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A new species of mailed catfish of genus *Rhadinoloricaria* (Siluriformes: Loricariidae: Loricariinae) from Rio Negro basin, Brazil

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Abstract

During a recent collection expedition to the Rio Negro, in the state of Amazonas, Brazil, eight individuals of an unknown species were collected, with a combination of characteristics that placed the species in the genus *Rhadinoloricaria*. Furthermore, the presence of two autapomorphic characteristics, including numerous elongated papillae on the lower lip and unbranched barbelets on the margin of lower lip, suggests that it is a new species. From morphological and phylogenetic analyses, including the sequencing of specific genes to calculate the maximum likelihood analyses, coupled with osteological computed tomography (CT) scan analyses, the authors corroborated that the specimens represent a new species of *Rhadinoloricaria*, described in the present study.

KEYWORDS
Cascudos, freshwater fishes, *Pseudohemiodon* group, South America

1 | INTRODUCTION

The subfamily Loricariinae includes the mailed catfishes, which lack an adipose fin and possess a depressed body and caudal peduncle. Members of this group exhibit high variation in external morphology, mainly in the number of teeth, caudal peduncle depth and colouration of the body, which may vary in association with habitat (Covain *et al*., 2016). The lips are highly diverse among genera, ranging from papillose to filamentous or smooth (Covain *et al*., 2016; Covain & Fisch-Muller, 2007; Isbrücker, 1979).

The systematics and classification of the Loricariinae had great contributions of some authors during the last century, including Regan (1904), Eigenmann (1924), Boeseman (1971; 1976), Rapp Py-Daniel (1997) and Isbrücker (1979), who classified the subfamily into four tribes and eight sub-tribes based on external morphology, and more recently, Covain and Fisch-Muller (2007) proposed the division of the 250 species of Loricariinae into two tribes: Harttiini and Loricariini. Furthermore, Covain and Fisch-Muller (2007) divided Loricariini into four sub-groups: the

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Pseudohemiodon, *Loricaria*, *Rineloricaria* and *Loricariichthys* groups. Covain *et al.* (2016) in a comprehensive multilocus molecular analysis using mitochondrial and nuclear genes, and Roxo *et al.* (2019) in a broad study of the family Loricariidae using phylogenomic ultraconserved elements (UCE), found similar results, and corroborated the classification of Covain *et al.* (2008) who found the Loricariini to comprise two sub-tribes: the Loricariina and the Farlowellina. In addition, in the Covain *et al.* (2016) study, the clade composed of *Dasyloricaria* + *Fonchiiloricaria* formed the sister group to other members of the sub-tribe Loricariina.

According to the study of Covain *et al.* (2016) the *Pseudohemiodon* group is monophyletic with strong statistical support and includes five genera: *Crossoloricaria*, *Apistoloricaria*, *Rhadinoloricaria*, *Pseudohemiodon* and *Planiloricaria*. Covain and Fisch-Muller (2007) also proposed the inclusion of the genera *Pyxiloricaria*, *Reganella* and *Dentectus*. Furthermore, Covain *et al.* (2016) found *Rhadinoloricaria* to be a paraphyletic group and proposed *Apistoloricaria* and the Cis-Andean *Crossoloricaria* species as synonyms of *Rhadinoloricaria*. The most recent study of subfamily Loricariinae (Londoño-Burbano & Reis, 2021) placed six genera in the *Pseudohemiodon* group, including *Dentectus*, *Reganella*, *Crossoloricaria*, *Planiloricaria*, *Pseudohemiodon* and *Rhadinoloricaria*, based on morphological and molecular characters. About the last genus, Provenzano-Rizzi and Barriga-Salazar (2020) described a new species of *Rhadinoloricaria* from Ecuador, and from a comparative morphological analysis, the authors suggested the revalidation of *Apistoloricaria*, including *Apistoloricaria condei*, *Apistoloricaria laani*, *Apistoloricaria listrorhinos* and *Apistoloricaria ommation*, and suggested that the generic allocation of *Rhadinoloricaria bahuaja* and *Rhadinoloricaria rhami* is uncertain.

This continued taxonomic changes in the subfamily Loricariinae, including the description of new species, suggests the importance of systematic reviews and indicates the needed revision of the *Pseudohemiodon* group to better understand generic and species limits. Herein the authors described a new species of *Rhadinoloricaria* based on molecular and morphological data from Rio Negro basin, Amazonas State, Brazil.

2 | MATERIALS AND METHODS

2.1 | Ethics statement

The specimens analysed in the present study were collected under licence number 13843-4, issued by the Brazilian Institute for the Environment and Renewable Natural Resources (IBAMA 13843-4). The animals in this research were used in accordance with Brazilian laws on animal welfare, and the present research was approved by the Ethics Committee on Animal Use of the Biosciences Institute of UNESP (licence number 2325010620).

2.2 | Conservation status

To access the conservation status of the new species, the authors performed two analyses, the extent of occurrence (EOO) and the area of occupancy (AOO), using GeoCat software (geocat.kew.org) (Bachman & Moat, 2012).

2.3 | Morphological analysis

Measurements follow Vera-Alcaraz *et al.*, 2012, with exception of the total length (TL) and superior caudal-fin ray (SCFR), because all specimens of the new species are with the superior caudal-fin ray broken. Body plate nomenclature follow Rapp Py-Daniel (1997) adapted and Paixão and Toledo-Piza (2009). Morphological data were recorded for the specimens using a digital calliper (0.01 mm). Dorsal-fin ray counts includes the first unbranched ray as "i" and zoological nomenclature follow the ICZN (International Commission on Zoological Nomenclature) (Ride, 1999). Type series was deposited at MZUSP (Museu de Zoologia, Universidade de São Paulo, São Paulo State, Brazil), LBP (Laboratório de Biologia e Genética de Peixes, São Paulo State, Brazil) and ANSP (Academy of Natural Sciences of Drexel University).

