

Original Article

## New records of the occurrence of *Megaleporinus macrocephalus* (Garavello & Britski, 1988) (Characiformes, Anostomidae) from the basins of the Itapecuru and Mearim rivers in Maranhão, Northeastern Brazil

Novos registros da ocorrência de *Megaleporinus macrocephalus* (Garavello & Britski, 1988) (Characiformes, Anostomidae) nas bacias dos rios Itapecuru e Mearim no Maranhão, Nordeste, Brasil

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### Abstract

The “piaussu”, *Megaleporinus macrocephalus* is an anostomatid fish species native to the basin of the Paraguay River, in the Pantanal biome of western Brazil. However, this species has now been recorded in a number of other drainages, including those of the upper Paraná, Uruguay, Jacuí, Doce, Mucuri, and Paraíba do Sul rivers. This study presents two new records of the occurrence of *M. macrocephalus*, in the basins of the Itapecuru and Mearim rivers in the state of Maranhão, in the Brazilian Northeast. The piaussu is a large-bodied fish of commercial interest that is widely raised on fish farms, and its occurrence in the Itapecuru and Mearim rivers is likely the result of individuals escaping from fish tanks when they overflow during the rainy season. Morphological analyses and sequences of the Cytochrome Oxidase Subunit I (COI) gene confirmed the taxonomic identification of the specimens as *M. macrocephalus*. The COI sequences were 99.66% similar to those of *M. macrocephalus* deposited in the BOLDSystems database. These records extend the known distribution of *M. macrocephalus* to the basins of the Itapecuru and Mearim rivers in the Brazilian Northeast, highlighting a new case of introduction of exotic fish species into Brazilian river basins.

**Keywords:** freshwater, Ichthyofauna, taxonomy, biological invasion, COI.

### Resumo

*Megaleporinus macrocephalus* é uma espécie de peixe anostomatídeo nativa da bacia do rio Paraguai, no bioma Pantanal do oeste do Brasil. No entanto, essa espécie já foi registrada em várias outras drenagens, incluindo as dos rios Alto Paraná, Uruguai, Jacuí, Doce, Mucuri e Paraíba do Sul. Este estudo apresenta dois novos registros da ocorrência de *M. macrocephalus*, nas bacias dos rios Itapecuru e Mearim, no estado do Maranhão, no nordeste brasileiro. O piaussu é um peixe de grande porte, de interesse comercial, amplamente criado em pisciculturas, e sua ocorrência nos rios Itapecuru e Mearim é provavelmente o resultado de indivíduos que escapam dos tanques quando transbordam durante a estação chuvosa. Análises morfológicas e sequências do gene da subunidade I do citocromo oxidase (COI) confirmaram a identificação taxonômica dos espécimes como *M. macrocephalus*. As sequências de COI foram 99,66% semelhantes às de *M. macrocephalus* depositadas no banco de dados BOLDSystems. Esses registros estendem a conhecida distribuição de *M. macrocephalus* às bacias dos rios Itapecuru e Mearim, no nordeste brasileiro, destacando um novo caso de introdução de espécies exóticas de peixes nas bacias hidrográficas brasileiras.

**Palavras-chave:** água doce, Ictiofauna, taxonomia, invasão biológica, COI.

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## 1. Introduction

The family Anostomidae is currently composed of 149 valid species, distributed in 15 genera (Fricke et al., 2020). Species of the family are endemic to the Neotropical region, and are widely distributed from north of Colombia to La Plata River, in Argentina (Garavello and Britski, 2003). The greatest diversity of anostomatids is found in the Amazon basin, where 91 species have been recorded to date (Dagosta and Pinna, 2019). The genus *Leporinus* Agassiz, 1829, is the most species-rich of Anostomidae (Burns et al., 2017).

Garavello and Britski (1988) described *Leporinus macrocephalus* based on 15 specimens collected in the Paraguay River basin in Brazil. The species was recently transferred to *Megaleporinus*, a genus described in 2017 by Ramirez et al. (2017), based on morphological, molecular, and cytogenetic data, including a unique system of ZZ/ZW sex chromosomes.

The fish of the genus *Megaleporinus* are relatively large in size (adult standard length typically over 35 cm), and are diagnosed among anostomids by having the following combination of exclusive features: premaxilla with three unicuspid teeth; dentary with three unicuspid teeth; and body with one to four dark midlateral blotches (Ramirez et al., 2017).

