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DISSERTAÇÃO

**BANCOS DE MACRÓFITAS COMO ÁREAS BERÇÁRIOS PARA PEIXES EM UMA
CASCATA DE PEQUENAS CENTRAIS HIDRELÉTRICAS NA REGIÃO
NEOTROPICAL**

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NEOTROPICAL**

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Palavras-chave: Cascata de reservatórios; Juvenis; Larvas; Migradores; Sedentários.

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Apresentação

No rio Sapucaí – Mirim, localizado no norte do Estado de São Paulo, há uma cascata de reservatórios de Pequenas Centrais Hidrelétricas (PCHs) selecionada para o presente estudo. Esse rio é um importante tributário do rio Grande (bacia do alto rio Paraná) e apresenta uma diversificada fauna de peixes, com cerca de 105 espécies já registradas. No trecho estudado há a ocorrência de fecundação e desova de peixes, comprovada através da presença de ovos e larvas em deriva (estágios embrionários iniciais – larval vitelínico e pré-flexão). Apesar dessa informação, existe uma lacuna de conhecimento sobre aonde estão ocorrendo os processos de desenvolvimento inicial da ictiofauna ao longo da cascata de reservatório, devido a inexistência de habitats laterais típicos que poderiam desempenhar a função de berçário, como lagoas marginais e desembocadura de tributários. A partir desta premissa, considera-se que os bancos de macrófitas são áreas de berçários potenciais para os peixes, devido suas características ecológicas favoráveis ao desenvolvimento de larvas e juvenis.

O trabalho foi redigido em formato de artigo científico, o qual será submetido posteriormente para publicação. O objetivo principal da dissertação foi avaliar o papel funcional dos bancos de macrófitas como áreas- berçário para peixes, através da sua associação com os primeiros estágios de vida dos peixes – larvas e juvenis.

Resumo

Barramentos de Pequenas Centrais Hidrelétricas (PCHs) modificam a estrutura física e o funcionamento do rio e, conseqüentemente, a biota aquática. Para a fauna de peixes, uma das características mais afetadas é o processo reprodutivo. O Rio Sapucaí-Mirim (São Paulo, Brasil) possui três Pequenas Centrais Hidrelétricas (PCHs) construídas em seu canal principal e dispostas em sistema em cascata. Este trecho do rio não possui habitats típicos para o desenvolvimento inicial da ictiofauna, como desembocaduras de tributários e lagoas marginais, assim, o estudo visou avaliar o papel dos bancos de macrófitas como áreas berçário para larvas e juvenis de peixes. A variação espacial e temporal desses estágios de peixes junto as macrófitas foi investigada. Para a amostragem dos peixes, utilizou-se uma peneira para coleta de larvas e juvenis pequenos e redes de espera para a captura de juvenis maiores e com maior capacidade de mobilidade. Simultaneamente, apenas adjacente aos bancos de macrófitas, os parâmetros limnológicos básicos foram medidos com uma sonda de água Horiba U-52. Trinta táxons, cerca de 30% das espécies de ictiofauna já inventariadas no Rio Sapucaí-Mirim, utilizam os bancos de macrófitas como área inicial de desenvolvimento. Apenas duas espécies não nativas ocorreram na área estudada. Foram capturados 88 larvas e 393 juvenis com diferentes estratégias reprodutivas, os não migradores ou migradores de curta distância com fertilização externa e cuidado parental (NEP) e não migradores ou migradores de curta distância com fertilização externa e sem cuidado parental (NEW) foram as estratégias predominantes. Também foi evidenciado estatisticamente que a organização espacial – posicionamento do reservatório na cascata e compartimentalização dos reservatórios internos, bem como as mudanças temporais ao longo do período reprodutivo, influenciaram a estrutura da comunidade de peixes (composição taxonômica, estágio de desenvolvimento, estratégia reprodutiva). Além disso, os bancos de macrófitas são efetivamente utilizados como áreas de berçário para peixes com diferentes estratégias reprodutivas, indicando a importância desses locais para a conservação/manejo da fauna local.

Palavras-chaves: Juvenis, Larvas, Migradores, Cascata de Reservatórios, Sedentários

Macrophyte banks as nursery areas for fish with different reproductive strategies in Neotropical Small Hydropower Reservoirs

Abstract

Damming for construction of Small Hydroelectric Power Plants (SHPs) modify the physical structure and functioning of the river and, consequently, of the aquatic biota. For fish fauna, one of the most affected characteristics is the reproductive process. The Sapucaí-Mirim River (São Paulo, Brazil) has three Small Hydroelectric Power Plants (SHPs) constructed on its main channel and arranged in a cascade system. This river stretch does not have typical habitats for the initial development of ichthyofauna, such as tributary mouths, marginal lagoons, thus, the study aims to evaluate the role of macrophyte banks as nursery areas for larvae and juvenile fish. The spatial and temporal variation of these fish stages with the macrophytes was investigated. For the sampling of fish, a sieve was used to collect larvae and small juveniles and gillnets for the capture larger juveniles with greater mobility capacity. Simultaneously, just adjacent to the macrophyte banks, the basic limnological parameters were measured with a Horiba U-52 water probe. Thirty taxa, about 30% of the fish fauna species already inventoried in the Sapucaí-Mirim River, use the macrophyte banks as an initial development area. Only two non-native species occurred in the studied area. Eighty-eight larvae and 393 juveniles of different reproductive strategies were captured, especially non-migrants or short-distance migrants with external fertilization and parental care (NEP) and non-migrant or short-distance migrant with external fertilization and without parental care (NEW) the predominant strategies. It was also statistically evidenced that the spatial organization – positioning of the reservoir in the cascade and internal reservoirs compartmentalization, as well as temporal changes along the reproductive period, influenced on the fish community structure (taxonomical composition, development stage, reproductive strategy). Therefore, the macrophyte banks are effectively used as nursery areas for fish with different reproductive strategies, indicating the importance of these sites for the conservation/management of local fauna.

Keywords: Juveniles, Larvae, Migrator, Reservoir Cascade, Sedentary

Introduction

In recent times, there has been a strong tendency to build hydroelectric plants, mainly small ones (SHPs), all over the world (Zarfl *et al.*, 2015), and in Brazil, particularly, there are a series of incentives and new regulations that contribute to the increase in the construction of this type of enterprise (Couto & Olden, 2018). According to ANEEL (2022), there are 430 Small Hydropower Plants (SHPs) in operation in Brazil, 29 under construction and another 81 planned. One of the reasons for the increase in the construction of SHPs around the world is due to the attempt to avoid the impacts caused by the construction of large dams. The scientific information about the negative environmental impacts resulting from the construction of Hydroelectric Power Plants (HPPs) is vast, but in case of SHPs information are still scarce (Kibler & Tullos, 2013; Ruocco *et al.*, 2019).

Although there are few studies that evaluate the impacts of SHPs, the ecological effects cannot be neglected. For large river systems, the hydrological and limnological changes of the original fluvial conditions, including the physical compartmentalization are noticeable and have a great influence on the restructuring of the aquatic community (Agostinho *et al.*, 2008; Nogueira *et al.*, 2008, 2010). Furthermore, HPPs and SHPs arranged in cascade systems can have cumulative effects, which can be even higher than the ones of an individual HPP (Kibler & Tullos, 2013; Nogueira & Pomari, 2019).

In SHPs reservoirs it may occur reduction in fish abundance, average length, total weight and condition factor (Benejam *et al.*, 2016). It has also been observed a reduction of populations of species with reproductive migration (Arcifa *et al.*, 2012; Bakken *et al.*, 2012); interruption of their free movement along the river (Bakken *et al.*, 2012; Kucukali, 2014) and alteration in ichthyoplankton dispersal processes (Suzuki *et al.*, 2011; Pelicice *et al.*, 2014; Brambilla *et al.*, 2020). Additionally, in case of cascade systems, the impacts may differ depending on the particular reservoir positioning, as already seen for large reservoirs (Ferrareze *et al.*, 2014; Loures & Pompeu, 2018; Pelicice *et al.*, 2018; Nobile *et al.*, 2019; Ganassin *et al.*, 2021).

Besides the direct impacts caused by the construction of SHPs, the seasonal variation of water flow in the reservoirs must be taken into account too, as they modify limnological

conditions and the fish community organization (Baumgartner *et al.*, 2018), especially with regard to different reproductive strategies (Vasconcelos *et al.*, 2014). Long-distance migratory species are the most affected by river regulation by dams. The main trigger for their reproduction is the increase in the water flow and speed (Vazzoler, 1982; Sanches *et al.*, 2006; Agostinho *et al.*, 2008), which modifies abiotic factors, increasing the availability of resources and decreasing the predatory activity (Sanches *et al.*, 2006; Gogola *et al.*, 2010; Suzuki & Pompeu, 2016). In these high-water periods, the reproductive activity is intense and, consequently, a higher drift rate of larvae, which reach the nursery areas faster, increasing the offspring survival (Lechner *et al.*, 2016).

