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Ecological notes on Rhinopetitia potamorhachia (Characidae: Stevardiinae) with extension of occurrence area at the Amana National Forest Reserve, Middle Rio Tapajós, Amazon Basin

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 $The geographic distribution of a small \ rare \ fish \ in the \ family \ Characidae \ (subfamily \ Stevardiinae) \ \textit{Rhinopetitia potamorhachia} \ Netto-like \ for \ fish \ final \ fish \ f$ Ferreira, Birindelli, Sousa & Menezes 2014, which was only known for its type locality in Rio Teles Pires (limits of Pará and Mato Grosso States), an area significantly affected by the construction of one hydroelectric plant. Currently the specie was found with endangered status according to the Brazil Red Book of Threatened Species of Fauna. Its distribution was extended to the middle Rio Tapajós, inside the National Forest of Amana (FLONA of Amana) and in the limits of the National Park of the Amazon, in the Municipality of Itaituba-Pará. This new record is more than 700 km apart from the previously registered location, in riverway distance, which provides relief for the endangered species. In addition to the new distribution record, brief information on feeding and reproduction of the species were provided.

Keywords: Characiformes; Rhinobrycon; distribution; anthropogenic impact; Amazon Eastern.

Nota ecológica sobre Rhinopetitia potamorhachia (Characidae: Stevardiinae) com extensão de área de ocorrência na Reserva Floresta Nacional do Amana, no Médio Rio Tapajós, Bacia Amazônica



A distribuição geográfica de um pequeno e raro peixe da família Characidae (subfamília Stevardiinae) Rhinopetitia potamorhachia Netto-Ferreira; Birindelli; Sousa & Menezes 2014, que era conhecida somente na sua localidade tipo, no rio Teles Pires (limites dos Estados do Pará e Mato Grosso) que é uma área significativamente afetada pela construção de uma usina hidrelétrica. Atualmente a espécie encontra-se com status de em perigo de extinção, de acordo com o Livro Vermelho de Espécies Ameaçadas da Fauna do Brasil. Sua distribuição é estendida para a bacia do médio rio Tapajós, dentro da Floresta Nacional do Amana (FLONA do Amana) e nos limites do Parque Nacional da Amazônia, no município de Italituba-Pará. Este novo registro está a mais de 700 km de distância da localização previamente registrada, em distâncias hidroviárias, fato importante, para uma espécie ameaçada de extinção. Além do novo registro de distribuição, breves informações sobre a alimentação e reprodução da espécie são fornecidas.

Palavras-chave: Characiformes, Rhinobrycon, distribuição, impactos antropogênicos, Amazônia Oriental.

Introduction

The Characidae generally composed of small Neotropical fishes, is the most diverse of Characiformes, currently containing more than 1,395 species recognized as valid (CAS, 2017), including in this amount a considerable number of species which are not yet described formally (REIS et al., 2003; CARVALHO et al., 2007). The Characidae is largely distributed in the freshwaters of the Neotropical region, from the Southern of United States of America to Argentinian Patagonia and, to tropical areas of Africa (NELSON et al., 2016). Characidae was recently redefined with based in 365 characters, most osteological, remaining ones come from coloration, external features and reproductive biology (MIRANDE, 2010). Most of the fish constituting this family are traditionally called tetras, which are now included in a group of genera denominated incertae sedis in Characidae, due to the uncertain relationships and dubious monophyly of the most genera (LIMA et al., 2003). Most of these small fishes usually have two rows of teeth in the premaxilla, eight branched rays in the dorsal fin, usually dimorphic hooks present in the pelvic and anal fin rays of mature males (LIMA et al., 2003).

Recent phylogenetic studies demonstrate that the monophyly of this group of small fish is valid (MIRANDE, 2010; OLIVEIRA et al., 2011), and are currently recognized as a subfamily denominated Stevardiinae. Besides the phylogeny, very little is known about the natural history of the most Stevardiinae species, their feeding, reproduction, distribution and natural habitats, which are still poorly known. In the Amazon basin, the species of this subfamily generally live in different environments, as small streams and, margin and beaches to the large rivers. Among the Stevardiinae fishes, are include about of 44 genera, and two species in genus Rhinopetitia.

The first, Rhinopetitia myersi described by Géry (1964), only known for its Holotype, with restricted distribution on the

Bananal Island, in the upper Rio Araguaia, a tributary of the Rio Tocantins, Brazil. The second, Rhinopetitia potamorhachia, Netto-Ferreira; Birindelli; Sousa & Menezes (2014), was described with restricted distribution to the Rio Teles Pires (and its small affluents), a tributaries of the upper Rio Tapajós, situated on the border of Pará and Mato Grosso States. Rhinopetitia (GÉRY, 1964), was considered a sister genus of Rhinobrycon (MIRANDE, 2010), a monotypic characid (Rhinobrycon negrensis) endemic to the Rio Negro, Brazil (GÉRY, 1964; 1977).

