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ECOLOGY

Fish associated with microhabitats of submerged fallen logs in a lake in the Brazilian Amazon

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ABSTRACT. Microhabitats represent an important factor for fish survival, providing food and shelter, in addition to good temperature conditions and dissolved oxygen levels. One of these temporary microhabitats formed is that of submerged logs, which, due to the flooding of adjacent areas, become available for fish. In this sense, the present study evaluates the composition and structure of fish assemblages associated with submerged logs on the banks of an Amazonian lake and identified the diversity of species according to the hydrological period. Field work was carried out on the banks of Tupé lake, in the lower Negro River, using scoop nets. The sampling was carried out in an area which measured 16 m² (4 m x 4 m) and, within this area, the effort was standardized with the presence of three researchers fishing for approximately one hour in each of the sampling sites. The capture of the fish was carried out with the aid of hand nets ("*puçá*" and "*rapiché*") and sieve. Absolute abundance (N), richness (S), Shannon-Wiener diversity index (H'), Berger Parker dominance (d) and equitability (E) were analyzed. Analysis of variance ($p < 0.05$) was used to verify the temporal variation. A total of 631 specimens were captured, which were distributed in eight families and 25 species, with the orders of Characiformes (67.8%) and Cyprinodontiformes (22.8%) predominating. In the high water period, the highest values of numerical abundance and dominance were observed, while in the low water period, the highest values of species diversity, richness and equitability were found. Therefore, the results presented here can serve as a basis for fisheries management, as well as for conservation measures in these environments that help to maintain fish assemblages.

Keywords: diversity; ichthyofauna; Negro river; small fish; stream.

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Introduction

Aquatic environments in the Amazon are subject to the influence of the flood pulse and this phenomenon is fundamental in large rivers with adjacent floodplains (Junk, Bayley, & Sparks,1989). In these environments, a lateral input of allochthonous materials (pieces of tree logs, branches, leaves, flowers, fruits, seeds and pollen) occurs as they fall from adjacent vegetation and form litter (Herdenson & Walker, 1986). This accumulation of materials (litter) on the margins of lentic water bodies, such as lakes and streams, form microhabitats, which are used by a variety of fish and invertebrate species, and serve as a place of feeding, reproduction (substrate for spawning) and shelter from predators (Mendonça, Magnusson, & Zuanon, 2005; Zuanon et al., 2015). According to Herdenson & Walker (1990), the micro and macroinvertebrates that exist in these microhabitats, feed and decompose up to 75% of the materials that make up the litter, and these, in turn, serve as food mainly for small fish.

Microhabitats represent an important environment for the survival of smaller aquatic fauna because provide they refuge from predators due to the entanglement of deposited materials (Miranda, Driscoll, & Allen, 2000). In addition to the litter, another microhabitat commonly formed in lakes and creeks are the logs. Some species adapt to these environments and often become typical of this physical space. However, in these environments, some factors influence the distribution of species, such as food availability, predation intensity and tolerance to the physical-chemical conditions of the environment (Lowe-McConnell, 1999; Vlach, Dusek, Svátora, & Moravec, 2005; Bonin, Srinivasan, Almany, & Jones, 2009).

In general, the evolutionary history of the species or the competitive interspecific interactions may explain the differences in the choice of microhabitat, varying from one place to another, whether for feeding, reproduction and/or refuge (Baltz, Vondracek, Brown, & Moyle, 1987; Tolimieri, 1995). However, it is common to observe that some species are frequently found in certain habitats and microhabitats, and that many of them present morphological adaptations that promote greater and better exploitation of a given resource (Rinne, 1992; Couto et al., 2015). Species of the genus *Panaque*, for example, have some morphological specializations, such as the presence of hypertrophied muscles between the mandibular teeth, which optimize mandibular strength during feeding (Schaefer & Stewart, 1993), as well as presenting robust unicuspid teeth, which allows them to forage around submerged fallen logs, where they can ingest wood particles (Schaefer & Stewart, 1993; Armbruster 2003; Lujan & Armbruster, 2011).

