sinistral, forming a sclerotized sinuous tube. Vitelline follicles random throughout trunk but absent in regions of gonads and copulatory complex.

Taxonomic Summary

Type-host: Prochilodus lineatus Valenciennes, 1837 (Characiformes: Prochilodontidae)  
Location: Batalha River, Tietê-Batalha River basin, São Paulo State, Brazil (21°53'17"S; 49°13'31"W)  
Site of infestation: Gills  
Infestation: Prevalence: 29.3%, Mean intensity: 13.2 ± 28.3  
Types: holotype n° INPA 809, paratypes nº INPA 810, 578L, 579L, 580L  
Etymology: The specific epipet takemotoi is a tribute to Dr. Ricardo Massato Takemoto for his significant contributions to the study of the monogenean c fish parasites, including those in Tereancistrum.

Remarks and Discussion

Nine species of Tereancistrum have been described after being found in Neotropical species of Characiformes. Except by Tereancistrum ornatus Kritsky, Thatcher & Kayton, 1980 reported in gills of Prochilodus reticulatus Valenciennes, 1850 from the Colombia, the other eight species were found in Brazil: Tereancistrum kerri Kritsky, Thatcher & Kayton, 1980 in Brycon melanopterus Cope, 1872; Tereancistrum parvus Kritsky, Thatcher & Kayton, 1980 in Leporinus fasciatus Bloch, 1794; Tereancistrum toksonum Lizama, Takemoto & Pavanelli, 2004; Tereancistrum curimba Lizama, Takemoto & Pavanelli, 2004; Tereancistrum pirassununguensis Cepeda, Ceccarelli & Luque, 2012 all in P. lineatus; Tereancistrum arcuatus Cohen, Kohn & Boeger, 2012 in Salminus brasiliensis Cuvier, 1816; Tereancistrum paranaensis Karling, Lopes, Takemoto & Pavanelli, 2014 in Schizodon borellii Boulenger, 1900, and Tereancistrum flabellum Zago, Yamada, Franceschini et al., 2017 in Leporinus spp. (Kritsky et al., 1980; Lizama et al., 2004; Cepeda et al., 2012; Cohen et al., 2012; Karling et al., 2014; Zago et al., 2017).

The new species is assigned to Tereancistrum genus due to the presence of distinctly spatulate accessory sclerites articulated to the tip of the superficial root of the ventral anchors, its two pairs of eyes, its overlapping gonads, and similar hooks (Kritsky et al., 1980; Kritsky & Boeger, 1989). The main characteristic that distinguishes this species from its congeners is the configuration of the ventral bar, which has an anvil-shaped and anterior projection with undulations and striations.

Regarding the male copulatory organ, Tereancistrum takemotoi n. sp. differs from T. kerri and T. arcuatus because these two species have a simple male copulatory organ, with a simple and unrolled conical tube, unlike Tereancistrum takemotoi n. sp. which exhibits a male copulatory organ in rings. The other species of the genus that also have a male copulatory organ arranged in rings, but with different number and direction of rings are: T. parvus with 3 1/2 rings counterclockwise, T. ornatus with 1 1/4 rings counterclockwise, T. paranaensis with 2 1/2 rings clockwise, T. flabellum with 3 1/2 ring clockwise, and T. curimba with 1 1/4 rings counterclockwise, while Tereancistrum takemotoi n. sp. has 2 1/4 rings arranged in counterclockwise.

Tereancistrum kerri and T. ornatus present an accessory piece articulated with the base of the male copulatory organ, while Tereancistrum takemotoi n. sp. and the other congeners exhibit an accessory piece not articulated with the base of the male copulatory organ.

Tereancistrum kerri and T. flabellum have a dextral dorsal vagina, while Tereancistrum takemotoi n. sp. and the other species of the genus have a sinistral vagina. In addition, for Tereancistrum takemotoi n. sp. the vagina is characterized as a sinuous and strongly sclerotic tube, unlike T. ornatus and T. pirassununguensis that have a slightly sclerotic vagina with a thin tube without sinuosity; T. parvus and T. flabellum which have a cone-shaped sclerotic vagina; and T. paranaensis and T. curimba that present sclerotized vagina forming a simple tube without coils.
A new species of *Tereancistrum* in Brazil

Regarding haptor structures, *Tereancistrum takemotoi* n. sp. has a Y-shaped dorsal bar and undulations in the anterior portion, while *T. kerri* has a U-shaped dorsal bar and *T. parvus* a V-shaped one. *T. paranaensis*, *T. arcuatus*, and *T. flabellum* exhibit a straight dorsal bar with slightly enlarged and rounded ends.

