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Factors affecting assemblage attributes of freshwater Oligochaeta in Neotropical shallow floodplain lakes

Fatores que afetam os atributos da assembleia de Oligochaeta em lagos rasos de planície de inundação Neotropical

Vanessa Ernandes de Amo[1]*, Jéssica Ernandes da Silva[1], Gisele Daiane Pinha[1], Flávio Henrique Ragonha[1] and Roger Paulo Mormul[2]

[1]Programa de Pós-graduação em Ecologia de Ambientes Aquáticos Continentais – PEA, Universidade Estadual de Maringá – UEM, Av. Colombo, 5790, Bloco G90, Jardim Universitário, CEP 87020-900, Maringá, PR, Brazil

[2]Departamento de Biologia – DBI, Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura – NUPELIA, Universidade Estadual de Maringá – UEM, Av. Colombo, 5790, Bloco H90, Jardim Universitário, CEP 87020-900, Maringá, PR, Brazil

*e-mail: vanessaernandes.amo@gmail.com

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Abstract: Aim: Identify the effects of sediment composition and water conditions on diversity, richness, evenness, density and composition of freshwater Oligochaeta in shallow floodplain lakes. **Methods:** We sampled 13 shallow floodplain lakes quarterly during the year 2010 in the Upper Paraná River floodplain. In each lake, four sediment samples were taken from the shore and central regions, three of them were used for biological analysis, and one for granulometric analysis. Concomitantly, temperature, dissolved oxygen, pH, conductivity, alkalinity, turbidity and chlorophyll-a were also measured. Initially, the biological samples were analyzed by a stereoscopic microscope. Oligochaeta individuals were identified under optical microscope at the lowest possible taxonomic level. For data analysis, we quantified density, richness, evenness and diversity index of freshwater Oligochaeta. In order to show differences between the months and the analyzed lakes, in relation to the percentages of coarse and fine organic material, the nonparametric Kruskal Wallis test was used. We also calculated the sediment granulometric diversity using the Shannon-Wienner index, using a simple regression analysis. We correlated assemblage attributes of Oligochaeta with sediment diversity and the assemblage species with the limnological variables using the Spearman correlation. **Results:** A total of 2,090 individuals were found distributed among 27 species. From the total individuals number, 57% were *Pristina americana*, followed by *Dero* (*Dero*) *righii* with 13%. Assemblage attributes were not significantly correlated with sediment diversity, and 7 of the 27 species recorded showed significant correlations with at least some of the abiotic variables. **Conclusions:** We verified that the abiotic variables of the water present greater influence on the attributes of the assemblage of freshwater Oligochaeta, when compared with sediment influences. Although we found low local diversity of this group, the wide range of water conditions offered by lakes culminates with a great species richness considering the whole landscape.

Keywords: diversity; benthic macroinvertebrates; Annelida; sediment heterogeneity.

Resumo: Objetivo: Identificar os efeitos da composição de sedimento e das condições abióticas da água sobre a diversidade, riqueza, equitabilidade, densidade e composição de espécies de Oligochaeta em lagoas rasas de planície de inundação. **Métodos:** Foram amostradas trimestralmente ao longo do ano de 2010, 13 lagoas isoladas na planície de inundação do rio Paraná. Em cada lagoa, foram coletadas quatro amostras de sedimento nas regiões marginais e centrais, sendo três delas destinadas às análises biológicas, e uma para granulométrica. Concomitantemente, também foram mensurados a temperatura, oxigênio dissolvido, pH, condutividade, alcalinidade, turbidez e clorofila. As amostras biológicas foram primeiramente analisadas em microscópio estereoscópico. Os indivíduos de Oligochaeta, foram identificados em microscópio óptico ao menor nível taxonômico possível. Para análise de dados, mensuramos os atributos da assembléia de Oligochaeta, sendo eles a densidade, riqueza, equitabilidade e o índice de diversidade. Para mostrar diferenças entre os meses e os lagoas analisados, em relação às porcentagens de material orgânico grosso e fino, utilizamos o teste não paramétrico Kruskal Wallis. Também calculamos a diversidade granulométrica do sedimento usando o índice de Shannon-Wienner, por meio uma análise de regressão simples. Relacionamos os atributos de Oligochaeta com a diversidade de sedimento e as espécies dessa assembleia com as variáveis limnológicas por meio de correlação de Spearman. **Resultados:** Foram encontrados 2.090 indivíduos distribuídos em 27 espécies, dentre as quais, 57% foram *Pristina americana*, seguida por *Dero* (*Dero*) *righii* com 13%. Os atributos de assembléia não foram significativamente correlacionados com a diversidade de sedimento, e 7 das 27 espécies registradas apresentaram correlações significativas com pelo menos alguma das variáveis abióticas da água. **Conclusões:** Verificamos que as variáveis abióticas da água apresentam maior influência sobre os atributos da assembleia de Oligochaeta, quando comparada com as influências do sedimento. Embora a diversidade local das lagoas rasas seja baixa, a ampla variedade de condições limnológicas oferecida, pode ter levado à maior riqueza de espécies quando se considera toda a paisagem.

