

http://www.uem.br/acta ISSN printed: 1679-9283 ISSN on-line: 1807-863X Doi: 10.4025/actascibiolsci.v37i1.22240

Relationship between bacterial density and abiotic factors at different sediment depths of lakes in the Upper Paraná River floodplain

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ABSTRACT. River-floodplain systems are known for their heterogeneity of habitats and the hydrological pulse, the main driving force, which increases nutrient concentrations at the onset of the flood due to leaching from the littoral region and decomposition. This decaying organic matter tends to be deposited in the sediment, where occur biogeochemical processes associated with microorganisms. This study aimed to characterize the vertical distribution of bacterial density in the sediment strata of six environments of the Upper Paraná River floodplain, checking similarity as for bacterial density and physical and chemical conditions. To this end, we analyzed the following factors: total phosphorus, total Kjeldahl nitrogen, levels of organic and inorganic material, oxidation potential and particle size. The results evidenced a gradient of vertical distribution of bacterial density, with predominance of cocci, which possibly indicates no nutrient limitation in this limnic compartment. The analysis of variance was applied to determine significant differences between the layers of the sediment and environments. It can be observed a tendency of deposition of total-P and MI within the upper layers of all environments. More reducing potentials in the initial layers indicate a higher bacterial activity, since this region possesses a greater availability of most easily decomposable material.

Keywords: bacterium, energy resources, phosphorus.

Relação entre densidade bacteriana e fatores abióticos em diferentes profundidades do sedimento em lagoas da planície de inundação do Alto Rio Paraná

RESUMO. Sistemas rios-planície de inundação são conhecidos pela sua heterogeneidade de habitats e pelo pulso hidrológico, principal função de força atuante, que aumenta as concentrações de nutrientes no início devido à lixiviação da região litorânea e à decomposição. Essa matéria orgânica tende a ser depositada no sedimento, onde ocorrem processos biogeoquímicos associados a microrganismos. O objetivo deste estudo foi caracterizar a distribuição vertical da densidade bacteriana em estratos do sedimento de seis ambientes da planície de inundação do Alto Rio Paraná, verificando a similaridade entre os mesmos em relação à densidade bacteriana e condições físicas e químicas. Foram analisados os seguintes fatores: Fósforo total (P-total), nitrogênio total Kjeldahl (NTK), material orgânico (MO) e inorgânico (MI), potencial oxidativo e granulometria. Os resultados mostraram um gradiente de distribuição vertical da densidade bacteriana, com predomínio da forma cocos, possivelmente indicando que não há limitação de nutrientes nos estratos. Análise multivariada mostrou grupamentos por ambientes, ressaltando a grande heterogeneidade de condições limnológicas e de habitats. Pôde-se observar uma tendência de deposição de P-total e MI nas ultimas camadas de todos os ambientes. Potenciais mais redutores nas camadas iniciais indicam maior atividade bacteriana, visto que nesta região há maior disponibilidade material mais facilmente decomponível.

Palavras-chave: bacteria, recursos energéticos, fósforo.

Introduction

In river-floodplain systems, the hydrological pulse, the main driving force, provides heterogeneous aquatic habitats, creating favorable conditions for the maintenance of high biodiversity (JUNK et al., 1989; NEIFF, 1990). On the other hand, the rising water level increases the connectivity between different habitats of the floodplain, favoring the exchange of biological, physical and chemical material, enhancing the similarity between habitats (THOMAZ et al., 2007).

In these systems, the concentrations of nutrients, especially nitrogen and phosphorus, increase early in the flood due to leaching from the littoral region and the decay of aquatic macrophytes (THOMAZ et al., 2004). This decaying organic matter tends to be deposited in the sediment, which comes to be the appropriate site to biogeochemical processes associated with microorganisms (D'HONDT et al., 2004; NELSON et al., 2007), and is crucial to the functioning of aquatic ecosystems (McDONALD et al., 2004).

The high deposition of organic matter in the littoral region of aquatic environments favors a rapid aerobic degradation in water-sediment interface, affecting the biogeochemical cycling of carbon and nutrients (HEDGES et al., 1999), and the process of decomposition and mineralization of this material is carried out by bacteria and supplies nutrients to primary producers (AZAM et al., 1983). Moreover, the bacteria consume a significant part of the total photosynthetic production (BRUM; ESTEVES, 2001).

The littoral sediment of floodplain lakes provides ample habitat for heterotrophic bacteria from the water/sediment interface until greater depths (SHIVAJI et al., 2011). Thus, these organisms contribute significantly to the organic matter cycling in aquatic ecosystems (RHEINHEIMER, 1984; SINSABAUGH et al., 1997).

During the flood occurs entrainment of water sediment with high levels of dissolved organic carbon, accompanied by high extracellular bacterial enzyme activity (BURNS; RYDER, 2001). Studies report that heterotrophic bacteria are more abundant in association with the organic matter (AZAM et al., 1983; GONZALES et al., 2006; KOLM et al., 2007). Being a limiting factor for the growth of heterotrophic microorganisms, organic carbon (O'LOUGHLIN; CHIN, 2004) and nitrogen (SCOW, 1990) are the major forms found in sediment layers deposited more recently.

