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Influence of tributaries on the periphytic diatom community in a floodplain

Influência dos tributários na comunidade de diatomáceas perifíticas em uma planície de inundação

Andressa Bichoff[1]*, Nicolli Cristina Osório[1], Daiane Trevisan Ruwer[1], Kisay Lorena Adame Montoya[1], Bárbara Dunck[2] and Liliana Rodrigues[1]

[1]Programa de Pós-graduacão em Ecologia de Ambientes Aquáticos Continentais – PEA, Universidade Estadual de Maringá – UEM, Av. Colombo, 5790, Bloco G-90, sala 08, CEP 87020-900, Maringá, PR, Brazil

[2]Programa de Pós-graduação em Ecologia, Universidade Federal do Pará – UFPA, R. Augusto Corrêa, 01, CEP 66075-110, Belém, PA, Brazil

*e-mail: andressabichoff@hotmail.com

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Abstract: In impacted floodplains, small tributaries have an essential role in the maintenance of ecosystem biodiversity. **Aim:** We aimed to characterise the periphytic diatom community (Bacillariophyta) in a protected area at the Upper Paraná River floodplain considering the importance of tributaries as a source of propagules. The purpose of this study was to understand the influence of the tributaries in the periphytic diatom community (Bacillariophyta) in a protected area at the Upper Paraná River floodplain. We expect that species present in the main river channel (mainly in the last stretch) would be a subset of the community present in the tributaries. **Methods:** The study area is located in the last non-dammed stretch of the Paraná River in this floodplain, with the contribution of several tributaries. Sampling was performed in 2014 in two tributaries and three sites at the Paraná River main channel, and the periphytic material was obtained from petioles of the aquatic macrophyte *Eichhornia azurea* (Sw.) Kunth. **Results:** We identified 115 diatom taxa. Higher values of species richness were registered in the tributaries, and higher densities in the tributaries and at sampling sites under the direct influence of the tributaries. We also observed a higher similarity between the tributaries, which are located in the preserved side of the floodplain. The results of beta diversity showed that closer sites had lower values, and that the species turnover was the component more important in this floodplain. **Conclusion:** In this way, these results reinforce the importance of tributaries mainly on the algae establishment and development reflected in the density of species. It demonstrates that sites in preserved area present a higher species richness and can serve as a pool of propagules for downstream environments.

Keywords: benthic algae; beta diversity; turnover; conservation.

Resumo: Em planícies de inundação impactadas, afluentes de pequeno porte tem um papel essencial na manutenção da biodiversidade do sistema. **Objetivo:** Este trabalho objetivou caracterizar a comunidade de diatomáceas (Bacillariophyta) perifíticas em área de proteção nacional na planície de inundação do Alto Rio Paraná considerando a importância dos tributários como fonte de recursos e propágulos. O objetivo deste estudo foi entender a influência dos tributários sob a comunidade de diatomáceas perifíticas de uma área protegida localizada na Planície do alto rio Paraná. Esperamos que as espécies presentes na calha principal do Rio (principalmente o último trecho) sejam um subconjunto da comunidade dos tributários. **Métodos:** A área de estudo está inserida no último trecho livre de barramentos nesta planície e conta com a contribuição de vários tributários. A amostragem foi realizada no ano 2014 em dois tributários e três pontos da calha principal do Rio Paraná, e o material perifítico obtido de pecíolos da macrófita aquática *Eichhornia azurea* (Sw.) Kunth. **Resultados:** Foram registrados 115 táxons de diatomáceas. Os maiores valores de riqueza de espécies foram registrados nos tributários, e as maiores densidades nos tributários e nos pontos que sofreram influência direta dos tributários. A maior similaridade foi observada entre os tributários, que são localizados na área preservada da planície. Os resultados de beta diversidade demonstraram que locais mais próximos tiveram menores valores, e que *turnover* de espécies foi o componente mais importante nesta planície. **Conclusão:** Dessa forma, esses resultados reforçam a importância dos tributários principalmente sobre o estabelecimento das algas e seu desenvolvimento refletido na densidade de espécies. Ainda demonstraram que ambientes em área preservada apresentam maior riqueza de espécies e podem servir como pool de propágulos para outros ambientes a jusante.