Palavras-chave: diversidade; macroinvertebrados bentônicos; Annelida; heterogeneidade do sedimento.

1. Introduction

Floodplains are widely recognized by their high environmental heterogeneity (Thomaz et al., 2007; Lansac-Tôha et al., 2009), that comes from the mosaic of aquatic, terrestrial and transitional habitats (Thomaz et al., 2004). The well-defined aquatic and terrestrial phases produce several adaptations in organisms inhabiting those areas (Junk et al., 1989), but the high spatial heterogeneity of such areas creates conditions that maintain rich natural communities (Siqueira-Souza & Freitas, 2004).

Lakes are very common environments in floodplain areas, which are characterized by high accumulation of mud and organic matter (Knoppers, 1994; Bilia et al., 2015). The content of this and the granulometric composition of the sediment exert great influence on aquatic biodiversity (Súarez et al., 2001; Takeda & Fujita, 2004; Príncipe & Del Corigliano, 2006) for differences in both complexity and environmental heterogeneity among lakes (Pressinatte Junior et al., 2016). The organic matter is important for its great contribution in the diet of most aquatic organisms (Darnell, 1964; Alongi, 1998), which acts as a determinant factor on the species composition and allows the occurrence of different communities among the floodplain lakes (Barreto, 1999; Assis et al., 2004; Townsend et al., 2010).

Benthic invertebrates compose a diverse and abundant community in the Upper Paraná River floodplain (Pinha et al., 2016). These organisms reflect sediment conditions (Bechara, 1996; Takeda & Fujita, 2004) and are strongly influenced by environmental conditions and show reduced mobility in the water (Würdig et al., 2007). Among the benthic invertebrates, Oligochaeta plays an important role representing one of the most abundant group (Takeda, 1999; Ezcurra de Drago et al., 2005; Takeda et al., 2017), which occur in almost all freshwater environments of Neotropical regions (Stevaux & Takeda, 2002; Behrend et al., 2009; Ragonha et al., 2013; Gonzáles, 2015; Flores & Aguirre, 2003).

Due to the potentially high densities of Oligochaeta (Brinkhurst & Jamieson, 1971; Ragonha et al., 2014), its wide distribution (Lafont, 1977; Takeda, 1999), its fully aquatic life cycle, its low mobility and its great capacity to penetrate sediments (Lafont et al., 2007; Ragonha & Takeda, 2014) this group represent an excellent tool for studies involving water management (Brinkhurst & Jamieson, 1971; Callisto et al., 2000). This is particularly important, given that their densities and species composition are indicative of water and sediment quality (Marchese & Drago, 1999; Alves & Strixino 2000). Therefore, the objective of this study was to identify the effects of sediment composition and water abiotic conditions on the assemblage attributes (diversity, richness, evenness, density and composition) of freshwater Oligochaeta in 13 shallow lakes from the upper Paraná River floodplain. Here, we evaluated whether sediment composition and water conditions affect assemblage attributes of Oligochaeta.

2. Material and Methods

2.1. Study area

The study area encompasses 13 lentic lakes, shallow and isolated environments of the Upper Paraná River floodplain (22º45 ‘S and 53º30’ W), in a region located downstream of the engineer Sergio Motta dam and upstream of the Itaipu reservoir. The isolated floodplain lakes are associated to three different rivers: Ivinhema, Baía and Paraná (Figure 1). Lakes differ in relation to surface area, depth, lake levee height, main river channel distance, composition of macrophytes, riparian vegetation coverage, as well as variations in the water physical and chemical characteristics (Thomaz et al., 2004).

