

Extension of the geographical distribution of *Plesiotrygon nana* Carvalho and Ragno 2011 (Elasmobranchii: Potamotrygonidae) for Rio Tarauacá, Upper Rio Juruá, Brazil

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ABSTRACI

The recently described freshwater stingray *Plesiotrygon nana* Carvalho and Ragno 2011 was observed in the Rio Tarauacá, a tributary of Rio Juruá, in the Brazilian Amazon. The specimens were similar to those examined in the original description of the species and are distinct from *Plesiotrygon iwamae* Rosa, Castello and Thorson 1987. The three specimens of *P. nana* have a dorsal coloration composed of fine rosettes and a very small spot on the disc edge, a circular disc which is small and has slightly rhomboidal spiracles, a distal coloration of the tail posterior to the caudal stings and high density of dermic denticles. This finding extends the geographic distribution of *P. nana* and reports the presence of this potamotrygonid species in small tributaries of white waters from the Upper Rio Iuruá.

Keywords: antenna ray; freshwater stingray; potamotrygonid; Chondrichthyes; Juruá basin.

Ampliação da distribuição geográfica para *Plesiotrygon nana* Carvalho e Ragno, 2011 (Elasmobranchii: Potamotrygonidae) no Rio Tarauacá, Alto Rio Juruá, Brasil

RESUMO

A recém descrita arraia de água doce *Plesiotrygon nana* Carvalho e Ragno 2011 é registrada no Rio Tarauacá, um tributário do Rio Juruá, na Amazônia brasileira. Os espécimes são similares aos analisados na descrição original da espécie, e diferentes de *Plesiotrygon iwamae* Rosa, Castello e Thorson 1987. Os três exemplares de *P. nana* têm coloração dorsal composta de rosetas finas, pequenas manchas na borda do disco, um disco circular, espiráculo pequeno e ligeiramente romboide, coloração na cauda posterior ao ferrão e alta densidade de dentículos dérmicos. Essa informação amplia a distribuição geográfica e relata a presença desse potamotrigonídeo em pequenos tributários de águas brancas do Alto Rio Juruá.

Palavras-chave: raia chicote, raia de água doce, potamotrigonídeos, Chondrichthyes, bacia do Juruá.

Introduction

Family Potamotrygonidae comprised four genera (Heliotrygon Carvalho and Ragno 2011; Paratrygon Duméril 1865, Plesiotrygon Rosa, Castello and Thorson 1987 and Potamotrygon Garman 1877) and lives exclusively in Neotropical freshwater environments (CARVALHO et al., 2003; ROSA et al., 2010; CARVALHO; LOVEJOY, 2011). Therefore, more recently a new genus (marine amphi-American stingrays), Styracura Carvalho, Loboda and Silva 2016 was included in the Family Potamotrygonidae (CARVALHO et al., 2016). Rosa et al. (1987) described the genus Plesiotrygon and it was monotypical until 2011. The type-locality was reported from Rio Solimões, above the municipality of Tefé. It is commonly cited that *P. iwamae* is restricted to the main Rio Amazonas/ Solimões channel (ROSA et al. 2010, DUNCAN; FERNANDES, 2010). In fact, several specimens of this freshwater stingray were collected in the Rio Solimões and the floodplain lakes (DUNCAN et al., 2015). P. iwamae is common ray found in shallow waters near the sand banks formed during the dry season in white-water rivers (DUNCAN; FERNANDES, 2010). Adults have a larger body size, a disk shape that is markedly oval and a long caudal filament (ROSA et al., 1987, CARVALHO; RAGNO, 2011). These characteristics are even found in the embryonic stage of development (DUNCAN et al., 2015).

The second species, *Plesiotrygon nana* Carvalho and Ragno 2011 was recently described. This species shares several morphological characteristics with *P. iwamae* but, in general, differs from its congener by an unusually small adult size, a dorsal coloration, a circular disk and distal coloration of the tail posterior to the caudal stings (CARVALHO; RAGNO, 2011).