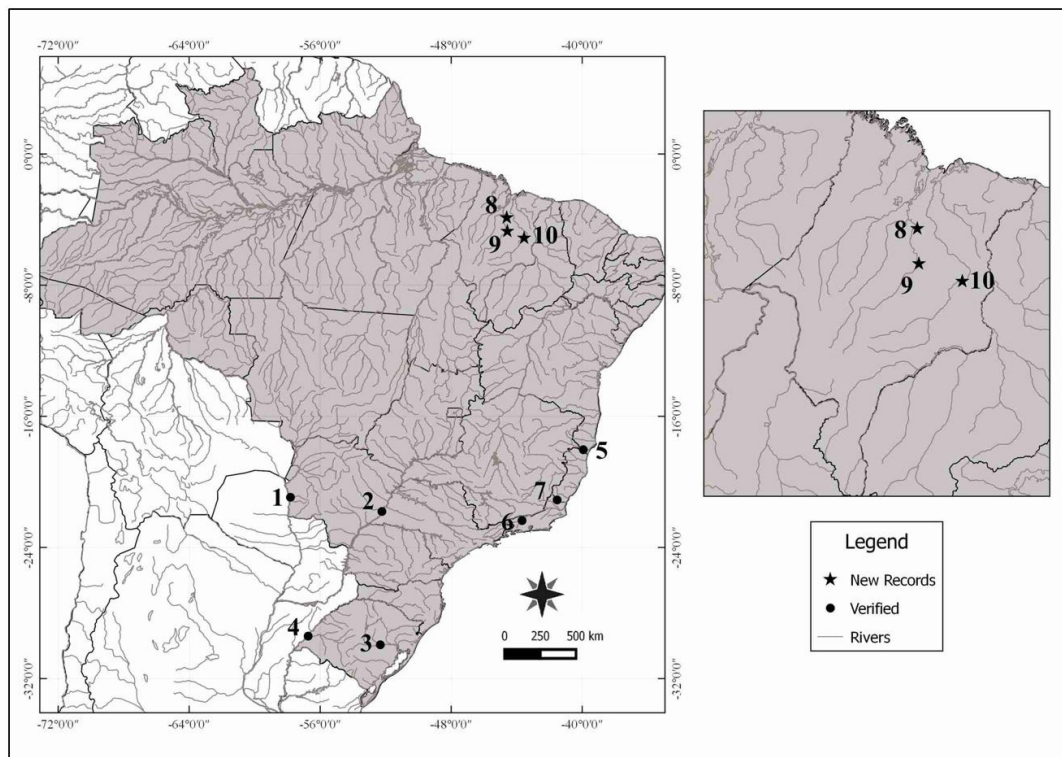
*Megaleporinus macrocephalus* (Garavello and Britski, 1988), known in Brazil as the “piaussu”, “piavuçu” or “piau-açu”, is an omnivorous fish that feeds on small fruit and seeds, as well as small fish and crabs (Peruca et al., 2000; Navarro et al., 2006). *M. macrocephalus* is distributed

in the Paraguay River in Argentina, Brazil, Bolivia and Paraguay (Garavello and Britski, 1988; Britski et al., 1999; Fricke, 2019). However, individuals escaped from fish farms have colonized a number of other South American basins, including that of the upper Paraná River (Graça and Pavanelli, 2007; Langeani et al., 2007), the Jacuí and Uruguay Rivers in the Brazilian state of Rio Grande do Sul (Bertaco et al., 2016), the Mucuri River (Gomes et al., 2015), and the Doce and Paraíba do Sul rivers (Alves et al., 2007). The piaussu is commercially important for sports fishing (Zeinad and Prado, 2012) and is widely raised on fish farms to supply markets in southern and central Brazil (Soares et al., 2000; IBGE, 2014).

The present study provides the first report of the occurrence of *M. macrocephalus* in northeastern Brazil, more specifically in the Itapecuru and Mearim rivers, in the state of Maranhão. The Mearim is the state's largest hydrographic basin, and plays an important role in the economy and subsistence of the region's riverside populations (Maranhão, 2011). These records are the first evidence of the occurrence of *M. macrocephalus* in the Itapecuru and Mearim Rivers, extending the limits of the known geographic distribution of the species.

## 2. Material and Methods

Specimens of *M. macrocephalus* were collected in the basins of the Itapecuru and Mearim rivers, in the state of Maranhão, Northeastern Brazil (Figure 1). The Mearim



**Figure 1.** Geographic distribution of *Megaleporinus macrocephalus* in Brazil. The area of the new registrations for the Itapecuru rivers in the municipality of Pé da Serra and Mearim in the locality Laje dos Currais, in São Mateus, and in the city of Pedreiras in the Northeast Region.