2.2. Data sampling

Samplings were carried out quarterly encompassing months of March, June, September and November of the year 2010, in order to consider the different seasons of the year. In each floodplain lake, three regions were determined, in an imaginary line from one margin to another (i.e., two marginal, and one central regions). At each region, four samples were performed with the modified Petersen bottom sampler (0.0345 m^2). Three samples were used for biological analysis and one for granulometric analysis.

All material collected for biological analysis was packed in containers and taken to the Nupélia / Porto Rico-PR field station, where samples were washed in a series of decreasing sieves (2.0 mm, 1.0 mm and 0.2 mm). The invertebrates retained in the first two meshes were immediately fixed in alcohol 80%, to be identified in the laboratory. The sediment retained in the last sieve was fixed with 92.6% alcohol for subsequent screening under a stereoscopic microscope at the laboratory.

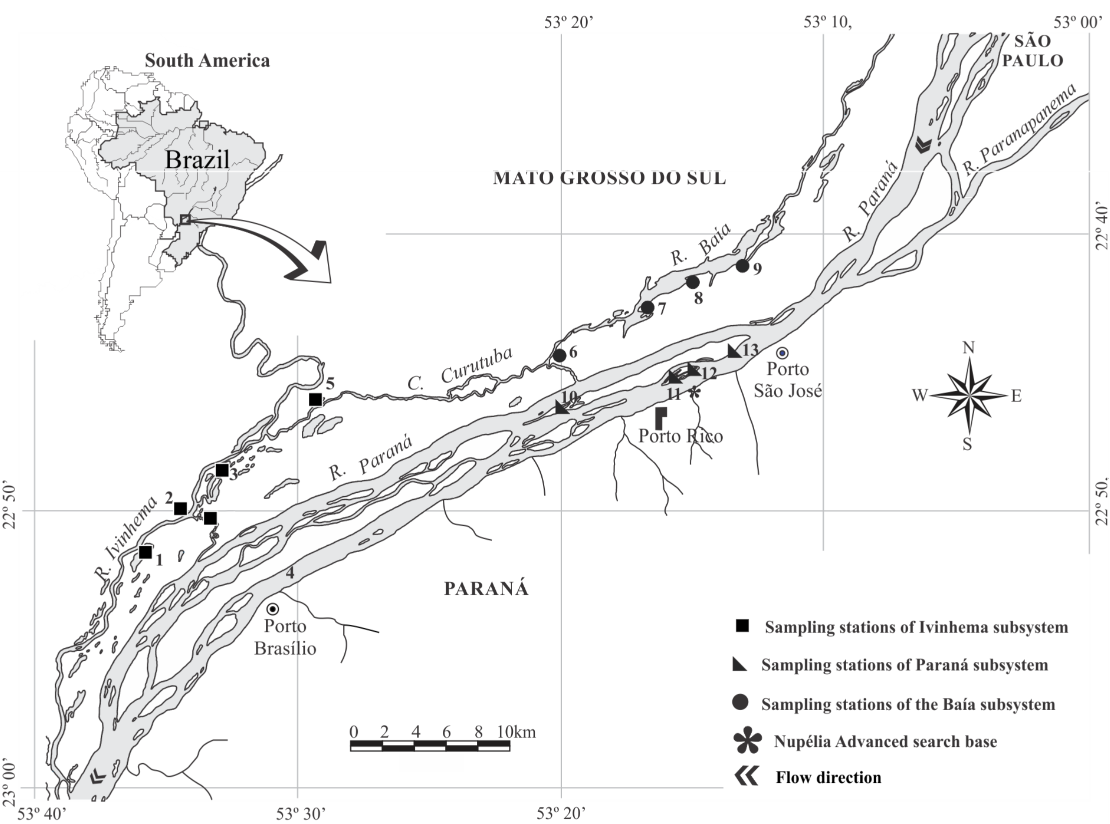

Figure 1. Map of the Upper Paraná River floodpain showing the locations of the 13 isolated floodplain lake sampled: (1) Ventura; (2) Zé do Paco; (3) Capivara; (4) Jacaré; (5) Cervo; (6) Traíra; (7) Fechada; (8) Pousada das Garças; (9) Aurélio; (10) Osmar; (11) Genipapo; (12) Clara and (13) Pousada.