Studies also reveal that biodegradation generates a large number of ionizable compounds, especially carboxylic and phenolic groups, which are difficult to degrade by microorganisms, and can provide particular characteristics to the aquatic environment 1986; (EPHRAIM; MARINSKY, TIPPING; HURLEY, 1992). In this way, the amount and origin of organic matter in aquatic environments may be decisive for the functioning of these ecosystems (McDONALD et al., 2004) because they influence the distribution of bacterial can communities. Furthermore. under anaerobic conditions, as in sediments, bacteria use humic substances as electron acceptors and energy source during carbon assimilation (COATES et al., 2002).

The greater the amount of decomposable organic material, the greater the activity of microorganisms, especially at the start of leaching (BLUM; MILLS, 1991), and more intense the Santana et al.

reduction processes in (LIIKANEN; MARTIKAINEN, 2003). The intensification of redox processes indicates greater precipitation of ions in more oxidized layers and interruption of the flow of ions from the sediment into the water column (ESTEVES, 2011). Thus, the redox potential can directly influence the flow of ions such as, for example, phosphorus forms (PENG et al., 2007). In particular the availability of phosphorus influences significantly the productivity of these environments (SCHINDLER, 1974).

The goal of this study was to characterize the vertical distribution of bacterial density in the sediment layers of lakes of the Upper Paraná River floodplain in relation to physical and chemical condition of the sediment, checking the similarity between the environments. To this end, the working hypothesis was that bacterial density is directly related to limnological conditions of the sediment, being influenced by a bottom-up process especially in relation to concentrations of TKN and total-P.

Materials and methods

Study area

The lakes selected for this study are located in the Upper Paraná River floodplain (Figure 1), where there are various biotopes formed by different topographic differences, hydrological regime of the main river and local characteristics such as rainfall and wind action, being the last dam-free stretch of the Paraná River, and the main tributaries are Ivinhema and Baía rivers.

The Patos Lake (S22°49'471" W053°33'26.8") is the largest lake sampled in this study, made up of small bays with an average depth of 3.5 m and area of approximately 113.8 ha, situated on the left bank of the Ivinhema River, with which has a communication through a channel with high diversity of macrophytes). The Ventura Lake (S22°51'23.7" W053°36'102") is on the left bank of the Ivinhema River, separated from it by a marginal levee of 3 m, and 200 m far from the river. It has an area of approximately 89.8 ha, average depth of 2.16m (AGOSTINHO et al., 2002).

The Guaraná Lake (S22°90'633" W053°16'5.54"') is located on the right bank of the Baía River and connected to it by a short channel with high density of macrophytes, has rounded shape with area of about 4.2 ha and average depth of 2.1.



1- Patos Lake, 2- Ventura Lake, 3- Guaraná Lake, 4- Fechada Lake; 5- Garças Lake, 6- Osmar Lake).

Figure 1. Location of sampling sites in the Upper Paraná River floodplain. Source: Jaime Luis Lopes Pereira-Nupelia/UEM

The Fechada Lake (S22°42'697"' W053°16'33.06") is on the left bank of the Baía River, inside the floodplain that separates it from the Paraná River, and shows a connection in its upper portion only during flood periods (AGOSTINHO et al., 2002).

The Garças Lake (S22°43'27.18" W053°13'4.56") is located on the right bank of the Paraná River, with direct connection with the channel of the main river through a channel, average depth of 2.0 m and area of 14.1 ha. Its banks are covered by several strata of riparian vegetation and macrophyte stands. The Osmar Lake lies on the Porto Rico Island (S22°46'26.64" W053°19'56.16") on the left bank of the Paraná River, average depth of 1.1 m and area of approximately 0.006 ha. It has elongated shape typical of fluvial islands of the region, and in periods of high waters connects with the channel of the Paraná River through its lower portion (AGOSTINHO et al., 2002).

Field Sampling

In March 2009, sediment samples were taken in triplicate, with the aid of a Corer sediment sampler with transparent acrylic tube, in the littoral region of the lakes Patos, Ventura, Guaraná, Fechada, Garças and Osmar, located in the Upper Paraná River floodplain (Figure 1). After collection, excess water was siphoned off from the collector using hose with 8 mm diameter. Using the same procedure, we siphoned off the water directly in contact with the surface of the sediment (3 cm above the sediment) (sediment-water interface) which was fixed with formaldehyde solution filtered (SHERR; SHERR, 1993).

The sediment from the collector taken from each environment was fractionated every 2 cm deep (0-2, 2-4, 4-6, 6-8, 8-10) to 10 cm, and packed in polyethylene pots, kept under refrigeration and protected from light for further analysis in the laboratory.

The oxidation-reduction potential (Eh) was obtained in situ with a portable digital potentiometer

(Digimed) in each aliquot fractionated. Sediment samples were taken with a modified Petersen grab for particle size analysis.

Also samples were collected in the subsurface of the water column near the littoral region of each environment for analysis of the density of planktonic bacteria and comparison with the vertical strata of the sediment. These samples collected in triplicate were fixed with the same formaldehyde solution filtered (SHERR; SHERR, 1993).

Laboratory

In order to estimate the bacterial biomass and density of total heterotrophic bacteria of the sediment, samples were diluted in deionized water to extract the bacteria following the methodology of Kolm et al. (2007): 15 ml of the supernatant was fixed with formaldehyde solution filtered (SHERR; SHERR, 1993). The water samples of the sediment surface, of the sediment and of the water column were quantified using the same methodology, where an aliquot (0.1 ml) was filtered through 0.2 μ m black polycarbonate membranes (Nuclepore®) stained with DAPI (fluorochrome-4-6-diamidino-2-phenylindole) FEIG, 1980) for (PORTER; subsequent

microscopic analysis. The bacteria were then quantified in an epifluorescence microscope at 1000 x magnification.