The authors examined the osteology of two specimens of the new taxon using computed tomography (CT). Scans were performed at the Microscopy and Imaging Facility at the American Museum of Natural History using a GE Phoenix v|tome|x with a 180 kV Nano Tube (General Electric, Fairfield, CT, USA). Scan resolution ranged from 10.6 to 24.3 µm. Beam energy was 120 kV and 166 mA. Scans were reconstructed using Phoenix datos|x (General Electric, Wunstorf, Germany) and were rendered and edited using VGStudio Max 3.3.4 (Volume Graphics, Heidelberg, Germany).

Buccal ornamentation is an important character to allocate the species of *Pseudohemiodon* group (Provenzano-Rizzi & Barriga-Salazar, 2020; Rojas-Molina *et al.*, 2019), and thus the authors designate the characteristics of lower lip of the new species described here (Figure 1a) comparing it to *Rhadinoloricaria* species (Figure 1b).

2.4 | Molecular analysis

2.4.1 | Taxon sampling

The authors used the data matrix from Covain *et al.* (2016), which included 360 terminal taxa, spanning 260 species. In-group sampling comprised 343 terminal taxa. Related taxa included 17 Hypostominae, 1 Hypoptopomatinae and 1 Rhinelepinae. The tree was rooted in *Pseudorinelepis genibarbis* (Loricariidae: Rhinelepinae). In addition, the authors included to the data matrix two samples of *Dentectus barbarmatus* and four samples of the new species that were sequenced in the present study (Supporting Information Table S1). As detailed in Covain *et al.* (2016) all the analysed samples are deposited at the Muséum d'histoire naturelle de la Ville de Genève (MHNG); Academy of Natural Sciences of Drexel University, Philadelphia (ANSP); Smithsonian Tropical Research Institute (STRI), Panama; Laboratório de Biologia de Peixes, Departamento de Morfologia, Universidade Estadual Paulista, Campus de Botucatu (LBP); Auburn University Museum, Auburn (AUM); and Museu de Ciências e Tecnologia of the Pontifícia Universidade Católica do Rio Grande do Sul (MCP), Porto Alegre. Tissue of both species included in the present study (*i.e.*, *D. barbarmatus* and the species described here) are deposited at the Laboratório de Biologia e Genética de Peixes (LBP), Unesp, Botucatu, São Paulo State,

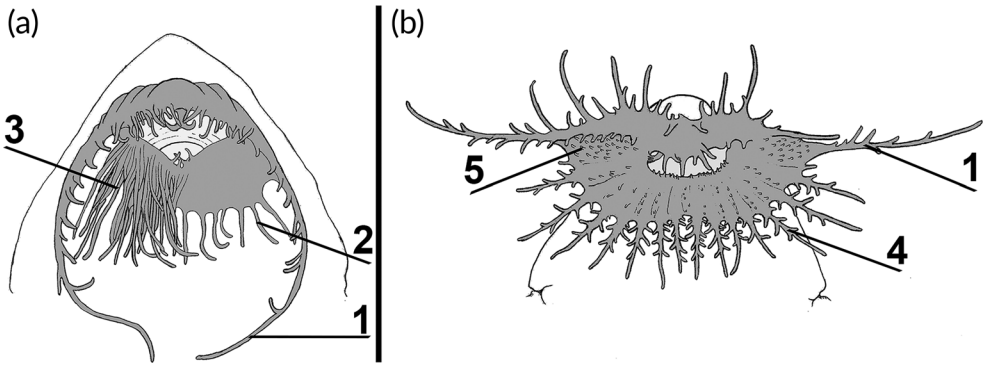

FIGURE 1 Scheme representing the buccal ornamentation of (a) *Rhadinoloricaria papillosa* (MZUSP uncat., *Rhadinoloricaria papillosa*, holotype) and (b) *Rhadinoloricaria bahuaja* (MUSM9916, holotype, Provenzano-Rizzi and Barriga-Salazar (2020) edit.). 1, maxillary barbel; 2, smooth barbelets; 3, elongated smooth papillae; 4, barbelets with lateral branches; 5, short papillae

Brazil. The complete list of tissues and voucher number used in this study is provided in Supporting Information Table S1. All sequences are deposited in GenBank.

2.4.2 | DNA extraction and sequencing

The authors used tissue from muscle and fin of specimens preserved in 95% alcohol and stored them at −20°C for DNA extraction, following the protocol described by Aljanabi and Martinez (1997). Partial sequences of the 16S genes–16S rRNA, forward 5′-ACG CCT GTT TAT CAA AAA CAT-3′, reverse 5′-CCG GTC TGA ACT CAG ATC ACG T-3′ (Kocher *et al*., 1989) were amplified by PCR.

The amplification was conducted in a total volume of 12.5 μl with 1.25 μl of 10× buffer (10 mM Tris–HCl + 15 mM $MgCl_2$), 0.5 μl of the dNTPs (200 nM of each), 0.5 μl of each 5 mM primer, 0.05 μl of platinum Taq polymerase (Invitrogen), 1 μl of template DNA (12 ng) and 8.7 μl of dd H_2O. The PCR reactions consisted of 30–40 cycles, 30 s at 95°C, 15–30 s at 48–58°C and 45–90 s at 72°C. All the PCR products were first identified visually on a 1% agarose gel and then purified using ExoSap-IT (USB Corporation) following the manufacturer's instructions. The purified PCR products were sequenced using the Big DyeTM Terminator v 3.1 Cycle Sequencing Ready Re-action kit (Applied Biosystems), purified by ethanol precipitation and loaded into a 3130-Genetic Analyser automatic sequencer (Applied Biosystems) available at LBP.