Microhabitat selection by fish can vary according to fish age, sex, reproductive stage, geographical area and environmental conditions (Karr, Toth, & Dudley, 1985). Henderson & Walker (1990) studied the banks of Amazonian streams with forest litter and reported that the individuals that inhabit these environments are either young or adults of small species, and the distribution of the species is predominantly localized within the sandbanks and that factors such as water depth and the size structure and age of the litter and the position of the site relative to the direction of the water flow are determinants for the species' choosing these sites. Carvalho, Fidelis, Arruda, Galuch, and Zuanon, (2013) studied the ichthyofauna of floating litter banks in Amazonian creeks and observed that most of the fish species were carnivorous/insectivorous with sedentary habits, representing the families Erythrinidae, Auchenipteridae, Pseudopimelodidae, among others, and that these environments are important points of refuge and feeding.

In the Central Amazon, fish assemblage studies are mostly focused on the microhabitats of aquatic herbaceous plants (Araujo-Lima, Portugal, & Ferreira, 1986; Soares, Freitas, & Oliveira, 2014), flooded forests (Loebens, Farias, Freitas, & Yamamoto, 2019), creeks (Beltrão & Soares, 2018), beach areas (Amaral, Anjos, & Yamamoto, 2020) and submerged litter (Henderson & Walker, 1990) and floating litter (Carvalho et al., 2013). Studies of the microhabitats of submerged logs as a research area in the Amazon are so far non-existent; therefore, the composition of fish assemblages that use these environments remains poorly understood. In this context, the present study evaluated the composition and structure of the fish assemblages associated with submerged logs on the shores of a lake in the Tupé Sustainable Development Reserve (SDR) and identified the diversity of species according to the hydrological period. We also sought to test the hypothesis that the fish assemblages around the logs present a similar pattern to other fish assemblages in the Amazon in relation to seasonal changes in the environment.

Materials and methods

The study was conducted in the Tupé lake, about 25 km upstream from the city of Manaus, capital of the state of Amazonas (Figure 1). The Tupé lake is bathed by the black waters of the Negro River (Sioli, 1984; Rios-Villamizar et al., 2020) and is located within the Tupé SDR, which occupies 12,000 hectares and has approximately 2 km of shoreline (Beltrão & Soares, 2018). According to Darwich, Aprile, and Robertson, (2005), in the of low-water period, when the river level is less than 19 m deep, the lake is isolated from the river and its waters are chemically similar to those of the inland streams that are located at the head of the lake. However, in the high water period, the lake is connected to the main channel of the river. The granulometric composition of the surface sediments of the Tupé lake is mostly sand (96.90 – 99.99%), and silt (0.01 – 2.74%), with clay (0 – 0.38%) in smaller percentages (Darwich et al., 2005).

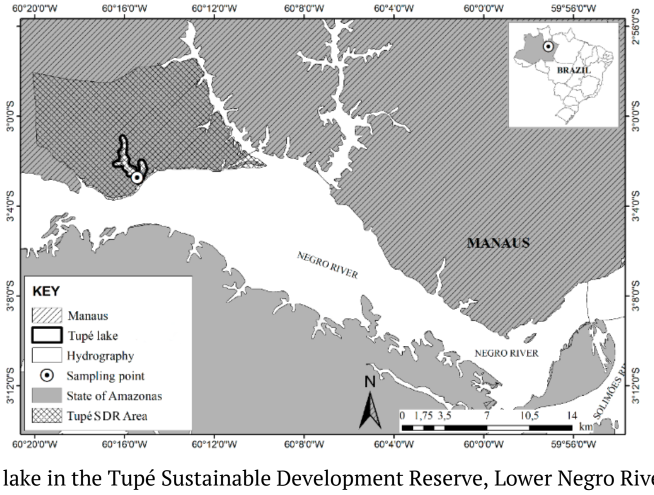

Figure 1. Location of Tupé lake in the Tupé Sustainable Development Reserve, Lower Negro River, Amazonas state, Brazil.

Data collection

The samplings were carried out during each phase of the hydrological cycle, in the months of September (falling water) and December (low water) of 2017 and March (rising water), April (high water) of 2018 (Bittencourt & Amadio, 2007), always in the same lakes, totaling four collection days. In each sampling, six sites were surveyed, comprising an area of approximately 16 m^2 (4 m x 4 m) around the naturally submerged fallen logs found on the shores of the lake (Figure 2), with a distance of 3 to 4 meters between the sampling sites.