For analysis of total Kjeldahl nitrogen (TKN) and total phosphorus (TP) in the sediment, samples were oven dried at 90°C and ground in a porcelain mortar. For analysis of total phosphorus, an aliquot of the dried material was subjected to nitroperchloric digestion and then diluted (ZAGATO et al., 1981), afterphosphorus concentration determined spectroscopically (GOLTERMAN et al., 1978). The total Kjeldahl nitrogen (TKN), was quantifield according by MacKereth et al. 1978.

The amounts of organic matter were measured gravimetrically after incinerated in a muffle furnace at 550°C according to Teixeira et al. (1965). To determine the granulometric texture, samples were dried at 80°C, following the Wentworth scale.

Statistical analysis

For determining the main factors that influence bacterial abundance, we performed a Principal Component Analysis (PCA) which was performed with the mean value of the variables for each pond and each layer. For the selection of axes used in the interpretation we followed the criteria of BrokenStick. In this analysis we used the program PC-ORD (McCUNE; MEFFORD, 1999). To test the observed clusters, a nonparametric analysis of variance (Kruskal-Wallis test) was run with scores of axes selected in the analysis.

Aiming to check for possible correlations and distribution patterns of biota in sediment of environments in the Upper Paraná River floodplain, Spearman correlations were applied between the scores of the selected PCA axes and bacterial density.

The analysis of variance was applied to determine significant differences between the layers of the sediment and the environments (two factor). Analyses of variance, correlations, and graphs were made by the package Statistica 7.1 (STATSOFT, 2005).

Results

The vertical distribution of bacterial density is illustrates by Figure 2 (log-density cm⁻³) with values of bacterioplankton ranging from 7.45 x 10^7 cels mL⁻¹ for the Patos Lake to 1.18 x 10^{10} cels mL⁻¹ for the Ventura Lake. Bacterial density in the water/ sediment interface the values varied between 5.88 x 10^{10} cels mL⁻¹ for the Patos Lake and 1.01 x 10^{13} cels mL⁻¹ for the Ventura Lake. For the sediment between 4.27 x 10^{10} cels mL⁻¹ for the Patos Lake and 6.19 x 10^{12} cels mL⁻¹ also in the Ventura Lake. All lakes showed the same trend of stratification, with a density reduction just below the upper four centimeters of the sediment, highlighting the still Patos lake had the lowest density in all layers.

Among the cells observed, cocci were the predominant form in all strata, with relative density higher than 80% (Figure 3). Among the lakes and layers studied, rods > 0.2 μ m³ were the less dense cell form especially in the upper strata.

The first two axes of the analysis explained 74.0% of the total variability of the data. The axis 1 (39.1%) was negatively influenced by fine sand and very fine sand (particle size less than 0.25 mm) and positively by granules and very coarse sand (particles 1-4 mm). The axis 2 (34.9%) was negatively influenced by TKN and total P, and positively by inorganic material and redox potential (Figure 4). This analysis allowed the visualization of the spatial distribution of the environment, proven by the nonparametric analysis of variance (Kruskal-Wallis test) which revealed significant differences between the lakes sampled (Axis 1: $H_{(5,90)}$: 84.32; Axis 2 $H_{(5,90)}$: 83.46, p < 0.05).

The analysis of variance evidenced that Patos and Guaraná lakes, both connected to the rivers (Ivinhema and Baía, respectively) showed no significant differences to each other with respect



PAT= Patos Lake; VENT= Ventura Lake; GUA= Guaraná Lake; FEC= Fechada Lake; GAR= Garças Lake; OSM= Osmar Lake.

Figure 2. Vertical distribution of bacterial density (log cels mL⁻¹) in plankton, in water-sediment interface and in sediment layers of lakes of the Upper Paraná River floodplain.

to the variables that influenced most the axis 1 (H(5, 30) = 84.32; p < 0.01) (Figure 5-a). The same behavior occurred with Fechada and Garças lakes (Baía and Paraná, respectively) and Patos and Ventura (Ivinhema).

Regarding the variables of greatest influence for axis 2, analysis of variance indicated no significant differences between the lakes Ventura and Garças (Ivinhema and Paraná) and Osmar and Guaraná (Paraná and Baía), even being environments with and without direct communication, respectively, with the main river channel ($H_{(5, 30)} = 83.46$; p < 0.01) (Figure 5-b). The same was observed for Fechada and Osmar lakes, both without direct communication with the main river channel (Baía and Paraná, respectively).

The scores of the PCA were positively correlated with bacterial density (Axis 1: $\rho = 0.26$, p <0.05 and Axis 2: $\rho = 0.52$, p < 0.05). According to the analysis, most influential variables for axis 2, i.e. nutrient concentrations, were the most correlated with the total density of heterotrophic bacteria.