2.4.3 | Sequencing and phylogenetic analysis

The individual sequences for the specimens were analysed in BioEdit 5.0.9 software (Hall, 1999), and a consensus sequence was obtained. After obtaining the consensus sequences the authors aligned the 16S from two samples of *D. barbarmatus* and four samples of the new species with the matrix of Covain *et al*. (2016) that included one nuclear gene [Fish Reticulon-4 receptor (*f-rtn4r*)] and two mitochondrial genes (12S and 16S). The matrix was aligned in MUSCLE (Edgar, 2004) using the default parameters and inspected visually. To evaluate the saturation of the matrix by substitution, the authors calculated the index of substitution saturation (Iss), as described by Xia *et al*. (2003) and Xia and Lemey (2009), and the transition/transversion rate, in DAMBE 5.2.31 (Xia & Xie, 2001). The Iss was calculated without taking gaps into account.

Maximum likelihood (ML) analyses were run in RAxML Web-Servers (Stamatakis *et al*., 2008). Bootstrap (BS) resampling (Felsenstein, 1985) was used to evaluate the support for each node, based on 1000 replicates. Random starting trees were used for each independent ML tree search, whereas all other parameters were set at the default values. The ML analyses used the GTR model of nucleotide substitution.

3 | RESULTS

3.1 | *Rhadinoloricaria papillosa* n. sp.

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Figures 2 and 3; Table 1.

3.2 | Holotype

MZUSP 127013, female, 73.92 mm SL, Brazil, municipality of Barcelos, Amazonas State, Rio Demeni, Rio Negro, Rio Amazonas basin, 00° 24′ 57.6″ S 62° 53′ 37.8″ W, 12 August 2018, col. Gabriel SC Silva, Maxwell Bernt, Brandon Waltz.

3.3 | Paratypes

All from Brazil, Amazonas State, Rio Amazonas basin, municipality of Barcelos, Amazonas State. LBP 26829 (2, 59.06–66.91 mm SL), collected with holotype.

LBP 26739, (1, 48.85 mm SL), Rio Negro, 01° 01′ 37.1″ S 62° 48′ 52.2″ W, 04 August 2018, col. Gabriel SC Silva, Maxwell Bernt, Brandon Waltz. LBP 26799, (1, 62.98 mm SL), Rio Demeni, 00° 38′ 28.8″ S 62° 53′ 47.5″ W, 10 August 2018, col. Gabriel SC Silva, Maxwell Bernt, Brandon Waltz. LBP 26812, (1, 40.77 mm SL), Rio Aracá, 00° 24′ 23.4″ S 62° 56′ 08.1″ W, 11 August 2018, col. Gabriel SC Silva, Maxwell Bernt, Brandon Waltz, ANSP 207807,

FIGURE 2 MZUSP 127013, *Rhadinoloricaria papillosa*, holotype, female, 74.46 mm standard length (SL) municipality of Barcelos, Amazonas State, Rio Demeni, Rio Negro basin. Photographed by Lais Reia

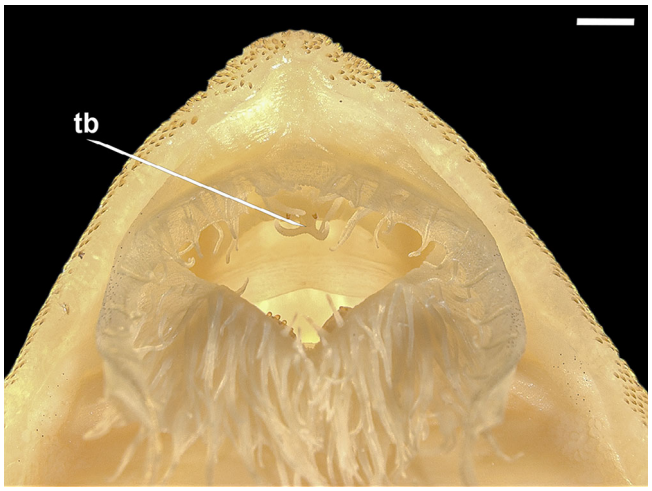

FIGURE 3 Image indicating thick barbelet inside premaxillae in holotype of *Rhadinoloricaria papillosa* n. sp. (MZUSP127013)

(1, 48.00 mm SL), Rio Negro, 01° 06′ 46.2″ S 62° 37′ 40.0″ W, 7 August 2018, col. Gabriel SC Silva, Maxwell Bernt, Brandon Waltz (Figure 5), ANSP 207862, (1, 63.40 mm SL), Rio Demeni, 00° 35′ 04.3″ S 62° 53′ 24.5″ W, 10 August 2018, col. M. Bernt, G. Costa Silva, B. Watz & R. Nilson.

3.4 | Diagnosis

In addition to the phylogenetic data, the new species were included in the genus *Rhadinoloricaria* by having the combination of characters: maxillary barbel surpassing the pectoral-fin base, few teeth in both jaws (1–2 in premaxilla; 4–6 in dentary), edge of lips with presence of barbelets, surface of lips with papillae and the presence of one thick barbelet behind premaxillae and other two barbelets next to each premaxillae, only in the holotype the barbelet behind premaxillae ramifies into two conical arms with equal size (Figure 3), whereas in paratypes this thick barbelet is not ramified. *R. papillosa* differs from all congeners by the presence of a unique pattern of the buccal ornamentation, with numerous elongated smooth papillae in irregular rows on lower lip, Figure 1a,3 (*vs.* papillae not elongated, Figure 1b,5). In addition, the new species differs from *Rhadinoloricaria macromystax*, *Rhadinoloricaria stewarti* and *Rhadinoloricaria listrorhinos* by having fewer premaxillary teeth 1–2 (*vs.* 4–6 in *R. macromystax* and *R. stewarti*, and 5–6 in *R. listrorhinos*) and by having a triangular-shaped head in dorsal view, Figure 4a [*vs.* spatula-shaped head, Figure 4b (*R. listrorhinos*) and Figure 4f (*R. macromystax* and *R. stewarti*)], and from *R. rhami* by maxillary barbel reaching the middle of the pectoral fin (*vs.* maxillary barbel not reaching the gill opening).

