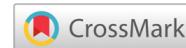


## Research Article

 Open Access


# Analysis of a new species of the *Astyanax orthodus* group (Characiformes: Characidae) from the upper Madre de Dios River, Amazon River Basin, Peru

## Abstract

Based on ten specimens, using morphometric, meristic and osteological characters, *Astyanax leoni* n. sp. is described from the upper Madre de Dios River drainage, Amazon River Basin of Peru. *A. leoni* n. sp. is a member of the *Astyanax orthodus* species group and is distinguished (except from *A. multidens*, *A. villwocki*, and *A. gandhaiae*) in having an inconspicuous rod-shaped stripe on the caudal peduncle that extends anteriorly on top of a silver lateral stripe. It differs from the species mentioned above as follows: from *A. multidens* by having shorter chevron marks that cover up to only two series of scales above and below the apex of the chevron (vs. chevrons taller, with extensions that cover three series of scales); from *A. villwocki* by having chevron markings along the entire body (vs. no chevrons on the caudal peduncle); It differs from *Astyanax gandhaiae* in having the caudal peduncle spot extending anteriorly to beyond the tip of the dorsal fin (vs. caudal peduncle spot not reaching anteriorly to tip of dorsal fin), and not of a uniform width as in *A. villwocki*. A dichotomous key to the species of *Astyanax orthodus* group is included.

**Keywords:** characin fish, tropical, osteology, pigmentation pattern, scales

## Introduction

The Neotropics hosts the greatest diversity in freshwater and estuarine fishes, with at least 6000 species estimated in the large river basins of South America.<sup>1-5</sup> While Characidae is the most diverse family of Neotropical fishes,<sup>5-8</sup> the group of characins exhibits an immense complexity of forms and occupies a vast range of niches in South and Central America.<sup>9</sup> The current state of taxonomy of the genus *Astyanax* makes it difficult to recognize it as a biological unit.<sup>10,11</sup> However, based on the description of widely distributed species and their morphological comparison between cryptic species, for example *Astyanax incaicus* Tortonese,<sup>12</sup> *A. rupununi* Fowler,<sup>13</sup> *A. siapae* Garutti,<sup>14</sup> and *A. utiariti*, Bertaco and Garutti,<sup>15</sup> it has been possible to form artificial groups of species based on external characters, which has made it easier to describe part of this complex diversity. One of these artificial groups is the *Astyanax orthodus* species group as defined by Ruiz-C. et al.,<sup>8</sup> which can be recognized by having a series of dark chevron-shaped marks along the sides of the body, positioned both dorsal and ventral to the horizontal septum, from the humeral region to the caudal peduncle. In addition to pigmentation, other internal morphological characters such as teeth on the maxilla, and a foramen above the teeth of the inner premaxillary row support the recognition of an assemblage that includes members of the *Astyanax orthodus* species group.

After examining material belonging to species of the *Astyanax orthodus* group, we describe here a new species from the Amazon River basin in Peru based on morphological characters and colour pattern.

## Materials and methods

Morphometric and meristic data were recorded from material of *Astyanax* listed in comparative examined (see below) and was accessible from institutions in Argentina (ILPLA, MLP), Bolivia (CBF, UMSS), Brazil (MNRJ, MCP), Colombia (IAvHP and IUQ), Mexico (UNAM), Peru (MUSM acronym of MHN.UNMSM), USA

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(INHS, AMNH, AUM and UF) and Venezuela (MCNG); acronyms used follow Sabaj-Perez.<sup>16</sup>

Osteological observations were done in cleared and stained (C&S) samples prepared with methods outlined in Taylor and Van Dyke;<sup>17</sup> only adult individuals were C&S to avoid confounding effects of ontogenetic disparity in the amount of ossification. The total vertebral number comprises those of the Weberian apparatus; while the compound ural centrum is usually counted as a single element Bone nomenclature follows Weitzman,<sup>18</sup> as modified by Mirande.<sup>6</sup>