According to these authors, the holotype is an adult male with a 247 mm disk width from the Río Pachitea, a tributary of Río Ucayali, up-river from the town of Puerto Inca, Peru. The other two paratypes were collected in the Rio Amazonas (near Tamshiyacu, Loreto Department) and Río Itaya, a tributary of Río Nanay (near Iquitos city), both located in Peru. A non-type specimen was reported from Rio Solimões, near the confluence with Rio Purus in Brazil. According to Carvalho; Ragno (2011), this ray was a juvenile or pre-adult female, with 118 mm disk width, collected by the Calhamazon Project (Arizona University/Museum of Zoology of the University of São Paulo) in 1996. As far as it is known, this was the first report of P. nana for Brazilian rivers. However, the site of capture was recorded as the main channel of Rio Solimões. Carvalho; Ragno (2011) clearly emphasize that this species is not restricted to the main Amazon channel as previously thought. To corroborate this statement, we examined specimens of the Neotropical freshwater stingray genus Plesiotrygon from Rio Tarauacá, a smaller tributary of Rio Juruá, southwest of state of Amazonas.

The Rio Tarauacá has muddy water and 700 km long that discharges into the right bank of the Rio Juruá. These rivers both meander (i.e. are highly sinuous) and have numerous oxbow lakes with muddy water, high dissolved solids and a circumneu-tral pH. The aquatic fauna is poorly described for Rio Tarauacá. To date, there are no records of potamotrygonid rays in the lower course of Rio Tarauacá. We reported the occurrence of *P. nana* to this river and provided a morphological comparison with the type specimens of *P. nana* and *P. iwamae*, as described by Carvalho; Ragno (2011) and Rosa et al. (1987), respectively.

Material and Methods

Fish collection and site description

The specimens of *Plesiotrygon nana* were collected in the Rio Tarauacá (07°43'25"S; 70°02'40"W) on July and November, 2015 by one of us (AEF), Municipality of Envira (Figure 1), State of Amazonas, near the border with State of Acre (Brazil). The stingrays were caught using a casting net near a sandy bank in the shallow water at 180 km above the confluence of Rio Juruá. The Rio Tarauacá has muddy water during rainy season; however, it is more transparent in the dry season.

Water environmental characteristics

The water analysis was carried out in same area of fish sampling. Five measurements of physicochemical parameters were analyzed using a Consort C535 multiparameter analyzer. In general, the white-waters had a pH of 6.3 \pm 0.7, electric conductivity of 156.1 \pm 20.4 μS cm $^{-1}$, dissolved oxygen of 3.7 \pm 2.1 mg L $^{-1}$, temperature of 28.5 \pm 3.8°C, and total dissolved solids of 83.7 \pm 9.5 mg L $^{-1}$. Additionally, dissolved ions as sodium (Na $^{+}$ 5.4 \pm 0.6 moles L $^{-1}$) and potassium (K $^{+}$ 2.6 \pm 0.04 moles L $^{-1}$) were also analyzed by flame photometry (Digimed DM-500), while calcium concentration (Ca $^{2+}$ 3.5 \pm 0.1 mmoles L $^{-1}$) was quantified by colorimetric methods (Labtest kit).

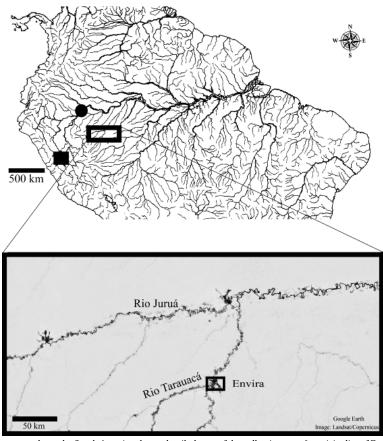


Figure 1. Map showing the study area. The rectangle on the South America shows detailed map of the collection area (municipality of Envira) of *Plesiotrygon nana* examined in this study. The symbols on the map indicate the collection sites of the type specimens, paratypes (circle) and holotype (square) according CARVALHO; RAGNO, 2011. / Figura 1. Mapa mostrando a área de estudo. O retângulo na América do Sul mostra o mapa detalhado da área de coleta (município de Envira) de *Plesiotrygon nana* examinado neste estudo. Os símbolos no mapa indicam os locais de coleta dos espécimes tipo, paratípos (círculo) e holótipo (quadrado) de acordo com CARVALHO; RAGNO, 2011.