3.5 | Description

Morphometric and meristic data in Table 1. Small to medium-sized loricariid (40.77–73.92 mm SL, mean = 58.75 mm SL).

In lateral view, dorsal profile of head straight and ascending from snout tip to area between orbitals, slightly concave and descending from that point to dorsal fin insertion; slightly concave and ascending to end of caudal peduncle. Ventral surface of body, slightly convex at head, straight to slightly concave from posterior end of head to pelvic-fin insertion and straight to origin of caudal fin. Snout tip pointed in dorsal view, lateral margins of head straight. Nostril relatively small. Odontodes of head and trunk small, thick and aligned in series (Figure 5a). Abdomen naked with a central and lateral series of platelets better defined in largest specimens available (>50 mm), Figure 2. Head depressed; dorsal portion convex; superior margin of orbits elevated forming pronounced ridges; area between orbitals flat. Posterior portion of head with two conspicuous lines of odontodes beginning on anterior margin of frontals, continued along the supraoccipital. Head and body plates covered with minute and evenly distributed odontodes. Tip and lateral margin of snout completely covered by minute odontodes. Eye dorso-laterally placed, relatively large (12.53%–16.19% in HL), rounded to slightly oval horizontally; shallow posterior notch present on orbit. Margin of lips with barbelets; lower lip surface with numerous elongated papillae. Lower lip larger than upper lip; upper lip with margins covered by barbelets. Maxillary barbel long, its tip reaching middle of pectoral fin. Dentary teeth larger than premaxillary teeth (Figure 5b); premaxillary teeth 1–2 (mode 1), dentary teeth 4–6 (mode 5), teeth bicuspid, spoon-shaped, with a larger central cuspid and a smaller lateral cuspid.

Greatest body depth on head at area between eyes; progressively narrowing anteriorly to snout tip and posteriorly to caudal peduncle. Body entirely covered by bone plates, except for ventral surface of head, abdomen and region between compound pterotic and opercular plate. Lateral series of plates 27–29 (mode 28).

TABLE 1 Morphometric data for *Rhadinoloricaria papillosa* sp. n.

Landmarks	Holotype	Minimum	Maximum	Mean	SD
Standard length (mm)	73.92	40.77	73.92	58.75	-
Percentage in SL					
Head length	22.00	22.00	24.34	23.14	1.03
Predorsal length	32.94	32.37	34.95	33.27	1.08
Postdorsal length	55.64	55.38	63.31	59.71	3.54
Prepectoral length	19.20	18.07	20.82	19.03	1.11
Postpectoral length	78.48	77.88	80.81	79.60	1.18
Prepelvic length	32.75	30.03	33.34	31.41	1.41
Postpelvic length	67.09	64.78	69.62	67.75	1.74
Pre-anal length	46.88	44.68	48.59	46.55	1.46
Post-anal length	50.27	45.23	53.79	49.91	3.08
Unbranched dorsal-fin ray	17.83	16.89	19.22	18.17	0.98
Unbranched pectoral-fin ray	18.01	18.01	20.10	16.04	7.91
Unbranched pelvic-fin ray	13.81	12.62	15.11	13.82	0.87
Unbranched anal-fin ray	13.72	12.70	16.68	12.32	6.22
Inferior unbranched caudal-fin ray	10.59	10.59	13.78	12.56	1.10
Thoracic length	16.80	15.39	16.80	15.92	0.49
Abdominal length	15.94	15.31	18.25	16.71	1.02
Cleithral width	19.93	18.82	21.19	19.98	0.99
Depth at dorsal-fin origin	6.53	4.67	6.53	5.65	0.71
Width at anal-fin origin	11.95	10.93	13.04	12.16	0.71
Caudal peduncle depth	1.30	1.05	1.37	1.21	0.14
Caudal peduncle width	1.83	1.83	2.25	2.07	0.14
Percentage of head length					
Snout length	53.08	45.33	53.23	50.97	2.94
Eye diameter	14.70	12.53	16.19	14.28	1.18
Maximum orbital diameter	23.06	15.90	23.06	19.87	2.73
Interorbital width	18.39	16.23	18.39	17.33	0.89
Internarial width	8.18	6.64	9.09	7.91	0.87
Head depth	29.46	20.35	30.53	27.03	3.55
Head width	84.07	75.11	84.07	81.74	3.42
Free maxillary barbel	53.51	24.81	53.51	42.42	11.10
Ventrorostral length	10.64	8.41	11.41	10.07	1.02
Lower lip length	22.94	22.46	27.35	24.10	1.75
Meristics	**Holotype**	**Minimum**	**Maximum**	**Mode**	**SD**
Right premaxillary teeth	1	1	2	1	-
Right dentary teeth	6	4	6	5	-
Left lateral scutes	28	27	29	28	-
Barbelets on margin of lower lip	7/7	7/7	7/7	7/7	-

Abbreviations: SD, standard deviation; SL, standard length.