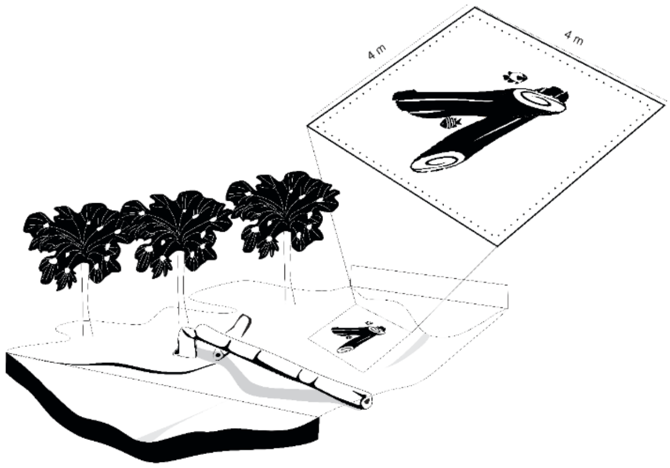

Figure 2. Representation showing the size of the sample site and naturally fallen logs on the shores of the lake.

The sampling was standardized, with the presence of three researchers who fished for approximately one hour at each sample site. The capture of the fish was carried out using active methods, i.e., hand nets (“*puçá*” and “*rapiché*”) and a sieve. When no more fish were seen swimming freely in the area of the sample site, the logs were removed from the water in order to investigate the presence of fish in crevices and holes. After removal of the fish, the logs were returned to their respective places of origin.

The fish were anesthetized with eugenol (as recommended by the American Veterinary Medical Association [AVMA], (2013). Subsequently, they were fixed in absolute alcohol and taken to the Ichthyology Laboratory of the Federal University of Amazonas (LABIC/UFAM), where they were identified with the use of taxonomic keys (Queiroz et al., 2013; Zuanon et al., 2015). The project was evaluated and approved by the Ethics Committee in the Use of Animals of the Federal University of Amazonas under No. 016/2017.

Analysis of the data

The fish assemblages were analyzed using measures of numerical abundance (N), richness (S), Shannon-Weiner diversity index (H’) (Krebs, 1989) and dominance (d) (Berger & Parker, 1970). To complement the interpretation of the Shannon-Weiner index, the equitability index (J) was calculated (Pielou, 1975). Sampling efficiency was estimated via the rarefaction curve (Chao et al., 2014), using the *iNEXT* package (Hsieh, Ma, & Chao, 2022).

The number of species that were common and exclusive to the seasonal periods (rising water, high water, falling water and low water) was illustrated via the construction of a Venn diagram, created using the *vennDiagram* package (Chen 2022). An ordering analysis was performed by means of non-metric dimensional scaling (NMDS), using a Euclidean index of dissimilarity to sort and verify patterns in the composition of the fish assemblage in relation to the hydrological period. Finally, to test the hypothesis of seasonal effects (falling water, low water, rising water and high water) on the composition of the fish assemblages, a permutational analysis of variance (PERMANOVA) was performed using the v*egan* package, which considered 999 permutations and had a significance level of $p<0.05$ (Oksanen et al., 2022). Likewise, a post-hoc test for PERMANOVA was also conducted using the *pairwiseAdonis* package (Martinez, 2017) with a significance level of $p<0.05$.

The computational system R, version 4.3.1 (R Development Core Team, 2023), was used in the statistical analysis.

Results

A total of 631 specimens were captured, and these were distributed in eight families and 25 species, with the orders Characiformes (67.82%) and Cyprinodontiformes (22.82%) being the most predominant, followed by Cichliformes (6.65%), Gobiiformes (2.21%) and Siluriformes (0.47%) (Table 1).

Table 1. Composition, numerical abundance and amplitude of standard length and total weight of species caught in naturally fallen logs in the periods of high and low water in the Tupé lake, Amazonas state, Brazil.