Morphometric and meristic data follow Vari and Siebert,<sup>19</sup> Fink and Weitzman<sup>20</sup> and Román-Valencia,<sup>21</sup> except for head length, which was measured from the anterior tip of the snout to the posterior margin of the subopercle and interorbital width, which was measured at the level of the epiphyseal bar. Measurements were taken using digital calipers with a precision of 0.1 mm. The mean and range of variation for 23 morphometric characters are expressed either as percentages of standard length or head length. Meristic data and sexually dimorphic characters are followed by the number of individuals examined in parentheses (=n: four). In material examined, the number of specimens both in alcohol and cleared and stained (C&S) is given after the lot number, followed by the range of standard length and location. In the descriptions, the number of individuals recorded for each count follows that value in parentheses (=n: four). In the text and tables, SD stands for standard deviation, H for Holotype, and n for the number of specimens examined.

## Comparative material examined

*Astyanax elachylepis*, Brazil. MCP 16054, one, 88.9, one C&S, 80.4 mm SL, Maranhão River, Goias. *Astyanax integer*, Colombia. IAvHP 3243, one, 59.7 mm SL, Tauramena, Casanare; IAvHP 3516, two, 90.9-109.4 mm SL, Orinoco River, Meta; IAvHP 5169, five, 49.2-66.1 mm SL, Urichare Creek inspección Puerto Caldas, Granada, Meta, Orinoco Basin River. IAvHP 7912, twelve, 54.1-64.7 mm SL, Cravo Sur River, Yopal, Casanare; IAvHP 11253, one, 70.6 mm SL, Caño Urichare, inspección Puerto Caldas, Granada, Meta,