The Rhinopetitia genus was diagnosed as a member of Characidae, which has a rudimentary upper lip, in that way exposing the outer pre-maxillary tooth row, also has a lateral line interrupted on the caudal peduncle area, exhibits the presence of a nonsetiform gill-rakers and an almost entirely closed anterior fontanelle (GÉRY, 1964). In addition to these characteristics, Géry (1977) supplemented the diagnostic characteristics for this genus, including: a pronounced and pointed nose with sub-inferior mouth, presence of adipose fin, oval eyes, external premaxillary tooth row with 6 to 7 tricuspid teeth, 15 rays in the anal fin and pale head and a body overall.

The two Rhinopetitia species are considered rare ones (NETTO-FERREITA el al. 2014). According to Netto-Ferreira et al. (2014), due to the lack of available sample material, their phylogenetic position within Stevardiinae group which has not yet been critically evaluated in the phylogenetic studies (MIRANDE, 2009; MIRANDE, 2010). The Rhinopetitia potamorhachia is easily diagnosed from its single congener (R. myersi) by having all premaxillary (outer and inner rows) with 7 to 9 cuspids teeth, a maxillary teeth with 7 to 8 cuspids ones, a distinct dark midlateral stripe on the body and a round humeral blotch (NETTO-FERREIRA et al., 2014). This species was only known previously from its type of locality at the Rio Teles Pires and Rio Tapajós basin, Brazil. Here we provide data for increasing this specie natural distribution range inside the Rio Tapajós basin, from specimens collected in the Montanha stream in the middle Rio Tapajós region.

Materials and Methods

Study area

The Reserve has an area of approximately 542,000 hectares and is located at the Itaituba municipality, in the Pará State (PA), neighboring with Amazonas State (AM), Brazil. In the central part of the Forest Reserve, there is a region of plateaus, which "put apart" the watercourses that drainage into the Rio Tapajós (PA), from those which drainage into the Rio Maués-Açú (AM), consequently this region is denominated as Tapajós-Madeira inter-fluvial area (STCP, 2008) Figure 1. In the west portion of the Tapajós-Madeira, inter-fluvial area, is bedded the Amana River basin, which from the Forest Reserve is named. In the eastern portion, the region presents a series of small to mid-sized streams flowing directly into the Rio Tapajós, of which the Montanha stream belongs (04°52'44 "S, 56°58'24" W) Figure 1. Montanha stream is about 20 to 30 meters wide, and it's is bordered by the areas of dense forests and it is apparently well preserved (Figure 2). This stream be existent a sequence of five rapids, all of them with strong water flow. The Rhinopetitia potamorhachia specimens were captured about 27 km from the mouth of the Tapajós River.

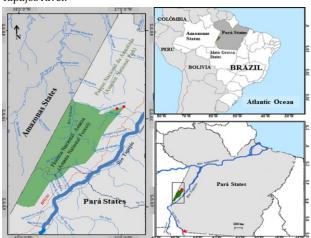


Figure 1. Records of *Rhinopetitia potamorhachia*. Red star represents the type locality in the Rio Teles Pires, bound in Pará and Mato Grosso States (Municipality of Jacareacanga); Red points represents the new record at Montanha stream, middle Rio Tapajós (Municipality of Itaituba) With details for the Forest Reserve (Floresta Nacional do Amana) (map modified of STCP, 2008).

Sample collections

The fish samples were conducted during an ichthyofauna survey occurred at the Forest Reserve (Floresta Nacional do Amana), where was collected the specimens of *Rhinopetitia potamorhachia* (= *Rhinopetitia* sp. "amana" in BELTRÃO et al. (2016)). The fishes were captured (by collection permit No. 13344-1; from IBAMA - Brazilian Institute of Environment and Renewable Natural Resources) in the period of April to August of 2008, by using a seine, in a mid-sized stream (Montanha stream) located inside of the Amana National Forest Reserve.



Figure 2. Montanha stream tributary of the middle Rio Tapajós, located in the National Forest Amana, habitats of the *Rhinopetitia potamorhachia*.

Part of the specimens captured, were submitted to the stomach contents analyzis, for frequency of occurrence and food index (KAWAKAMI; VAZZOLER, 1980), besides the gonadal analysis (VAZZOLER, 1996). The remaining fishes were deposited in the fish collections of the National Institute of Amazonian Research – INPA (voucher number INPA-53209), and of the Federal University of Amazonas – UFAM (voucher number UFAM-207 and UFAM-208).