Morphometric characters

The terminology and measurements follow Carvalho; Ragno (2011). The morphometric parameters were taken from the fresh specimens using a digital caliper. The data was presented in millimeters and then transformed into % in disk width. These data were used for comparisons with the specimens of *P. nana* and P. iwamae reported by Carvalho; Ragno (2011). It is important to emphasize that type-series (holotype and paratype) of P. nana and P. iwamae are catalogued at Museum of Zoology of the University of São Paulo. In this study, after measurements, the animals were preserved in 70% ethanol. This work was based on the examination of three specimens of P. nana collected in the Rio Tarauacá. Voucher specimens were catalogued at Instituto Nacional de Pesquisas da Amazônia (INPA-ICT 049941) and at the Laboratory of Functional Morphology (LMF-194 and LMF-195), Federal University of Amazonas (UFAM), Manaus, Brazil. The specimens were collected with the permission of the Chico Mendes Institute for Biodiversity Conservation (ICMBio/ SISBIO #7466-1 to WPD), and all protocols were conducted in accordance with institutional guidelines for the protection of animal welfare (Ethical Committee of Animal Experimentation, CEAA-UFAM protocol N°.070/2012).

Results

The linear measurements of the three specimens of *Plesiotrygon nana* collected in the Rio Tarauacá are presented in Table 1. The specimen identified as INPA-ICT 049941 was a juvenile male weighing 150 g, while LMF-194 and LMF-195 were adult males weighing 332 and 318 g, respectively. The dorsal coloration was characterized by a dark brown background color with yellow rosette-like pattern. The caudal filament had a dark purplish-brown color (Figure 2) and possessed a ventral tail fold. These two features allowed us to identify the specimen as *P. nana*. Numerous, very small, yellow spots in a dark brown background color were observed near the

disk's edge (Figures 3A and 3E). The disk had a more circular shape and spiracles were small and had a slightly rhomboidal shape (Figure 3B). The mouth was narrow (Figures 3C and 3D) and smaller ranging from 4.7 to 5.0% disk width. The specimens LMF-194 and LMF-195 have rigid and firm claspers indicating that both rays were sexually mature (Figure 3F).

The dermal denticles were similar to those reported in the description of *P. nana* above. We observed three types of dermal denticles over the disc and tail: numerous small denticles with stellate bases, larger denticles with a wider stellate base and several small denticles in vertical position as spine-like (Figure 3G).

Table 1. Morphometric characters (as minimum-maximum) of specimens of *Plesiotrygon nana* (INPA-ICT 049941, LMF-194 and LMF-195) collected in the Rio Tarauacá, Upper Rio Juruá. The data were transformed in percentage of disc width (in %, DW) to allow comparisons with specimens of *P. nana* and *P. iwamae* described by Carvalho; Ragno (2011). / **Tabela 1.** Caracteres morfométricos (como mínimo-máximo) de espécimes de *Plesiotrygon nana* (INPA-ICT 049941, LMF-194 e LMF-195) coletados no Rio Tarauacá, no Alto Rio Juruá. Os dados foram transformados em porcentagem de largura do disco (em %, DW) para permitir comparações com espécimes de *P. nana* e *P. iwamae* descritos por Carvalho; Ragno (2011).

