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ORIGINAL ARTICLE / ARTÌCULO ORIGINAL

A NEW SPECIES OF *RHINOXENOIDES* (DACTYLOGYRIDAE) PARASITIZING *TRIPORTHEUS ANGULATUS* (SPIX & AGASSIZ, 1829) (CHARACIFORMES) FROM THE JURUÁ RIVER BASIN, BRAZIL

UNA NUEVA ESPECIE DE *RHINOXENOIDES* (DACTYLOGYRIDAE) QUE PARASITA A *TRIPORTHEUS ANGULATUS* (SPIX & AGASSIZ, 1829) (CHARACIFORMES) EN LA CUENCA DEL RÍO JURUÁ, BRASIL

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ABSTRACT

A new species of *Rhinoxenoides* was described parasitizing the gill filaments of neotropical triportheid fishes. The new species was described from *Triportheus angulatus* (Spix & Agassiz, 1829) from the Juruá River, State of Acre, Brazil. *Rhinoxenoides cruzeirensis* n. sp. resembles *Rhinoxenoides horacioschneideri* Santos Neto, Costa, Soares & Domingues, 2018, mainly by presenting a coiled male copulatory organ (MCO) with clockwise rings; accessory piece with articulation process extending within the coils to the base of MCO; dorsal anchor with well-developed superficial root, and dorsal bar absent. However, it differs from this by number of the coils of the MCO; accessory piece formed by a straight piece that expands in the middle, where it folds over itself; ventral bar short and robust; dorsal anchor with a well-developed superficial root twice as long as the deep root, with a small wing-shaped extension in the distal portion.

Keywords: Dactylogyridae – Neotropical region – Parasites of fishes – *Rhinoxenoides* – South America – Triportheidae

RESUMEN

Se describió una nueva especie de *Rhinoxenoides* parasitando los filamentos branquiales de peces triportheidos neotropicales. La nueva especie fue descrita de *Triportheus angulatus* (Spix & Agassiz, 1829) del río Juruá, estado de Acre, Brasil. *Rhinoxenoides cruzeirensis* n. sp. se asemeja a *Rhinoxenoides horacioschneideri* Santos Neto, Costa, Soares & Domingues, 2018, principalmente por presentar organo copulatorio masculino (OCM) con anillos espirales en sentido horario; pieza accesoria con proceso de articulación que se extiende dentro de espirales hasta la base de OCM; ancla dorsal con raíz superficial bien desarrollada y barra dorsal ausente. Pero se diferencia de este por el número de espirales de la OCM; pieza accesoria formada por una pieza recta que se expande en el centro, donde se pliega sobre sí misma; barra ventral corta y robusta; ancla dorsal con una raíz superficial bien desarrollada dos veces más larga que la raíz profunda con una pequeña extensión en forma de ala en la porción distal.

Palabras clave: Dactylogyridae – Parásitos de peces – Región neotropical – *Rhinoxenoides* – Sudamérica – Triportheidae

INTRODUCTION

The Amazon biome boasts the largest hydrographic system in the world and the largest freshwater reserve, containing around 20% of the global freshwater, and is considered one of the richest ecosystems on the planet. The Amazon hydrographic region represents 49.29% of the Brazilian territory, covering all or part of the states of Acre, Amazonas, Roraima, Rondônia, Pará, Amapá, Maranhão, Tocantins, and Mato Grosso (Tosta & Coutinho 2015; IBGE, 2019; Silva & Bampi, 2019).

Parasite diversity in Amazonian fishes is likely underestimated, given the region's rich ichthyofauna and high levels of endemism. This unique combination supports a remarkable variety of helminth species (Luque *et al*., 2017). Understanding fish parasite diversity could be an important tool for conserving global biodiversity (Luque *et al*., 2017).