Concomitant to biological samples we took samples of limnological variables such as temperature (°C), dissolved oxygen (mg.l^{-1}), conductivity (µS.cm^{-1}), pH, turbidity (NTU), chlorophyll (µg.l^{-1}) and alkalinity (mEq.l^{-1}). The granulometric composition of each lake sediment was determined using the Wentworth scale (Wentworth, 1922). The estimated organic matter content of the sediment was obtained by the burning of 20g of dry sediment, in a muffle at 560 °C for about four hours. The difference between the initial and final weights indicates the amount of organic matter that was present in the substrate.

2.3. Screening and identification of Oligochaeta species

The screening and counting of Oligochaeta individuals were realized under a stereoscopic microscope. All individuals were organized on slides in glycerin medium and later identified at the lowest possible taxonomic level according to specialized identification key of Brinkhurst & Marchese (1991) and Righi (1984).

2.4. Data analysis

To analyze the assemblage attributes we calculated the density of Oligochaeta (ind.m^{-2}), species richness (S), Shannon-Wiener Diversity Index (Pielou, 1975) and evenness. Shannon index was also used to calculate the sediment diversity.

In order to evaluate differences between months and analyzed lakes, in relation to the percentages of coarse and fine organic material (COM and FOM), the nonparametric Kruskal Wallis test was used. These classes of organic material size were selected due to their predominance in the sampled environments.

In order to verify the relationship between sediment diversity and the species richness, evenness, density and diversity of Oligochaeta, a simple regression analysis was performed. Sediment diversity was calculated using Shannon index, and represents a surrogate of sediment heterogeneity of organic matter composition. The regression was performed in the software Statistica 7.1 (Statsoft, 2005).

To verify the relationship between biological data with environmental variables, Oligochaeta species were correlated with abiotic variables: water temperature (°C), dissolved oxygen, conductivity, pH, turbidity, chlorophyll and alkalinity using Spearman's correlations. All associations were inferred with significant values for $\alpha \leq 0.05$.

3. Results

A total of 27 species was identified among the 2,090 Oligochaeta individuals registered, of which 57% were *Pristina americana* Cernosvitov, 1937, followed by *Dero* (*Dero*) *righii* Varela, 1990, with 13%. Genipapo, Clara and Pousada lakes showed the highest density and species richness (Figure 2). The most common species were *D.* (*D.*) *righii*, *Aulodrilus piguetti* Kowalewski, 1914, *P. americana* and *Chaetogaster diastrophus* Gruithuisen, 1828, while the other species had restricted occurrence. Capivara, Jacaré, Traíra, Fechada and Aurelio floodplain lakes did not present any records of Oligochaeta (Figure 2).

The nonparametric Kruskal Wallis test showed significant differences between the concentration of coarse organic material over the months ($H=17.1802$, $p<0.001$; Figure 3A), and the percentages of this variable were higher in the flood months (March), followed by initiation of flood (December). We observed that in relation to the coarse organic matter the month of March differed from the months of June and September, whereas for the fine organic matter there were no differences between the months. Significant differences were also observed between the lakes (Figure 3B) in relation to both organic matter size classes, coarse organic material (COM - $H=47.316$, $p<0.001$) and fine (FOM - $H=40.0523$, $p<0.001$).

In relation to the coarse and fine organic matter, it was possible to observe some significant relations between the lakes (Table 1). The Traira lake differed from the lakes Aurelio, Clara, Osmar, Pousada das Garças, Ventura and Zé do Paco, and the Capivara lake differed from the lakes Osmar and Zé do Paco, taking into consideration the coarse organic matter. In relation to fine organic matter, it was possible to

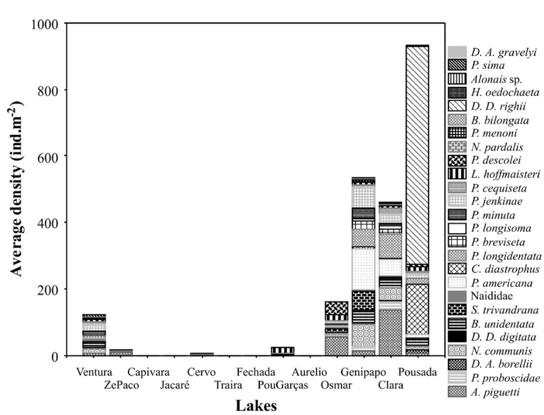

Figure 2. Mean densities of Oligochaeta species in each floodplain lake.