Results

A total of 78 specimens of *R. potamorhachia* were captured during the study, from this amount 56 specimens were collected through the rainy season (in April) and 22 specimens sampled during the dry season (in August) of 2008. A number of 40 individuals were chosen to stomach contents and gonadal analysis. The remaining (38 specimens) were deposited in the fish collections at the INPA, and UFAM.

Among the diagnostic characteristics of *R potamorhachia* mentioned in the species description (NETTO-FERREIRA et al., 2014), the population found in Montanha stream had similar in the round humeral blotch, where half of the individuals presented a slightly conspicuous and poorly diffuse humeral blotch (Figure 3). The dark range along the mid-lateral body, as mentioned in the description of the species, is quite conspicuous. This dark longitudinal range is characterized by the presence of small black chromatophore dots, which was dispersed in the medial humeral region of the fish body. As a result, it's gradually decrease in quantity and taper to the mid-lateral body (region of the last rays of the dorsal fin), whose median dark range is narrow and quite defined up to the fish caudal peduncle (Figure 3A). Together with the dark range, a longitudinal black line was also observed in specimens preserved in alcohol (Figure 3A).

Other diagnostic characteristics were observed, such as two rows of the premaxillary teeth; an outer row with six (6) teeth (7 to 9 cuspids) and an inner row with four (4) teeth (7 to 9 cuspids). A maxillary with 2 small teeth (7 to 8 cuspids) and dentary with 5 teeth, being the first 3 teeth larger (7 to 8 cuspids) and the last two much smaller (5 to 7 cuspids) Figure 3B and 3C.

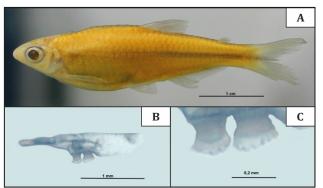


Figure 3. (A) Rhinopetitia potamorhachia Montanha stream (male 3.7 cm CP) deposited in INPA fish collection (voucher number INPA-53209). (B) Maxillary with 2 small teeth (7 to 8 cuspids) and (C) Details of the teeth of the maxillary, morphologically as mentioned in the species description.

The females captured during the rainy season were in the process of gonadal development, had gonads in stage B, oocyte development, according to the macroscopic scale proposed by (VAZZOLER, 1996), a light yellow color and oocytes clearly observed in the stereo microscope (Figure 4A). In males, testicles were observed in the form of filaments (Figure 4B), and small bone hooks were present in the pelvic and anal fins (Figure C and D), similarly as diagnosed in the description of the species. As for the specimens captured during the dry period, gonads and testicles presented as small filaments, often almost imperceptible even in stereomicroscope, suggesting that the specimens were in the period of gonadal resting (VAZZOLER, 1996).

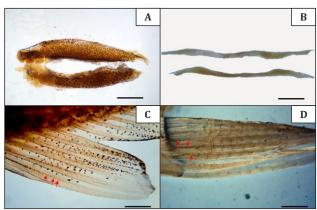


Figure 4. (A) Gonads of *Rhinopetitia potamorhachia* in full oocyte development and, (B) testicle of sexually mature males. (C) anal and (D) pelvic fins who had small bony hooks of male sexually mature (3.4 cm of Standard Length). Scale bars = 1 mm.

Rhinopetitia potamorhachia has principally insectivorous habit. Its diet consists of at least five main groups of food items (n=28, analyzed specimens) of autochthonous and allochthonous origins. Terrestrial insects, especially Hymenoptera (species of the family Formicidae, "ants") (Figure 5A and 5B), Isoptera (termites), Coleoptera (beetles) and Vespidae (wasp). Also were found in the stomach content filamentous and conjugated algae (mainly Bacillariophyceae-filaments) that had a predominant role in their diet (Figure 5C and 5D). Plant material, aquatic insects and detritus were also part of their diet (Figure 5E to 5H).

The highest volume of food items consumed by *R. potamorhachia* were from allochthonous origin. The frequency of occurrence and the relative volume of food items indicate that the food of major importance in the diet of this fish population, found in Montanha stream are: terrestrial insects (50%, mainly Formicidae), algae (30%), plant material (10%) aquatic insects and detritus (5% each). The intestine length is on average 92.4 % (\pm 5 SD - Standard Deviation) of the standard length (SL), with an intestine diameter of 1.3 mm (\pm 0.1 SD).

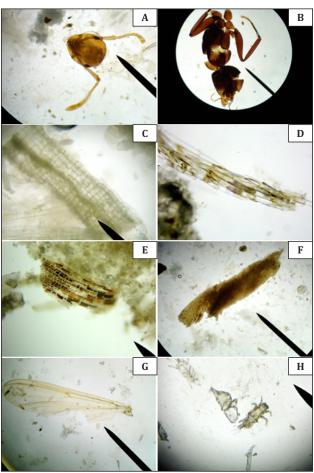


Figure 5. Characteristics of the main food items found in the stomachs of *R. potamorhachia*: (A and B) Ants (Hymenoptera), (C and D) Filamentous algae, (E and F) plant fragments, (G) wasp wing (Vespidae) and (H) aquatic invertebrates (stereoscope increased 40x).

