



## Fish fauna from a fragmented river in the Atlantic Forest

Paulo Roberto Monteiro de Brito<sup>1</sup>, Luiz Henrique Garcia Pereira<sup>2</sup>, Fábio Fernandes Roxo<sup>3</sup>,

Jefferson Henriques Monteiro<sup>3</sup>, Claudio Oiveira<sup>4</sup> & Guilherme José da Costa Silva<sup>5\*</sup>

<sup>1</sup>Universidade Santo Amaro, Departamento de Biologia, São Paulo, SP, Brasil.

<sup>2</sup>Universidade Federal da Integração Latino-Americana, Foz do Iguaçu, PR, Brasil.

<sup>3</sup>Universidade Estadual Paulista, Instituto de Biociências, Botucatu, SP, Brasil.

<sup>4</sup>Instituto Federal de São Paulo, São Paulo, SP, Brasil.

<sup>5</sup>Universidade Estadual Paulista, Botucatu, SP, Brasil.

<sup>6</sup>Universidade Santo Amaro, Pós Graduação em Medicina Veterinária, São Paulo, SP, Brasil.

\*Corresponding author: [costasilvagj@gmail.com.br](mailto:costasilvagj@gmail.com.br)

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**Abstract:** This study sought to identify the ichthyofauna composition of the Muzambinho River, an upland tributary of the Paraná River. We also investigate whether waterfalls in the region can serve as barriers to the dispersal of fish species. For this purpose, collections were carried out at 34 points on the Muzambinho River using different techniques. In all, 37 species were recorded, some of which were endemic, and the majority were native. Among these species, some are predominant in degraded places and can be used as bioindicators. The results also demonstrate zonation in the ichthyofauna composition in Muzambinho that segregates the fauna into three sessions separated by waterfalls: Lower Muzambinho, Upper Muzambinho, and Sao Domingos. We conclude that, for the ideal preservation of the ichthyofauna of the Muzambinho River, it is necessary to preserve its sections independently, which would guarantee the maintenance of naturally isolated strains.

**Keywords:** Upper Parana; Biodiversity; Ictiofauna; Mantiqueira.

## Fauna de peixes de um rio fragmentado na Mata Atlântica

**Resumo:** Este estudo buscou identificar a composição da ictiofauna do rio Muzambinho, um tributário de terras altas do Rio Paraná. Também buscamos investigar se as cachoeiras da região podem servir de barreiras para dispersão de espécies de peixes. Para isso, foram realizadas coletas em 34 pontos do Rio Muzambinho, com diferentes técnicas. Ao todo foram registradas 37 espécies de peixes, sendo algumas endêmicas e a maioria nativa. Dentre essas espécies algumas são predominantes em locais degradados e podem ser usadas como bioindicadores. Os resultados também demonstram que há uma zonação na composição ictiofaunística no Muzambinho que segrega a fauna em três sessões separadas por cachoeiras, Baixo Muzambinho, Alto Muzambinho e São Domingos. Concluímos que para a ideal preservação da ictiofauna do rio Muzambinho é necessária a preservação independente de suas sessões o que garantiria a manutenção de linhagens naturalmente isoladas.

**Palavras-chave:** Alto Paraná; Biodiversidade; Ictiofauna; Mantiqueira.

## Introduction

The Neotropical freshwater ichthyofauna accounts for almost 30% of the world's freshwater fish species, with more than 6000 described species and potentially a further uncataloged 3000 species (Reis et al. 2016). Most of these species are distributed in the Amazon, Orinoco, and Paraná-Paraguay river basins, which are among the largest rivers worldwide. However, several researchers have pointed out that a significant percentage of this biodiversity is found in small headwater streams (Castro 1999, Langeani et al. 2007).