Cuenca del Orinoco. Venezuela. MBUCV 33 123, five, 45.9-58.6 mm SL, Guatiré Creek, afl. Orinoco, Anzoategui; MCNG 48480, one, 53.1 mm SL, Creek afl. of Apure River, Portuguesa; MCNG 52153, five, 51.2-57.2 mm SL, Rabipelado Creek, affluent of the Apuro, Portuguesa. *Astyanax lineatus*, Argentina. ILPLA 318, three, 55.5-60.2 mm SL, de agua Caliente Creek, San Francisco River, Santa Barbara, 23°44'South, 64°38'West; ILPLA 1423, four, 52.2-72.1 mm SL, Saladillo River, Guames, Salata, 24°40'South, 65°03'West. ILPLA 1446, río de las Conchas RN 34 y 9.5 Km al norte de Metán (loc. 51) Juramento Basin River 25°28'S- 64°57'W, Salta; IUQ 1647, two, 77.6-80.3 mm SL, three C&S, 46.6-59.4 mm SL, río Bermejo sobre la ruta Prov. 19. Oran, Salta, Paraná River. Bolivia. CBF 2606, three, 46.7-94.2 mm SL, San Bernardo River on the road the camino Yucumo, Rurrenabaque, Beni; CBF 2810, twelve, 57.4-89.1 mm SL, Ixiamas, a 15 Km al oeste de Ixiamas camino al alto Madidi, La Paz; CBF 7185, four, 40.8-70.4 mm SL, parque nacional y área de manejo integrado Amboro, Ichilo, Santa cruz; CBF 7448, six, 45.8-50.8 mm SL, UF 167447, one, 50.5 mm SL, Santa Cruz, Quise river, San Pablo drainage, Santa Cruz; UF 167471, one, 73.8 mm SL, Piray river, Mamoré drainage; UF 167504, seven, 50.9-67.6 mm SL, Heredia Parapeti River, Chuquisaca, 19°48'11.3724 South, 64°0'41.6556 West; UF 167 515, two, 67.9-73.0 mm SL, Mamoré River, Cochabamba, 18°7'48.8532 South, 64°53'41.9504 West; UMSS 8928, twenty, 52.7-85.6 mm SL, Tacuaral Creek, Tohomonoco, Alto Beni, Madeira, 15°45'46.33 South, 67°44'84.66 West; UMSS 10012, 20, 56.4-86.1 mm SL, Tacuaral Creek, Tohomonoco, Alto Beni, Madeira; Perú. MUSM 11301, four, 58.4- 75.3 mm SL, río Tambopata, Madre de Dios. *Astyanax maximus*, Colombia. IUQ 616, two, 104.3-105.8 mm SL, Río Guayuyaco, en la población de Miraflor, Cauca, sobre la vía Villa Garzoa- San Jose de Fragua; IUQ 971, two, 98.9-115.9 mm SL, Inchiyaco River, on the road Villa Garzon-San Jose de Fragua; IUQ 972, six, 59.8- 130.3 mm SL, one C&S, 53.2 mm SL, Tambor River, on the road Villa Garzón-San José de Fragua, Caquetá; IUQ 1067, two, 86.3-113.9 mm SL, Nabueno Creek sector de Miraflor Cauca, on the road Villa Garzón, San Jose del Fragua; IUQ 1426, one, 112.3 mm SL, La Vio Creek en el parador el pomar vía Florencia, La Montañita, Vereda Citarca, Caqueta, 1° 31'10'' North, -75° 28' 19'' West, 403 m.a.s.l.; IUQ 1598, two, 114.6- 140.3 mm SL, Orteguaza River, Casqueta. *Astyanax mexicanus*, USA. INHS 80766, eight, 47.4-59.7 mm SL, 2 C&S, 53.0-63.7 mm SL, Devils River, Texas. México. UNAM -P 12235, two, 31.1- 35.6 mm SL, Tonalá River, San José, Tancuchapa, Choapas. *Astyanax ruberrimus*, Colombia. IUQ 253, five, 62.5-.75.6 mm SL, boca del Río Yamunde, on Telembi River, 1 Km debajo de Barbacoas, Nariño, IUQ 1237, two, 62.7-74.6 mm SL, Patia River, refugio UPESIA, Municipio de Patía, departamento de Cauca, cuenca del pacífico, 2° 2' 36'' North, 77° 06' 5'' West, 620 m.a.s.l. *Astyanax abramis*, Argentina. ILPLA 1, one, 85.6 mm SL, Arroyo El Pescado, departamento Anta, Salta, 24°42'S-64°38'W; ILPLA 359, one, 90.5 mm SL, one C&S, 89.0 mm SL, Laguna La Mira, Santa Fe; ILPLA 874, one, 93.2 mm SL, Paraná River basin, Corrientes; MLP 1630, one, 93.9 mm SL, one C&S, 81.6 mm SL, Provincia de Corrientes; MLP 9427, 1, 105.5 mm SL to 1 Km. de la desembocadura del Iguaçú, Provincia Misiones. *Astyanax altiparanae*, Brazil. MCP 27862, twenty one, 105.0-110.6 mm SL, one C&S, 101.3 mm SL, Piracicaba river, Sao Paulo, 22°43'19" South, 47°39'21"West. *Astyanax atratoensis*, Colombia. IAHP 7140, 21, 55.0-117.9 mm SL, Atrato River; IAHP 7187, two, 78.4-82.3 mm SL, Turbo, Antioquia, Atrato River basin, caño Muerto, última Ciénaga, vereda El Cuarenta; IAHP 7206, three, 71.2-91.8 mm SL, Antioquia, Turbo, Atrato River basin, ciénaga Tumarado, vereda El cuarenta; IUQ 84, two, 85.7-88.2 mm SL, two C&S, 83.6-84.9 mm

SL, Chocó, Quibdó, Atrato River on Malecón de Quibdó; IUQ 696, three, 90.2-110.7 mm SL, Chocó, Atrato River basin, Negro River, vereda El Ganado; IUQ 745, two, 68.9-76.0 mm SL, 1 C&S, 64.8 mm SL, Chocó, Atrato River basin, Negro River, ciénaga de Achuara. *Astyanax bimaculatus*, Essequibo-Guyana. AMNH 14 434, one, 44.9 mm SL, one C&S, 68.0 mm SL, Amacuro River, Santa Rosa; AMNH 14 549, one, 52.5 mm SL, Yaraquita River; AMNH 221021, one, 27.3 mm SL, Georgetown, Demerara. *Psalidodon chico*, Argentina. IUQ 1646, three, 56.6-70.5 mm SL, one C&S, 68.3 mm SL, Salta, Campo Largo, Arroyo tributario del río Itau. *Psalidodon endy*, Argentina. IUQ 3817, nine, 45.2-61.1 mm SL, Salta, Orán, Cuenca Río Bermejo, Río Blanco en El Oculto. *Psalidodon fasciatus*, Brasil. MNRJ 20 716, five, 84.9-101.4 mm SL, Claro River, reservatorio de Ribeirao das Legesbraco que recebe o rio da Prata, Rio de Janeiro.