*Tereancistrum takemotoi* n. sp. exhibits a robust accessory sclerite with a spatulated end and a small deep groove opening at one end, unlike *T. arcuatus*, which has a thin accessory sclerite, with a longitudinally present groove on the entire surface; and *T. pirassununguensis*, which present a narrow accessory sclerite, without expansions in the extremities. Moreover, *T. parvus*, *T. paranaensis*, *T. flabellum* and *T. curimba* have and intermuscular structure between sclerites, while *Tereancistrum takemotoi* n. sp. and the other congeners do not have this structure.

*Tereancistrum kerri*, *T. parvus*, *T. ornatus*, *T. paranaensis* and *T. curimba* have filaments in the ventral and dorsal anchors, while *Tereancistrum takemotoi* n. sp. and the other species of *Tereancistrum* do not have filaments in either anchor. Regarding the hooks, *T. kerri* and *T. arcuatus*, have hooks of unequal sizes, while *Tereancistrum takemotoi* n. sp. and the other species of the genus have hooks of the same size.

*Tereancistrum toksonum* is the morphologically closest species to *Tereancistrum takemotoi* n. sp. In addition to parasitizing the same host species, they are similar due to the morphology of their dorsal bars (Y-shaped), their dorsal anchors with roots (superficial and deep) very distant from each other, and they both have an elongated deep root; and also, they both exhibit thickening in the posterior margin of the ventral bar, resulting in a bent angle in this bar portion; the accessory piece is not articulated with the male copulatory organ, and they both exhibit a strongly sclerotized sinistral vagina forming a sinuous and evident tube. However, these species differ in that *T. toksonum* exhibits 1¼ rings of the male copulatory organ running counterclockwise (in *T. takemotoi* is 2¼ rings). Although the two species’ dorsal bars have the same Y-shape, the dorsal bar in *T. takemotoi* have undulations in the anterior margin. The species also exhibit differences in the ventral bars: it is anvil-shaped in *T. takemotoi* and arched in *T. toksonum*.

This study presents the fourth species of the *Tereancistrum* described as a parasite of *P. lineatus*. The data obtained in this work broaden the geographic distribution of the genus and increase the knowledge on the parasitic diversity of Brazilian fish in freshwater environments.

**Acknowledgements**

The authors would like to thank Shelly Favorito de Carvalho from the Center for Electronic Microscopy in the Department of Biosciences of UNESP Botucatu for her assistance with the Confocal Laser Scanning Microscope. Vanessa D. Abdallah was supported by a Research fellowship from the São Paulo Research Foundation, or FAPESP (2012/23655-0), as was Rodney K. de Azevedo (2014/12862-0).

**References**

Agostinho AA, Júlio HF Jr, Gomes LC, Bini LM, Agostinho CS. Composição, abundância e distribuição espaço-temporal da ictiofauna. In: Vazzoler AEAM, Agostinho AA, Hahn NS, editors. *A planície de inundação do alto rio Paraná: aspectos físicos, biológicos e socioeconômicos*. Maringá: EDUEM; 1997. p. 179-208.

Boeger WA, Vianna RT. Monogenoidea. In: Thatcher VE, editor. *Amazon fish parasites*. Sofia: Pensoft Publishers; 2006. p. 42-116. (vol. 1).

Bush AO, Lafferty KD, Lotz JM, Shostak AW. Parasitology meets ecology on its own terms: Margolis et al. revisited. *J Parasitol* 1997; 83(4): 575-583. http://dx.doi.org/10.2307/3284227. PMID:9267395.

Cepeda PB, Ceccarelli PS, Luque JL. A new species of *Tereancistrum* (Monogenea, Dactylogyridae) parasitic on *Prochilodus lineatus* (Valenciennes, 1837) (Characiformes) from Mogi-Guaçu river, Brazil. *Neotrop Helminthol* 2012; 6(2): 205-210.

Cohen SC, Kohn A, Boeger WA. Neotropical Monogenoidea. 57. Nine new species of Dactylogyridae (Monogenoidea) from the gill of *Salminus brasiliensis* (Characidae, Characiformes) from the Paraná river, State of Paraná, Brazil. *Zootaxa* 2012; 3049(1): 57-68. http://dx.doi.org/10.11646/zootaxa.3149.1.3.

Gomori G. A rapid one-step trichrome stain. *Am J Clin Pathol* 1950; 20(7): 661-664. http://dx.doi.org/10.1093/ajcp/20.7_ts.661. PMID:15432364.