Headwater streams (perennial water bodies of the first and second order) (Fagan 2002, Meyer et al. 2007, Richardson 2019) are a prominent

feature of the Neotropical region. The Headwater streams provide small areas of suitable habitat for residents species, resulting in relatively small isolated populations and ultimately leading to population segregation and speciation (Richardson 2019, Richardson & Danehy 2007). Furthermore, physical barriers, such as waterfalls and temperature gradients, can constitute impediments to the dispersal of stream species (Kurylyk et al. 2015, Torrente-Vilara et al. 2011), which can in turn contribute to the formation of refugia and eventually to the maintenance of relictual lineages (Buckup 2011), regardless of their taxonomic distinction. This segregation can also promote accelerated adaptive radiation, as small populations tend to be more susceptible to genetic drift-related factors, such as natural selection and local extinction (Frankham et al. 2004).

Located in the Brazilian Shield, the Muzambinho River is a typical Atlantic Forest river, which are commonly characterized by their high variation between lotic and lentic stretches. They are also home to a high diversity of endemic fish species whose evolutionary history is generally closely associated with the evolution of the rivers themselves (Castro 1999). The Muzambinho is a fourth-order river that can be divided into three sections separated by two waterfalls. 1) The Kita waterfall, locates at the mouth of the São Domingo stream, separates this tributary from the Muzambinho river. 2) The Usina waterfall separates the lower from the upper Muzambinho (Figure 1). This region, in which human activities are based on agriculture and livestock rearing, is characterized by an incredible diversity of plants and terrestrial animals (Domingos, 2014) typical of high-altitude Atlantic Forest areas and biodiversity hotspots. However, the fish fauna inhabiting the Muzambinho River in this region have yet to be systematically studied.

Given the growing anthropogenic pressure on natural areas and the lack of knowledge regarding the fish fauna of rivers and streams that occur in the Muzambinho basin, we conducted a detailed survey of the ichthyological components of the Muzambinho River (upper Paraná) to establish whether the waterfalls that separate the main stretches of this river can serve as barriers to the dispersal of species, and consequently provide havens for species inhabiting the upper reaches.