In the Neotropics, lateral habitats associated with rivers and reservoirs, such as marginal lagoons and entrance of tributaries, are extremely important for the reproduction and initial development of fish fauna (Vianna & Nogueira, 2008; Ferrareze & Nogueira, 2011; Ávila-Simas *et al.*, 2014; Silva *et al.*, 2015, 2019; Agostinho *et al.*, 2016). Mainly in marginal lagoons, larvae and juveniles of fish find refuge from predators and essential resources for survival and ending of the early stages of development (Orsi *et al.*, 2016). However, due to the small size of SHP reservoirs and low spatial complexity, there is usually a simplification of their lateral habitats and consequently the influence of these lateral environments on the structure and ecological functioning of the system. Thus, the association with other kind of habitats or microhabitats, for example the macrophyte banks, may be essential for the offspring development.

Macrophyte banks, when not excessive in coverage and biomass (Pompêo, 2017), increase the complexity of the habitat contributing to the maintenance of fish diversity (Agostinho *et al.*, 2003, 2007; Pelicice *et al.*, 2005; Dibble & Pelicice, 2010; Hermes-Silva & Zaniboni-Filho, 2012). These aquatic plants provide a high availability of shelters from predators, substrate for the development of several organisms, especially macroinvertebrates, used in the feeding of most species of fish (small size and initial development stages), besides to serving as spawning substrate for certain species of fish (Agostinho *et al.*, 2003; Ferrareze & Nogueira, 2011).

Within this context, three SHPs arranged in a cascade system in the Sapucaí-Mirim River (SP) were selected for the study. This river is an important tributary of the Grande River (upper Paraná River basin) and has a diverse fish fauna, with at least 105 species already recorded (Freitas-Souza *et al.*, 2016; Oliveira *et al.*, 2016; Brambilla *et al.*, 2018; Diniz *et al.*, 2019). In the considered river stretch there is the occurrence of successful spawning and

fecundation events, reported from the analysis of drifting of eggs and larvae (ichthyoplankton samples) (Brambilla *et al.*, 2020). Nevertheless, there is a lack of knowledge about where the processes of initial development of the different species of fish along the cascade of reservoirs occurs. Moreover, there is no typical lateral environments that could play the role of natural nurseries, such as marginal lagoons and tributary mouths, in the river stretch occupied by the cascade of reservoirs.

Based on the premise that littoral habitats play a fundamental role in the survival of the offspring, the objective of the study was to evaluate the importance of lateral macrophyte banks as nursery areas for fish in the SHPs reservoirs of Sapucaí-Mirim River (preliminary observations by Kimura *et al.*, 2021). The association between the first stages of fish development (larvae and juveniles) with the littoral plants in the cascade of reservoirs was evaluated. The main hypothesis is that macrophyte banks are habitats used for the early stages of fish development. Variation in terms of fish community structure and reproductive strategies was also expected, both, spatially, among the three reservoirs (positioning in the cascade) and within each reservoir (internal compartments), as well as temporally, along the reproductive period.

Material and Methods

Study area

The Sapucaí-Mirim River springs are located in the Atlantic Plateau (Serra da Mantiqueira), between the states of São Paulo and Minas Gerais. The river mouth is located in the left margin of Porto Colômbia Reservoir, middle stretch of Grande River (Paiva, 1982). Sapucaí-Mirim is a plateau river, with several rapids and small waterfalls in the upper section, becoming less turbulent with decreased water velocity in the lower section. Another distinctive characteristic of this river is the presence of several knickzones mesohabitats, bed rock (basaltic) platforms popularly known as “pedrais” (Brambilla *et al.*, 2018; Ruocco *et al.*, 2019). The river total length is 290 km, with its course entirely in the State of São Paulo (Paiva, 1982).

In the main channel of the Sapucaí-Mirim River it was constructed three SHPs arranged in a reservoir cascade system, Palmeiras, Anhanguera and Retiro, which started to operate in 2011, 2009 and 2013, respectively. The reservoirs have different areas and lengths, Palmeiras

has 2.6 km² and 7.5 km, Anhanguera has 1.8 km² and 5 km and Retiro has 2.8 km² and 11 km. Despite these differences, they are all considered small reservoirs and run-of-river systems, with an average of just three days of water retention time. Due to these conditions, these reservoirs respond immediately to rainfall events, instantly reflected in the outflow (Brambilla *et al.*, 2020). The three reservoirs have the same outflow characteristics, represented in Figure 1, whose data were obtained on the system's website of the *Sistema Integrado de Bacias Hidrográficas – Departamento de Água e Energia Elétrica de São Paulo*.

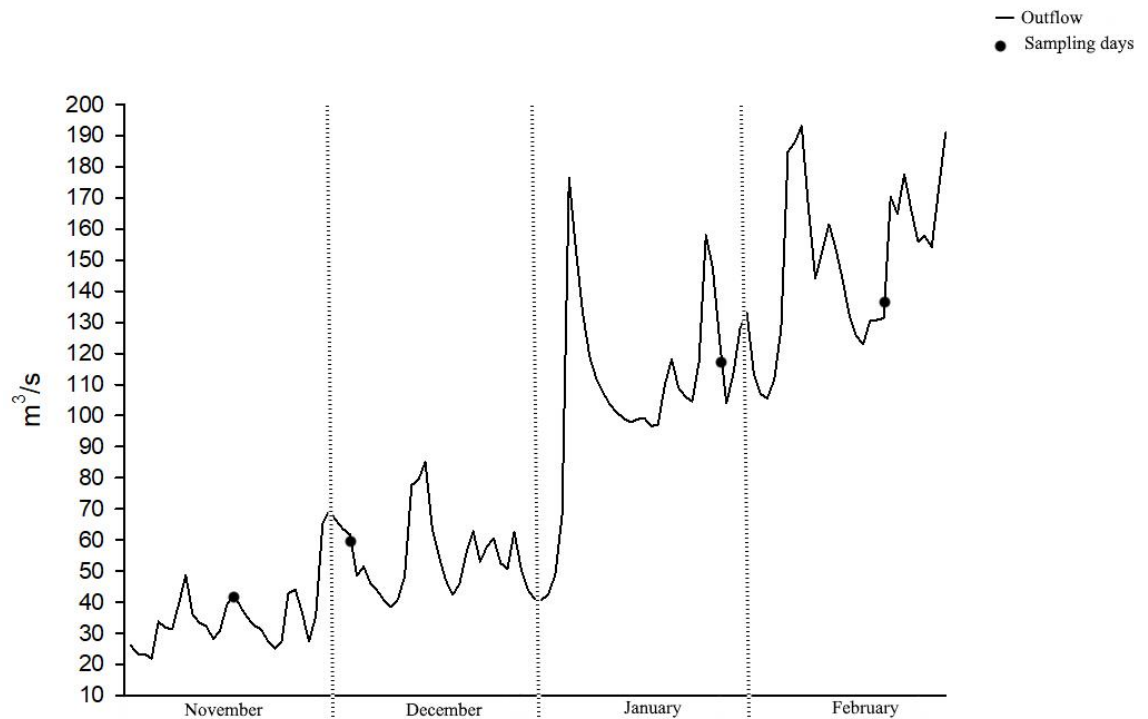


Figure 1. Outflow variation during the sampling period in a reservoir cascade on Sapucaí - Mirim river (SP, Brazil). Dots correspond to the sampling days

For this study, the intermediate and lentic compartments of the three reservoirs were selected for sampling (Figure 2). In the upper stretches, spawning activities predominate, rather than initial development (Brambilla *et al.*, 2020), and macrophyte banks are scarce because of the intense flow conditions.