The two factor analysis of variance with the variables measured in the study, concentrations of TKN (from 4.88 mg g^{-1} in the Fechada Lake to 0.36 mg g^{-1} in the Ventura Lake), total-P (from 12.92

mg g⁻¹ in the Patos Lake to 1.50 mg g⁻¹ in the Ventura Lake), content of organic matter (21.41% in the Patos Lake and 3.84% in the Ventura Lake) and inorganic matter (96.15% in the Ventura Lake and 78.59% in the Patos Lake), redox potential (-206 mV in the Fechada Lake and 24.8 mV in the Garças Lake) and bacterial density (log-transformed data) (Figure 6), was performed to check for significant differences between environments and depths of the sediment analyzed. As for concentrations of total-P (Figure 6-a), the lakes Ventura and Osmar showed no significant difference between strata examined (p < 0.05). In the Ventura and Garças lakes, it was observed a decrease in concentrations of phosphorus in the lower layers of the sediment. Unlike, Patos and Fechada lakes were the environments that tended to accumulate total-P (Figure 6a).

The concentrations of total and nitrogen phosphorus in the sediment were higher in the lakes Patos and Fechada (Figure 6a and b). Concentrations of TKN, in the lakes Ventura and Fechada showed no significant differences between the layers (Figure 6b). For the other environments, concentrations were highest in the surface layers, which may indicate a recent deposition of organic nitrogen forms.



Figure 3. Distribution of relative densities of bacterial forms found at different sediment depths; \blacksquare =Vibrio; \blacksquare =Rod > 0.2 μ m³; \blacksquare = Cocci > 0.05 μ m³; \Box Cocci < 0.05 μ m³.

In the analysis of organic and inorganic matter at different layers of the sediment (Figure 6c and d), for all environments, the inorganic matter had higher values, with a nearly homogeneous distribution between the different depths. However in the Patos Lake, we verified a trend of deposition of this matter. The same behavior was registered for the values of organic matter, the Ventura Lake was the most distinct environments, with the lowest value.

The environments were statistically different from each other for the values of redox potential (Eh) of the sediment. The Garças Lake presented more oxidized sediment (higher values of Eh), and the Fechada Lake more reduced sediment (lower values of Eh) (Figure6e). This may suggest that in this latter environment there was a greater amount of decomposable organic material, thus favoring a higher activity of microorganisms, compared with other environments. Another factor is the tendency of lakes Ventura, Guaraná and Fechada in presenting lower oxidative potential in the deeper layers of the sediment.



PAT= Patos Lake; VENT= Ventura Lake; GUA= Guaraná Lake; FEC= Fechada Lake; GAR= Garças Lake; OSM= Osmar Lake.

Figure 4. Scatterplot of the principal components analysis (a) and (b) ordination of the environments of the Upper Paraná River floodplain, according to the sediment.



PAT= Patos Lake; VENT= Ventura Lake; GUA= Guaraná Lake; FEC= Fechada Lake; GAR= Garças Lake; OSM= Osmar Lake.

Figure 5. Nonparametric analysis of variance (Kruskal-Wallis test), with the scores selected for interpretation from the principal component analysis. (a) principal component 1 and (b) principal component 2.

Significant differences were detected between the environments and the sediment layers for bacterial density (Figure 6f). In all lakes, the greatest densities were observed in the upper layers (0-4 cm). It is worth noting the Patos Lake as the environment with lower bacterial densities.

In all environments, concentrations of total-P in the sediment analyzed were greater than NTK (mean: 8.06 mg g⁻¹ total-P, 2.69 mg g⁻¹ TKN). For Patos and Garças lakes, concentration of total-P was approximately 2.5 times greater than total-N and in the Osmar Lake, only 1.1 (Figure 6g).

Discussion

Rheinheimer et al. (1989) studied marine environments and reported higher bacterial activity in compartments with more favorable conditions for theirdevelopment, like oxygen and organic matter. This was corroborated in the present study by the vertical distribution of heterotrophic bacteria in sediments of different environments of the Upper Paraná River floodplain. In these environments, characterized by high availability of organic matter, the aerobic oxidation prevails at high rates in the surface layers of the sediment, providing carbon, nitrogen, and phosphorus, resulting in a more efficient combination for bacterial productivity (FARJALA et al., 2002). Even in the absence of oxygen, bacteria are in high density for being organisms that are extremely versatile in energy uptake (NEALSON, 1997).

In the present study we detected higher bacterial density in the sediment-water interface and in vertical strata, compared to the water column. This is



PAT= Patos Lake; VENT= Ventura Lake; GUA= Guaraná Lake; FEC= Fechada Lake; GAR= Garças Lake; OSM= Osmar Lake.

Figure 6. Two factor analysis of variance between sampled environments and sediment depths: O = 0.2 cm; $\Box = 2.4 \text{ cm}$; $\bullet = 4.6 \text{ cm}$; $\blacksquare = 6.8 \text{ cm}$; $\blacktriangle = 8-10 \text{ cm}$; because the sediment presents higher concentrations of organic carbon, which stimulates bacterial growth (SHIVAJI et al., 2011) besides having high diversity (FENG et al., 2009) and activity of bacteria (GANTZER; STEFAN, 2003).

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In all environments studied, the highest densities of bacteria found in the sediment-water interface are due to constant exchanges between these compartments, and to conditions of oxygenation promoted by the water column. According to Liikanen and Martikainen (2003) this region is extremely favorable to the development of heterotrophic bacteria. The highest values of bacterial density obtained at the surface layers (0-4 cm) followed by a marked decrease in density toward the deeper layers can be a result of the transition from an oxide environment to an anoxic condition, which occurs in a vertical profile (GAST; GOCKE, 1988; SHIVAJI et al., 2011).