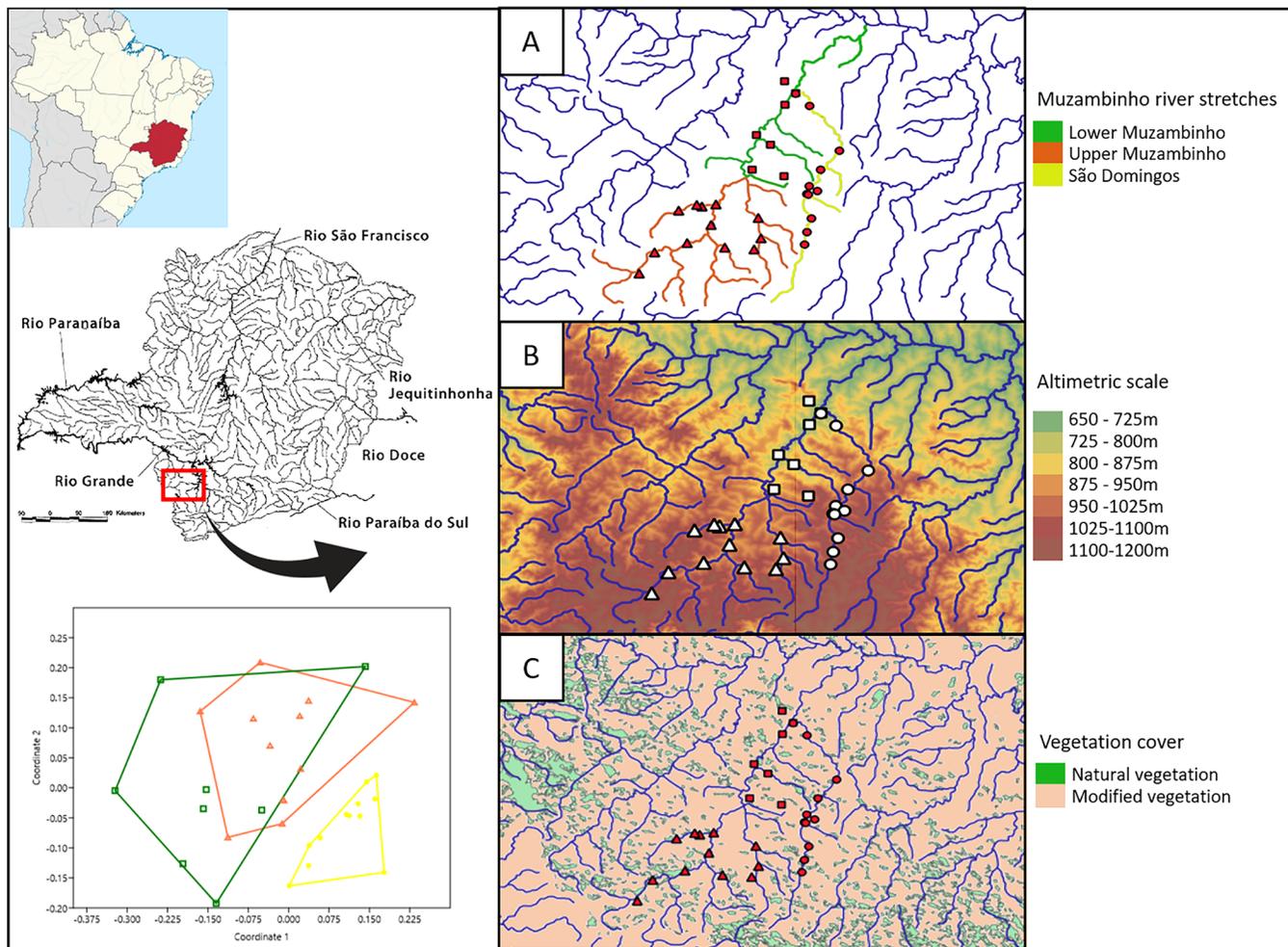
## Material and methods

### 1. Study area

The Muzambinho river presents both lentic and lotic systems. The is entirely present in the Serra da Matiqueira region, in the south of Minas Gerais. The Muzambinho flows into the Muzambo river, an important tributary of the Grande river. Small agricultural companies and enterprises mostly occupy the surroundings of the Muzambinho River. The climate is typically tropical at altitude, and the predominant vegetation is Atlantic forest. The sampling points were divided into three groups, which are herein defined as sections: (1) the São Domingos stream (SD - P1 to P14), (2) the lower Muzambinho (LM - P15 to P22), and (3) the upper Muzambinho (UM - P23 to P34). The definition of sections is based on the location of two waterfalls mentioned above, the Kita (coordinate  $21^{\circ}17'37.94''S$   $46^{\circ}29'6.33''W$ ) and Usina (coordinate  $21^{\circ}21'0.79''S$   $46^{\circ}31'2.87''W$ ), and on the premise that these waterfalls represent physical barriers to the dispersal of populations (Figure 1).

### 2. Sampling design

Sampling was conducted at 34 collection points approximately 100 m from the main river channel based on sweeping at sampling points.



**Figure 1.** Study area map and Muzambinho river sections characteristics. A. A hydrographic map of the Muzambinho River basin. B. An altimetric map of the Muzambinho River basin. C. A map showing the vegetation cover of the Muzambinho River basin. Triangles, squares, and circles denote collection points in the Alto Muzambinho, Baixo Muzambinho, and São Domingos sections, respectively. The nMDS graphic shows the isolation between river section communities, mainly the São Domingos (yellow) from Upper Muzambinho (red) and Lower Muzambinho (green).

We applied a combination of active and passive capture methods to obtain samples that were as representative as possible of the total ichthyofauna present in each area sampled. Sampling was performed using manually deployed nets, trawls, and gillnets. Each sampling point was georeferenced, and the collection team assessed the corresponding environmental characteristics, such as water speed, type of bottom substrate, and state of preservation of the riparian forests by visual estimation. The sampling team consisted of four members, totaling approximately 384 hours of total sampling effort (about 2.5 hours of collection effort per sampled point). All three field trips were carried out in the rainy summer period.