Taxonomy	N	Standard length range (cm)	Weight range (g)	Periods			
				Rising water	High water	Falling water	Low water
Characiformes							
Erythrinidae							
Hoplias malabaricus Bloch 1794	1	2.80	0.20	-	-	-	1
Lebiasinidae							
Copella callolepis (Steindachner 1876)	11	0.70 - 2.00 (1.15 ± 0.39)	0.01 - 0.30 (0.05 ± 0.09)	-	-	11	-
Copella nattereri (Steindachner 1876)	20	1.10 - 3.40 (2.07 ± 0.48)	0.01 - 0.21 (0.10 ± 0.05)	5	3	-	12
Nannostomus digrammus (Fowler 1913)	9	1.10 - 2.10 (1.79 ± 0.30)	0.03 - 0.10 (0.06 ± 0.02)	-	7	2	-
Nannostomus eques Steindachner 1876	4	1.10 - 2.00 (1.48 ± 0.39)	0.02 - 0.10 (0.06 ± 0.04)	1	1	2	-
Nannostomus unifasciatus Steindachner 1876	25	0.70 - 3.50 (1.90 ± 0.93)	0.01 - 0.40 (0.11 ± 0.12)	-	6	19	-
Acestrorhynchidae							
Acestrorhynchus nasutus Eigenmann 1912	2	1.50 - 2.20 (1.85 ± 0.49)	0.02 - 0.02 (0.02 ± 0.00)	1	1	-	-
Characidae							
Hemigrammmus analis Durbin 1909	278	0.70 - 3.90 (2.02 ± 0.64)	0.01 - 3.40 (0.18 ± 0.27)	16	219	20	23
Hemigrammus coeruleus Durbin 1908	1	1.70	0.05	-	1	-	-
Hemigrammus levis Durbin 1908	48	0.80 - 3.60 (2.19 ± 0.79)	0.01 - 0.80 (0.20 ± 0.22)	-	3	13	32
Hyphessobrycon copelandi Durbin 1908	29	0.80 - 3.00 (1.76 ± 0.57)	0.01 - 0.70 (0.16 ± 0.18)	-	-	29	-
Siluriformes							
Loricariidae							
Farlowella spp.	1	5.60	0.08	-	1	-	-
Pseudoloricaria laeviuscula (Valenciennes 1840)	1	1.40	0.04	-	-	-	1
Sturisoma spp.	1	3.70	0.10	-	-	1	-
Gobiiformes							
Eleotridae							
Microphilypnus ternetzi Myers 1927	14	0.80 - 1.80 (1.10 ± 0.28)	0.01 - 0.10 (0.02 ± 0.02)	-	-	14	-
Cichliformes							
Acarichthys heckelii (Müller and Troschel 1849)	12	1.40 - 3.00 (1.88 ± 0.42)	0.03 - 0.50 (0.14 ± 0.13)	-	-	-	12
Acaronia nassa (Heckel 1840)	5	1.10 - 2.00 (1.56 ± 0.42)	0.02 - 0.17 (0.08 ± 0.06)	-	-	-	5
Apistogramma spp.	12	0.70 - 2.90 (1.94 ± 0.56)	0.01 - 0.50 (0.18 ± 0.16)	1	-	3	8
Caquetaia spectabilis (Steindachner 1875)	1	3.70	1.00	-	-	-	1
Cichla temensis (Humboldt 1821)	1	4.70	1.60	-	-	-	1
Geophagus proximus (Castelnau 1855)	4	1.20 - 2.00 (1.55 ± 0.33)	0.03 - 0.20 (0.09 ± 0.08)	-	-	-	4
Mesonauta festivus (Heckel 1840)	4	1.00 - 12.60 (4.03 ± 5.72)	0.01 - 7.50 (1.92 ± 3.72)	-	-	-	4
Mesonauta insignis (Heckel 1840)	1	1.60	0.05	-	1	-	-
Taeniacara candidi Myers 1935	2	1.70 - 1.90 (1.80 ± 0.14)	0.08 - 0.11 (0.10 ± 0.02)	-	-	-	2
Cyprinodontiformes							
Poeciliidae							
Fluviphylax zonatus Costa 1996	144	0.50 - 1.80 (1.03 ± 0.21)	0.01 - 0.04 (0.01 ± 0.01)	7	98	5	34

Hemigrammus analis Durbin, 1909 and *Fluviphylax zonatus* Costa, 1996 were the most abundant species in the high water period; with 64 and 28%, respectively. The sampling effort, analyzed using the species rarefaction curve, was sufficient for the high water and falling water periods, because the curve tends to stabilization. However, the curve obtained for the low-water and rising water periods did not present a tendency to stabilization, and showed an increasing pattern in the number of species collected according to the number of individuals captured (Figure 3), and thus additional samplings may increase the number of species.