The Juruá River Basin belongs to the great Amazon Basin and is located between the states of Acre and Amazonas. It is drained by extensive rivers in a general southwest-north-east direction, with rivers of great importance throughout its course, such as the Juruá River, which rises in the Peruvian Andes and flows into the Solimões River. This river is one of the main tributaries of the right bank of the Amazon River and is considered one of the most winding in the world, forming huge flood grids and thousands of lakes (ACRE, 2012; Costa *et al*., 2012; Sousa & Oliveira, 2016, Mota da Silva, 2020).

Although there is significant diversity of helminth species recorded in the Amazon, knowledge about parasite richness remains incomplete in certain areas. Most research focuses on the Central and Eastern Amazon, while the southwestern Amazon has received limited attention. Studies in the Juruá River basin have primarily documented parasitic fauna in aquaculture systems, leading to a substantial knowledge gap regarding the biodiversity of parasites associated with fish in natural systems in this southwestern Amazon region (Virgilio *et al*., 2021).

Recent research on the Juruá River has identified five species of monopisthocotylan, including three *Cosmetocleithrum* spp. parasitizing the catfish *Oxydoras niger* (Valenciennes, 1821) in the Juruá River: *Cosmetocleithrum basicomplexum* Silva, Meneses, Martins, Cohen, Costa & Justo, 2023, *Cosmetocleithrum confusus* Kritsky, Thatcher & Boeger, 1986, *Cosmetocleithrum sacciforme* Silva, Meneses, Martins, Cohen, Costa & Justo, 2023, and *Unibarra juruaensis* Justo, Martins & Cohen, 2023 and *Unibarra paranoplatensis* (Suriano & Incorvaia, 1995) parasitizing *Pimelodus blochii* Valenciennes, 1840 (Justo *et al*., 2023; Silva *et al.,* 2023).

Within the order Characiformes, Triportheidae includes five genera and 23 species (Froese & Pauly, 2024), among which *Triportheus angulatus* (Spix & Agassiz, 1829), a benthopelagic freshwater fish, distributed in the Amazon River basin in South America and exclusive to the Neotropical region (Malabarba, 2004, Froese & Pauly, 2024). These species are exploited by subsistence fishing and have become an alternative source of fish since the natural stocks of other commercially important fish species in the Amazon have declined over time (Cajado *et al*., 2023).

To date, 29 species of Monopisthocotyla have been described in South America from Triportheidae hosts, with the majority of these species classified under the genus *Anacanthorus* Mizelle & Price, 1965 (see Table).

In continuation of the studies carried out in the Juruá River Basin in Brazil, aiming to describe the fauna of helminths parasitizing freshwater fish, a new species of *Rhinoxenoides* Santos Neto, Costa, Soares & Domingues, 2018 was found infesting the gills of *T. angulatus*. The new species described here enhances our understanding of the diversity of monopisthocotylan parasites in triportheid fishes, as well as offering valuable insights into the region's biodiversity, and underscoring the importance of expanding research across the Amazon basin.

MATERIAL AND METHODS

In January 2024, two specimens of *T. angulatus* were captured with gill nets and a hook and line from Jurua River, Acre, Brazil (7°40′34.1″S, 72°39′39.5″W) (Fig. 1). All collections obtained environmental licensing through the Biodiversity Authorization and Information System (SISBIO, 396871-1). The gills were removed and placed in vials containing hot water (~65ºC) and they were then vigorously shaken to detach the parasites from the gill filaments. Absolute ethanol was added to reach a concentration of 70%. The parasites were collected from the sediment and gill arches in the laboratory with the aid of a stereoscopic microscope. The specimens were mounted unstained in Hoyer's medium for the study of the sclerotized parts. Photomicrographs and drawings were taken using a Zeiss® Axioskop microscope micrographic system, with a differential interference contrast (DIC) apparatus and an Olympus BX 41 microscope with phase contrast, equipped with a camera lucida. All measurements are in micrometers, and the means are followed by the range in parentheses and by the number of specimens measured when more than two. Dimensions of organs and other structures represent the greatest distance; lengths of curved or bent structures (bars and accessory piece) represent the straight-line distances between extreme ends. The numbering of hook pairs follows

Mizelle & Price (1963). Holotype and paratypes were deposited in the helminthological collection of the Instituto Oswaldo Cruz (CHIOC).