Palavras-chave: algas bentônicas; diversidade beta; *turnover*; conservação.

1. Introduction

The quality of water resources has been one of the most worrying issues for society in recent decades, due to an increase in demand, intensification of anthropogenic activities and pollution of water bodies, which affect the trophic levels, deteriorate drinking water conditions and possibly causes diseases (Wetzel, 2001; Tundisi, 2008). Aquatic organisms may be used as bioindicators of this artificial eutrophication, and help to evaluate the trophic state and characteristics of different types of habitats, such as marine, estuarine, freshwater, lotic or lentic (Loeb, 1992; Lowe & Pan, 1996; Bere & Tundisi, 2010).

Among aquatic communities, periphytic organisms are considered particularly sensitive to environmental changes. Periphyton is defined as a complex community of organisms attached to submerged substrate (organic, inorganic, dead or alive) (Wetzel, 1983), and algae are the most studied component. The development and establishment of periphytic algal communities is the result of complex interactions between hydrological, biotic and abiotic factors in aquatic systems (Biggs & Kilroy, 2000).

In periphytic algal communities, diatoms (Bacillariophyta) display high abundances, cosmopolitan distribution and a wide tolerance limit to different types of habitat (Round et al., 1990). Diatoms have been used as bioindicators of water quality, climate change, acidification and eutrophication. It is because they have a short life cycle, sessile life style and relatively low mobility, which generates rapid response in the face of environmental alterations (Maidana et al., 2005; Passy, 2007; Jüttner et al., 2010; Burliga & Schwarzbold, 2013; Bellinger & Sigee, 2010; Lobo et al., 2002). These algae are considered more efficient bioindicators than macroinvertebrates in certain types of environments, and allow monitoring lightly or heavily impacted environments (Soininen & Könönen, 2003; Taylor et al., 2007).

Brazil harbours a great variety of aquatic ecosystems, distributed in several distinct biomes, such as floodplains. These are characterised as transitional habitats originated from the flooding of river in adjacent areas, exhibiting temporary or permanently flooded environments, and are considered important areas for the maintenance of the regional biodiversity (Thomaz et al., 2004). Fluctuations in the water level cause great temporal variations in physical, chemical and biological characteristics, maintaining the high structural diversity of those habitats (Junk et al., 1989; Neiff, 1990; Petry et al., 2004; Gubiani et al., 2007).

The Upper Paraná River floodplain constitutes a highly complex ecological system, crucial to the maintenance of diversity and environmental stability. It is located downstream Porto Primavera hydroelectric power station and upstream Itaipu hydroelectric power plant, and constitutes the last non-dammed stretch in Paraná River basin, with many conservation units and protected natural areas (Ilha Grande National Park, Environmental Protection Area of the Islands and Várzeas of the Paraná River and Ivinhema State Park). In this stretch, the Paraná River main channel is fed by different streams and tributaries, which are important habitats providing areas for refuge, reproduction and development of several species, besides exhibiting high diversity and a high degree of endemism (Agostinho et al., 2005; Thomaz et al., 2007).

Studies performed in the Upper Paraná River floodplain highlight the importance of small tributaries in maintaining biodiversity, allowing the reproduction, spawning and development of fish eggs and larvae (Pavanelli & Caramaschi, 1997; Nakatani et al., 2001). These tributaries serve as migration routes to the fish stock of the Paraná River, due to reservoir construction (Baumgartner et al., 2004), and contribute to the maintenance of species in a reservoir, by preserving the original characteristics of a lotic system and consequently reducing the impact of damming (Hoffmann et al., 2005). Moreover, the influence of tributaries contributes to the reestablishment and recovery of the attributes and natural conditions of lotic environments such as the river-floodplain system, which are modified or affected by damming and anthropogenic activities (Stanford & Ward, 2001).