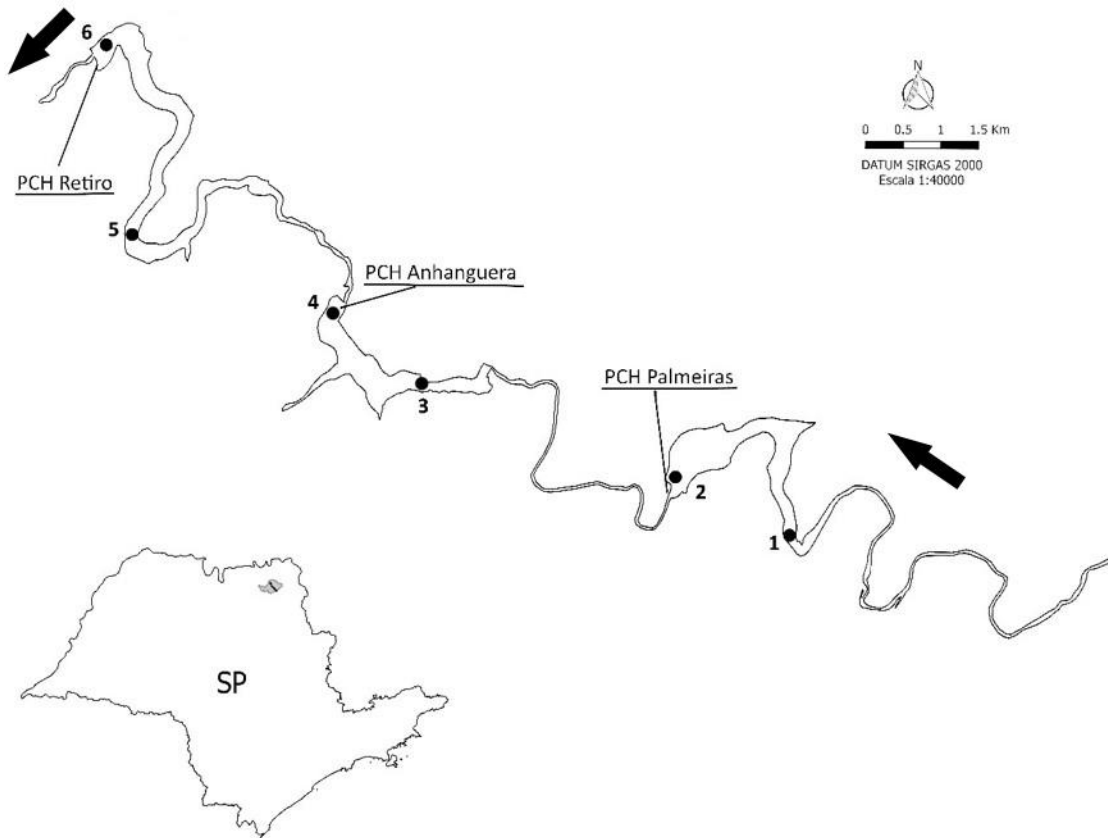


Figure 2. Map of the Sapucaí-Mirim River (SP, Brazil) stretch, where are located the studied Small Hydropower Plants (SHPs) reservoirs – Palmeiras, Anhanguera and Retiro, with indication of the sampling sites: 1 – Intermediate Palmeiras; 2 – Lentic Palmeiras; 3 – Intermediate Anhanguera; 4 – Lentic Anhanguera; 5 – Intermediate Retiro and 6 Lentic Retiro. Dots correspond to the sampling sites and arrows the river course direction.

Sampling

Samplings were performed monthly, in November and December 2019, January and February 2020. Sampling should also have included March 2020, however, due to the pandemic situation (COVID-19), it was not possible to collect in that month. The period corresponds to the reproduction season of most fish species in upper Paraná River basin (Vazzoler 1982). In each intermediate and lentic compartment of Palmeiras, Anhanguera and Retiro reservoirs (Figure 1), three distinct macrophyte banks (nine per reservoir) were sampled in triplicates (Table 1), resulting in a total of 18 samples per month. The macrophyte banks were randomly chosen, within each compartment, considering the recurrent/dominant species determined through visual inspection (Table 1). It is important to mention that submerged macrophytes do not occur in these reservoirs, probably due to the excessive water turbidity mainly observed in the summer-rainy season.

Table 1. The sampling points geographic coordinates for Palmeiras, Anhanguera and Retiro reservoirs, Sapucaí-Mirim River (SP, Brazil) and the main macrophytes observed during the fieldwork.

	PALMEIRAS		ANHANGUERA		RETIRO	
Sampling Area	Main aquatic macrophytes	Geographic coordinates	Main aquatic macrophytes	Geographic coordinates	Main aquatic macrophytes	Geographic coordinates
Intermediate 1	<i>Eichhornia crassipes</i> , <i>Salvinia</i> spp.	20°33'50.2 "S 47°47'20.9 "W	<i>Salvinia</i> spp., <i>Eichhornia</i> spp.	20°30'51.7 "S 47°50'58.4 "W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°28'04.7 "S 47°52'06.5 "W
Intermediate 2	<i>Eichhornia crassipes</i> , <i>Salvinia</i> spp.	20°33'51.8" S 47°47'22.9" W	<i>Salvinia</i> spp., <i>Eichhornia</i> spp., Ciperaceae, <i>Pistia</i> spp.	20°30'52.0" S 47°50'57.7" W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°28'05.9" S 47°52'05.6" W
Intermediate 3	<i>Eichhornia crassipes</i>	20°33'52.4" S 47°47'23.9" W	<i>Salvinia</i> spp., <i>Eichhornia</i> spp., Ciperaceae	20°30'53.8" S 47°50'52.5" W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i> , <i>Pistia stratiotes</i>	20°28'08.7" S 47°52'03.7" W
Lentic 1	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°32'35.0" S 47°48'10.0" W	<i>Eichhornia</i> spp., Ciperaceae	20°29'41.3" S 47°51'52.7" W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i> , <i>Pistia stratiotes</i>	20°26'08.4" S 47°52'44.0" W
Lentic 2	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°32'35.6" S 47°48'09.4" W	<i>Eichhornia</i> spp.	20°29'42.5" S 47°51'53.4" W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°26'12.6" S 47°52'45.9" W
Lentic 3	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i>	20°32'36.1" S 47°48'08.3" W	<i>Eichhornia</i> spp.	20°29'44.8" S 47°51'54.8" W	<i>Salvinia</i> spp., <i>Eichhornia crassipes</i> , <i>Oxycaryum cubense</i>	20°26'16.1" S 47°52'46.7" W

For fish collection a sieve of 0.287 m² area with a 1 mm mesh and dimensions of 0.7 x 0.4 m was used, which was manually introduced, bellow the marginal aquatic vegetation and quickly raised at three different points of the same macrophyte bank. Samplings were carried out in the afternoon period. The sieve technique is mainly directed for collection of both larvae and juvenile. It was assumed that most fish larvae with exogenous feeding, therefore with active

swimming (not in passive drift), should be captured with a 1 mm mesh size. Sampling was carried out from the forward part of the boat (bow), which was moving (engine propulsion) frontally towards the aquatic plants, without traversing spaces, all the effort being standardized. In addition, one gillnet of 1.5 mm mesh (2 m high and 10 m long) was installed next to each selected bank. It remained exposed for about 3 hours between dusk and early evening. The gillnet technique was also employed aiming to collect larger juveniles with higher mobility capacity. The collected material was stored in plastic bags and eugenol solution was immediately added to euthanize the organisms (Vidal *et al.*, 2008). Subsequently, the samples were fixed in 4% formalin solution with calcium carbonate for preservation of the bone structures. Sampling was performed under the license SISBIO 13.794-1 (M. G. Nogueira). In February the gillnet sampling was not performed due to logistic problems. Simultaneously to the fish samplings, the following environmental variables were measured adjacent each macrophyte bank, in the water sub-surface: temperature, pH, electrical conductivity (K), turbidity, dissolved oxygen (DO), oxide-reduction potential (ORP) and TDS, with a previously calibrated Horiba U-52 multiparameter probe.

Sorting and identification

Larger fishes were immediately separated for posterior identification. The detailed sorting for larvae and early juveniles was made in laboratory using a Bogorov acrylic plate under stereomicroscope ZEISS (STEMI SV 6), and, for juveniles, through a visual inspection using a transparent plastic tray with illumination from below. The captured individuals were classified into two stages (larval and juvenile) and identified to the lowest taxonomic level possible. Larvae were identified following Ahlstrom & Moser (1976) and Nakatani *et al.* (2001). Juveniles were identified following specialized literature (Castro *et al.*, 2004; Graça & Pavaneli, 2007; Langeani & Rego, 2014; Ota *et al.*, 2018).