The predominance of small sized coccoid bacteria in all environments and sediment layers suggests that bacterial growth in the studied environments is not limited by nutrient availability, corroborating studies of Sigee (2005) and Øvreas et al. (2003). Another factor that enables the dominance of this cell shape is the surface/volume ratio, which directly involves the absorption capacity of nutrients (SIGEE, 2005).

Despite high bacterial densities found in different environments, the Patos Lake differed from others by low values in all samples and layers. This assumes the occurrence of a relationship between quantity and quality of carbon once Azevedo et al. (2008) observed that this water environment presents a great amount of allochthonous carbon, especially fulvic acids, both in the water column and sediment. Furthermore, studies report that increased bacterial densities are obtained in the presence of more labile carbon (TEIXEIRA et al., 2011).

The clear separation of the environments indicated by principal component analysis, proven by analysis of variance, points out that although environments belonged to the same sub-basin, local factors may be more important for the dynamics and functioning (ROCHA; THOMAZ, 2004). Thus, these peculiarities have high relevance contributing to the maintenance of high diversity in riverfloodplain systems (AGOSTINHO et al., 2000).

Moreover, the sediment grain size seems to play an important role in the structure of environments, as evidenced by the results of the lakes Fechada and Garças, which were similar as for the scores of the axis 1 of the principal component analysis, which was affected by this variable, even differing in the degree of communication (with and without communication, respectively) with the main river (Baía and Paraná).

The weak correlation between bacterial density and the scores of the first axis of the principal component analysis corroborates the idea that bacterial activity is more intense in the presence of finer particles (JACKSON; VALLAIRE 2007). This probably occurs due to the presence of more labile organic compounds in a certain stage of decomposition (BERTILSSON; TRANVIK 2000) which becomes easily assimilated by bacteria (D'HONDT et al., 2004; TEIXEIRA et al., 2011).

As both PCA axis were positively correlated with bacterial density, the results indicate dependence of the increase in the number of cells on physical and chemical properties of the sediment, as reported by Coveney and Wetzel (1992) and Rejas et al. (2005), which reported a limitation of bacterial growth by the concentration of inorganic nutrients. However, the data did not determine what the limiting factor for the bacterial growth in the sediment of studied environments.

According to Toolan et al. (1991) and Morris and Lewis Jr. (1992) the availability of nutrients can control the bacterial density. Therefore, the distinction between the environments especially in relation to the concentrations of TKN and total-P may be due to the biodegradation and influence of decomposition, as there are differences in the release of nutrients depending on the stage of the process (PADIAL; THOMAZ 2006). This process originates many compounds difficult to degrade bv microorganisms, and can provide particular characteristics to aquatic environments (EPHRAIM; MARINSKY, 1986; TOOLAN et al., 1991).

There was a tendency for deposition of concentrations of total phosphorus in the deeper layers of the sediment (6-10 cm), except for the Garças Lake. The cycling of this element, limiting factor in the productivity of aquatic environments (SCHINDLER, 1974), is ruled by the physical and chemical environment at the sediment-water interface and by the activity of microorganisms in the sediment (GACHTER; MEYER, 1993), thus indicating conditions that contribute to the conservative characteristics of this compartment. The lowest concentrations of this nutrient were observed in environments that showed smaller particle size, contradicting the idea that larger particles would facilitate the release of this nutrient to the water column (GAINSWIN et al., 2006).

Another factor was the low availability of TKN in different sediment layers and environments, resulting in a low N:P ratio (about 2 times more phosphorus than nitrogen). It should be emphasized that this ratio is only an estimate, since for such a relationship were not obtained inorganic forms of nitrogen.

The low availability of NTK, which is the organic and ammonia fraction of nitrogen forms, can be explained by the preferential use of amino acids by bacterial community during the warmer season (sampling period) (CHRISTIAN; LIND 2007). This leads to a reduction in the availability of nitrogen forms, mainly in regions of greater bacterial density, the first few centimeters of the sediment in this study.

The environments that had higher nutrient concentrations showed more reducing redox potential (Eh) in the sediment, thus obtaining a negative correlation between Eh and concentrations of nutrients (total-P and TKN). This indicates that the deposition of organic matter favors the development of organisms that utilize the energy involved in the oxidation process of this material, thus releasing metal ions to the water column. Liikanen and Martikainen (2003) reported that the smaller the environment, possibly more intense is the microbial activity, thus explaining the correlation obtained. Probably this may have occurred in the Garças Lake, which showed more oxidative potential and high bacterial densities. Yet this lake has a high degree of connectivity with the Paraná River, also presenting high levels of dissolved oxygen in the water column (ROBERTO et al., 2009).

Conclusion

In short, our results revealed that the predominance of coccoid bacterial forms may indicate the high availability of energy resources to heterotrophic bacteria in the sediment of lakes in the Upper Paraná River floodplain. Bacterial density was correlated with the physical and chemical properties of the sediment; moreover, high bacterial density especially in the surface layers can interfere with the values of redox potential, and with the deposition and/or uptake of nutrients in this region. It was also noted evidence of deposition of phosphorus forms mainly in the lower layers. Moreover, the spatial distribution of the environments studied emphasized once again the high heterogeneity of habitats present in river-floodplain systems.

Acknowledgements

To Nupélia - Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura for the support, to PEA-UEM, to Capes, for the scholarship. The authors are tankful to Gazo, Tato, João Latini, for help in field.