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

RESULTS

TAXONOMY

Class Monopisthocotyla Brabec, Salomaki, Kolısko, Scholz & Kuchta, 2023

Order Dactylogyridea Bychowsky, 1937

Dactylogyridae Bychowsky, 1933

Rhinoxenoides Santos Neto, Costa, Soares & Domingues, 2018

Rhinoxenoides cruzeirensis n. sp. (Fig. 2 A-G)

Type-host: *Triportheus angulatus* (Spix & Agassiz, 1829) (Characiformes: Triportheidae).

Site in host: Gill lamellae.

Type-locality: Juruá River, Acre, Brazil (7°40′34.1″S, 72°39′39.5″W).

Parasitological indices: Total number of hosts: 2; number of infected hosts: 1; total number of parasites: 11.

Deposited material: Holotype (CHIOC 40615a); Paratypes (CHIOC 40615b-i).

Etymology: The specific name is dedicated to the municipality of Cruzeiro do Sul, in the state of Acre, Brazil, where the parasite was recovered.

Description (Based on 11 specimens, mounted in Hoyer's medium). Body fusiform, elongated, comprising cephalic region, trunk, and haptor. Tegument thin, smooth. Body 509 (415–625; n = 10) long, 74 (60–85; n = 11) wide at the level of germarium. Anterior region with four cephalic lobes well developed, two terminal, two bilateral; three pairs of head organs; cephalic glands indistinct. Two pairs of eyespots, posterior pair larger and closer together than anterior pair; accessory granules absent (Fig. 2A). Pharynx spherical 20 (18–22; n = 6) in diameter. Esophagus short. Two intestinal ceca confluent posteriorly to testis, lacking diverticula. Gonads intercecal, tandem. Germarium ventral to testis, 74 (70–80; n = 4) long. Vagina and vaginal canal slightly sclerotized; vaginal aperture sinistroventral. Seminal receptacle spherical. Mehlis' gland, uterus, oviduct, and ootype not observed. Egg 72 long by 45 wide (n = 1). Vitellaria extends throughout the trunk, except in areas of other reproductive organs. Prostatic reservoir elongated posterior to male copulatory organ (MCO), 41 (40–42; n = 3) long. Copulatory complex comprising MCO and accessory piece. Coiled MCO with 1½ clockwise rings, 17 (15–20; n = 5) proximal ring diameter; accessory piece formed by a straight piece that expands in the middle, where it folds over itself; in the proximal portion it articulates with the base of the MCO, 40 (35–44; n = 4) long, through a copulatory ligament (Fig. 2B). Peduncle inconspicuous. Haptor subtriangular, 65 (52–73; n = 8) wide (Fig. 2A). Ventral bar short and wide, 19 (15–22; n = 9) long (Fig. 2C). Dorsal bar absent. Ventral and dorsal anchor dissimilar in size and shape. Ventral anchor with superficial root elongated, well developed; deep root short, rounded; evenly curved shaft, point not passing from the level of the type of superficial root, 69 (65–72; n = 16) long, base 28 (24–31; n = 13) (Fig. 2D). Dorsal anchor with a well-developed superficial root, slender, twice the size of the deep root and a small wing-shaped extension in the distal portion; deep root elongated, rounded at the tip; straight shaft, short point hook-shaped, 55 (53–58; n = 17) long, base 10 (8–11; n = 7) (Fig. 2E). Hooks similar in shape, shank divided into two subunits, distal part of shank inflated; filamentous hook (FH) loop extending close to the beginning of shank dilation, erect thumb, curved long shaft, delicate point. Hook pairs 2, 3, 4, 6, and 7, 21 (20–22; n = 8) long (Fig. 2F), hook pairs 1 and 5, 16 (15–16; n = 4) long (Fig 2G).