Dorsal-fin rays i,7; dorsal-fin originating and posteriorly to pelvic-fin; distal margin slightly convex. Pectoral-fin rays i,6 (Figure 5); middle portion convex; unbranched pectoral-fin ray reaching pelvic-fin origin; unbranched and branched pectoral-fin rays covered with large and pointed odontodes. Pelvic-fin rays i,5; fin slightly convex; tip of adpressed pelvic-fin almost reaching anal-fin origin; unbranched pelvic-fin ray not covered with odontodes. Anal-fin rays i,5; fin straight; unbranched anal-fin ray not

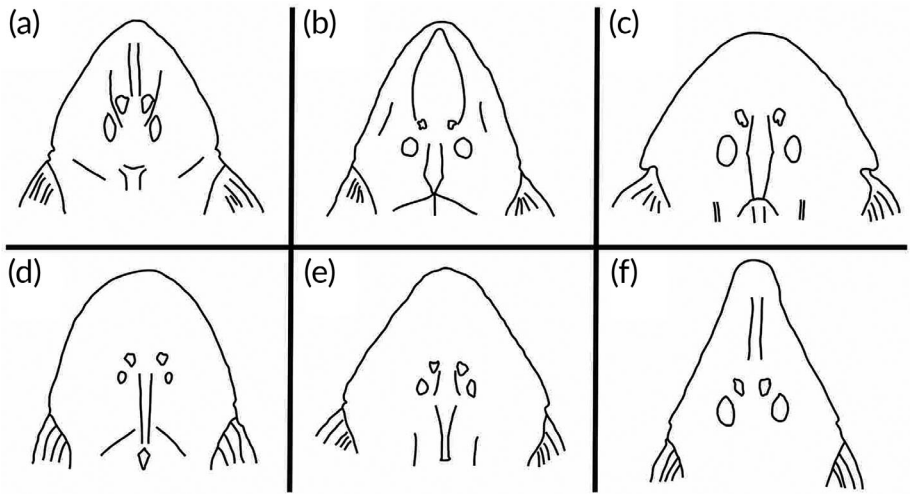

FIGURE 4 Scheme representing the shape of head in dorsal view of (a) *Rhadinoloricaria papillosa* (MZUSP127013, holotype); (b) *Rhadinoloricaria listrorhinos* (ANSP 131482, holotype); (c) *Dentectus barbamatus* (MBUCV-v-12985, paratype); (d) *Planiloricaria cryptodon* (ZFMK/1/66/1717, holotype); (e) *Pseudohemiodon unillano* (IAvH-P19034, holotype); and (f) *Rhadinoloricaria macromystax* (BMNH1869.5.21.8, holotype)

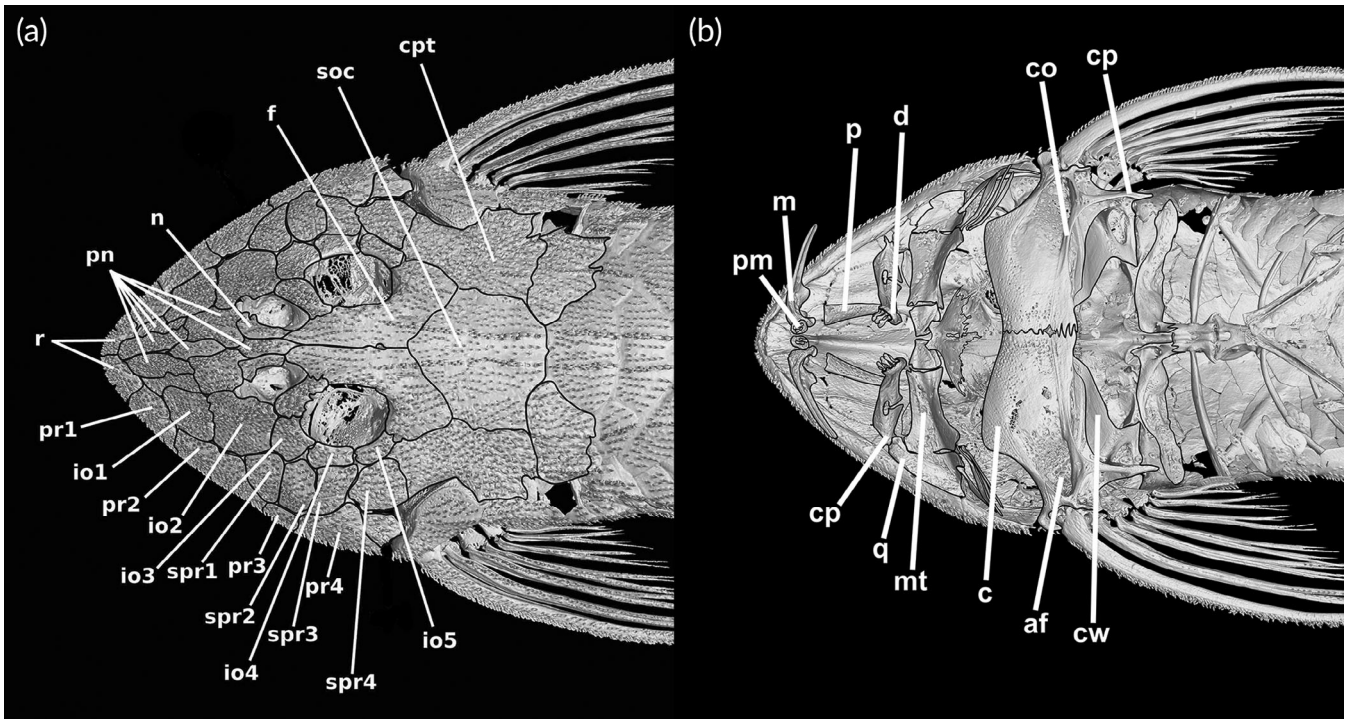

FIGURE 5 Image of (a) dorsal and (b) ventral head plates of *Rhadinoloricaria papillosa* n. sp. (ANSP207807) obtained by scan from a microscopy and imaging facility at the American Museum of Natural History

covered with odontodes. Caudal-fin truncated, with five upper rays and five lower rays (i,10,i), with long, thin filament on dorsal caudal-fin ray (usually broken) at least two-thirds of standard length; unbranched rays of dorsal and upper caudal-fin covered by odontodes (Figure 5); lower unbranched ray caudal fin not covered by odontodes.