Therefore, the purpose of this study was to understand the influence of the tributaries on the periphytic diatom community (Bacillariophyta) in a protected area at the Upper Paraná River floodplain. We hypothesised that the influence of tributaries enhances the diatom community attributes (richness and density) in the main river sites. As well as, we expect that species present in the main river channel (mainly in the last stretch) would be a subset of the community present in the tributaries.

2. Methods

2.1. Study area

The study area is located at the Upper Paraná River floodplain, downstream Porto Primavera hydroelectric power station and upstream Itaipu hydroelectric power plant, in a stretch situated between Mato Grosso do Sul and Paraná states. It covers around 230 km extension of a non-dammed fragment, with many important tributaries (Ivinhema, Amambaí and Iguatemi rivers) associated to the right margin of the Paraná River (Agostinho et al., 2008), most of them non-dammed.

2.2. Sampling

Sampling was performed in November 2014 at five sampling sites along the right margin of the Paraná River, encompassing the Ilha Grande National Park area. Three sampling sites are located at the main river channel and the other two at the mouth of the main tributaries: Amambaí and Iguatemi rivers (Figure 1). All sampling sites are lotic, Amambaí (T1) and Iguatemi (T2) rivers are located inside the side of the Ilha Grande National Park, and P1-P3 are located in the Paraná River channel. The two tributaries had higher values of total solid materials (6-8.5 mg L^{-1}) than others sites (mean 3.5 mg L^{-1}) (Santana et al., 2017, in this volume). Mean depth in the sampling sites was 1.6 m (P1), 4.2 m (T1), 2.5 m (T2), 4.8 m (P2) and 2.2 m (P3). Sampling sites in the Paraná River had lower values of nutrients (Santana et al., 2017, in this volume).

2.3. Water level of the Paraná River

Samplings were performed on November 4th, 2014 (P2- Paraná River 2, T2- Iguatemi River and P3- Paraná River) and on November 7th, 2014 (T1- Amambaí River and P1- Paraná River 1). In November, there was a high water level fluctuation, varying from 1.5 m to 2.7 m (Figure 2). Samples were taken during days showing increased water levels of the Paraná River (Figure 2). Prior to the first

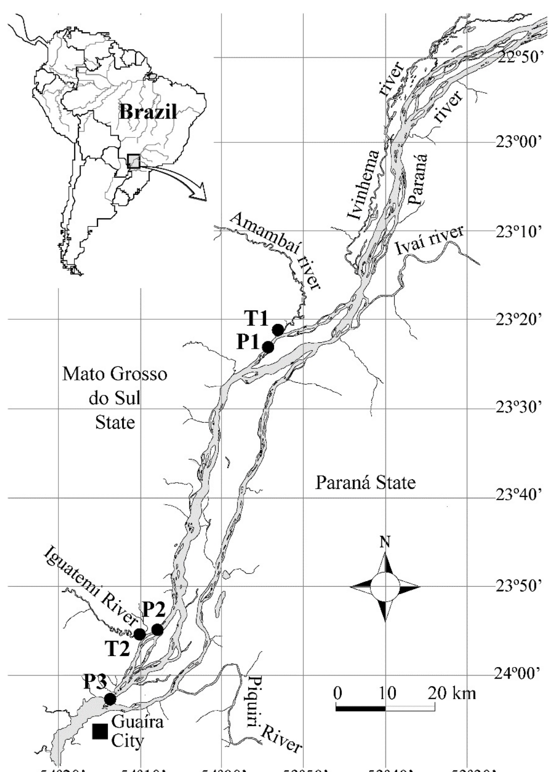

Figure 1. Location of sampling sites at the Upper Paraná River floodplain. Sampling sites: T1- Amambaí River, P1- Paraná River 1, P2- Paraná River 2, T2- Iguatemi River, P3- Paraná River 3.