Table 1. Significant results of "z" obtained through analysis of multiple comparisons of the Kruskal Wallis test.

	Aurélio	Capivara	Cervo	Clara	Fechada	Genipapo	Jacaré	Osmar	P.Garças	Pousada	Traíra	Ventura	ZéPaco
Aurélio											3.618		
Capivara								3.416					
Cervo													
Clara											4.033		
Fechada													
Genipapo													
Jacaré													
Osmar											4.505		
P.Garças											3.817		
Pousada													
Traíra				**4.245**		**4.078**						3.816	4.567
Ventura													
ZéPaco											**4.45**		

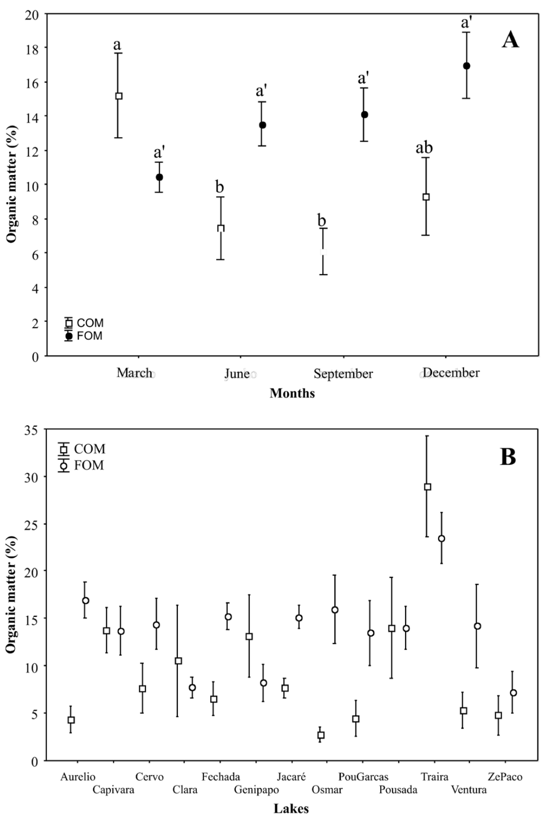

Figure 3. Mean and standard deviation of the organic matter percentage in each months (A) and lake (B) analyzed. COM: Coarse Organic Matter; FOM: Fine Organic Matter.

observe that the Traira lake differed from the Clara, Genipapo and Zé do Paco.

The simple linear regression analysis performed between the Oligochaeta assembly attributes and the sediment diversity showed no significant relationship (Table 2), indicating that the habitat structural components are not the main predictors of the attributes of this assembly in those shallow floodplain lakes. However, Oligochaeta community attributes correlated significantly with some limnological variables (Table 3).

Considering Oligochaeta species individually, Spearman's correlation showed some significant correlations between species and limnological variables (Table 4). It was observed that *Dero* (*Aulophorus*) *borellii* Michaelsen, 1900, and *Bratislavia unidentata* Harman, 1973, had positive correlations with conductivity and alkalinity, whereas the species *Nais pardalis* Piguet, 1906, and *Stephensoniana trivandrana* Aiyer, 1926, had a positive correlation only with chlorophyll. *P. americana* showed positive correlation with conductivity, turbidity, chlorophyll and alkalinity, while *Nais communis* Piguet, 1906, showed a positive correlation with pH and turbidity. *A. piguetti* showed positive correlation with only dissolved oxygen. It was observed that the temperature has negative correlation with some of the Oligochaeta species found.