The taxonomic framework followed Fricke *et al.* (2019), the term “larvae” refers to the period of development between the egg hatching and the total formation of the fins (with rays) and scales (depending on the taxonomical group). The “juvenile” term refers to the period between the total development of fin and scales and the sexual maturity (Nakatani *et al.*, 2001). For differentiation between juveniles and adults, it was used the length at first maturity (LS50) (Weyl & Hecht, 1998; Santos & Fontoura, 2000; Teresa & Gonçalves-de-Freitas, 2011;

Langeani & Rego, 2014; Barros *et al.*, 2016), considering the difference between females and males. For individuals between the female/male range, the maturation of the gonads was analyzed. The standard length of fish was obtained, using a caliper (Mitutoyo), snout tip to the end of the caudal fin, except for Gymnotiformes and Synbranchiformes. Individuals with standard length smaller than first maturation size (LS50) described in the literature were classified as juveniles.

After identification, the individuals were classified according to the reproductive strategy into four groups, according to Vasconcelos *et al.* (2014): Long-distance migrants with external fertilization but no parental care (LME), non-migrants or short-distance migrants with external fertilization but no parental care (NEW), non-migrants or short-distance migrants with external fertilization and parental care (NEP) and non-migrants or short-distance migrants with internal fertilization, but without parental care (NIF).

Data Analysis

A principal component analysis (PCA) was performed in PRIMER 6 & PERMANOVA to ordinate the spatial and temporal trends according to the limnological conditions (Temperature, pH, ORP, electrical conductivity, turbidity, dissolved oxygen and TDS). A correlation matrix was used and the data were previously transformed using $\log(x + 1)$ (except pH) and normalized.

For all fish fauna analyses, a standardization of effort per capture for different sampling gears (sieve and gillnet) was performed, with the method of Multigear Mean Standardization (MGMS) (Gibson-Reinemer *et al.*, 2016). To perform the MGMS, it was used sieve data in density and gillnet data in capture per unit of effort (CPUE), ind./m²/3h.

Descriptive analyses of fish fauna, were performed in order to discriminate spatial and temporal tendencies. In addition to the descriptive graphics, a PERMANOVA and a PERMIDISP were performed in PRIMER 6 & PERMANOVA, using the Bray-Curtis dissimilarity, to test the statistical differences of fish composition, among different reservoirs, compartments and months.

A non-metric multidimensional scaling analysis (NMDS), based on abundance and Bray Curtis distance, was performed in the R program using abundance to spatially cluster the fish community. The ggplot2 (Wickham, 2016) and vegan (Oksanen *et al.*, 2020) packages were used for the analysis.

Results

The first two axes of Principal Component Analysis (PCA) (Figure 3) applied to the limnological conditions explained 83.2% of data variability, considering in the first component (58%) and in the second component (25.2%). All variables, except temperature and ORP, showed a high correlation (> 0.4) with at least one of the two first components (Table 2). The values of the limnological variables (mean and standard deviation) measured in the different compartments of the reservoirs, were presented in the complementary material. PCA ordination mainly indicates the temporal variation. In axis one, there was a clear separation of November in relation to the other months, influenced mainly by pH, conductivity and TDS. For axis two there was not a clear separation. The spatial variation was less evident; however, it was possible to observe, mainly for November, that the lentic compartments of Palmeiras and Retiro were separated on the negative side of axis 1 and 2, by pH and DO. The intermediate compartments of Palmeiras and Retiro were correlated with turbidity and ORP, respectively, on the positive side of axis two. The Anhanguera compartments showed no separation.

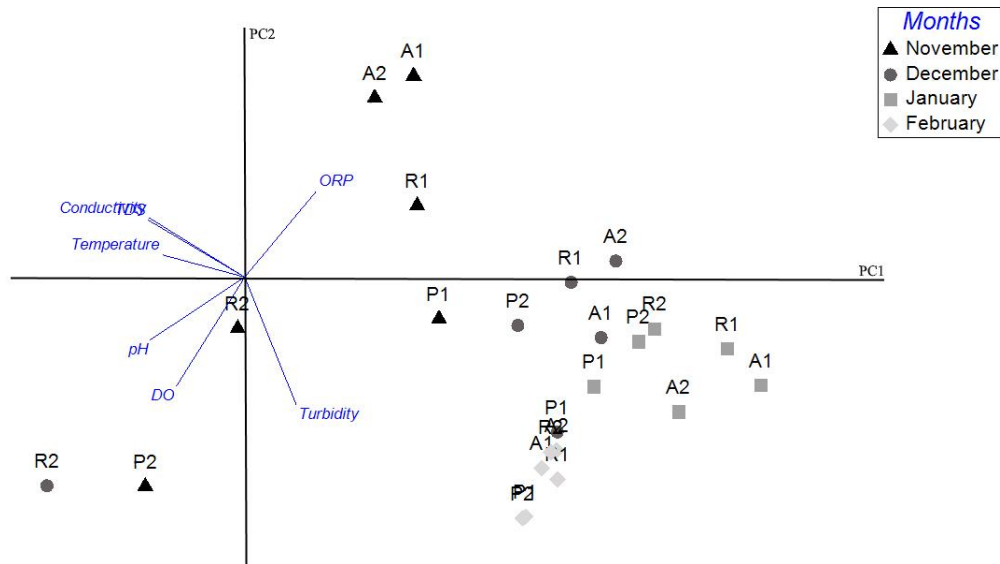


Figure 3. Graphic results of principal component analysis (components 1 and 2), based on the limnological characteristics, adjacent to macrophyte banks, in intermediate and lentic compartments of Palmeiras, Anhanguera and Retiro SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil). P1: Palmeiras Intermediate, P2: Palmeiras Lentic, A1: Anhanguera Intermediate, A2: Anhanguera Lentic, R1: Retiro Intermediate, R2: Retiro Lentic.

Table 2. Correlation values (scores) among limnological variables, adjacent to macrophyte banks, and components 1 and 2 of principal component analysis. Data from intermediate and lentic compartments of Palmeiras, Anhanguera and Retiro SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil). Higher values in bold.

Variable	PC1	PC2
Temperature	-0.379	0.070
pH	-0.424	-0.324
ORP	0.341	0.386
Conductivity	-0.444	0.264
Turbidity	0.290	-0.557
DO	-0.280	-0.542
TDS	-0.449	0.259

A total of 481 individuals were collected in the macrophyte banks of the three reservoirs along the entire study. Eighty eight larvae and 313 juveniles were collected with a sieve and 80 juveniles were collected with a gillnet. Thirty taxa were identified, including 24 at species level,

five at genera level and only one individual at the family level (Table 3). Limitation in identification was due to the early development stage of some sampled specimens.

In Palmeiras the richness was represented by 24 taxa, with 13 taxa found only in the intermediate compartment, five only in the lentic and six in both compartments (Figure 4, Table 3). In Anhanguera the richness was represented by 18 taxa, with five taxa found only in the intermediate compartment, four only in the lentic, and nine in both compartments (Figure 4, Table 3). In Retiro the richness was represented by 22 taxa, with four taxa found only in the intermediate compartment, eight only in the lentic and ten in both compartments (Figure 4, Table 3). Comparing the fish composition of each reservoir, we found 11 taxa present in all reservoirs and nine taxa were exclusive, with five taxa unique to Palmeiras (*Astyanax lacustris*, *Apareiodon affinis*, *Apareiodon* spp., *Coptodon rendalli*, and *Hoplosternum littorale*), two unique to Anhanguera (Cichlidae and *Rhamdia quelen*), and two unique to Retiro (*Eigenmannia trilineata* and *Pimelodus maculatus*) (Table 3).

PERMANOVA and PERMDISP indicated that for the composition of the fish fauna associated to the macrophyte banks there was a statistical difference among reservoirs, between reservoirs compartments and among months and for the interactions between reservoir/compartment and reservoir/months (Table 4).

Table 3. Taxonomic list of fish fauna associated to macrophyte banks in the distinct compartments of Palmeiras, Anhanguera and Retiro SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil), their respective reproductive strategies, development stages and length.