Remarks: The new species was allocated in *Rhinoxenoides* as it shares characteristics of the genus, such as MCO formed by a coiled tube with clockwise rings articulated to the accessory piece by copulatory ligament; dorsal anchor with superficial root twice the size of the deep root; and by the absence of dorsal bar. *Rhinoxenoides cruzeirensis* n. sp. is similar to *Rhinoxenoides horacioschneideri* Santos Neto, Costa, Soares & Domingues, 2018, the only species of the genus, by presenting a well-developed ventral anchor with a superficial truncated root; prostatic reservoir elongated, not divided into regions; vagina sinistral; Coiled MCO with clockwise rings; accessory piece with articulation process extending within coils to the base of MCO; dorsal anchor with well-developed superficial root comprising three times the total length, and dorsal bar with enlarged ends. The new species differs from *R. horacioschneideri* by presenting an Coiled MCO with 1½ clockwise rings; accessory piece formed by a straight piece that expands slightly at its center, where it bends into the shape of an elbow (2½ coils; comprising variable distal sheath in *R. horacioschneideri*); ventral bar short and robust (V-shaped in *R.*

horacioschneideri); dorsal anchor with a well-developed superficial root twice as long as the deep root and a small wing-shaped extension in the distal portion (three times longer than the deep root; small wing-shaped extension in the distal portion absent in *R. horacioschneideri*) and by the hook size (1-3 and 5 smaller than the others in *R. horacioschneideri*). In the original description, the authors stated that the species present the dorsal anchor with superficial root twice the size of deep root, but in the original drawing the superficial root is shown as three times as long as deep root, feature confirmed by examining the type material deposited in the Helminthological Collection of Oswaldo Cruz Institute CHIOC 39025a (holotype); 39025b-c (paratypes); 39026 (paratype), and 39027a-d (vouchers).

DISCUSSION

Rhinoxenoides Santos Neto, Costa, Soares & Domingues, 2018 was erected by Santos-Neto *et al.* (2018) to accommodate *R. horacioschneideri* described from *Acestrorhynchus falcatus* (Bloch, 1794). It is characterized by the morphology of copulatory complex, composed of a coiled tube with clockwise rings articulated to the accessory piece by copulatory ligament; by the morphology of haptoral sclerites as dorsal anchor with the superficial root twice as long as the deep root; straight shaft, curved point; and by the absence of dorsal bar (Santos-Neto et al., 2018). The monotypic genus presents shared characteristics, such as the presence of a copulatory ligament and dorsal anchors with a straight long shaft and absence of a dorsal bar, with *Rhinoxenus* Kritsky, Boeger & Thatcher, 1988, but differ by the coiled MCO with counterclockwise coils, absence of superficial and deep roots in both pairs of anchors, and dorsal anchor with elongate, straight shaft in *Rhinoxenus*. *Protorhinoxenus* Domingues & Boeger, 2002, a genus closely related to *Rhinoxenus* was proposed by Domingues & Boeger (2002). Both genera share morphological features such as a coiled MCO with counterclockwise coils, absence of superficial and deep roots in both pairs of anchors, and dorsal anchors with elongated and straight shafts. *Rhinoxenus* and *Rhinoxenoides* share the character of the absence of a dorsal bar, differing from *Protorhinoxenus* in this feature. According to Santos-Neto et al. (2018), the absence of dorsal bar could be interpreted as an independent secondary loss in *Rhinoxenoides* and *Rhinoxenus*, or its absence could be shared by the three genera as a synapomorphy with secondary acquisition in *Protorhinoxenus*. These authors proposed a cladistic hypothesis, in which species of *Protorhinoxenus*, *Rhinoxenoides*, and *Rhinoxenus* share the presence of a copulatory ligament and dorsal anchors with a straight long shaft. These features are also observed in the new species of *Rhinoxenoides* proposed herein. The authors stated that species of *Protorhinoxenus*, *Rhinoxenoides*, and *Rhinoxenus* seem to be exclusively found infecting characiform fishes from South America. According to our results, by proposing another species of *Rhinoxenoides* parasitizing a characiform fish belonging to Triportheidae, while the previously reported species of the genus was collected from Acestrorhynchidae, propose that members of these three other genera are not restricted to members of one host family within Characiformes. The finding shows that the parasite faunas of fishes need to be further characterized through additional sampling for monopisthocotylan of species of all characiform families, and that cladistics studies are necessary, to elucidate host-parasite relationships in the freshwater fishes of the Neotropical Region.