4. Discussion

We found no correlation between Oligochaeta community attributes and habitat heterogeneity, indicating that habitat structural components are not the main predictor of Oligochaeta assemblage in these shallow floodplain lakes. Through the nonparametric Kruskal Wallis test, it was possible to observe that the concentration of coarse organic matter in the lakes was higher in the flood months (March and December). In the flood months, the sediment present in the unconnected lakes may suffer great influence from the main rivers (Bilia et al., 2015), which changes the substrate type and granulometric composition, factors that influence the presence or absence of benthic invertebrates (Rosin & Takeda, 2007; Rosin et al., 2010). The lakes presented significant differences in relation to the size class of coarse and fine organic matter, due to the dense arboreal vegetation verified in the surroundings of these lakes, in which it allows the increase in the entrance of branches and leaves that become part of the sediment of these bodies of water and mainly with the distance between this lake and the main river (Bilia et al., 2015). Benthic invertebrates use the plant material already in the senescence phase as a food resource (Oertli & Lachavanne, 1995; Mormul et al., 2006), which may explain the increase of the fine organic matter after the flood period.

We found many correlations between abundance of Oligochaeta species and limnological variables, indicating a stronger influence of limnogical factors than habitat components in structuring Oligochaeta assemblage. Some species were negatively correlated with temperature, as also found in studies conducted by Iliopoulou-Georgudaki et al. (2003), Davanso & Henry (2006) and Fulan & Henry (2006). According to Bechara (1996), many species of Oligochaeta are sensitive to chemical changes in the environment such as conductivity, alkalinity, pH and turbidity, and these changes are seasonally present in floodplain due to flooding pulse, which promotes relevant limnological changes (Thomaz et al., 2007).

In relation to the Oligochaeta assemblage, Pristininae, considered a cosmopolitan subfamily (Martin et al., 2008), was the most representative in number of species. Genipapo, Clara and Pousada lakes presented a high density of species, sharing the occurrence of *P. americana*. Although Montanholi-Martins & Takeda (1999) correlate the occurrence of *P. americana* with high values of organic matter, mud and low oxygen conditions, the correlations found here between this species with conductivity, turbidity and chlorophyll exemplifies the ability of this species to survive in a wide range of environmental conditions.

Tubificinae subfamily had *A. piguetti* as the representative with greatest abundance. Although the occurrence of *A. piguetti* is also related to high values of organic matter, mud and low oxygen contents (Montanholi-Martins & Takeda, 2001). *A. piguetti* was dominant in different environments, where occurred high levels of oxygen (e.g., Osmar and Clara lakes) which, similarly to *P. americana,* demonstrates the environmental plasticity of the species. In recent years, *A. piguetti* has become more frequent in several floodplain environments (Petsch et al., 2013; Ragonha et al., 2013; Ragonha & Takeda, 2014). The ability of this species to occur in habitats with very different conditions may be an indication of high environmental tolerance, and these characteristics may have favored the increase of its occurrence throughout the floodplain in the last decades.

In general, many species of Oligochaeta tolerate environments with low concentrations of dissolved oxygen (Fischer & Beeton, 1975). However, *A. piguetti* presented negative correlation with oxygen which suggests a sensibility of these individuals to the conditions of hypoxia and contrasts with some previous pieces of information about this species (e.g., Montanholi-Martins & Takeda, 2001).

N. pardalis, *S. trivandrana* and *P. americana* showed a positive correlation with chlorophyll, showing the dependence of benthic organisms on food resources derived from primary production (Jonasson, 1972; Brinkhurst, 1974). There was no positive correlation between species with

Table 2. Results obtained by the simple linear regression analysis for the attributes of the Oligachaeta assemblage, using as a predictor variable the sediment diversity.

	R^2	GL	F	P
Richness	0.009894	1	0.519924	0.474372
Evenness	0.019596	1	0.039016	0.844234
Diversity Index	0.012186	1	0.398058	0.531026
Density	0.028261	1	2.425088	0.125976

Table 3. Spearman correlation results between the abiotic variables and the Oligochaeta attributes.

Attributes	DO	Cond	Secchi	Alk
Richness	-0.004141	**0.481729**	**-0.339316**	**0.540432**
Evenness	**0.286124**	0.217073	-0.043529	0.176438
Diversity Index	0.063027	**0.465212**	**-0.311155**	**0.513934**
Density	-0.023972	**0.300326**	-0.217825	**0.361260**

DO = Dissolved Oxygen; Cond = Conductivity; Secchi = Secchi disk; Alk = Alkalinity. **Bold numbers** represent significant correlations ($p<0.05$).