## Results

### *Astyanax leoni* n. sp.

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Figure 1 *Astyanax leoni* n. sp. Holotype, Scale bar = 1 cm.

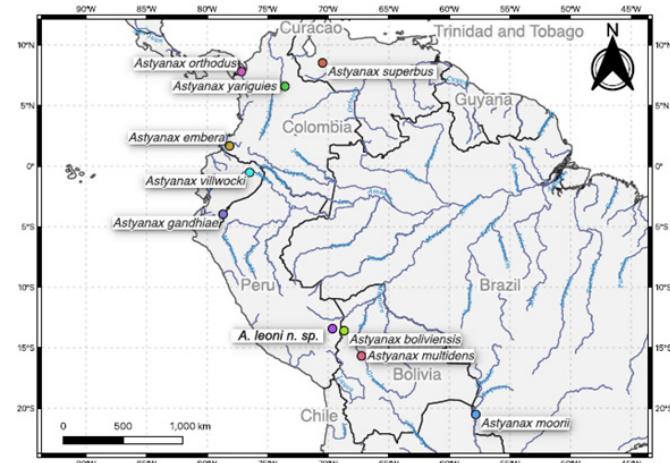


Figure 2 Distribution model for species of the *Astyanax orthodus* group.

**Holotype:** MUSM 46844, 96.2 mm SL, Río Tavará a 2 Km de Q. Grande. Tambopata-Candamo, Sandia, Puno, Perú.

**Paratypes:** All from Perú: MUSM 2457: one (108.5 mm SL), río Manu, Parque Nacional Manu, Pakitza, MUSM 3822: 3 (79.5-115.6 mm SL), río Madre de Dios, Manu, 340m. MUSM 9076: one (56.0 mm SL), quebrada Martín Pescador, Parque Nacional Manu, Pakitza, Madre de Dios, 345 m. MUSM 4662: four (99.4-81.9 mm SL), quebrada Picaflor, Parque Nacional Manu, Pakitza, Madre de Dios, 350 m.