River has a total extension of 832.18 km, and has a number of tributaries, in particular the Pindaré River, which joins the Mearim only 20 km from the mouth of the Grajaú River, a second important tributary of the Mearim River. The Itapecuru River is located wholly within the state of Maranhão, and has three distinct stretches – the upper river (from the source to the municipality of Colinas), the middle river (from Colinas to the municipality of Caxias), and the lower Itapecuru, from Caxias to the mouth, in São José Bay (Alcântara, 2004). The Itapecuru River, which is 852.71 km long, has eight major tributaries on its right margin, in particular the Pirapemas and Itapecuruzinho Rivers, and the Seco, do Ouro, Gameleira, Cachimbo, and Guariba streams (UEMA, 2016).

The specimens were collected using gill, drag, and cast nets. The voucher specimens were taken to the Laboratory of Genetics and Molecular Biology (GENBIMOL) at the Center for Higher Studies at Maranhão State University (CESC/UEMA), fixed in 10% formalin, and conserved in 70% alcohol. The prepared specimens were sent to the Zoology Museum at the State University of Londrina (MZUEL), Paraná, Brazil, for morphological identification and cataloguing (voucher MZUEL: 15357 – Mearim river) e (voucher MZUEL: 20221 – Itapecuru river).

The collection of specimens was authorized by the Brazilian Federal Institute for the Environment and Renewable Natural Resources (IBAMA) through licence number 02012.004159/2006, and ICMBio/MMA licence 42119-2, from the Chico Mendes Institute for Biodiversity Conservation.

Measurements were taken following Britski and Garavello (1978) and Birindelli et al. (2013). Morphometric data were obtained with a digital caliper with accuracy of 0.01 mm. Counts of scales and rays were taken according to Birindelli et al. (2013).

Samples of muscle tissue were extracted from the specimens for the genetic analyses. The total DNA was extracted using the Promega Wizard Genomic DNA purification kit, following the manufacturer's instructions. The Cytochrome Oxidase Subunit I (COI) was then amplified by Polymerase Chain Reaction, using the universal primers COI FishF1: 5' -TCAACCAACCACAAAGACATTGCCAC - 3'; and COI FishR1: 5' - TAGACTTCTGGGTGCCAAAGAATCA - 3', as described by Ward et al. (2005). The samples were sequenced by Sanger et al. (1977) dideoxyterminal method, using a Big Dye kit in an ABI Prism™ 3500 automatic sequencer (Applied Biosystems, USA). The sequences were edited and aligned in BIOEDIT 7.0 (Hall, 1999), and plotted in the BOLD Systems v4 platform (BOLD, 2020) to evaluate their similarity with existing sequences (Ratnasingham and Hebert, 2007).

### 3. Results

On the Mearim River, one *M. macrocephalus* specimen was collected on the lower course of the river at Laje dos Currais, in the municipality of São Mateus, during the flood season, while two specimen was obtained on the middle course of the river in the town of Pedreiras during the dry (low water) season. The *M. macrocephalus* specimen was

collected at the locality of Pé da Serra in the municipality of Caxias during the dry season.

The four *M. macrocephalus* specimens (Table 1) present a characteristic coloration pattern, with a darker superior half of the body, three large black spots on each flank, and dark lines between the rows of scales (Ramirez et al., 2017). The body is robust, and the mouth terminal, with three teeth in the pre-maxillary bone and three teeth in

**Table 1.** Morphological data of *Megaleporinus macrocephalus*.

Characters	MZUEL 20221	MZUEL 15357
	Itapecuru	Mearim
Standard Length (mm)	387.1	175.2
<b>Percentages of standard length</b>		
Predorsal distance	41.10	43.83
Dorsal-fin origin to adipose-fin origin	41.37	37.76
Prepelvic distance	43.40	47.62
Body depth	28.14	28.06
Caudal-peduncle length	11.41	12.50
Caudal-peduncle depth	16.93	15.45
Anal-fin lobe length	10.59	10.04
Head length	22.32	27.26
<b>Percentages of head length</b>		
Preopercle length	75.81	64.68
Snout length	35.78	29.45
Head depth	96.92	85.41
Eye diameter	13.33	23.51
Bony interorbital	58.88	54.53
<b>Meristics</b>		
Lateral-line scales	43	42
Scale rows between lateral line and dorsal-fin origin	7	7
Scale rows between lateral line and anal-fin origin	6	6
Scale rows around caudal peduncle	16	16
Predorsal scales	13	13
Dorsal-fin unbranched rays	2	2
Dorsal-fin branched rays	10	10
Pectoral-fin branched rays	14	15
Pelvic-fin branched rays	8	8
Anal-fin unbranched rays	3	3
Anal-fin branched rays	8	8
Upper caudal-fin branched rays	9	9
Upper caudal-fin branched rays	8	8
Teeth on left premaxilla	3	3
Teeth on left dentary	3	3