Referências

AGOSTINHO, A. A.; THOMAZ, S. M.; MINTE-VERA, C. V.; WINEMILLER, K. O. Biodiversity in the high Paraná River floodplain. In: GOPAL, B.; JUNK, W. J.; DAVIS, J. A. (Ed.). **Biodiversity in wetlands**: assessment, function and conservation. Leiden: Backhuys Publishers, 2000. p. 89-118. Santana et al.

AGOSTINHO, A. A.; THOMAZ, S. M.; RODRIGUES, L.; GOMES, L. C. **A planície de inundação do alto rio Paraná**: Site 6 PELD/CNPq. Maringá: Nupélia/ PELD/CNPq, 2002. (Relatório anual).

AZAM, F.; FENCHEL, T.; FIELD, J. G.; GRAY, J. S.; MEYER-REIL, L. A.; THINGSTAD, F. The ecological role of water-column microbes in the sea. **Marine Ecology Progress Series**, v. 10, p. 257-263, 1983.

AZEVEDO, J. C. R.; TEIXEIRA, M. C.; SANTOS, A. M.; LEANDRINI, J. A.; PAGIORO, T. A. Caracterização espectroscópica da matéria orgânica dissolvida da planície de inundação do Alto Rio Paraná. **Oecologia Brasilienses**, v. 12, n. 1, p. 66-77, 2008.

BERTILSSON, S.; TRANVIK, L. J. Photochemical transformation of dissolved organic matter in lakes. **Limnology and Oceanography**, v. 45, n. 4, p. 753-762, 2000.

BLUM, L. K.; MILLS, A. L. Microbial growth and activity during the initial stages of seagrass decomposition. **Marine Ecology Progress Series**, v. 70, p. 73-82, 1991.

BRUM, P. R.; ESTEVES, F. A. Changes in abundance and biomass of the attached bacterial community throughout the decomposition of three species of aquatic macrophytes. In: FARIA, B. M.; FARJALLA, V. F.; ESTEVES, F. A. (Ed). **Aquatic microbial ecology in Brazil**. Rio de Janeiro: UFRJ, 2001. p. 77-96.

BURNS A.; RYDER D. S. Response of bacterial extracellular enzymes to inundation of floodplain sediments. **Freshwater Biology**, v. 46, n. 10, p. 1299-1307, 2001.

CHRISTIAN, B. W.; LIND, O. T. Multiple carbon substrate utilization by bacteria at the sediment-water interface: seasonal patterns in a stratified eutrophic reservoir. **Hydrobiologia**, v. 586, p. 43-56, 2007.

COATES, J. D.; COLE, K. A.; CHAKRABORTY, R.; O'CONNOR, S. M.; ACHENBACH, L. A. Diversity and ubiquity of bacteria capable of utilizing humic substances as electron donors for anaerobic respiration. **Applied and Environmental Microbiology**, v. 68, n. 5, p. 2445-2452, 2002.

COVENEY, M. F.; WETZEL, R. G. Effects of nutrients on specific growth rate of bacterioplankton in oligotrophic lake water culturest. **Applied and Environmental Microbiology**, v. 58, n. 1, p. 150-156, 1992.

D'HONDT, S.; JORGENSEN, B. B.; MILLER, D. J.; BATZKE, A.; BLAKE, R.; CRAGG, B. A.; CYPIONKA, H.; DICKENS, G. R.; FERDELMAN, T.; HINRICHS, K. U.; HOLM, N. G.; MITTERER, R.; SPIVACK, A.; WANG, G. Z.; BEKINS, B.; ENGELEN, B.; FORD, K.; GETTEMY, G.; RUTHERFORD, S. D.; SASS, H.; SKILBECK, C. G.; AIELLO, I. W.; GUERIN, G.; HOUSE, C. H.; INAGAKI, F.; MEISTER, P.; NAEHR, T.; NIITSUMA, S.; PARKES, R. J.; SCHIPPERS, A.; SMITH, D. C.; TESKE, A.; WIEGEL, J.; PADILLA, C. N.; ACOSTA, J. L. S. Distributions of microbial activities in deep subseafloor sediments. **Science**, v. 306, n. 5705, p. 2216-2221, 2004.

EPHRAIM, J.; MARINSKY, J. A. A unified physicochemical description of the protonation and metal ion complexation equilibria of natural organic acids

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(humic and fulvic acids). 3. Influence of polyelectrolyte properties and functional heterogeneity on the copper ion binding equilibria in an Armadale Horizons Bh Fulvic Acid Sample. **Environmental Science Technolology**, v. 20, n. 4, p. 367-376, 1986.

ESTEVES F. A. **Fundamentos de limnologia**. 3. ed. Rio de Janeiro: Interciência, 2011.

FARJALA, V. F.; ESTEVES, F. A.; BOZELLI, R. L.; ROLAND, F. Nutrient limitation of bacterial production in clear water Amazonian. **Hydrobiologia**, v. 489, n. 1-3, p. 197-205, 2002.

FENG, B. W.; LI, X. R.; WANG, J. H.; HU, Z. Y.; MENG, H.; XIANG, L. Y.; QUAN, Z. X. Bacterial diversity of water and sediment in the Changjiang estuary and coastal area of the East China Sea. **FEMS Microbiology Ecology**, v. 70, n. 2, p. 236-248, 2009.

GÄCHTER, R.; MEYER, J. S. The role of microorganisms in mobilization and fixation of phosphorus in sediments. **Hydrobiologia**, v. 253, n. 1-3, p. 103-121, 1993.