**Table I** Morphometric and meristic data of *Astyanax bolense*, *Astyanax leoni* n. sp. and *A. superbus*

(mm)	<i>A. leoni</i> n. sp. (n:10)			<i>A. superbus</i> (n:7)		
	SD	H	Range media	SD	Range media	SD
Standard length (SL)		96.2	80.7-117.3 (95.0)			42.5-87.0 (67.3)
Percents of standard length (SL)						
Body depth	1.3	36.1	32.0-37.9 (35.8)	2.1	33.0-34.9 (34.0)	0.6
Head length	1.1	27.3	27.1-28.4 (27.9)	0.5	26.6-29.0 (27.5)	0.9
Predorsal distance	1.1	49.1	47.1-50.4 (48.7)	1.2	46.0-49.8 (48.8)	1.4
Prepectoral distance	0.9	26.1	25.9-27.4 (26.8)	0.5	24.6-27.1 (25.6)	0.8
Prepelvic distance	0.6	46.4	47.0-48.5 (47.6)	0.5	44.0-47.7 (45.2)	1.4
Dorsal-pectoral distance	1.3	40.4	37.7-41.2 (39.6)	1.3	36.9-39.0 (37.6)	0.8
Preanal distance	1.6	62.2	62.7-65.0 (63.8)	0.7	57.5-63.9 (60.4)	2.4
Dorsal-fin length	2.5	28.2	25.0-30.2 (26.9)	1.8	28.8-30.4 (29.1)	0.6
Pectoral-fin length	1.1	20.5	20.3-23.7 (21.7)	1.2	20.3-22.6 (21.3)	0.9
Pelvic-fin length	3.6	16.9	12.8-20.2 (16.8)	2.2	14.2-18.0 (16.0)	1.2
Anal-fin length	1.5	15.7	13.3-16.6 (15.2)	1.4	17.1-20.1 (19.0)	1.3
Dorsal-anal distance	1.1	37.6	33.4-39.4 (37.4)	2.1	34.8-36.4 (35.6)	0.6
Caudal peduncle depth	0.5	11.7	9.4-11.4 (10.7)	0.7	10.5-12.1 (11.6)	0.7
Caudal peduncle length	1.2	13.8	10.7-13.0 (11.4)	0.8	11.0-11.8 (11.4)	0.3
Percents of head length (HL)						
Snout length	1.7	26.2	24.5-28.6 (26.0)	1.3	23.3-26.3 (25.2)	1
Orbital diameter	2.6	24	22.4-26.0 (24.8)	1.5	26.3-28.8 (27.4)	0.9
Postorbital length	2.2	53.6	49.8-55.2 (52.3)	1.6	47.5-52.6 (50.4)	1.9
Maxillary length	1.7	27	25.7-32.3 (29.6)	2.2	29.7-33.0 (31.3)	1.4
Interorbital width	1.2	36.1	36.0-38.5 (37.4)	0.9	28.4-32.3 (30.3)	1.7
Upper jaw length	1.4	28.5	25.4-30.7 (28.5)	1.9	24.0-28.8 (26.5)	1.6

## Diagnosis

*Astyanax leoni* n. sp. is a member of the *Astyanax orthodus* species group and differs from other members of that group except for *A. multidens*, *A. villwocki* and *A. gandhiae*<sup>8</sup> in having an inconspicuous rod-shaped stripe on the caudal peduncle that extends anteriorly on top of a silver lateral stripe (vs. blotch restricted to caudal peduncle or absent). *A. leoni* n. sp. differs from *A. multidens* in having shorter chevrons that cover up to two scale series above and below the vertex of the chevron (vs. chevrons taller, with extensions that cover three series of scales above and below the vertex of the chevron). It differs from *A. villwocki* in having the chevrons distributed along the entire length of the body (vs. chevrons absent on caudal peduncle). It differs from *Astyanax gandhiae* in having the caudal peduncle spot extending anteriorly to beyond the tip of the dorsal fin (vs. caudal peduncle spot not reaching anteriorly to tip of dorsal fin) and not of a uniform width as in *A. villwocki*.

## Description

Body compressed, greatest body depth at or anterior to dorsal-fin origin. Mouth terminal, dorsal profile of head sigmoid between tip of snout and posterior margin of supraoccipital spine. Predorsal profile arched as is the postdorsal region from the last ray of the dorsal fin to the origin of the adipose fin. Predorsal with complete series of scales along dorsal midline and keel. Ventral profile convex between tip of snout to pelvic fin origin.

Pored scales of lateral line 40-42 (n=18), scales from lateral line to dorsal-fin origin 8 (n=18), scales from lateral line to anal-fin origin 5-8 (n=18), scales from lateral line to pelvic-fin origin 6-7 (n=18). Dorsal-fin rays iii, 9, first unbranched ray reduced in size, only visible in cleared and stained specimens, second unbranched ray about half the length of third unbranched ray. Distal margin of dorsal fin slightly convex. Adipose fin origin anterior to vertical through insertion of last anal-fin ray. Pectoral-fin rays i, 10-12, ii. Pelvic-fin rays i, 7. Anal-fin rays iii-iv, 25-27 (Table 1), and anterior unbranched rays only visible in cleared and stained material. Anal-fin origin posterior to vertical through insertion of last dorsal-fin ray. Total number of vertebrae 36-38; precaudal vertebrae 17-18, the three posterior are transition: vertebra 16 with the pleural rib folded, not attached to center of vertebra, vertebra 17 without true pleural rib (elongated transverse process) not attached to center of vertebra and vertebra 18 without lateral process or developed pleural rib; 19-20 caudal vertebrae; 21 or 22 epipleurals and 34 epineurals. Caudal skeleton with seven hypurals; four hypurals of dorsal lobe support eleven main rays and ten procurent rays; three hypurals of ventral lobe supports ten main rays and nine procurent rays.