Snout tip composed of two square-shaped rostral plates (r) (Figure 5a). Pre-nasals (pn) positioned posteriorly of rostral plates (r), formed by square or rectangular-shaped plates (Figure 5a). Nasal plate (n) L-shaped forming anterior medial nostril margin in contact posteriorly with frontals (*f*) and anteriorly and laterally with pre-nasals (pn) (Figure 5a). Dorsal portion of head composed by compound pterotic (cpt), parieto supraoccipital (soc) and frontal (f) (Figure 5a). Posterior rostrum plates pr1-pr3 small, and rectangular shaped; pr4–pr2 largest, and rectangular shaped. Infraorbital plate series complete (io1–io5), present just above posterior rostrum series, all covered by latero-sensory canal system; io2 largest and io5 smallest; io3, io4 and io5 forming inferior orbital margin of eyes (Figure 5a). Four square-shaped supra-rostral plates (spr1–spr4) present just above rostral plates (Figure 5a). Ventral of head (Figure 5b) composed by premaxilla (pm), maxilla (m), palatine (p), dentary (d), quadrate (q), metapterygoid (mt). Pectoral-girdle composed by

cleithrum (c), abductor fossae (af), cleithrum walls (cw), coracoid (co) and coracoid process (cp) (Figure 5b). Abductor fossae completely opened, opening relatively large, extending laterally towards base of pectoral fin (Figure 5b).

3.6 | Colour in alcohol

Ground colour of body yellow with few dark chromatophores overall the dorsal surface of body (most evident in specimens above 66 mm SL) and forming dark transversal bars on caudal peduncle. Light brown odontodes at surface of bone plates and at dorsal face of unbranched and branched pectoral-fin rays, and on unbranched dorsal-fin and superior caudal-fin rays. Ventral surface completely yellow with light brown odontodes at surfaces of bone plates. Ground colour of fins hyaline, with dark chromatophores on dorsal surface of branched and unbranched pectoral-fin rays forming difuse bars and on caudal-fin base forming a difuse spot. The pigmentation is most observed in the largest specimens (>60 mm SL), whereas small specimens (<60 mm SL) are mostly pale.

3.7 | Colour in Life

Similar to pattern described for alcohol individuals, but with ground color light yellow, almost translucent, and pectoral and pelvic fins hyaline, with a large dark bar on its distal portion (Figure 6).

3.8 | Sexual dimorphism

Adult males are differentiated from females by presence of papilla at urogenital opening (*vs*. absence of papilla at urogenital opening) (Figure 7).

3.9 | Habitat

The new species occurs at the bottom of Rio Negro, between the states of Amazonas and Pará, Brazil (Figure 8).

3.10 | Etymology

From Latin, "papillosa" refers to numerous elongated papillae present on lower lip of specimens examined.

3.11 | Conservation status

R. papillosa n. sp. was found in six points along Rio Negro basin, including Demeni, Aracá and Rio Negro channel. Despite the known occurrence in six points, only eight specimens were collected in 10 days of expedition, suggesting that is a rare species or very difficult to collect. Furthermore, considering the distribution of *R. papillosa*, the authors recommended it to be categorised as endangered (EN), according to the EOO (which pointed 843.917 km^2 of extent of occurrence) and AOO (which pointed 24.000 km^2 of area of occupancy) analysis, following the IUCN criteria (IUCN Standards and Petitions Subcommittee) (International Union for Conservation of Nature (IUCN), 2022).

FIGURE 6 Live specimen of *Rhadinoloricaria papillosa* n. sp. (ANSP 207862), 63.40 mm SL, Rio Demeni, 00° 35′ 04.3″ S 62° 53′ 24.5″ W, 10 August 2018, col. M. Bernt, G. Costa Silva, B. Waltz & R. Nilson. Photographed by Maxwell J. Bernt

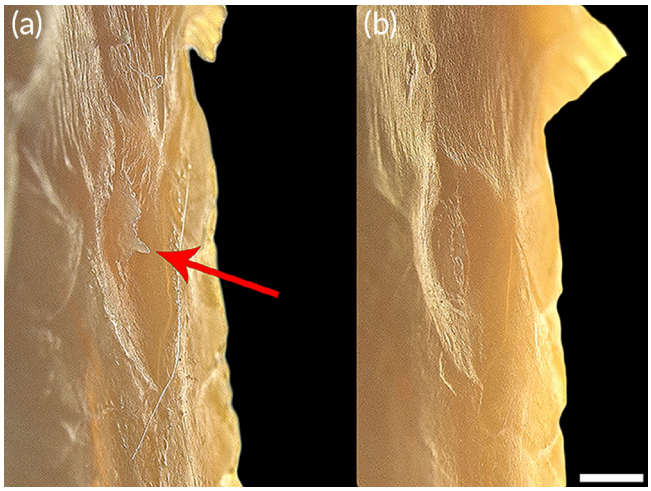

FIGURE 7 Sexual dimorphism in *Rhadinoloricaria papillosa* n. sp. Red arrow indicates urogenital papillae present in males (a – LBP26829), that is absent in females (b – MZUSP127013)

3.12 | Molecular analysis

The combined sequence data of the three genes resulted in a matrix with 8238 base pairs (bp). The estimated Iss performed in DAMBE 5.2.31 (Xia & Lemey, 2009; Xia and Xie, 2001) showed that the data