| Parameters | <i>Plesiotrygon nana</i> examined in this study | | Type-series of P. nana | Specimens of <i>P. iwamae</i> |
|--------------------------------------|---|---------------|------------------------|-------------------------------|
| | (mm) | %DW | %DW | %DW |
| Total length (TL) | 652.0 - 995.0 | - | - | - |
| Disc length (DL) | 153.0 - 210.0 | 97.7 - 102.0 | 98.4 - 112.5 | 99.7 - 107.0 |
| Disc width (DW) | 150.0 - 215.0 | - | - | - |
| Interorbital distance | 9.9 – 24.6 | 6.6 – 11.6 | 10.5 - 16.7 | 11.1 - 11.4 |
| Interspiracular distance | 21.6 - 28.1 | 13 - 14.4 | 12.6 - 25.0 | 13.1 - 14.0 |
| Eye length | 4.4 - 7.0 | 2.6 – 3.3 | 1.8 - 5.6 | 1.4 - 1.8 |
| Spiracle length | 6.7 - 7.4 | 3.4 - 4.5 | 2.8 - 6.9 | 5.9 - 8.0 |
| Preorbital length | 33.2 - 42.9 | 19.1 - 22.1 | 20.0 - 22.7 | 27.5 - 28.8 |
| Prenasal length | 24.3 - 30.6 | 13.8 - 16.2 | 12.1 - 18.1 | 18.0 - 19.6 |
| Preoral length | 30.2 - 39.8 | 17.2 - 20.1 | 16.2 - 22.2 | 24.9 - 27.9 |
| Internarial length | 8.8 - 11.2 | 4.9 - 5.9 | 6.1 - 8.3 | 7.6 - 10.0 |
| Mouth width | 7.0 – 10.6 | 4.7 - 5.0 | 6.5 - 8.3 | 8.1 - 13.3 |
| Distance between 1st gill slits | 34.6 - 47.3 | 22.0 - 23.1 | 19.4 - 29.2 | 21.9 - 25.6 |
| Distance between 5^{th} gill slits | 27.8 - 40.1 | 18.5 – 18.7 | 16.6 - 20.8 | 17.4 - 19.6 |
| Branchial basket length | 19.5 - 23.1 | 10.5 - 13.0 | 10.9 - 12.9 | 11.5 - 14.3 |
| Pelvic fin anterior margin length | 36.7 - 50.0 | 22.9 - 24.5 | 23.5 - 30.6 | 23.4 - 27.6 |
| Pelvic fin width | 83.3 - 106.0 | 47.6 - 55.5 | 48.6 - 57.6 | 47.9 - 53.8 |
| Clasper external length | 7.4 – 21.7 | 4.9 - 10.3 | 6.5 - 8.8 | 7.6 - 8.9 |
| Clasper internal length | 13.7 - 46.6 | 9.1 – 21.7 | 15.4 - 16.5 | 3.4 - 21.4 |
| Distance between cloaca and tail tip | 520.0 - 820.0 | 346.7 - 390.5 | 311.2 - 540.3 | 153.0 - 527.4 |
| Tail width | 15.9 – 31.8 | 10.6 - 14.8 | 11.1 - 15.3 | 9.6 - 12.5 |
| Snout to cloaca distance | 125.0 - 170.0 | 79.1 - 83.3 | 84.2 - 94.4 | 84.7 - 97.2 |
| Pectoral to posterior pelvic length | 16.5 - 31.4 | 11.0 - 14.6 | 22.2 - 30.0 | 15.3 - 23.2 |
| Distance from cloaca to sting origin | 111.5 - 150.0 | 69.0 - 74.3 | 64.0 - 79.2 | 56.8 - 63.3 |
| Sting length | 22.3 - 37.9 | 14.9 – 17.6 | 21.8 - 22.2 | 17.5 - 27.0 |
| Sting width | 3.8 - 5.7 | 2.5 – 2.7 | 1.2 - 2.1 | 1.3 - 1.9 |

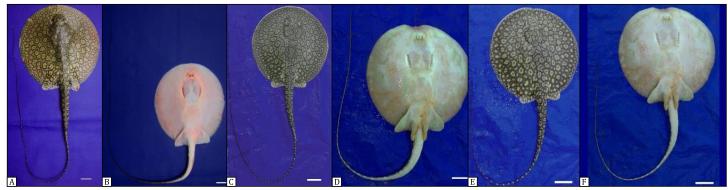


Figure 2. Dorsal and ventral views of *Plesiotrygon nana* from Rio Tarauacá, Brazil. (A and B) juvenile male (INPA-ICT 049941; 150 mm DW). (C and D) adult male (LMF-195; 210 mm DW). (E and F) adult male (LMF-194; 215 mm DW). Scale bars in A and B = 20 mm; C, D, E and F = 50 mm. / Figura 2. Vista dorsal e ventral de *Plesiotrygon nana*, Rio Tarauacá, Brasil. (A e B) do sexo masculino juvenil (INPA-ICT 049941; 150 mm DW). (C e D) macho adulto (LMF-195; 210 mm DW). (E e F) macho adulto (LMF-194, 215 mm DW). Barras de escala em A e B = 20 mm; C, D, E e F = 50 mm.