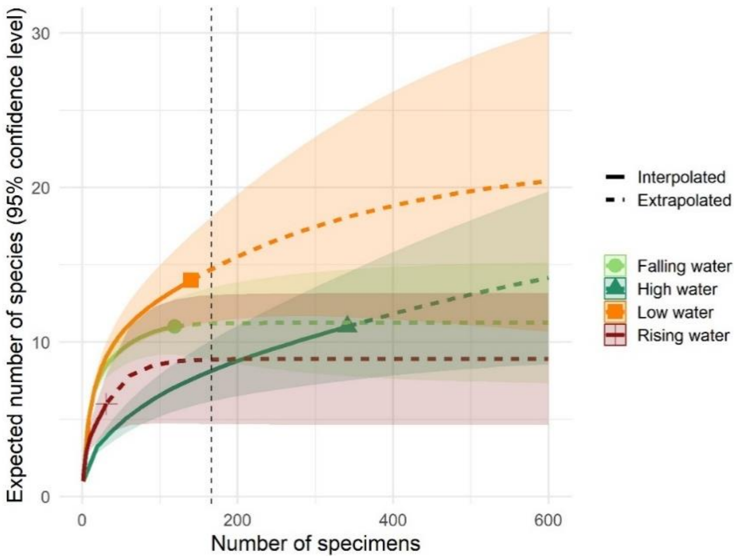

Figure 3. Rarefaction curve for fish species caught near or under submerged logs in the Tupé lake, Amazonas, Brazil.

In the high water period, higher values of numerical abundance and dominance were obtained, while the low water period presented higher values of species richness, diversity and equitability (Table 2). As for the composition, the Venn diagram shows the exclusive occurrence of four species in the high water period and 13 species in the low water period, the others being common in both periods (Figure 4).

Table 2. Ecological indices obtained for fish captured around submerged fallen logs in the Tupé lake, Amazonas, Brazil.

Ecological indices	Rising water	High water	Falling water	Low water
Abundance (N)	31	341	119	140
Richness (S)	6	11	11	14
Diversity (H')	1.30	0.96	2.05	2.09
Equitability (J')	1.68	0.92	1.97	1.82
Dominance (d)	0.52	0.64	0.24	0.24

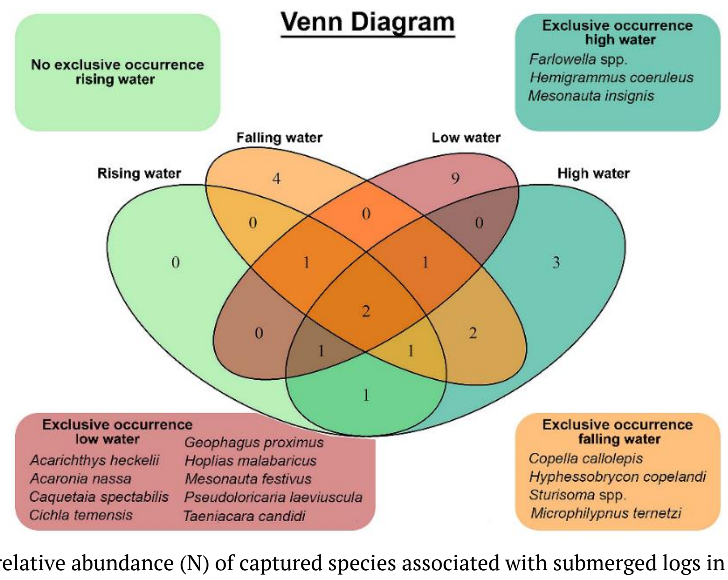

Figure 4. Venn diagram of relative abundance (N) of captured species associated with submerged logs in high- and low-water periods in the Tupé lake, Amazonas, Brazil.

The results of ordering analyses (NMDS| stress= 0.13) based on abundance data indicate that only the high water period presented a different species composition (Figures 5). This difference was confirmed with the PERMANOVA (Pseudo-F = 6.7777; df= 3;20; *p* (perm) = 0.001), indicating a difference in species composition between the pairs: rising water vs high water; rising water vs falling water; high water vs falling water; high water vs low water ($p<0.05$; Table 3: Figure 5).