Author contributions: CRediT (Contributor Roles Taxonomy)

SCC = Simone Chinicz Cohen
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Conceptualization: SCC
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Writing – review & editing: SCC, MCNJ

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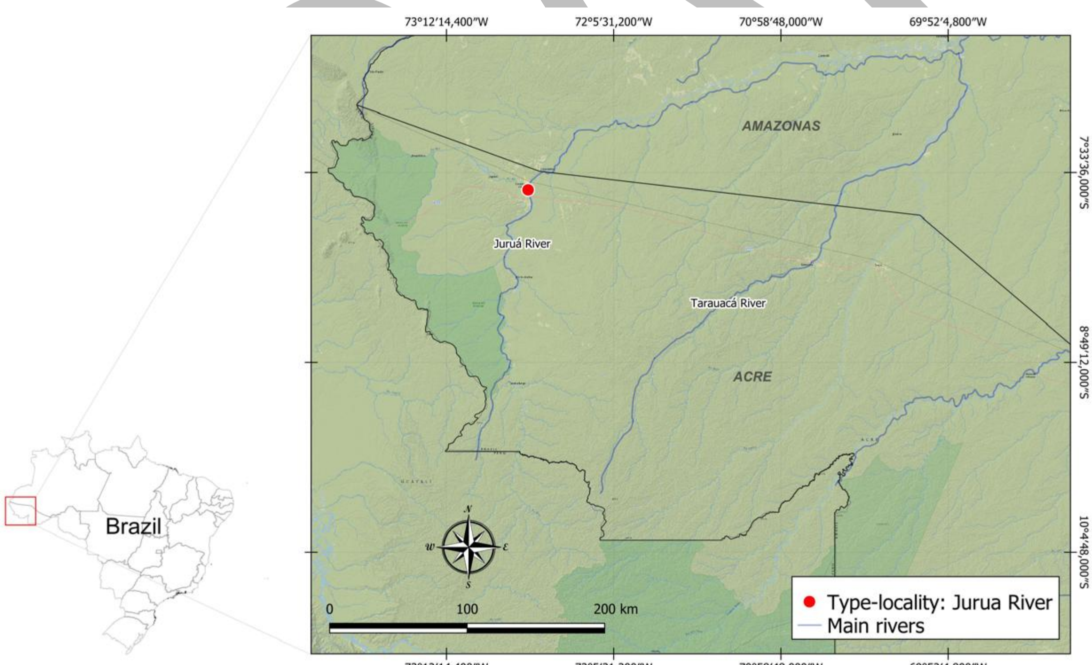

Figure 1. Location of the study area, Jurua River, Acre, Brazil (7°40′34.1″S, 72°39′39.5″W) Cruzeiro do Sul, Acre State, Brazil.