Table 4. Correlation results between the abiotic variables and the Oligochaeta species.

Oligochaeta Species	PT	NT	Chloro	Alk	Turb	Cond	pH	DO	Temp	Hsed
A. piguetti	0.178169	**-0.33675**	0.186483	0.197070	-0.11685	0.035273	-0.07865	**-0.37730**	-0.07590	**-0.33942**
D.A. borelli	0.073961	0.215895	0.117696	**0.279223**	0.125124	**0.322966**	0.121008	0.069490	0.109671	0.098842
N. communis	-0.26027	-0.12588	-0.23088	0.057933	**-0.42932**	0.104847	**0.341765**	0.128113	-0.11350	0.081921
B. unindentata	0.054084	-0.01080	-0.04365	**0.389011**	-0.06943	**0.351914**	0.221769	0.111986	-0.18757	-0.03918
S. trivandrana	-0.12830	**-0.34589**	-0.26149	0.026866	-0.20672	-0.00808	-0.03439	0.012860	-0.05471	0.169278
P. americana	-0.19825	**-0.39424**	**-0.32357**	**0.304663**	**-0.42193**	**0.280057**	0.061652	-0.08176	-0.26603	-0.05781
C. diastrophus	-0.05053	-0.14531	-0.17183	**0.279515**	-0.10091	**0.292934**	0.101083	-0.05617	0.005449	**0.282059**
P. longidentata	-0.06761	-0.16161	-0.16872	**0.308095**	-0.19041	0.260613	0.157203	0.181094	**-0.28463**	0.047101
P. breviseta	-0.06351	-0.12573	-0.20480	**0.329617**	**-0.28667**	**0.362594**	0.172114	-0.05736	-0.20402	0.010521
P. minuta	-0.11173	-0.02631	-0.16691	0.203425	-0.17574	0.221331	0.265680	0.168336	**-0.34796**	0.199556
P. jenkinae	-0.03733	-0.22026	-0.14056	**0.424112**	**-0.28258**	**0.370666**	0.168286	0.078797	**-0.30514**	-0.13423
Tubificidae	**0.287761**	**0.369571**	0.239760	-0.09331	**0.418150**	-0.04194	0.038509	0.139493	-0.04021	0.108881
L. hoffmaisteri	**0.340670**	0.052430	0.267419	0.131543	0.212880	0.069810	-0.24381	-0.21221	-0.00778	-0.15578

PT = Total Phosphorus; NT = Total Nitrogen; Chloro = Chlorophyll; Alk = Alkalinity; Turb = Turbidity; Cond = Conductivity; DO = Dissolved Oxygen; Temp = Temperature; and Hsed = sediment diversity. **Bold numbers** represent significant correlations at $p<0.05$.

temperature, although this extrinsic variable being considered the most important for these organisms, mainly for controlling the asexual growth rates of many Oligochaeta species (Judet et al., 1989).

Oligochaeta species reflect the conditions necessary for their occurrence in certain habitats. When some species found their set of ideal conditions in a same place, it tends to favor co-occurrence of species, which increases the diversity, richness and evenness of the assemblage. From this, it was expected that the availability of various sizes of sediment particles and organic particles would favor the diversity of the Oligochaeta assemblage, similar to found by Fomenko (1972), but this idea was not corroborated by our simple linear regression analysis. Through the contradiction between our data and the literature findings, it can be inferred that the limitations imposed by the limnological variables could mask the effects of sediment heterogeneity.

In this study, an ideal set of variables was not identified for the development of a very diverse local assemblage of Oligochaeta, but it can be considered that the wide range of habitats can increase the diversity of these organisms at the landscape level (Williams et al., 2003). Thus, limnological factors were more determinant for the occurrence of Oligochaeta species than the sediment particle size and organic matter composition of the sediment. Despite of showing low values at the local level, the Oligochaeta richness at landscape scale is high, which demonstrate the importance of maintenance of the species at the wide range of lakes of the Upper Paraná River floodplain, where there is a great heterogeneity in limnological variables.

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