Gray P, Wess G. The use of polyvinyl alcohol and its derivates as microscopical mounting media. Part I. Water miscible mounting media. *J Microsc* 1950; 70(3): 287-291. PMID:24538706.
A new species of Tereancistrum in Brazil

Karling LC, Lopes LPC, Takemoto RM, Pavanelli GC. New species of Tereancistrum (Dactylogyridae) monogenean parasites of Schizodon borellii (Characiformes, Anostomidae) from Brazil, and emended diagnosis for T. parvus. Acta Sci Biol Sci 2014; 36(3): 365-369. http://dx.doi.org/10.4025/actascibilsci.v36i3.20216.

Kritsky DC, Boeger WA, Thatcher VE. Neotropical Monogenea. 7. Parasites of the pirarucu, Arapaima gigas (Cuvier), with descriptions of two new species and redescription of Dawestrema cycloancistrium Price and Nowlin, 1967 (Dactylogyridae: ancyrocephalinae). Proc Biol Soc Wash 1985; 98(2): 321-331.

Kritsky DC, Boeger WA. The phylogenetic status of the Ancyrocephalidae Bychowsky, 1937 (Monogenea: dactylogyroidea). J Parasitol 1989; 75(2): 207-211. http://dx.doi.org/10.2307/3282767. PMid:2926588.

Kritsky DC, Mizelle JD. Studies on monogenetic trematodes. XXXV. Some new and previously described North American species of Gyrodactylus. Am Midl Nat 1968; 79(1): 205-215. http://dx.doi.org/10.2307/2423166.

Kritsky DC, Thatcher VE, Kayton RJ. Neotropical Monogenoidea. 3. Five new species from South America with the proposal of Tereancistrum gen. n. and Trinibaculum gen. n. (Dactylogyridae: ancyrocephalinae). Acta Amazon 1980; 10(2): 411-417. http://dx.doi.org/10.1590/1809-43921980102411.

Leite LAR, Pelegrini LS, Agostinho BN, Azevedo RK, Abdallah VD. Biodiversity of the metazoan parasites of Prochilodus lineatus (Valenciennes, 1837) (Characiformes: Prochilodontidae) in anthropized environments from the Batalha River, São Paulo State, Brazil. Biota Neotrop 2018; 18(3): e20170422. http://dx.doi.org/10.1590/1676-0611-bn-2017-0422.

Lizama MAPL, Takemoto RM, Pavanelli GC. New species of Tereancistrum Kritsky, Thatcher & Kayton, 1980 (Monogenea: Dactylogyridae: Ancyrocephalinae) from the gills of Prochilodus lineatus (Osteichthyes: Prochilodontidae) from the upper Paraná River floodplain, Brazil. Syst Parasitol 2004; 57(1): 45-49. http://dx.doi.org/10.1023/B:SYPA.0000010684.67784.6e. PMid:14739674.

Lizama MAPL, Takemoto RM, Pavanelli GC. Influence of host sex and age on infracommunities of metazoan parasites of Prochilodus lineatus (Valenciennes, 1836) (Prochilodontidae) of the Upper Paraná River floodplain, Brazil. Parasite 2005; 12(4): 299-304. http://dx.doi.org/10.1051/parasite/2005124299. PMid:16402561.

Lizama MAPL, Takemoto RM, Pavanelli GC. Influence of the seasonal and environmental patterns and host reproduction on the metazoan parasites of Prochilodus lineatus. Braz Arch Biol Technol 2006; 49(4): 611-622. http://dx.doi.org/10.1590/S1516-89132006000500011.

Oyakawa OT, Menezes NA, Shibatta OA, Lima FCT, Langeani F, Pavanelli CS, et al. Peixes de água doce. In: Bressan PM, Kierulff MCM, Sugieda MAS, editors. Fauna ameaçada de extinção no Estado de São Paulo. São Paulo: Fundação Parque Zoológico de São Paulo; 2009. p. 107-123.

Rosa RS, Lima FCT. Os peixes brasileiros ameaçados de extinção. In: Machado ABM, Drummond GM, Paglia AP, editors. Livro vermelho da fauna brasileira ameaçada de extinção. Brasilia: MMA; 2008. p. 9-285.

Zago AC, Yamada FH, Franceschini L, Bongiovani MF, Yamada POF, Silva RJ. A new species of Tereancistrum (Monogenea, Dactylogyridae) from the gills of three Leporinus species (Characiformes, Anostomidae) and a revised description of Tereancistrum parvus. An Acad Bras Cienc 2017; 89(2): 1121-1131. http://dx.doi.org/10.1590/0001-3765201720160628.