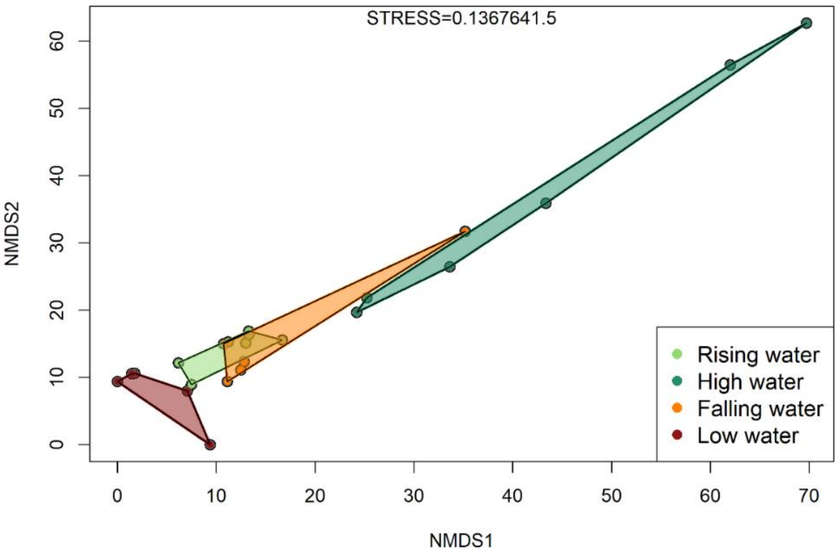

Figure 5. NMDS of the composition of species according to the hydrological period: falling water (orange circle), low water (dark red circle), rising water (light green circle) and high water (dark green circle) in the Tupé lake, Amazonas state, Brazil.

Table 3. Summary of the post-hoc PERMANOVA conducted to assess differences in the composition of fish species found in and around submerged fallen logs in the four hydrological periods in the Tupé lake, Amazonas. *significance level: $p<0.05$

Pairs	df	SumsOfSqs	F.Model	R^2	p-value
Rising water vs High water	1	4,132.6667	10.1481	0.5036	0.003*
Rising water vs Falling water	1	145.3333	2.0781	0.1720	0.026*
Rising water vs Low water	1	175.9167	1.0815	0.0976	0.411
High water vs Falling water	1	4,143.8333	9.1583	0.4780	0.002*
High water vs Low water	1	3,649.7500	6.6945	0.4010	0.007*
Falling water vs Low water	1	259.7500	1.2494	0.1110	0.171

Discussion

In general, fish found in and around fallen logs do not seem to have the same distribution pattern of the fish assemblage expected for other habitats, with greater abundance in the high water period. In addition, the species that inhabit these environments are mostly small. Small fish species are distributed mainly in streams and creeks and can represent up to 50% of the fish assemblages of these environments (Castro, 1997; Casatti, Langeani, & Castro, 2001; Castro et al., 2003; Castro & Polaz, 2020).

The most abundant species in the study was *Hemigrammus analis* and *Fluviphylax zonatus*. The abundance of *H. analis* has already been reported before for other environments, such as lakes, creeks and beach environments (Beltrão & Soares, 2018; Castanho et al., 2020; Olentino, Furtado, & Yamamoto, 2020; Pereira, 2019), and this finding can be explained by the fact that the species forms a school. The species of the genus *Hemigrammus* are distributed throughout much of the Amazon basin and can be easily found in this region (Marinho, Carvalho, Langeani, & Tatsumi, 2008). Generally, these species inhabit marginal areas, which have a variety of microhabitats, such as submerged logs and roots, which serve as refuge, shelter and feeding areas, and provide areas with slow flow of water (Dos Anjos, Yamamoto, & Magalhães, 2017). Unlike *H. analis*, *Fluviphylax zonatus* has a geographic distribution that is restricted to the lower Negro River (Costa, 1996; Souza, 2008), and few studies have recorded this species as being abundant in creeks (Beltrão & Soares, 2018) and in or around submerged logs (Olentino et al., 2020). This species inhabits a wide range of microhabitats such as the banks of tributaries of rivers, streams, flooded forests and black-water creeks (Souza, 2008).