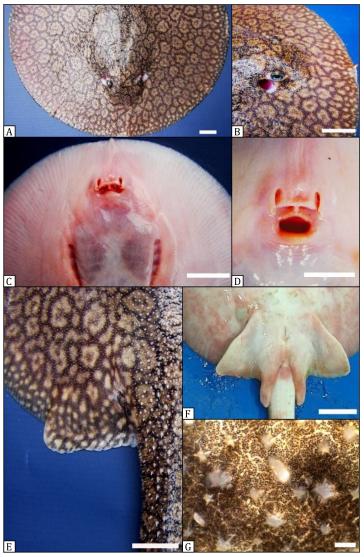


Figure 3. Details of the anterior disc in the juvenile male (INPA-ICT 049941) of *Plesiotrygon nana* (A). Morphological details of spiracle (slightly rhomboidal) and very small eyes (B). Ventral view (C) and details of the nostril region (D). Dorsal view of pelvic fin and base of tail (E). Pelvic fins and clasper viewed on the ventral side of adult specimen (F). Details of dermal denticles on the dorsal disc (G). Scale bars in A and C = 20 mm; B, D and E = 10 mm; F = 50 mm; G = 0.5 mm. / Figura 3. Details do disco anterior no macho juvenil (INPA-ICT 049941) de *Plesiotrygon nana* (A). Details morfológicos do espiráculo (ligeiramente romboidal) e olhos muito pequenos (B). Vista ventral (C) e detailse da região da narina (D). Vista dorsal da nadadeira pélvica e base da cauda (E). Nadadeiras pélvicas e clasper vistos no lado ventral do espécime adulto (F). Detailse de dentes dérmicos no disco dorsal (G). Barras de escala em A e C = 20 mm; B, D e E = 10 mm; F = 50 mm; G = 0,5 mm.

Discussion

It was suggested that Plesiotrygon nana was one of the smallest known freshwater stingrays. According to Carvalho; Ragno (2011), this species appeared to be smaller than the cururu ray (Potamotrygon wallacei Carvalho, Rosa e Araújo 2016), a new species recently described from the Rio Negro. P. wallacei seems to reach sexual maturity with 160 mm disk width (DW) for males and 170 mm for females (CHARVET-ALMEIDA et al., 2005). The two adult males were around 21 cm DW and weighed approximately 300 g. These dimensions are similar to those reported for the adult form of the cururu ray. Based on the morphological features, such as dermic denticles, caudal stings and clasper size, the specimen identified as INPA-ICT 049941 was not a neonate. Although the clasper was not well developed, the morphometric parameters suggested an immature individual. We believe that the P. nana was born with a DW less than 100 mm. In contrast, the congener species, Plesiotrygon iwamae, had a DW at birth which is greater than 120 mm because embryos of P. iwamae were recorded with a DW greater than 100 mm (DUNCAN et al., 2015). All the stingrays were morphologically similar to the Peruvian specimens and their morphological parameters are within the range reported for the species. P. nana from Rio Tarauacá had a

dorsal color with a rosette-like pattern, similar to rays collected in the Peruvian basin, although some polychromatic color can be found in several specimens, as reported by Carvalho; Ragno (2011). Other common features were numerous dermic denticles over the dorsal disc, including the tail. The tail coloration was also similar to the Peruvian specimens. A distal coloration of the tail posterior to the caudal stings was observed. This feature was likely responsible for the term "black-tailed," as this ray is called in the ornamental market (ROSS; SCHÄFER, 2000). All of these characteristics (numerous dermal denticles, dorsal color and tail coloration) were reported as distinctive of P. iwamae, according to the authors cited above.

The occurrence of *P. nana* to other areas was already expected for some tributaries of the Rio Amazonas/Solimões (CARVALHO; RAGNO, 2011). These authors reported that *P. nana* is not restricted to the main river channel, as can be seen for *P. iwamae*. We suggest that abundance of *P. nana* is high in the headwater of small tributaries of the largest muddy (white) water rivers. The occurrence of this species in the Rio Tarauacá (Upper Rio Juruá basin) can be explained by dispersal routes through the main channel of the major rivers, since the muddywater rivers have similar physicochemical and ecological characteristics (DUNCAN; FERNANDES, 2010). Another

possible explanation may be associated with historical headwater capture events between major drainages. However, the Juruá basin is separated from the Peruvian Ucayali basin on the western side by the Cenozoic basement (Serra do Divisor). The eastern side is also separated from Marañon and Solimões basin by the Late Jurassic Envira Arch (CAPUTO, 1991). In the past, these sub-basins originally formed a single major basin (CAPUTO, 2014). Therefore, it can be expected that *P. nana* can be found in the tributaries from the Upper Rio Purus. If this species is confirmed for this drainage, it is suggestive to speculate that this ray may have evolved in the areas of the ancient Pebasian Sea.

We conclude that the specimens collected in Rio Tarauacá belong to the dwarf species of freshwater stingray, *Plesiotrygon nana*. All animals were collected at the same site, suggesting the occurrence of a well-defined local population in that river. This finding extends the geographic distribution of this species and also reports their presence in small tributaries of white-water from Upper Rio Juruá in the Brazilian Amazon.

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