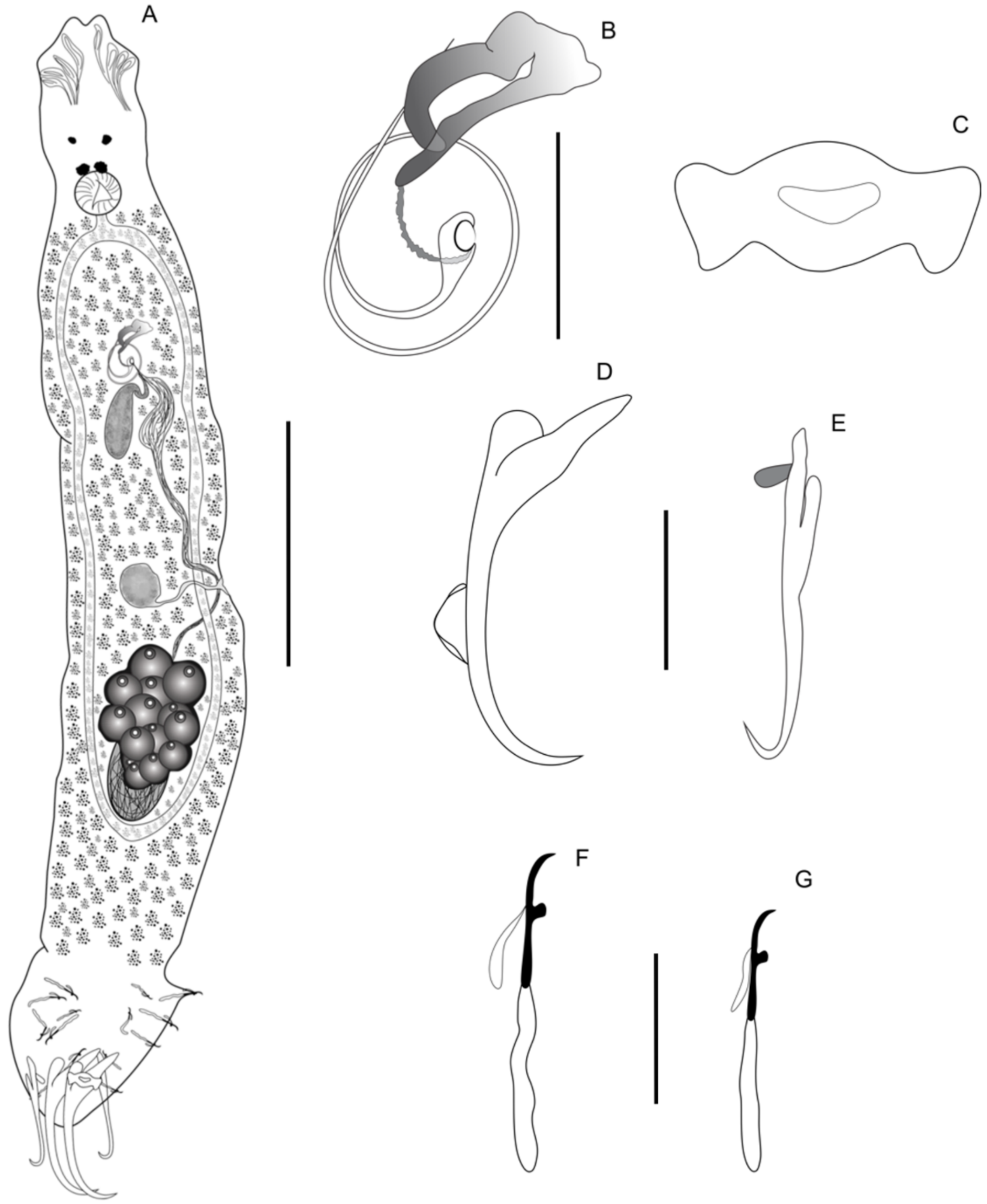

Figure 2. *Rhinoxenoides cruzeirensis* n. sp. from *Triportheus angulatus*. A, Total ventral view (composite); B, Copulatory complex ventral view; C, Ventral bar; D, Ventral anchor; E, Dorsal anchor; F, Hook pairs 2-4,6,7. G, Hook pairs 1,5. Scale bars: A, 100 µm, B, 20 µm, C, F,G, 10 µm, D, E, 30 µm.

Table 1. Species of Monopisthocotyla reported in South America from *Triportheus* spp. hosts.