**Color in alcohol:** Sides of body yellowish, with silver band along median portion; dorsal region of head and body light chestnut. Sides of head light silvery, ventral portion of body similar. Humeral spot black, conspicuous, polygon-shaped, located from first or second to fifth scale of lateral series, covering two horizontal scale rows above lateral line. Deep melanophores present between myosepta over silver lateral stripe, forming a chevron-pattern (<) that extend from humeral region to caudal peduncle.

Pectoral, pelvic, dorsal and anal fins hyaline; pigments in interradial membranes of median caudal-fin rays continuous with lateral band. Rod-shaped peduncle blotch does not extend anteriorly to tip of anal fin, however in some individuals (depending on fixation process), it extends as thin dark line on to the silver lateral stripe.

**Sexual dimorphism:** Males of *Astyanax leoni* n. sp. have small hooks at ventral end of second to fourth branched anal-fin rays; hooks are absent from rays of other fins.

**Distribution:** This species is found in the upper Madre de Dios River drainage of Perú (Figure 2).

**Etymology:** This species epithet *A. leoni* is named to honor Ing. RNR Oscar Leon Mata, researcher MCNG, Guanare, Venezuela. A noun in the genitive case.

**Key to species of the *Astyanax orthodus* group (see Figure 2)**

- 1a. Predorsal midline with a continuous series of scales covering more than three-fourths of the distance between posterior tip of supraoccipital spine and dorsal-fin origin.....2
- 1b. Predorsal midline with non-continuous scales covering less than three-fourths of the distance between the posterior tip of the supraoccipital spine and the origin of the dorsal fin, with isolated scales or short series, especially along the most posterior portion of predorsal midline.....3
  - 2a. Caudal peduncle blotch restricted to caudal-fin base.....4
  - 2b. Caudal peduncle blotch not restricted to caudal-fin base, extending anteriorly along sides of body.....5
    - 3a. Caudal peduncle blotch short, not extending anteriorly to vertical through adipose fin; body with reticulate pattern from back to pelvic region... *A. yariguies* Torres-Mejía et al. 2012 (Magdalena River Basin)
    - 3b. Caudal peduncle blotch long, extending anteriorly beyond vertical through adipose fin; body without reticulate pattern... *A. superbus* Myers 1942 (Apure River Basin, Orinoco)
  - 4a. Caudal blotch restricted to middle axis of body, not extending to dorsal and ventral margins of caudal peduncle or on to bases of caudal-fin rays... *A. orthodus* Eigenmann 1907 (Atrato River basin, Caribbean coast of Colombia)
  - 4b. Caudal peduncle blotch not restricted to middle axis of body, extending to dorsal and ventral margins of the caudal peduncle and on to bases of caudal-fin rays... *A. embera* Ruiz-C., Román-Valencia, Taphorn, Buckup and Ortega<sup>8</sup> 2018 (Patia River basin, Pacific coast of Colombia)
  - 5a. Caudal peduncle blotch extending anteriorly beyond vertical through dorsal-fin origin.....6
  - 5b. Caudal peduncle blotch extending anteriorly not reaching vertical through dorsal-fin origin.....7
    - 6a. Caudal peduncle blotch extended anteriorly as thin dark line of continuous width that reaches second humeral spot... *A. villwocki* Zarske and Géry 1999 (drainages of the Ucayali, Beni and Mamoré rivers, Amazon River basin)
    - 6b. Caudal peduncle blotch extended anteriorly as dark line that gradually tapers anteriorly as it reaches second humeral spot... *A. leoni* n. sp. (Madre de Dios River drainage, Perú).
  - 7a. Chevrons overlying silver lateral stripe short, not extending vertically from more than two series of scales above and below vertex of chevron; caudal-peduncle spot not conspicuous... *A. gandhiae* Ruiz-C., Román-Valencia, Taphorn, Buckup and Ortega<sup>8</sup> 2018 (Cenepa River drainage, upper Amazon River Basin)