TAXA	INTERMEDIATE	LENTIC	REPRODUCTIVE STRATEGIES	DEVELOPMENT STAGES	LENGHT (cm)	
					Min.	Max.
Characiformes						
Anostomidae						
<i>Leporinus friderici</i> (Bloch 1794)	Palm	Ret	NEW	Juveniles	2.45	15.9
<i>Megaleporinus obtusidens</i> (Valenciennes 1837)	Palm	Anh/Ret	LME	Juveniles	2.21	16
<i>Schizodon nasutus</i> Kner 1858	Palm/Anh/Ret	Palm/Anh/Ret	NEW	Larvae/ Juveniles	2.58	13.67
Characidae						
<i>Astyanax lacustris</i> (Lütken 1875)	Palm		NEW	Juvenile	3.28 ¹	-
<i>Galeocharax gulo</i> (Cope 1870)	Palm/Anh/Ret	Palm/Ret	NEW	Juveniles	6.06	14
<i>Hyphessobrycon eques</i> (Steindachner 1882)	Anh/Ret	Ret	NEW	Larvae/ Juveniles	1.32	1.59
<i>Knodus moenkhausii</i> (Eigenmann & Kennedy 1903)	Palm/Anh	-	NEW	Juveniles	1.44	1.49
<i>Psalidodon bockmanni</i> (Vari & Castro 2007)	Palm	Ret	NEW	Juveniles	1.79	3.6
Erythrinidae						
<i>Hoplias malabaricus</i> (Bloch 1794)	Palm/Anh	Palm/Anh/Ret	NEP	Larvae/ Juveniles	2.9	5.69
Parodontidae						
<i>Apareiodon affinis</i> (Steindachner 1879)	Palm	-	NEW	Larvae	-	-
<i>Apareiodon</i> spp.	Palm	-	NEW	Juveniles	2.12	2.52
Serrasalminidae						
<i>Serrasalmus maculatus</i> Kner 1858	Palm/Anh/Ret	Palm/Anh/Ret	NEP	Larvae/ Juveniles	1.31	6.92
Cichliformes						
Cichlidae						
<i>Cichla kelberi</i> Kullander & Ferreira 2006 *	Ret	Palm/Ret	NEP	Juveniles	2.57	8.72
<i>Cichlasoma paranaense</i> Kullander 1983	Palm/Anh/Ret	Anh/Ret	NEP	Juveniles	1.45	4.49
Cichlidae	-	Anha	NEP	Larvae	-	-
<i>Coptodon rendalli</i> (Boulenger 1897) *	-	Palm	NEP	Juveniles	2.19	5.89
<i>Crenicichla britskii</i> Kullander 1982	Anh/Ret	Palm/Anh/Ret	NEP	Juveniles	2.5	6.53
<i>Crenicichla</i> sp.	Ret	Palm	NEP	Juveniles	2.34	1.94
<i>Geophagus brasiliensis</i> (Quoy & Gaimard 1824)	Palm	Anh/Ret	NEP	Juveniles	1.55	4.15
<i>Laetacara araguaiae</i> Ottoni & Costa 2009	-	Palm/Ret	NEP	Juveniles	2.36	3.85
Gymnotiformes						
Gymnotidae						
<i>Gymnotus sylvius</i> Albert & Fernandes-Matioli 1999	Palm/Anh/Ret	Palm/Anh/Ret	NEP	Juveniles	2.01	18.1
<i>Gymnotus</i> sp.	Palm/Anh/Ret	-	NEP	Larvae/Juvenile	1.91 ¹	-
Sternopygidae						

<i>Eigenmannia trilineata</i> López & Castello 1966	Ret	-	NEW	Juvenile	8.57 ¹	-
<i>Eigenmannia</i> sp.	Palm/Anh/Ret	-	NEW	Larvae	-	-
Siluriformes						
Auchenipteridae						
<i>Tatia neivai</i> (Ihering 1930)	Palm/Anh/Ret	Anh/Ret	NIF	Larvae	-	-
Callichthyidae						
<i>Hoplosternum littorale</i> (Hancock 1828)	Palm	-	NEP	Juvenile	1.45 ¹	-
Heptapteridae						
<i>Rhamdia quelen</i> (Quoy & Gaimard 1824)	-	Anh	NEW	Larvae	-	-
Loricariidae						
<i>Hypostomus</i> spp.	Anh	Anh/Ret	NEP	Juveniles	1.36	3.34
Pimelodidae						
<i>Pimelodus maculatus</i> Lacepède 1803	-	Ret	LME	Juvenile	9.66 ¹	-
Synbranchiformes						
Synbranchidae						
<i>Synbranchus marmoratus</i> Bloch 1795	Palm/Anh/Ret	Palm/Anh/Ret	NEP	Larvae/Juveniles	2.6	28.9

*: Non-native species, Palm: Palmeiras, Anh: Anhanguera, Ret: Retiro, LME: Long-distance migrants with external fertilization but no parental care, NEW: Non-migrants or short-distance migrants with external fertilization but no parental care, NEP: Non-migrants or short-distance migrants with external fertilization and parental care, NIF: Non-migrants or short-distance migrants with internal fertilization, but without parental care, ¹: unique individual.

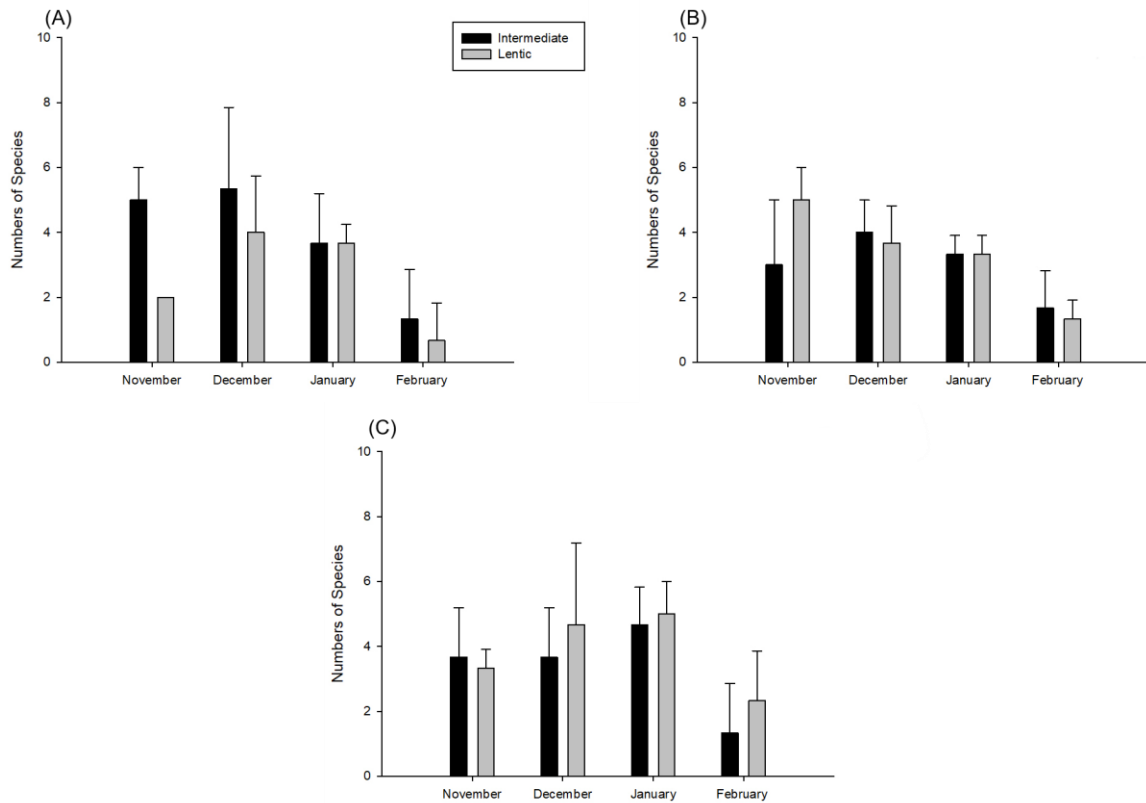


Figure 4. Spatial and temporal variation of richness values (mean and standard deviation) for the fish fauna associated to macrophyte banks in Palmeiras (A), Anhanguera (B) and Retiro (C) SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil).

For the relative abundance per developmental stage, in Palmeiras the abundance of larvae was higher in the intermediate compartment in all months and for juveniles, with the exception of November, higher values were found in the lentic compartment (Figure 5). In Anhanguera, larvae were more abundant, except in November, in the intermediate compartment, and juveniles in the lentic compartment in all months (Figure 5). For Retiro, the larvae showed higher abundance in the intermediate compartment in the months of December and January and in the other months higher values were found in the lentic compartment, for juveniles higher values were found in the lentic compartment for all months (Figure 5). PERMANOVA and PERMDISP showed that there were statistical differences among reservoirs and months, for the interactions between reservoir/compartment and reservoir/months (Table 4).