In the high water period, the high values of relative abundance reflect the dominance of *Hemigrammus analis* (N=219), since this species occurs in marginal areas amid fallen logs and roots (Dos Anjos et al., 2017), and it uses association in shoals to enhance prey identification, predator detection and swim more efficiently (Classon, Godin, & Abrahams, 1988; Day, Macdonald, Brown, Laland, & Reader, 2001). In the low water period, there was greater diversity and richness of species associated with the submerged logs, while the highest equitability was achieved for the falling water period. Although diversity is considered high in the present study, it is lower when compared to the study by Silva, Ferreira, and De Deus, (2010), who cite na H' of 1.54 – 2.60 for creeks in the flood zone of the lower Purus River, in the period of low water. Other factors that influence abundance and diversity are habitat structures. According to Gois, Antonio, Gomes, Pelicice, and Agostinho, (2011), environments with submerged trees present higher catches per unit of effort (CPUEn) when compared to less structured environments. The same pattern was observed for floating litter banks, which harbor a rich assembly of fish. These habitats serve as refuges for fish during the peak of the high water period (Carvalho et al., 2013) and also during low water periods when predation intensifies.

The homogeneous distribution that is indicated by the equitability and the low dominance value found can be explained by the fact that the falling water period is a critical period for many species due to the increase in predation with the decrease of the water level (Junk et al., 1989; Matthews & Marsh-Matthews, 2003; Petry, Gomes, Piana, & Agostinho, 2010). Therefore, as well as aquatic herbaceous plants, which serve as a shelter for immature fish from predators and young forms of sedentary species such as *Hoplias* and small species such as *Hemigrammus* and *Moenkhausia* (Agostinho, Gomes, & Júlio Jr, 2003; Hahn & Loureiro-Crippa, 2006; Bittencourt, Zacardi, Monteiro, Nakayama, & Queiroz, 2020; Oliveira, Cajado, Dos Santos, De Lima Suzuki, & Zacardi, 2020), submerged logs seem to play the same role, since it is difficult for medium to large predators to enter the cavities in the logs.

Most of the fish that are associated with the submerged logs are small individuals, which have pelagic habits, elongated fusiform bodies that allow them mobility throughout the water column, and are always active, i.e., the small Characiformes, which are commonly found in streams (Beltrão & Soares, 2018; Silva, 1993). Furthermore, although three species of Loricariidae were collected, no other species of the order Siluriformes were found. The absence of more specimens of this taxon is due to the fact that the collections were carried out during the day, when there was still daylight. According Sabino and Zuanon (1998) and Zuanon & Ferreira (2008), in general, the Siluriformes are usually more active at night. The presence of species of the families Loricariidae (Silva, 1993;1995; Casatti & Castro, 1998) and Auchenipteridae are commonly associated with submerged logs, since most of the species live in crevices of logs and roots in the beds of lakes and streams (Queiroz et al., 2013).

Submerged logs near the shores of lakes seem to represent an important refuge for small ichthyofauna in the four periods of the hydrological cycle. Our results demonstrate that the ichthyofauna observed among the submerged logs is usually formed by small Characiformes (*piabas*), and this is very similar to the ichthyofauna found in the litter. Henderson & Walker (1986) demonstrated that, despite the low fish richness found in the litter, its occupants seem to occupy these banks of leaves for quite some time, and this also seems to occur with the species of fish associated with logs.

Conclusion

The results show that a rich and diverse composition of species are associated with submerged logs, and species of the order Characiformes prevail, with a greater prevalence of *Hemigrammus analis*, especially during the high water period.

In general, most species (especially small ones) seem to use submerged logs as shelter. However, future studies are needed to verify the use of these trunks for food and reproduction, as well as to relate them to the environmental parameters of the water. This information will contribute to enriching our knowledge about the distribution of the fish species and their use of submerged logs. Furthermore, among these species found in the study, we can highlight some with ornamental potential, such as species of the genera *Hemigrammus*, *Nannostomus* and *Copella*. The results presented here can serve as a basis for fisheries management, as well as for conservation measures in these environments that help to maintain fish assemblages.

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