GAINSWIN, B. E.; HOUSE, W. A.; LEADBEATER, B. S. C.; ARMITAGE, P. D.; PATTEN, J. The effects of sediment size fraction and associated algal biofilms on the kinetics of phosphorus release. **Science of the Total Environment**, v. 360, n. 1-3, p. 142-157, 2006.

GANTZER, C. J.; STEFAN, H. G. A model of microbial activity in lake sediments in response to periodic watercolumn mixing. **Water Research**, v. 37, n. 12, p. 2833-2846, 2003.

GAST, V.; GOCKE, K. Vertical distribution of number, biomass and size-class spectrum of bacteria in relation to oxic/anoxic conditions in the Central Baltic Sea. **Marine Ecology Progress Series**, v. 45, p. 179-186, 1988.

GOLTERMAN, H. L.; CLYMO, R. S.; OHMSTAD, M. A. M. Methods for physical and chemical analysis of fresh water. Oxford: Blackwell Scientiic, 1978.

GONZALES, A. M.; PARANHOS, R.; LUTTERBACH M. S. Heterotrophic bacteria abundance in Rodrigo de Freitas lagoon (Rio de Janeiro, Brazil). **Brazillian Journal of Microbiology**, v. 37, n. 4, p. 428-433, 2006.

HEDGES, J. I.; HU, F. S.; DEVOL, A. H.; HARTNETT, H. E.; TSAMAKIS, E.; KEIL, R. G. Sedimentary organic matter preservation: a test for selective degradation under oxic condictions. **American Journal of Scienc**, v. 299, n. 7-9, p. 529-555, 1999.

JACKSON, C. R.; VALLAIRE, S. C. Microbial activity and decomposition of fine particulate organic matter in a Louisiana cypress swamp. **Journal of the North American Benthological Society**, v. 26 n. 4, p. 743- 754, 2007.

JUNK, W. J.; BAYLEY, P. B.; SPARKS, R. E. The flood pulse concept in river-floodplain systems. In: DODGE, D. P. (Ed.). Proceedings of the International large Rivers Symposium (LARS). **Canadian Special Publication**, **Fish Aquatic Sciences**, v. 106, p. 110-127, 1989.

KOLM; H. E.; SANTOS, P. R. N. M.; SAUTTER, K. D. Bacteria in water and sediments of Guaratuba bay, Paraná, Brazil; **Tropical Oceanography**, v. 35, n. 1, p. 51-69, 2007.

LIIKANEN, A.; MARTIKAINEN, P. J. Effect of ammonium and oxygen on methane and nitrous oxide fluxes across the sediment–water interface in a eutrophic lake. **Chemosphere**, v. 52, n. 8, p. 1287-1293, 2003.

MacKERETH, F. Y. H.; HERON, J.; TALLING, J. F. Water analysis: some revised methods for limnologists. **Freshwater Biological Association**, v. 36, p. 1-120, 1978.

McCUNE, B.; MEFFORD, M. J. PC-ORD. **Multivariate analysis of ecological data, version 4.0**. Gleneden Beach: MjM Software Design, 1999.

McDONALD, M.; BISHOP, A. G.; PRENZLER, P. D.; ROBARDS, K. Analytical chemistry of freshwater humic substances. **Analytica Chimica Acta**, v. 527, n. 2, p. 105-124, 2004.

MORRIS, D. P.; LEWIS JR., W. M. Nutrient limitation of bacterioplankton growth in Lake Dillon, Colorado. **American Society of Limnology and Oceanography**, v. 37, n. 6, p. 1179-1192, 1992.

NEALSON, K. H. Sediment bacteria: who's there, what are they doing, and what's new? **Annual Review of Earth and Planetary Sciences**, v. 25, p. 403-434, 1997.

NEIFF, J. J. Ideas para la interpretacion ecologica del Paraná. **Interciência**, v. 15, n. 6, p. 424-441, 1990.

NELSON, D. M.; OHENE-ADJEI, S.; HU, S. F.; CANN, I. K. O.; MACKIE, R. I. Bacterial diversity and distribution in the Holocene Sediments of a northern temperate lake. **Microbial Ecology**, v. 54, n. 2, p. 252-263, 2007.

O'LOUGHLIN, E. J.; CHIN, Y. P. Quantification and characterization of dissolves organic carbon and iron in sedimentary porewater from Green Bay, WI, USA. **Biogeochemistry**, v. 71, n. 2, p. 371-386, 2004.

ØVREAS, L.; BOURNE, D.; SANDAA, R. A.; CASAMAIOR, E. O.; BENLLOCH, S.; GODDARD, V.; SMERDON, G.; HELAS, M.; THINGSTAD, T. F. Response of bacterial and viral communities to nutrient manipulations in seawater mesocosms. **Aquatic Microbial Ecology**, v. 31, n. 2, p. 109-121, 2003.

PADIAL, A. A.; THOMAZ, S. M. Effects of flooding regime upon the decomposition of *Eichhornia azurea* (Sw) Kunth Measured on a tropical flow-regulated floodplain (Paraná river, Brazil). **River Research and Applications**, v. 22, n. 7, p. 791-801, 2006.

PENG, J. F.; WANG, B. Z.; SONG, Y.; YUAN, P.; LIU, Z. Adsorption and release of phosphorus in the surface sediment of a wastewater stabilization pond. **Ecological Engineering**, v. 31, n. 2, p. 92-97, 2007.