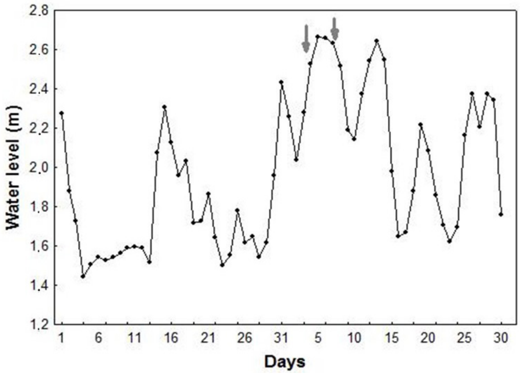

Figure 2. Water level of the Paraná River between October and November 2014. Arrows indicate sampling days (November 4th and 7th).

sampling day, the water level was approximately 1 m lower. This water level is lower than that commonly registered for this month in other years, month that commonly is characterized as high water period (Thomaz et al., 2007). In each sampling site, we measured abiotic variables (Santana et al., 2017, in this volume).

2.4. Periphytic diatom community

Samples for periphytic diatom community analysis were obtained by scraping mature petioles of the aquatic macrophyte *Eichhornia azurea* (Sw.) Kunth. Two petioles were collected in each sampling site, placed in 150 ml Wheaton bottles and kept cool until further removal of the periphytic biofilm, which was performed using a stainless-steel blade wrapped in aluminium foil and jets of distilled water. After removal, periphyton was fixed and preserved in 5% acetic lugol solution (Bicudo & Menezes, 2006). The area of the scrapped substrate (cm^2) was calculated from length and width measurements of each petiole.

Diatoms were quantified using sedimentation chambers in an inverted microscope following the Utermöhl (1958) method, by counting random fields until reaching at least 100 individuals of the predominant taxon and the stabilization of the species accumulation curve, according to Bicudo (1990). Taxonomic analysis was performed by mounting temporary slides and analysing it in a binocular optical microscope with ocular micrometres at 400× and 1000× magnification (Bicudo & Menezes, 2006). For species identification, material was oxidized according to the technique proposed by Simonsen (1974), modified by Moreira-Filho & Valente-Moreira (1981), and further mounted on permanent slides. Taxa were identified to the lowest taxonomic level possible, using classical literature and regional studies. We adopted the Round et al. (1990) classification system, except for the genus after this work.

2.5. Data analyses

Density (ind cm^{-2}) was calculated from an equation proposed by Ros (1979). Species richness was obtained through quantitative analyses. The similarity between sites were evaluated by cluster analysis with Bray-curtis. Beta diversity (βsor) and its components (βsim and βnes) were calculated according to Baselga & Orme (2012). Those values were calculated through pairwise comparisons between habitats. Differences between components of beta diversity (βsim and βnes) and between stretches of the studied area (first stretch: T1 and P1, and second stretch: P2, T2 and P3) were tested using T-tests. For this analysis, we used the package *betapart* (Baselga et al., 2013) in R software.

All analyses were performed using R software (R Development Core Team, 2014), and graphs using Statistica software v. 7.1 (StatSoft, 2005).

3. Results

Taxonomic analysis allowed the identification of 115 diatom taxa. Higher values of species richness were verified in the tributaries (T1, T2), however, species richness variation was low (Figure 3A).

We registered 18 genus (Figure 3B). *Gomphonema*, *Eunotia* and *Encyonema* were the genus showing higher taxa richness and density (Figure 3B, C). Only five species were common to all environments, *Achnanthidium minutissimum* Kützing complex, *Eunotia incisa* W.Smith ex W.Gregory complex, *Eunotia meridiana* Metzeltin & Lange-Bertalot, *Eunotia pseudosudetica* Metzeltin and *Gomphonema lagenula* Kützing complex.

Some genus were exclusively found in the tributaries, such as *Planothidium* (T1 and T2) and *Pinnularia* (T2), whereas *Neidium* was exclusively found in the Paraná River (P1). Other genus occurred in most sampling sites, such as *Achnanthes* (except at T2) and *Cocconeis* (except at P1) (Figure 3B).