**7b.** Chevrons overlying silver lateral stripe tall, extending vertically by prolongations from dorsal and ventral tips over three horizontal rows of scales; caudal-peduncle spot conspicuous..... 8

**8a.** Teeth present along two-thirds of the length of the maxilla... *A. multidens* Pearson 1924 (upper Madeira River drainage).

**8b.** Teeth present along less than two-thirds of the length of the maxilla with teeth..... 9

**9a.** Lateral line with 43 or fewer scales... *A. boliviensis* Ruiz-C., Román-Valencia, Taphorn, Buckup and Ortega (upper Madeira River drainage, Amazon River Basin)

**9b.** Lateral line with 44 or more scales... *A. moorii* Boulenger 1892 (Upper Paraguay River basin)

## Discussion

The systematics of the genus *Astyanax* requires much additional investigation. Mirande<sup>6,7,22</sup> has pointed out that the genus is paraphyletic. Until a phylogeny of the genus *Astyanax* based on morphometric, osteological and molecular characters is available, we are limited to recognizing novel species using traditional characters. Nonetheless, as Ornelas-Garcia et al.,<sup>23</sup> have shown for the *Astyanax* species of Mesoamerica, the prevalence of multiple morphological homoplasies complicates this type of traditional approach.

There is morphological<sup>22</sup> and molecular evidence,<sup>7,23</sup> indicating that the species identified in the *orthodus* group,<sup>8</sup> previously named *Astyanacinus*, form a clade, possibly related to the *bimaculatus* species group (*Astyanax*, *Poecilurichthys*), so it could represent a natural group not yet understood, with a wide distribution in the basins of South America, represented by a series of characters that could indicate that they are part of a natural group: chevron marks on the sides of the body, a foramen on the internal series of the premaxilla, as well as a horizontally elongated, roughly oval humeral spot with sharp well delimited margins and bordered anteriorly and posteriorly by lighter regions, mostly devoid of melanophores.<sup>8</sup>

Species in the *Astyanax orthodus* species group show a variation in the character states used to define it. This can be observed in the degree of lateral stripe pigmentation in the Amazonian species where the width of the dark lateral band present is reduced in *A. leoni* n. sp. compared with *A. villwocki* given that in both species the length of the band is the same (e.g., both species have the lateral line extending anteriorly to the origin of the dorsal fin, but the stripe's thickness is not the same; in *A. leoni* n. sp. the stripe gradually tapers anteriorly; while in *A. gandiae* its width is the same along the entire length, and abruptly ends without extending beyond the anal fin. Another character of interest is the low visibility of the chevron marks on the lateral band in *A. leoni* n. sp., which is also the case in *A. gandiae* and *A. multidens*, while *A. boliviensis* has an evident lateral band superimposed on the well-defined and well-pigmented chevron marks. Species of the *Astyanax orthodus* group could represent an ancestral group since they occur throughout the circum-Amazon arc, made up of the Ucayali, Mamoré, Madeira, Madre de Dios, Paraguay basins (*Astyanax villwocki*, *A. boliviensis*, *A. gandiae*, *A. multidens*, *Astyanax leoni* n. sp. and *Astyanax moorii*), in addition to the independent drainages of the Pacific sea slope (*Astyanax embera*) and the Caribbean sea (*A. orthodus*) as well as in the Magdalena River (*A. yariguies*) and Orinoco River (*A. superbus*).

## Participation of authors

R IRC= conception, experimental design, data collection, analysis, and writing of document. CRV= conception, experimental design, data collection, analysis and writing of the document. DCT= Analysis and writing of document.

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## Conflicts of interest

The authors declare that there are no conflicts of interest.

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