MONOPISTHOCOTLA	HOST	COUNTRY	REFERENCE
Anacanthorus acuminatus Kritsky, Boeger & Van Every, 1992	*T. albus, T. angulatus, T. elongatus*	BR	Kritsky *et al.* (1992); Moreira *et al.* (2017)
Anacanthorus alatus Kritsky, Boeger & Van Every, 1992	*T. albus, T. elongatus*	BR	Kritsky *et al*. (1992)
Anacanthorus andersoni Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al*. (1992)
Anacanthorus bellus Kritsky, Boeger & Van Every, 1992	*T. albus, T. elongatus, Triportheus* sp.	BR	Kritsky *et al.* (1992)
Anacanthorus calophallus Kritsky, Boeger & Van Every, 1992	*T. elongatus*	BR	Kritsky *et al.* (1992)
Anacanthorus carinatus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al*. (1992)
Anacanthorus chaunophallus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al.* (1992); Moreira *et al.* (2017)
Anacanthorus chelophorus Kritsky, Boeger & Van Every, 1992	*T. angulatus, Triportheus* sp.	BR	Kritsky *et al.* (1992); Moreira *et al.* (2017)
Anacanthorus cornutus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al.* (1992)
Anacanthorus euryphallus Kritsky, Boeger & Van Every, 1992	*T. albus, T. angulatus, T. elongatus*	BR	Kritsky *et al.* (1992); Moreira *et al.* (2017)
Anacanthorus formosus Kritsky, Boeger & Van Every, 1992	*T. elongatus, Triportheus* sp.	BR	Kritsky *et al.* (1992)
Anacanthorus furculus Kritsky, Boeger & Van Every, 1992	*T. elongatus, T. rotundatus*	BR	Kritsky *et al.* (1992); Santos & Tavares-Dias (2017)
Anacanthorus glyptophallus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al.* (1992)
Anacanthorus lygophallus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al.* (1992); Moreira *et al.* (2017)

Anacanthorus nanus Kritsky, Boeger & Van Every, 1992	*T. angulatus*	BR	Kritsky *et al.* (1992)
Anacanthorus pelorophallus Kritsky, Boeger & Van Every, 1992	*T. elongatus*	BR	Kritsky et al. (1992)
Anacanthorus pithophallus Kritsky, Boeger & Van Every, 1992	*T. angulatus*, *T. curtus*, *T. rotundatus*	BR	Kritsky *et al*. (1992); Oliveira *et al.* (2016); Moreira *et al.* (2017), Santos & Tavares-Dias (2017)
Anacanthorus quinqueramus Kritsky, Boeger & Van Every, 1992	*T. albus, T. elongatus, Triportheus sp.*	BR	Kritsky *et al.* (1992)
Anacanthorus ramulosus Kritsky, Boeger & Van Every, 1992	*T. albus, T. elongatus*	BR	Kritsky *et al.* (1992)
Anacanthorus strongylophallus Kritsky, Boeger & Van Every, 1992	*T. elongatus*	BR	Kritsky *et al.* (1992)
Anacanthorus tricornis Kritsky, Boeger & Van Every, 1992	*T. angulatus, T. elongatus*	BR	Kritsky *et al.* (1992)
Ancistrohaptor falcatum Agarwal & Kritsky, 1998	*T. elongatus*	BR	Agarwal & Kritsky (1998)
Ancistrohaptor falciferum Agarwal & Kritsky, 1998	*T. albus, T. angulatus, T. elongatus, Triportheus* sp.	BR	Agarwal & Kritsky (1998); Moreira *et al*. (2017)
Ancistrohaptor falcunculum Agarwal & Kritsky, 1998	*T. albus, T. angulatus, T. elongatus*	BR	Agarwal & Kritsky (1998); Moreira *et al.* (2017)
Ancistrohaptor forficata Diniz, Sousa, Yamada & Yamada, 2025	*T. signatus*	BR	Diniz *et al.* (2025)
Jainus iquitensis Morey, Viana, Chota & Chero, 2025	*T. angulatus*	PE	Morey *et al.* (2025)
Jainus loretoensis Morey, Viana, Chota & Chero, 2025	*T. angulatus*	PE	Morey *et al.* (2025)
Jainus sardinae Morey, Viana, Chota & Chero, 2025	*T. angulatus*	PE	Morey *et al.* (2025)
Rhinoxenus anaclaudiae Domingues & Boeger, 2005	*T. angulatus, T. nematurus, Triportheus* sp.	BR	Domingues & Boeger (2005); Moreira *et al.* (2017)

BR: Brazil; PE: Peru.