Discussion

Rhinopetitia potamorhachia were only recognized just in a restricted area, the species was included with the endangered status in the Brazil Red Book of Threatened Species of Fauna (ICMBIO, 2016). Considering the absence of records at most of the ichthyofauna surveys previously performed before the construction of the hydroelectric plant and closing to the floodgates in 2015 (SILVA; HOJO, 2015), could be related with a very small population size or with rarity of the species (NETTO-FERREIRA et al., 2014).

Netto-Ferreira et al. (2014) mentioned that the area where the species was described in Rio Teles Pires, downstream of Sete Quedas rapids, where Teles Pires hydroelectric plant is being built, and the fish fauna of this region is threatened, including the only known population of *R. potamorhachia*.

The two species of the genus *Rhinopetitia* are considered rare (NETTO-FEREIRA et al., 2014). As an example *Rhinopetitia myersi*,

only known for its Holotype, with restricted distribution in the Bananal Island, in the Upper Rio Araguaia, an affluent of the Tocantins River, Brazil (GERY, 1964, 1977), after its description there was no more record of the species in the literature. A further expansion and distribution of *Rhinopetitia potamorhachia* should be celebrated because, although it is found in the same drainage basin, and this enlargement in the species distribution area helps to reduce in the status of threatened species.

Rhinopetitia potamorhachia was only known for being present on beaches on the margin of Rio Teles Pires and affluent stream, located in upper Rio Tapajós (NETTO-FERREIRA et al. 2014). The species name (potamorhachia) Greek potamo, meaning river, and rhachia, meaning beach, was denominated referring to the environment where the species was usually caught, sand beaches on the margin of the river, hence the adjective. In the present study, its distribution was expanded and included the Montanha stream at the basin of middle Rio Tapajós, into the municipality of Itaituba (PA), which suggests that its range of distribution may be in a broader area, at least 700 km from its type locality, at the Rio Teles Pires

In Montanha stream, the *R. potamorhachia* specimens were usually captured on the shallow margin of the stream, about 27 km upstream from the confluence of Rio Tapajós. The margin of the Montanha stream presented large areas with canopy cover, little sand, and large litter bank, showing that the species does not live only in a specific environment such as the beaches, but it can also be found in densely shaded streams.

The dataset confirmed also that the *R. potamorhachia* has an insectivorous habit due to its allochthones food items found into the specimens intestine. Furthermore, the gonadal analysis from male and female individuals suggests that the reproductive period of *R. potamorhachia* occurs during the rainy season, although, eventually, more detailed analyzes of sampling proceeds throughout the hydrological cycle have to be done to confirm this hypothesis.

In this way, long-term monitoring is need to confirm if the presence or absent of *R. potamorhachia* sub-population are related to the impacts of Teles Pires hydroelectric plant establishment in the river course. The current scenario for the *R. potamorhachia* from the Rio Teles Pires is concerning. Even including new sites from the present report, the species conserves a very small range of occurrence, mostly affected by several threats, such as deforestation and river dam.

With the extension of the distribution of this species, it opened a way for its conservation, since the new distribution area is within a perimeter between two Forest Reserves, the Amana National Forest and the Amazon National Park. In this way, the present specie occurrence report could be a great deal for the conservation and maintenance of the R. potamorhachia populations. Although, of course, this fish species survival and conservation depends on the preservation of its habitats and the maintenance of the water quality and course, in this region, which is little known by the scientists community, and also is already greatly altered by anthropic actions (as mentioned for the Rio Teles Pires region). Without the good integrated conditions of habitats and its connectivity, the probability of the occurrence of this fish species population local extinctions and the probable disappearance of the others unknown fish species, are greatly increased (HURD et al., 2016) and becomes even more worrying when facing the modifications in the aquatic environment (damming) that also contributes to local climate changes (MALHI et. al., 2008) and the disappearances of small and rare resident fish species (MERELES, et al., 2017).

Conclusions

This study reported a new location presence of *Rhinopetitia potamorhachia* in the National Forest of Amana at the middle Rio Tapajós region, bordering the Amazon National Park (Municipality of Itaituba - Pará State) it's extending more than 700 km (river way distance) apart from the previously registered type locality, in the Rio Teles Pires (limits of Pará and Mato Grosso States). In addition, the data matched that *R. potamorhachia* has an omnivorous habit, and possible do its reproductive period during

the rainy season. This finds, could be used to support management's plans in order to preserve the *R. potamorhachia* populations in the Amazon basin, and putout this fish species from the endangered list as informed by the Brazil Red Book of Threatened Species of Fauna.

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