Higher periphytic diatom densities were registered at P1 (Paraná River 1) and P3 (Paraná River 3), sampling sites which were influenced by the tributaries, and lower values were registered at P2 (Paraná River 2) (Figure 3C). A higher density of *Eunotia* was observed in all sampling sites, except at P2, in which *Gomphonema* dominated (Figure 3D). Dominance by these genus were due to the presence of *Eunotia meridiana, E. incisa* and *G. lagenula* at T1 (Amambaí River),

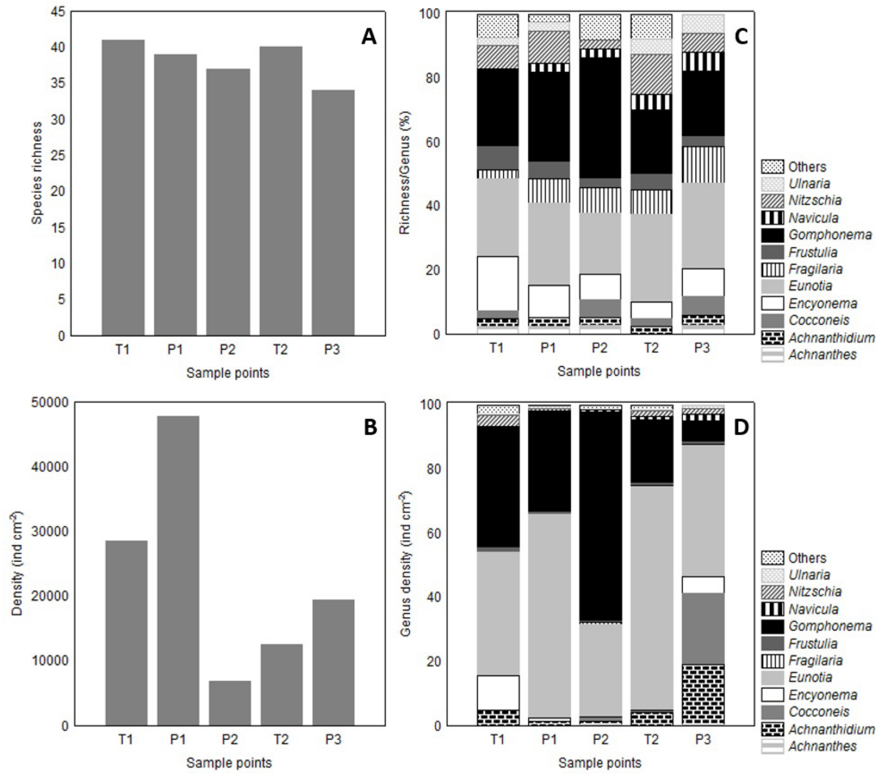

Figure 3. The Bacillariophyta taxa at the sampling sites. (A) Species richness; (B) Relative richness of taxa within each diatom genera; (C) Species density; (D) Relative density of taxa within each diatom genera (T1. Amambaí River; P1. Paraná River 1; P2. Paraná River 2; T2. Iguatemi River; P3. Paraná River 3).

E. pseudosudetica, E. incisa and *G. lagenula* at P1 (Paraná River 1), *E. pseudosudetica, E. incisa* and *Gomphonema angustatum* (Kützing) Rabenhorst at P2 (Paraná River 2), *E. meridiana, E. incisa* and *G. lagenula* at T2 (Rio Iguatemi), *E. incisa* and *E. pseudosudetica* at P3 (Paraná River 3). An increase in the density of *Cocconeis* genus (Figure 3D) was also observed at the last sampling site (P3), and it was related to the species *Cocconeis fluviatilis* J. H. Wallace.

Regarding the other genus, *Encyonema* showed higher densities at T1 (Amambaí River) and *Achnanthidium* at the downstream site P3 (Paraná River 3) (Figure 3D).