Small and medium-sized fish (up to 15 cm in length) were fixed and preserved in 95% ethanol. Larger fish (over 15 cm) were fixed in 10% formaldehyde and transferred to 70% ethanol. Whenever feasible, collected specimens were identified to the lowest possible taxonomic level with the aid of identification keys and the assistance of specialists in the field. All specimens have been deposited in the fish collection of the Botucatu Fish Biology and Genetics Laboratory (LBP).

### 3. Similarity analysis

To visualize differences between river sections' faunal compositions, we performed an similarity analysis—"Multidimensional Scaling (nMDS)" in the

PAST 3 software (Hammer *et al.* 2001). In this analysis, we used the Bray-Curtis similarity index, and for the other parameters, we used the Default.

## Results

Different types of environments characterized each sampled section. The bottom substrates varied considerably from sandy to muddy, sandy-muddy, gravel, pebbles, rocks, and slabs. The preservation status of riparian vegetation ranged from well-preserved stretches to those that had been substantially degraded and converted to pastures, plantations, or urban areas. We collected 4,101 individual fish belonging to six orders, 24 genera, and 37 species (Table 1). The species were not homogeneously distributed along the sampled sections, with some showing a notably restricted distribution within the basin (Table 1). The similarity analysis of nMDS reinforces this hypothesis showing that the fish communities of the sections are distinct. Especially when we compare the São Domingos community to the other two studied, this result suggests that waterfalls, especially Kita, can act as a barrier to the dispersion of species. (Figure 1)

Section LM was found to be characterized by the highest species richness (27 species), among which only 11 species were also found in other sections. The lowest richness (15 species) was recorded in section UM, with four species being characterized as exclusive, whereas 17 species were collected from section SD, of which five were section specific (see Table 1).

**Table 1.** List of species collected in the demarcated sections of the Muzambinho River. (UM) Upper Muzambinho section; (LM) Lower Muzambinho section, (SD) São Domingos stream. The first number in each cell represents the total of individuals sampled in each section and the value in parentheses indicates the number of sample points at which the species was captured.

Order/Family	Species ID	UM	LM	SD
Characiformes				
Characidae				
	<i>Astyanax altiparanae</i> Garutti & Britski, 2000		1 (1)	
	<i>Psalidodon fasciatus</i> (Cuvier, 1819)		21 (5)	
	<i>Psalidodon paranae</i> (Eigenmann, 1914)	22( 5)	25 (5)	46 (4)
	<i>Bryconamericus stramineus</i> Eigenmann, 1908		56 (2)	
	<i>Piabina argentea</i> Reinhardt, 1867		54(5)	
Anastomidae				
	<i>Leporinus marcgravii</i> Lütken, 1875		2(1)	
	<i>Leporinus striatus</i> Kner, 1858		5(1)	
Prochilodontidae				
	<i>Prochilodus lineatus</i> (Valenciennes, 1837)		1(1)	
Crenuchidae				
	<i>Characidium</i> aff. <i>zebra</i> Fowler, 1914		45 (5)	
	<i>Characidium</i> aff. <i>gomesi</i> Travassos, 1956	8(3)	36 (2)	96 (19)
	<i>Characidium gomesi</i> Travassos, 1956			12(1)
Erythrinidae				
	<i>Hoplias malabaricus</i> (Bloch, 1794)	13(3)	7(4)	19(6)
Gymnotiformes				
Gymnotidae				
	<i>Gymnotus</i> sp.	32(3)	1(1)	3(2)
Sternopygidae				
	<i>Eigenmannia</i> sp.		2(2)	

## Discussion

The Muzambinho River is home of at least 37 species of fish, which corresponds to 11% of the total species inhabiting the Alto Paraná, within an area less than 0.04% of the total (Langeani et al. 2007). Compared with other rivers of similar size in this region, such as the Sapucaí River inhabited by 24 species (Azevedo-santos *et al.* 2019), the Muzambinho River could be considered relatively rich in species. The ichthyofauna of the Muzambinho River brings together species such as *Neoplecostomus langeanii* and *Cetopsorhamdia* sp., which appear to be exclusively distributed in this basin, indicating that it may be a region of high endemism, a characteristic of brook rivers (Richardson 2019).