**Figure 2.** *Megaleporinus macrocephalus*, MZUEL 20221, 387.10 mm SL.

**Table 2.** Similarity percentage obtained by comparing the sequences of the COI gene on the BOLDSystems platform (The Barcode of Life Data System) for the *M. macrocephalus* species from the Itapecuru and Mearim rivers, Maranhão, Brazil, identified morphologically and which were confirmed with the molecular data.

Código	N	Identificação	
		Taxonômica	Molecular: Bold
MEA 831	1	<i>M. macrocephalus</i>	<i>M. macrocephalus</i> (99.66%)
MEA 835	1	<i>M. macrocephalus</i>	<i>M. macrocephalus</i> (99.66%)
MEA 836	1	<i>M. macrocephalus</i>	<i>M. macrocephalus</i> (99.66%)
LEP 158	1	<i>M. macrocephalus</i>	<i>M. macrocephalus</i> (99.66%)

the dentary bone (Figure 2). The specimens have 42–43 scales in the lateral line, seven rows of scales between the lateral line and the origin of the dorsal fin, six rows of scales between the lateral line and the origin of the pelvic fin, and 16 rows of scales around the caudal peduncle. These characteristics, together with the morphometry of the specimens, are fully consistent with the descriptions of *M. macrocephalus* published Garavello and Britski (1988) and Britski et al. (1999), and leave no doubt with regard to the identity of the taxon.

Four COI sequences were obtained, one from the specimen collected from the Itapecuru River, and three from the specimens obtained from the basin of the Mearim River. Comparisons with the sequences available in the BOLDSystems database confirmed the morphological identification of the specimens. The COI sequences obtained from the specimens collected in the present study were more than 99% similar to that of *M. macrocephalus* from the basin of the Paraná River (Table 2). This value is well within the 2% threshold for the identification of fish species by DNA barcoding (Ward et al., 2009; April et al., 2011; Carvalho et al., 2011; Mabragna et al., 2011; Castro Paz et al., 2014).

#### 4. Discussion

No records of *M. macrocephalus* species were obtained during previous studies in the Itapecuru and Mearim rivers (Soares, 2005; Barros et al., 2011; Fraga et al., 2012; Nascimento et al., 2016; Abreu et al., 2019). It seems likely that the species was exhausted accidentally during the rainy season, when the tanks of local fish farms overflow and become connected with the tributaries of the two basins.

The species *M. macrocephalus* is used in fish pisciculture, larger and has economic importance for fishing in some regions, in particular in the Pantanal Mato-grossense (Navarro et al., 2007). *M. macrocephalus* is well-suited to fish pisciculture, which has led to its accidental introduction into a number of different Brazilian river basins (Godinho and Godinho, 2003; Barbosa and Soares, 2009; Vieira, 2010).

It is interesting to note that *M. macrocephalus* has not been recorded in recent surveys of other, nearby basins, in the Brazilian Northeast, including the Munim and Parnaíba in Maranhão/Piauí (Matavelli et al., 2015), the Parnaíba in Maranhão/Piauí (Ramos et al., 2014), and the São Francisco, in Bahia (Carvalho et al., 2011).

The records of *M. macrocephalus* presented here represent an important extension of the known distribution of the species, whose occurrence is now confirmed in the Itapecuru and Mearim rivers of Maranhão, in the Brazilian Northeast. It is nevertheless premature to confirm that *M. macrocephalus* has established viable populations in these rivers, and what effects its presence may have on the resident species. However, the introduction or transfer of exotic fish species into a river basin, whether intentional or otherwise, almost always has negative implications for the resident native species (Pelicice et al., 2014), and any invasion of this type requires carefully-planned management strategies to guarantee the long-term integrity of the local biota.

#### 5. Conclusion

The combined analysis of morphological and molecular analysis confirmed the occurrence of *M. macrocephalus* in the Itapecuru and Mearim Rivers, state of Maranhão.



This substantially amplifies the known distribution of this species to the Brazilian Northeast.

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