Cluster analysis showed that the tributaries (Amambaí-T1 and Iguatemi-T2 rivers) were more similar to each other, regarding diatom composition and density, than other sampling sites located at the Paraná River. Moreover, Paraná River sampling sites directly influenced by the tributaries were more similar to each other than those sites that were not under a direct influence (Figure 4). P2 is the most different site.

Beta diversity values (βsor) varied from 0.57 to 0.75. The first stretch (T1 and P1) showed lower mean values of beta diversity (0.64, standard deviation=0.09) than the second stretch (P2, T1 and P3 mean value 0.67, standard deviation=0.08). Beta diversity partitioning into βsim and βnes evidenced that beta diversity was driven by species turnover (t=13.92, p=0.01, Figure 5).

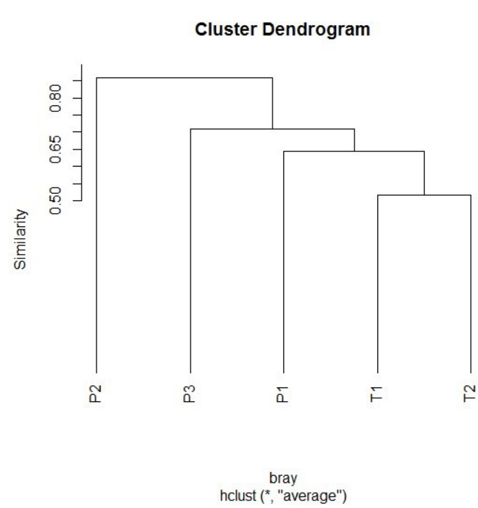

Figure 4. Similarity based on diatom densities between the sampling sites (T1. Amambaí River; P1. Paraná River 1; P2. Paraná River 2; T2. Iguatemi River; P3. Paraná River 3).

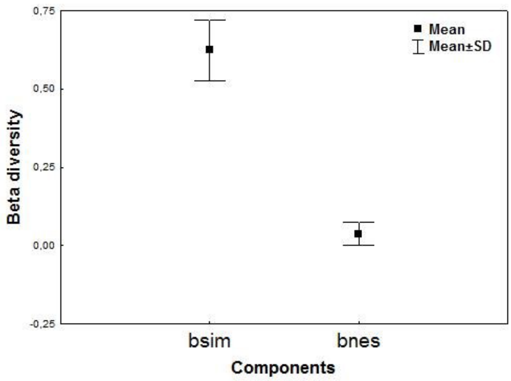

Figure 5. Beta diversity partitioning of diatom communities into turnover (βsim) and nestedness (βnes) components.

4. Discussion

Higher values of diatom species richness were found in the tributaries (T1 and T2), however, species richness variation between sites was low. Species densities were the attribute that most vary in this study, with higher values in sites under direct influence of the tributaries (P1 and P3), and in the tributary (T1). We also observed a higher similarity between the tributaries, which are located in the preserved side of the floodplain, showing similar composition and density of diatoms; whereas P2 was the most different site due this site was not directly influenced by the tributaries. Moreover, results of beta diversity showed that closer sites (T1 and P1) had lower values, and that the species turnover was more important component of beta diversity over nestedness in this floodplain.

Fluctuations in the water level lead to changes in abiotic, and consequently, biotic factors of the system, due to modifications in depth and water velocity (Roberto et al., 2009). Therefore, the intensity and duration of these changes in the water level could be considered as a disturbance to the system, and consequently, to biotic communities, such as the periphytic community (Biggs & Thomsen, 1995; Rodrigues & Bicudo, 2004). In this study, periphytic algae sampling was performed during a period of increased water level, mainly in the downstream sampling sites. In this way, we observed a clear increase in prostrate taxa at the last sampling site, such as *Cocconeis* and *Achnanthidium,* which are considered more resistant to this type of disturbance due to their better adherence properties (Biggs et al., 1998).