Some of our observations in the present study have raised concerns regarding the preservation of this biodiversity. For example, we recorded the occurrence of alien species introduced in the region, such as *Synbranchus marmoratus* and *Gymnotus* sp., whereas residents in this region have indicated that other nonnative species, such as *Oreochromis niloticus* and *Cyprinus carpio*, were not sampled in the study. In addition, at certain sampling points, we noted the disproportionate prevalence of bioindicator species such as *Poecilia* sp. and *Phalloceros harpagos*, which tend to proliferate in disturbed environments (Vieira et al. 2007). Collectively, these observations may serve to indicate that the degradation of habitats and the introduction of alien species in this region are contributing to a loss of diversity and local extinction (Reis et al. 2016).

This scenario is of particular concern if the populations in question are endemic and isolated, as thus, there may be little or no opportunity to restore populations in the event of local extinction (Bizerril 1998, Richardson 2019). Our findings showing that migratory and widely distributed species, such as *Leporinus* spp., *Iheringichthys labrosus*, *Hoplosternum littorale*, *Psalidodon fasciatus*, and *Astyanax altiparanae* (Langeani et al. 2007), do not occur in the UM and SD regions tend to indicate that the two aforementioned waterfalls delimit these sections, effectively isolating these stretches from section LM (Figure 1), and acting as barriers to species dispersal. The nMDS analysis confirms segregation, at least the separation of SD from the other two sections (Figure 1). Further evidence of the efficacy of these waterfalls as physical barriers is the presence of several exclusive species (11 in total) in the stretches of river upstream of these barriers.

*Neoplecostomus langeanii*, a highly rheophilic species (Bressman et al. 2020; Menezes et al. 2007), reinforces the assumed segregational role played by the Kita and Usina waterfalls as barriers to dispersal. It is predicted that constant gene flow between populations would promote population homogeneity (Frankham et al. 2004). However, *N. langeanii* has undergone population segregation for thousands of years (Roxo et al. 2012). These data are even more evident when considering that this is a highly rheophilic species and might be one of the species most adept at negotiating barriers of this type.

Modern preservationist approaches tend to be based on the maintenance of the most significant possible number of strains, regardless of taxonomic rank (Frankham et al. 2004). Accordingly, given that the stretches of river surveyed in the present study appear to be characterized by a relatively distinctive ichthyofauna, they should ideally be preserved independently; local extinctions could represent the total extinction of distinct lineages.

## Supplementary Material

The following online material is available for this article:

**Table S1.** Species per sampled point.

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## Authors' Contributions

Paulo Roberto Monteiro de Brito: Contribution to data analysis and interpretation, contribution to manuscript preparation, and contribution to critical revision, adding intellectual content.

Luiz Henrique Garcia Pereira: Contribution to manuscript preparation, substantial contribution in the concept and design of the study contribution to data collection.

Fábio Fernandes Roxo: Contribution to manuscript preparation, substantial contribution in the concept and design of the study contribution to data collection.

Jefferson Henriques Monteiro: Contribution to manuscript preparation, substantial contribution in the concept and design of the study contribution to data collection.

Claudio Oliveira: Contribution to manuscript preparation, substantial contribution in the concept and design of the study contribution to data collection.

Guilherme José da Costa Silva: Contribution to data analysis and interpretation, contribution to manuscript preparation, substantial contribution in the concept and design of the study contribution to data collection, and contribution to critical revision, adding intellectual content.

## Conflicts of interest

The authors declares that they have no conflict of interest related to the publication of this manuscript.

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