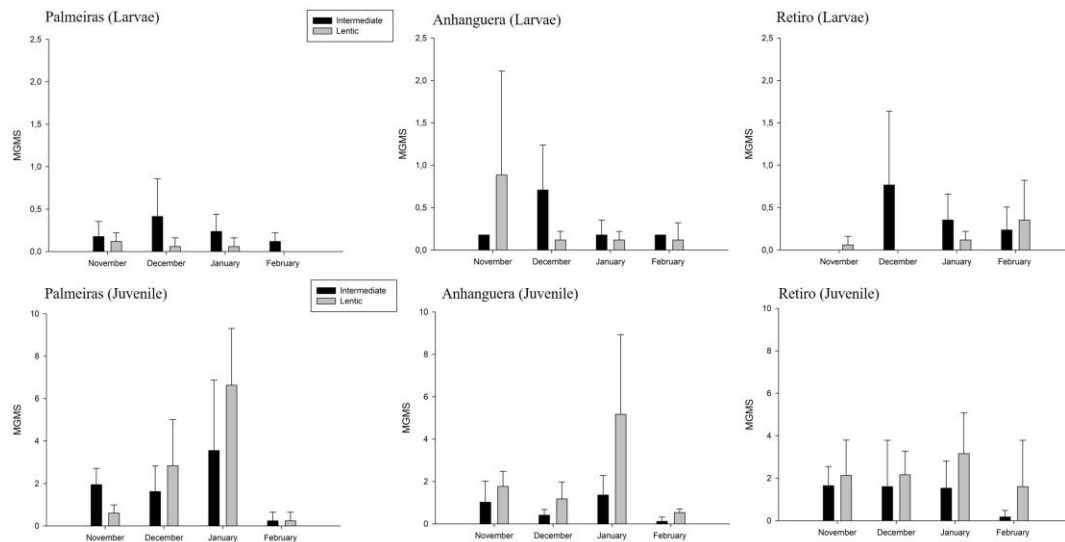


Figure 5. Spatial and temporal variation of abundance (MGMS) per development stage for the fish fauna associated to macrophyte banks in the distinct compartments and sampling months of Palmeiras , Anhanguera and Retiro SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil).

Data on the relative abundance per reproductive strategy indicated that in Palmeiras predominated non-migrants or short-distance migrants with external fertilization but no parental care (NEW) (Figure 6 A). In Anhanguera, the predominant strategy was non-migrants or short-distance migrants with external fertilization and parental care (NEP) (Figure 6 B). For Retiro there was a predominance of the NEP and NEW strategies, respectively (Figure 6 C). Long-distance migrants with external fertilization but no parental care (LME) was only found in January across the entire cascade. PERMANOVA and PERMDISP indicated that there were differences among the months and for interactions between reservoir/compartiment (Table 4).

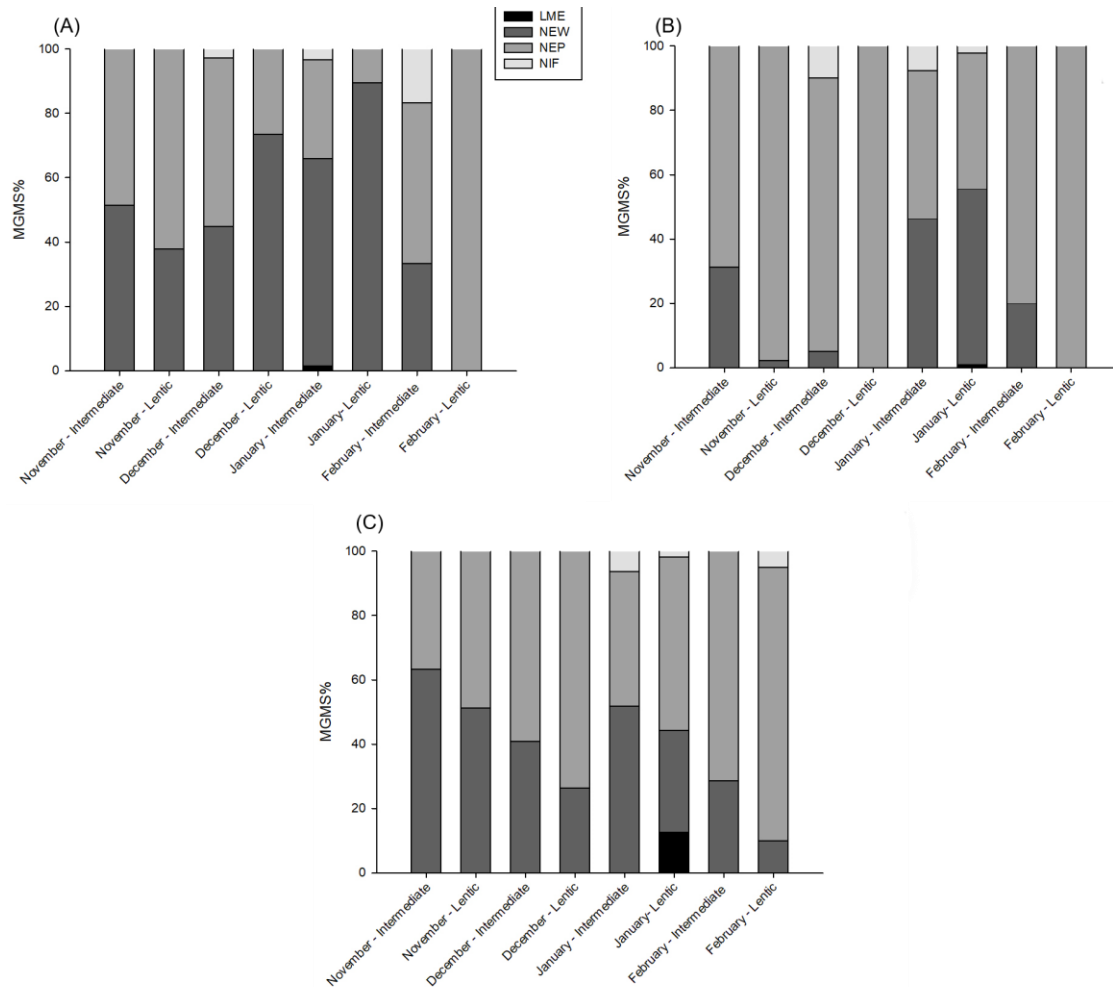


Figure 6. Relative abundance values of each reproductive strategy of the fish fauna associated to macrophyte banks in the distinct compartments of Palmeiras (A), Anhanguera (B) and Retiro (C) SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil). LME: Long-distance migrants with external fertilization but no parental care, NEW: Non-migrants or short-distance migrants with external fertilization but no parental care, NEP: Non-migrants or short-distance migrants with external fertilization and parental care, NIF: Non-migrants or short-distance migrants with internal fertilization, but without parental care.

The NMDS analysis, for the abundance, showed a stress value of 0.171209, indicating that Palmeiras presents the higher dissimilarity in relation to the other reservoirs, while Retiro is the one that most similar the other reservoirs (Figure 7). Furthermore, the analysis PERMANOVA and PERMDISP, for the abundance, indicated that there were statistical differences among reservoirs and between compartments and for the interactions between reservoir/compartment and reservoir/months.

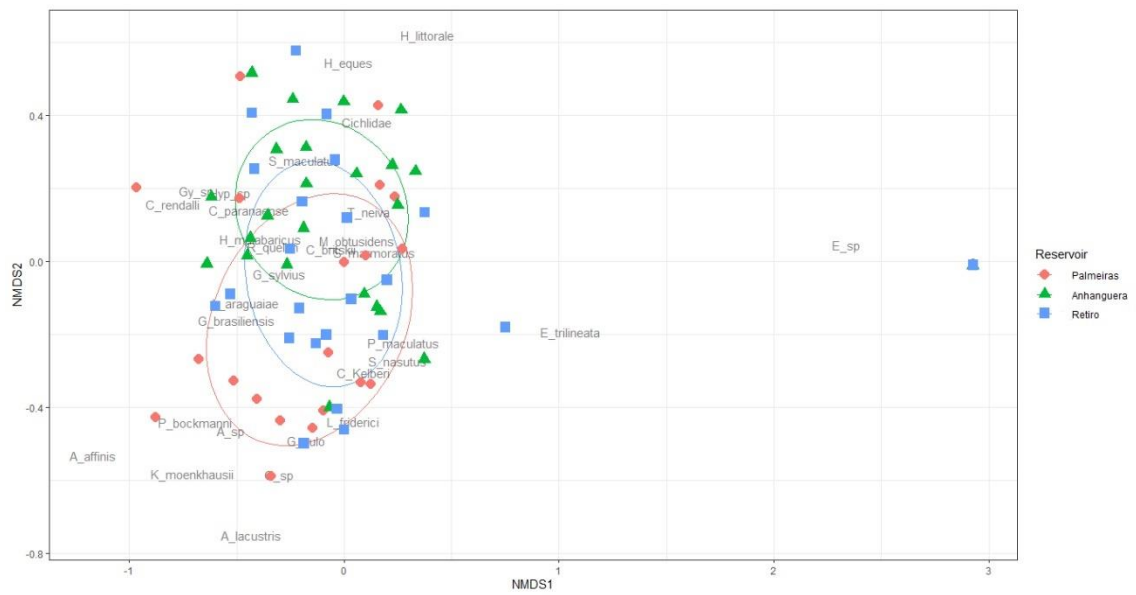


Figure 7. Graphical results of the Non-Metric Multidimensional Scaling Analysis, based on abundance and Bray Curtis distance, considering the distribution of fish species spatially in the Palmeiras, Anhanguera and Retiro SHPs reservoirs, Sapucaí-Mirim River (SP, Brazil).