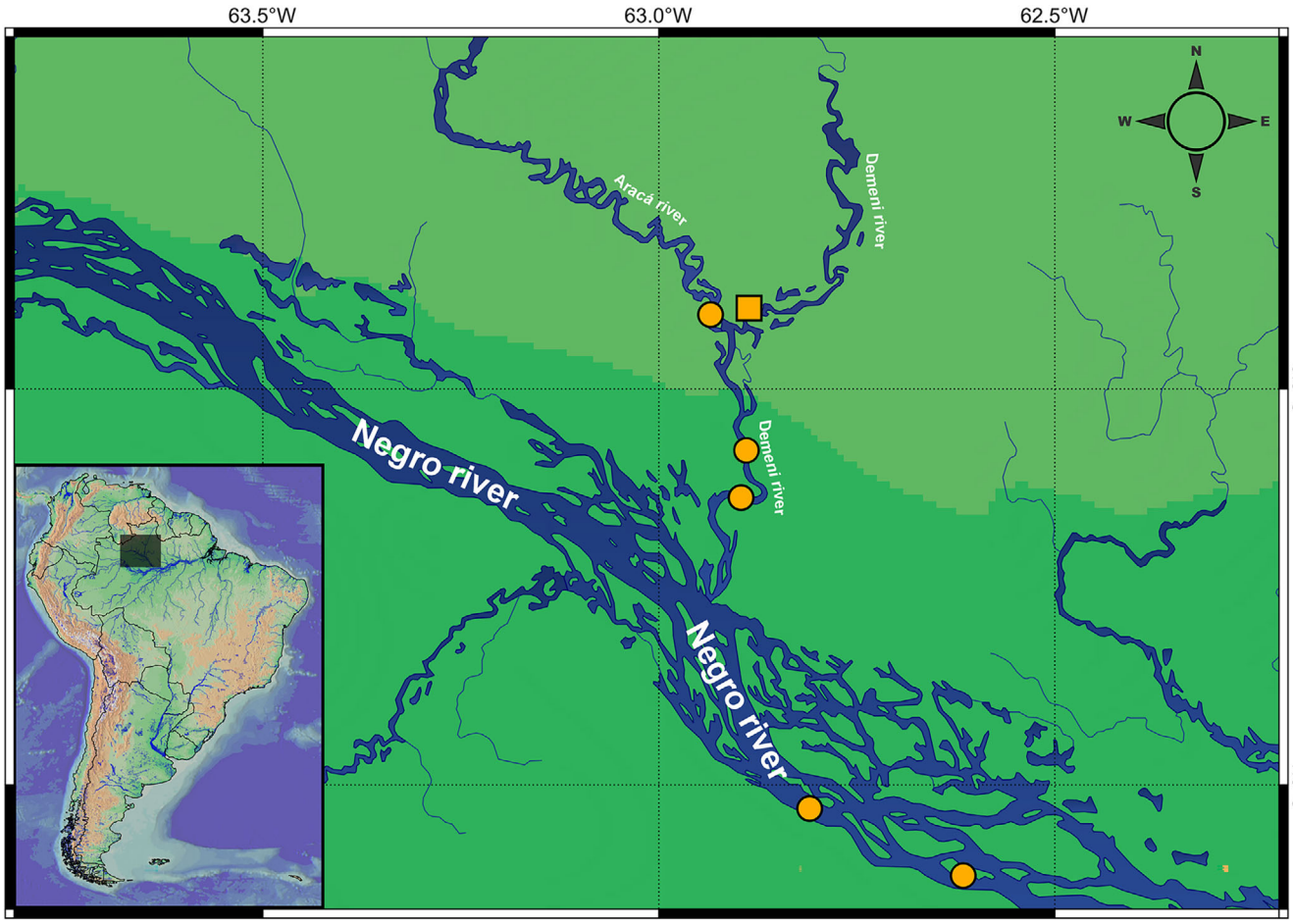

FIGURE 8 Distribution of *Rhadinoloricaria papillosa* n. sp. through drainages of Rio Negro. Square represents the type-locality at municipality of Barcelos, Amazonas State, Rio Demeni (S 00° 24′ 57.6″ W 62° 53′ 37.8″)

were not saturated (*i.e.*, Iss.c value greater than Iss). The total matrix includes 372 specimens representing 227 loricariid species.

The RAxML phylogenetic analyses among members of Loricariinae are similar to those presented by Covain *et al.* (2016), except for the inclusion of the species *D. barbamatus* and *R. papillosa*. The new species *R. papillosa*, was found to be the sister taxon to *Rhadinoloricaria* "Orinoco" (identified as *Rhadinoloricaria* sp. "Orinoco" in Covain *et al.*, 2016), and this clade appears as sister to a group composed of *Rhadinoloricaria* species, within in the *Pseudohemiodon* group (Figure 9). With the uncertain diagnosis of the genus *Rhadinoloricaria* and the addition of *D. barbamatus* in the phylogeny, the authors suggest which *Pseudohemiodon* group need a taxonomic revision.

4 | DISCUSSION

This phylogenetic analysis placed the new species *R. papillosa* within the *Pseudohemiodon* group, as the sister to an undescribed species of *Rhadinoloricaria* from the Orinoco River basin. Both species (*i.e.*, *R. papillosa* n. sp. and *Rhadinoloricaria* "Orinoco") are sister of a clade composed of *R. macromystax*, *R. ommation*, *R. bahuaja* and *R. rhami*. Recently, Provenzano-Rizzi and Barriga-Salazar (2020) suggest the revalidation of the genus *Apistoloricaria* (previously synonymised with *Rhadinoloricaria* by Covain *et al.*, 2016) based on putative derived characters shared among *Apistoloricaria* species. The results of this study are in accordance with Covain *et al.* (2016), which found *R. ommation* nested inside *Rhadinoloricaria* and recovered *Rhadinoloricaria* as a monophyletic group.