In the absence of major floods, floodplain environments regulated by dams are impaired by a decrease in the degree of connectivity; however, adjacent tributaries distributed along the floodplain (preserved environments) have a determinant role in minimizing the effects of anthropogenic activities on the dynamics of those ecosystems, and may help restructuring the environmental conditions and aquatic communities (Braghin et al., 2015). For those reasons, lateral connectivity with those preserved tributaries may serve as a source of resource to the main river channel. Indeed, we registered higher diatom richness in the tributaries, evidencing the importance of those preserved environments as a source of environmental resources and propagules to ecosystem regulation.

Our results revealed that some diatom genera were exclusively found in the tributaries (*Planothidium* and *Pinnularia*). Other genera occurred in almost all sampling sites located upstream (*Cocconeis*). This demonstrates that the establishment of this genera at the downstream sites (P3) is an upstream propagule dispersal result (T1, P1 and T2), further evidencing the relevance of tributaries in structuring periphytic algae along the Paraná River.

High diatom densities found in the Paraná River sampling sites (P1 and P3), which had a greater influence of the tributaries (T1 and T2), were possibly affected by the input of propagules originating from those tributaries. Studies previously reported the direct positive influence of tributaries to the main river in a hydrographic basin (Rice et al., 2001). Considering this stretch of the Paraná River floodplain, tributaries are beneficial to several communities, as a source of food resources and nutrients, and areas for refuge and fish spawning (Ragonha et al., 2014; Braghin et al., 2015). These two tributaries are located in the preserved area, had higher values of total solid materials and nutrients (Santana et al., 2017, in this volume). This also can explain the higher similarity of communities (cluster analysis), and the higher dissimilarity of P2 (Paraná River).

Genera *Eunotia* and *Gomphonema* were predominant in all sampling sites. These genera are considered cosmopolitan, since their species have a wide range of tolerance for various abiotic conditions. *Gomphonema* species are relatively common in freshwater environments (Tremarin et al., 2009). They form branched, mucilaginous stalks attached to solid substrates (Round et al., 1990; Wojtal, 2003), ensuring good substrate adherence, better nutrient exchange and better light capture. *Eunotia* is a genus restricted to freshwaters and usually abundant in the epiphyton and metaphyton of oligotrophic and acid waters (Round et al., 1990). Those organisms are mostly cosmopolitan, however there are some endemic species in tropical and subtropical regions (Metzeltin & Lange-Bertalot, 1998; Bicca & Torgan, 2009).

In addition, results show an interchange in the abundance of genera *Gomphonema* and *Eunotia* at P2 (Paraná River 2), mainly as a function of the establishment of *Gomphonema angustatum.* This species is considered cosmopolitan, with a preference for mesotrophic environments (Krammer & Lange-Bertalot, 1986; Wojtal, 2003), corroborating the results found in this study (Santana et al., 2017, in this volume).

The lower values of beta diversity in closer sites (T1 and P1) show the importance of distance and local forces in the structuring communities (Soininen et al., 2007). This pattern can occur due to the similarity in environmental variables and to dispersion limitation of the organisms.

The diversity partitioning evidenced a species turnover, and that species present in the last stretches are not a subset of upstream communities. High species turnover indicates that established communities are arriving through dispersal from several environments, and not only from specific sites. This demonstrates the importance of conserving several habitats. High species turnover may be provided by environmental restrictions or spatial barriers (Villéger et al., 2013), and in our case it can be provided by the sum of all hydrographic basin downstream influence. A higher contribution of the βsim component (species turnover) in periphytic communities in this floodplain was also demonstrated by Dunck et al. (2016) and for periphytic diatoms by Algarte et al. (2016). Those studies suggest a great alteration occurring in these communities due to flood pulses, and that after Porto Primavera reservoir construction, the diatom community went through major changes, mainly indicating great environmental fluctuations provided by damming in a floodplain.

In summary, our results presented the great importance of the tributaries in the diatom density. The data highlight the great importance of conserving several tributaries, which showed higher values of species richness, and can be source of propagules for the establishment of downstream communities, for maintenance of primary productivity and food source for the trophic web.

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