Table 4. PERMANOVA and PERMDISP values (Pseudo-F and P) on Bray-Curtis distance matrixes for fish fauna in different approaches characterized by reservoirs, compartments and months of the Sapucaí-Mirim River (SP, Brazil).

	Composition				Abundance				Development stages				Reproductive strategies			
	PERMANOVA		PERMDISP		PERMANOVA		PERMDISP		PERMANOVA		PERMDISP		PERMANOVA		PERMDISP	
	Pseudo F	P	Pseudo F	P	Pseudo F	P	Pseudo F	P	Pseudo F	P	Pseudo F	P	Pseudo F	P	Pseudo F	P
Reservoirs	2.671	0.006	3.4531	0.074	2.3308	0.005	1.7415	0.226	5.565	0.002	1.6796	0.178	2.0487	0.095	-	-
Compartments	3.582	0.003	4.4867	0.058	2.2907	0.037	1.7642	0.218	0.64156	0.532	-	-	1.3185	0.294	-	-
Months	4.5383	0.001	2.0512	0.211	4.5328	0.001	3.7714	0.032	5.5006	0.002	0.15915	0.929	2.7128	0.015	6.1466E-2	0.99
ReXCo	3.1133	0.001	-	-	2.4142	0.002	-	-	5.0911	0.003	-	-	2.8308	0.027	-	-
ReXMo	1.6443	0.025	-	-	1.5072	0.034	-	-	2.4105	0.027	-	-	1.554	0.11	-	-
CoXMo	1.5688	0.076	-	-	1.5876	0.054	-	-	0.67587	0.643	-	-	0.53184	0.816	-	-
ReXCoXMo	1.2923	0.179	-	-	1.3311	0.089	-	-	1.9457	0.062	-	-	1.4348	0.167	-	-

P<0.05, RE: Reservoirs, CO: Compartments and Mo: Months.

Discussion

The PCA analysis evidenced temporal differences along the cascade, mainly for November, month with the lowest outflow. The posterior increase in the outflow (towards the summer season) was probably responsible for more homogeneous conditions among reservoirs. In November it was also possible to observe a trend of spatial separation between the compartments of the Palmeiras and Retiro reservoirs, in the other months the separation was unclear. This occurs because the Sapucaí-Mirim is a typical middle size plateau river and the reservoirs of this cascade respond instantly to rainfall events (Brambilla *et al.*, 2020).

Despite sampling small and punctual habitats –the macrophyte banks colonizing the banks of the reservoirs cascade, during a single reproductive period, thirty taxa were found, belonging to five orders and 14 families. This composition represents about 30% of the species inventoried in this river (Oliveira *et al.*, 2016; Brambilla *et al.*, 2018; Diniz *et al.*, 2019). These data indicate that fish with different evolutionary histories, biological and ecological characteristics (Reis *et al.*, 2013) are using these habitats as nursery areas, validating that macrophyte banks really play an important role for the initial development of the fish fauna.

Species richness in the area of influence of the Sapucaí-Mirim River cascade followed the general pattern found in the upper Paraná River basin, which has large numbers of species of Siluriformes and Characiformes (Langeani *et al.*, 2007). Another similarity, comparing to large reservoirs of the HPPs of the upper Paraná River basin, is the predominance of sedentary species, with or without parental care, generalists, with high tolerance to environmental changes and capable to complete their life cycle entirely within the reservoirs, such as some species of small Characiformes, Cichliformes and Siluriformes, in addition to some predators (Agostinho *et al.*, 2008).

A relevant finding, may be intriguing, is that even after a decade of the construction of these SHPs, the richness of non-native species is low, with only two species (*Cichla kelberi* and *Coptodon rendalli*) of the 30 found in macrophyte banks. This characteristic is distinct from those found in the HPPs of the upper Paraná River basin, that have a high contamination with non-native species (Gubiani *et al.*, 2018; Pereira *et al.*, 2018), such as in the Paranapanema River reservoirs where about 26.6% of its fish fauna is composed by invasive species (Jarduli

et al., 2020). This particularity may be associated with these reservoirs presenting characteristics more similar to unbarred rivers.

In general, for abundance per developmental stage, larvae showed higher abundance values in the intermediate compartment and the juveniles in the lentic compartment. This trend is explained by the fact that spawning events, in drift, in these small reservoirs occurs mainly in the upstream lotic compartment, and the predominant fast flow make possible larvae in very initial stages reach the reservoirs transitional (lotic-lentic) compartments (Brambilla *et al.*, 2020), in addition, macrophyte banks are rare in the lotic compartments in the cascade due to their intense flow conditions. This is similar to undammed rivers, where fish dispersion occurs faster due to higher water velocity in the rainy season (Vianna & Nogueira, 2008). Temporally, the abundance of larvae was higher in the beginning of the cycle and juveniles were more abundant in the final months.

The taxa captured by Brambilla *et al.*, (2020), in their investigation on the dispersion of fish eggs and larvae in drift in this same reservoir cascade, were also found in the present study (larvae and/or juveniles), with the exception of *Bryconamericus* spp., a pelagic species (Bialecki *et al.*, 2005) and *Prochilodus lineatus*. In the mentioned previous study, most larvae were found in the early stages of development (yolk sac and pre-flexion), whereas in the macrophyte banks it was sampled larvae in more advanced stages (flexion and post-flexion), most in post-flexion. This comparison reinforces that macrophytes banks are suitable areas for recruitment of the local fish fauna.

Higher abundance of larvae corresponded to *Serrasalmus maculatus*, a sedentary species with parental care that presents partial spawning and adhesive eggs that are deposited in the aquatic vegetation (Langeani & Rego, 2014). It is considered an opportunistic species, with rapid growth (Suzuki *et al.*, 2004). The species *Synbranchus marmoratus* was the most abundant when considered the juveniles. It is characterized by sedentary habits, with parental care and for building nest during the reproductive periods (Langeani & Rego, 2014). It also shows adaptations appropriate for living in macrophyte banks, such as elongated body shape, camouflage pattern between plants, young individuals feeding on insects and tolerance to low concentrations of oxygen (Henderson & Hamilton, 1995; Crampton & Hopkins, 2005; Bulla *et al.*, 2011).

It is important to emphasize that specimens of all groups of reproductive strategies, following the classification of Vasconcelos *et al.* (2014), were found in the macrophytes banks.

The more abundant reproductive strategies were non-migrants or short-distance migrants with external fertilization and parental care (NEP) and non-migrant or short-distance migrant species with external fertilization and without parental care (NEW). This finding reflects the existing knowledge, mainly based on adults captures, that the fish fauna of the Sapucaí-Mirim River is predominantly composed of small and medium-sized non-migrant/short-distance migrant species such as some species of Anostomidae and Pimelodidae (Freitas-Souza, 2014; Oliveira *et al.*, 2016; Brambilla *et al.*, 2020).

Another relevant result was that two, *Megaleporinus obtusidens* and *Pimelodus maculatus*, of the three long-distance migratory species commonly found in these reservoirs (Freitas-Souza, 2014; Oliveira *et al.*, 2016), were found in the sampled macrophyte banks. According to Agostinho *et al.*, 2008, analyzing a consistent set of 75 reservoirs in the Neotropics, only 5% have more than three species of long-distance migrators in their area of influence, because this kind of the species is the most affected by large dams. For SHPs the patterns do not seem to be different, having a reduced diversity of long-distance migratory fish. *Prochilodus lineatus* is the other species commonly found in the Sapucaí-Mirim river (Oliveira *et al.*, 2016; Diniz *et al.*, 2019), but not sampled in the macrophyte banks, despite the presence of eggs and larvae in drift (Brambilla *et al.*, 2020). Probably larval/juvenile recruitment of this species is taking place downstream the last dam (Retiro). Future investigation is necessary to answer this question.