The new species described here is placed within *Rhadinoloricaria* as it possesses some characteristics used to recognise the genus, such as long maxillary barbel extending beyond pectoral-fin base; lower lip with broad surface in numerous elongated papillae; both jaws with small teeth; orbital with shallow posterior notch. In addition, the placement of *R. papillosa* in *Rhadinoloricaria* is supported by the molecular phylogeny. Despite its placement in *Rhadinoloricaria*, the new taxon possesses two autapomorphic characters related to buccal morphology not found in others *Rhadinoloricaria* species.

The first character is the filamentous papillae on lower lip. The new species possesses its lower lip covered with numerous papillae that are modified in longer filaments, all surpassing the posterior border of the lower lip (Figure 1a,3). In contrast, all other species of *Rhadinoloricaria* have the lower lip covered with short papillae (Figure 1b,5). *D. barbamatus*, a member of the *Pseudohemiodon*

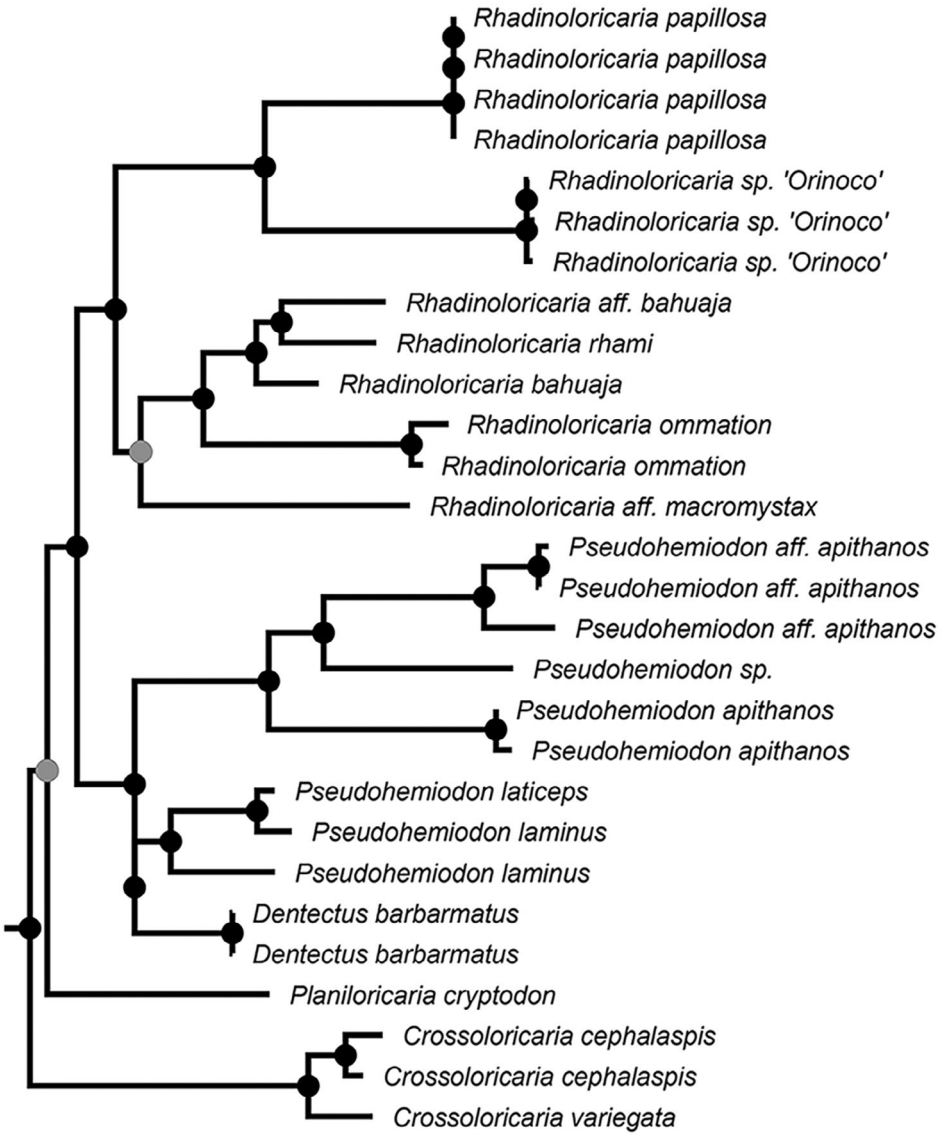

FIGURE 9 Maximum likelihood (ML) analysis focused in *Pseudohemiodon* group (modified from Covain *et al*., 2016. ●, BS > 75%; ●, BS > 50%

group, also possesses filamentous papillae extremely elongated (Martín Salazar *et al*., 1982), as found in the new species. In this phylogenetic analysis *D. barbamatus* is not closely related with the new species, but with some species of *Pseudohemiodon*. Thus, the authors propose here that the elongated papillae is a derived condition that occurs homoplastically in *D. barbamatus* and in the new species *R. papillosa*.

The second character is the unbranched barbelets on the lower lip border. The new species possesses seven barbelets unbranched on the edge of its lower lip (Figure 1a,2). In all other species belonging to the *Pseudohemiodon* group, including *Rhadinoloricaria* "Orinoco," the barbelets are branched (Figure 1b,4).

Provenzano-Rizzi and Barriga-Salazar (2020) stated that the species of *Rhadinoloricaria* are rare. *Loricaria macromystax* (Günther, 1869) (now *R. macromystax*) was described from a single specimen and *R. stewarti* was described with just six specimens. During 10 days of expedition in the deep channel of the Rio Negro and Rio Demeni, with several points sampled, only eigth specimens of the new species *R. papillosa* were caught, also indicating that this species is rare. Despite the low number of specimens analysed, they showed strong distinct morphological and molecular characters in comparison with its congeners that support these specimens collected in the Rio Negro basin as a new species of *Rhadinoloricaria*.

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CONFLICT OF INTEREST STATEMENT

This study was done without confict of interest.

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SUPPORTING INFORMATION

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