PORTER, K.; FEIG, Y. S. The use of DAPI for identifying and counting aquatic microflora. **Limnology Oceanography**, v. 25, n. 5, p. 943-948, 1980.

REJAS, D.; MUYLAERT, K.; DE MEESTER, L. Phytoplankton-bacterioplankton interactions in a neotropical floodplain lake (Laguna Bufeos, Bolívia). Hydrobiologia, v. 543, n. 1, p. 91-99, 2005.

RHEINHEIMER, G. Bacterial ecology of the North and Baltic Seas. **Botanica Marina**, v. 27, n. 1, p. 277-299, 1984.

RHEINHEIMER, G.; GOCKE, K.; HOPPE, H.-G. Vertical distribution of microbiological and hydrographicchemical parameters in different areas of the Baltic Sea. Marine Ecology Progress Series, v. 52, n. 1, p. 55-70, 1989.

ROBERTO, M. C.; SANTANA, N. F.; THOMAZ, S. M. Limnology in the Upper Paraná River floodplain: large-scale spatial and temporal patterns, and the influence of reservoirs. **Brazilian Journal of Biology**, v. 69, n. 2-Suppl., p. 631-637, 2009.

ROCHA, R. R. A.; THOMAZ, S. M. Variação temporal de fatores limnológicos em ambientes da planície de inundação do alto rio Paraná (PR/MS – Brasil). Acta Scientiarum. Biological Sciences, v. 26, n. 3, p. 261-271, 2004.

SCHINDLER, D. W. Eutrophication and recovery in experimental lakes: implications for lake management. **Science**, v. 184, n. 4139, p. 897-899, 1974.

SCOW, K. M. Rate of biodegradation. In: LYMAN, W. J.; REEHL, W. F; ROSENBLATT, D. H. (Ed.). **Handbook of chemical propriety estimation methods**. Washington: American Chemical Society, 1990. Chapter 9.

SHERR, E. B.; SHERR, B. F. Preservation and storage of samples for enumeration of heterotrophic protests. In: KEMP, P. F.; SHERR, B. F.; SHERR, E. B.; COLE, J. J. (Ed.). Handbook of methods in Aquatic Microbial Ecology. Florida: Boca Raton, 1993. p. 207-212.

SHIVAJI, S.; KUMARI, K.; KISHORE, K. H.; PINDI. P. K.; RAO, P. S.; SRINIVAS, T. N. R.; ASTHANA, R.; RAVINDRA, R. Vertical distribution of bacteria in a lake sediment from Antarctica by culture-independent and culture-dependent approaches. **Research in Microbiology**, v. 162, n. 2, p. 191-203, 2011.

SIGEE, D. C. Freshwater microbiology biodiversity and dynamic interactions of microorganisms in the aquatic environment. Manchester: John Wiley Sons LTD., 2005.

SINSABAUGH, R. L.; FINDLAY S.; FRANCHINI, P.; FISCHER, D. Enzymatic analysis of riverine bacterioplankton production. Limnology and Oceanography, v. 42, n. 1, p. 29-38, 1997.

STATSOFT. **Statistica for Windows**. Version 7.1. Tulsa: Statsoft Inc., 2005.

TEIXEIRA, C.; TUNDIZI, J. G.; KUTNER, M. B. Plankton studies in a mangrove. II: the standing-stock

and some ecological factors. **Boletim do Instituto Oceanografico**, v. 24, n. 1, p. 23-41, 1965.

TEIXEIRA, M. C.; SANTANA, N. F.; AZEVEDO, J. C. R.; PAGIORO, T. A. Bacterioplankton features and its relations with DOC characteristics and other limnological variables in Paraná river floodplain environments (PR/MS-Brazil). **Brazillian Journal of Microbiology**, v. 42, n. 3, p. 897-908, 2011.

THOMAZ, S. M.; CHAMBERS, P. A.; PIERINI, S. A.; PEREIRA, G. Effects of phosphorus and nitrogen amendments on the growth of Egeria najas. **Aquatic Botany**, v. 86, n. 2, p. 191-196, 2007.

THOMAZ, S. M.; PAGIORO, T. A.; BINI, L. M.; ROBERTO, M. C.; ROCHA, R. R. A. Limnological characterization of the aquatic environments and the influence of hydrometric levels. In: THOMAZ, S. M.; AGOSTINHO, A. A.; HAHN, N. S. (Ed.). **The upper Paraná river and its floodplain**: physical aspects, ecology and conservation. Leiden: Backhuys Publishers, 2004. p. 75-102.

TIPPING, E.; HURLEY, M. A. A unifying model of cation binding by humic substances. **Geochimica et Cosmochimica Acta**, v. 56, n. 10, p. 3627-3641, 1992.

TOOLAN, T.; WEHR, J. D.; FINDLAY, S. Inorganic phosphorus stimulation of bacterioplankton production in a meso-eutrophic lake. **Applied and Environmental Microbiology**, v. 57, n. 7, p. 2074-2078, 1991.

ZAGATO, E. A. G.; JACINTO, A. O.; REIS, B. F.; KRUG, F. G.; BERGAMIN, F. H.; PESSENDA L. C. R.; MORTATTI, J.; GINÉ, M. F. Manual de análises de plantas e águas empregando sistema de injeção de fluxo. Piracicaba: Universidade de São Paulo, Centro de Energia Nuclear na Agricultura, 1981.

Received on October 23 2013. Accepted on April 30 2014.

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