The NMDS showed that abundance of Palmeiras (upstream in the cascade) presents the highest dissimilarity in relation to the other reservoirs, while the abundance of Retiro (downstream in the cascade) exhibited more similarities among reservoirs. PERMANOVA and PERMDISP corroborated that there are differences among the abundance of reservoirs in the cascade and in this case, specifically, the position of the SHP in the cascade may have influenced the distribution of fish fauna.

In conclusion, we can assume that the studied macrophyte banks are effectively used by different fish species as nursery areas. These banks were chosen randomly and without distinction of plant composition to represent the different compartments of the three SHP reservoirs. No specific association between the aquatic plant composition of the macrophyte banks and the fish fauna was tested. However, regardless of their composition, macrophyte banks increase habitat complexity, thus enhancing the diversity of aquatic fauna (Agostinho *et al.*, 2003, 2007; Pelicice *et al.*, 2005, 2008; Dibble & Pelicice, 2010; Ferrareze & Nogueira, 2011). Nevertheless, macrophyte overgrowth can disrupt the ecological functioning of aquatic

ecosystems and cause major problems in terms of water quality and hydropower reservoir management (Thomaz & Bini, 2003; Pompêo, 2017).

The results obtained are valuable for the conservation and management of fish species found in the Sapucaí-Mirin River reservoir cascade, giving the absence of typical nursery areas (*e.g.* tributary entrances and marginal lagoons). A representative percentage of the local fauna was found in the study, including fish with different reproductive strategies, corroborating the hypothesis that these habitats are of great relevance for reservoirs, as nursery areas for fish. Furthermore, the position of the reservoirs in the cascade, the compartmentalization and the temporal variation influenced the distribution of larvae and juveniles along the cascade.

Acknowledgment

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Supplementary Material

Table. Mean values and standard deviations (in parentheses) of limnological variables measured in different compartments of the Palmeiras SHP reservoir, Sapucaí-Mirim River (SP, Brazil).

Month	Compartment	Temp (°C)	pH	ORP (mV)	Cond. ($\mu\text{S cm}^{-1}$)	Turbidity (NTU)	DO (mg L^{-1})	TDS (g L^{-1})
November	Intermediate	26.49 (0.03)	6.9 (0.08)	234.77 (7.82)	70 (0)	45.46 (1.76)	6.45 (0.87)	0.046 (9.31E-10)
	Lentic	30.75 (0.21)	9.14 (0.26)	163.33 (10.85)	75 (1.8E-03)	43.61 (4.85)	10.96 (1.14)	0.048 (1.32E-03)
December	Intermediate	26.94 (0.06)	6.92 (0.03)	236.44 (26.97)	55 (0)	165.22 (5.19)	6.17 (0.33)	0.035 (3.33E-04)
	Lentic	28.37 (0.30)	7.22 (0.43)	247 (14.81)	56 (0.86)	53.13 (8.58)	4.71 (1.91)	0.036 (5.00E-04)
January	Intermediate	25.55 (0.09)	6.9 (0.13)	296.11 (11.77)	55.66 (0.5)	111.55 (7.17)	6.39 (0.19)	0.036 (5.27E-04)
	Lentic	27.21 (0.22)	6.62 (0.02)	300 (15.01)	46 (0)	49.13 (1.95)	5.45 (0.35)	0.03 (0)
February	Intermediate	24.94 (0.16)	7.38 (0.11)	169.88 (4.88)	56 (6.67E-04)	224.88 (11.66)	6.35 (1.31)	0.037 (6.58E-10)
	Lentic	27.77 (0.15)	7.22 (0.23)	173 (20.95)	47 (6.0E-04)	132 (15.77)	6.92 (0.38)	0.031 (3.33E-04)

Table. Mean values and standard deviations (in parentheses) of limnological variables measured in different compartments of the Anhanguera SHP reservoir, Sapucaí-Mirim River (SP, Brazil).

Month	Compartment	Temp. (°C)	pH	ORP (mV)	Cond. ($\mu\text{S cm}^{-1}$)	Turbidity (NTU)	DO (mg L^{-1})	TDS (g L^{-1})
November	Intermediate	28.71 (0.38)	6.42 (0.10)	272.22 (7.69)	73.44 (5.27E-04)	2.9 (4.22)	4.22 (0.67)	0.047 (5.00E-04)
	Lentic	29.36 (0.36)	6.75 (0.22)	237.22 (21.95)	73.55 (1.33E-03)	3.51 (3.95)	3.95 (1.22)	0.048 (8.66E-04)
December	Intermediate	26.84 (0.029)	6.74 (0.086)	276.55 (13.35)	53.66 (0.5)	86.74 (7.4)	4.82 (0.22)	0.035 (0)
	Lentic	26.85 (0.048)	6.64 (0.093)	300.22 (3.73)	56 (0)	57.9 (4.65)	3.81 (0.86)	0.036 (6.58E-10)
January	Intermediate	25.67 (0.36)	5.86 (0.16)	306.87 (25.88)	42 (0.75)	138.12 (5.66)	5.14 (0.15)	0.027 (4.63E-04)
	Lentic	27.31 (0.81)	6.02 (0.13)	208.2 (20.67)	43 (0)	134.8 (5.73)	4.47 (0.37)	0.028 (0)
February	Intermediate	26.34 (0.34)	6.78 (0.44)	180.37 (8.97)	52 (0)	107.87 (4.79)	6.57 (0.18)	0.034 (0)
	Lentic	26.32 (0.25)	7.2 (0.14)	202.1 (10.35)	50.6 (6.99E-04)	102.31 (4.65)	5.89 (0.31)	0.033 (3.16E-04)

Table. Mean values and standard deviations (in parentheses) of limnological variables measured in different compartments of the Retiro SHP reservoir, Sapucaí-Mirim River (SP, Brazil).

Month	Compartment	Temp. (°C)	pH	ORP (mV)	Cond. ($\mu\text{S cm}^{-1}$)	Turbidity (NTU)	DO (mg L^{-1})	TDS (g L^{-1})
November	Intermediate	28.31 (0.075)	6.86 (0.06)	235.11 (5.73)	73 (9.31E-10)	21.5 (1.78)	4.35 (0.37)	0.047 (5.27E-04)
	Lentic	27.38 (0.033)	7.91 (0.45)	166.77 (28.63)	83 (0)	18.78 (2.46)	7.28 (0.82)	0.054 (0)
December	Intermediate	26.81 (0.25)	6.9 (0.16)	274.11 (19.25)	60.77 (0.97)	81.72 (2.19)	3.83 (0.41)	0.039 (5.0E-04)
	Lentic	33.64 (0.74)	9.63 (0.11)	160.66 (15.63)	81.33 (3.53)	57.31 (8.37)	11.08 (0.71)	0.052 (2.33E-03)
January	Intermediate	25.87 (0.81)	6.31 (0.05)	326.55 (7.28)	44 (0)	86.47 (3.11)	4.87 (0.42)	0.028 (5.0E-04)
	Lentic	28.37 (0.05)	6.21 (0.17)	308.44 (18.92)	48 (0.86)	104.27 (16.73)	4.73 (0.78)	0.031 (5.0E-04)
February	Intermediate	25.51 (0.005)	7.29 (0.05)	205.77 (6.7)	50.22 (4.41E-04)	102.85 (7.46)	6.87 (0.21)	0.033 (0)
	Lentic	27.51 (0.69)	7.02 (0.25)	196.33 (22.43)	48.22 (1.20E-03)	87.63 (8.03)	6.09 (1.54)	0.031 (0.001)

The following images illustrate the activities of the distinct phases of the study

	
General view of the macrophyte banks	General view of the macrophyte banks
	
Sampling with sieve	Material sampled with the sieve for laboratory analysis
	
Sampling with gillnet	Material sampled with gillnet
	
Measurement of limnological parameters	Sorting of the material sampled with the sieve



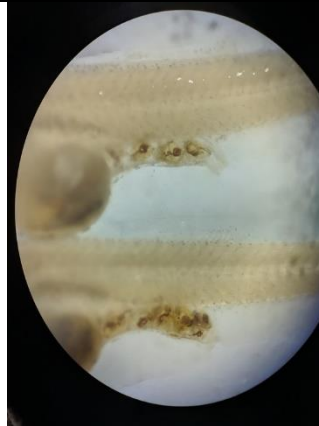
Larval identification



Juveniles identification and biometrics



Larvae of *Serrasalmus maculatus* in flexion



Typical cladocerans from littoral environments