

## OCOLOGIA BRASILIENSIS

Gianotti, E.P. & Santos, J.E. 2001. Denitrification in a oxbow lake in tropical floodplain river. pp. 13-20. In: Faria, B.M.; Farjalla, V.F. & Esteves, F.A. (eds). *Aquatic Microbial Ecology in Brazil*. Series Oecologia Brasiliensis, vol. IX. PPGE-UFRJ. Rio de Janeiro, Brazil.

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### DENITRIFICATION IN A OXBOW LAKE IN TROPICAL FLOODPLAIN RIVER

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#### Resumo

Foram estimadas as taxas máximas de desnitrificação (método do bloqueio pelo acetileno) em “condições reais” (nitrito endógeno) e em “condições potenciais” (fornecimento suplementar de nitrito) para sedimento (regiões litorânea e limnética) e raízes de macrófita aquática *Scirpus cubensis* (região litorânea) da Lagoa do Infernã, na perspectiva de avaliar qualitativa e quantitativamente o significado ecológico do processo de desnitrificação para o sistema ambiental em questão. Em “condições reais” as taxas de desnitrificação para sedimentos variaram de  $5,45 \cdot 10^{-10}$  a  $2,66 \cdot 10^{-9}$  moles  $N_2O/g/h$  na Estação 1, e de  $4,35 \cdot 10^{-10}$  a  $1,96 \cdot 10^{-9}$  moles  $N_2O/g/h$  para Estação 2. Em “condições potenciais” as taxas para sedimento variaram de (Estação 1)  $3,77 \cdot 10^{-10}$  a  $1,35 \cdot 10^{-7}$  moles  $N_2O/g/h$  de (Estação 2)  $1,06 \cdot 10^{-9}$  a  $5,39 \cdot 10^{-8}$  moles  $N_2O/g/h$ . Em raízes de *Scirpus cubensis* as taxas variaram de  $1,73 \cdot 10^{-7}$  a  $1,08 \cdot 10^{-6}$  moles  $N_2O/g/h$ . Maiores valores de taxas de desnitrificação e do número de bactérias desnitrificantes foram observados durante o período chuvoso, provavelmente em decorrência do aporte de matéria orgânica e de nutrientes, associado a ocorrência do período hidrológico na planície de inundação do Rio Mogi-Guaçu.

Palavras-chave: desnitrificação; sedimento; macrófitas aquáticas; redução do acetileno.

#### Abstract

The qualitative and quantitative importance of the denitrification process in sediment and aquatic macrophyte in an oxbow lake in tropical floodplain river have been assessed. The denitrification rate was estimated using acetylene to block reduction of  $N_2O$  to  $N_2$  and denitrifying bacteria numbers were estimated by the Most Probable Number technique. The denitrification rates at “real condition”, taking into account endogenous nitrogen, ranged from  $5.45 \cdot 10^{-10}$  to  $2.66 \cdot 10^{-9}$  moles  $N_2O/g/h$  for sediment (sampling station 1) and  $4.35 \cdot 10^{-10}$  to  $1.96 \cdot 10^{-9}$  moles  $N_2O/g/h$  for sampling station 2. The “potential” denitrification rate, with supplementary nitrate, ranged from  $3.77 \cdot 10^{-10}$  to  $1.35 \cdot 10^{-7}$  moles  $N_2O/g/h$  and  $1.06 \cdot 10^{-9}$  to  $5.39 \cdot 10^{-8}$  moles  $N_2O/g/h$  for sediment at sampling station 1 and 2, respectively; and  $1.73 \cdot 10^{-7}$  to  $1.08 \cdot 10^{-6}$  moles  $N_2O/g/h$  for macrophyte roots. The highest denitrification rates and bacteria numbers were observed in the rainy season, which was correlated to an increase of organic matter and nutrient input as a result of the hydrological period in Mogi-Guaçu floodplain river.

Key-words: denitrification; sediment; aquatic macrophytes; acetylene reduction.

## Introduction

The ecological importance of denitrification in aquatic ecosystems lies in the removal of nitrogen, principally from sediment in an oxygen-free state. This interferes with primary productivity, where nitrogen is one of the principal limiting factors (Wetzel, 1981).

The Infern o Lake is one of a group of 14 lakes on the Mogi-Gua u floodplain river, at the Ecological Station of Jatai, Municipality of Lu z Ant nio, State of S o Paulo, Brazil. The lake has a surface area of 3.05 ha and a maximum depth of 4.9 m (Nogueira *et al.*, 1996). The hydrological cycle is of fundamental importance in its effect on ecological conditions. During the rainy season, more reductive conditions are established (Ballester, 1994; Gianotti & Santos, 1997) and consumption of nutrients is greater (Dias Jr., 1990). These processes lead to different responses at certain stages in nitrogen dynamics (Ballester & Santos, 1997; Feresin, 1991; Obara & Santos, 1996; Gianotti & Santos, 1997). The aim of this work is to assess the qualitative and quantitative ecological importance of the denitrification process in sediment and among the roots of aquatic macrophytes, to understand the nitrogen dynamics in a shallow oxbow lake in a riverine wetland system.

## Material and Methods

Two sampling sites were chosen for this study. One, on the shore zone (Station 1), covered with aquatic macrophytes. From this site samples of sediment and roots of *Scirpus cubensis* were taken. The other site (Station 2), located in open water with no vegetation cover, provided just sediment samples. Collections were made at monthly intervals during two distinct hydrological periods: the rainy season (January, February, March, October, November, December 1992 and February, March 1993), and the dry season (May, June, August 1992 and July 1993). The numbers of denitrifying bacteria in the sediment and among the roots of *Scirpus cubensis* were estimated by the Most Probable Number (MPN) technique, described by Tiedje (1982) and adapted for this study. The denitrification rates were estimated by using acetylene to block reduction of  $N_2O$  to  $N_2$  (Yoshinari & Knowles, 1976; Knowles, 1979). Two situations were studied: 1. the "real condition", taking into account the endogenous nitrogen, and 2. the "potential condition", with supplementary nitrate. Samples of 100 ml of sediment (the first 10cm of the core) and 100 g (fresh weight) of roots were used; these were incubated anaerobically in 400 ml reaction flasks with 100 ml of water taken from the sampling site and/or with a solution of  $NaNO_3$  0.5 mM (0.36 mM of  $NO_3^-$ ). The flasks were subjected to a flow of  $N_2$  (200 ml/min) for 5 minutes and sealed with rubber stoppers; 10% of the gas phase was replaced by acetylene (Tiedje, 1982) at a partial pressure of 10 Kpa. Incubation was carried out in darkness, at 25 C during the rainy

period and at 20°C during the dry period. The N<sub>2</sub>O withdrawn during the gas phase with a gastight syringe, was quantified by electron capture gas chromatography (the <sup>63</sup>Ni detector operating at 290°C, 10<sup>-9</sup> amperes using N<sub>2</sub> at 30 ml/min) in a "Porapak Q" column (stainless steel, 3 m in length, by 1/8"), vaporizer at 80°C and over isothermy at 53°C. Experimental results relative to N<sub>2</sub>O production and taking endogenous nitrate into account were adjusted to a first order model, and under potentialized conditions to a logistic model (sigmoid curve) (Gianotti, 1994). Statistical interpretation of results was carried out by variance analysis and Tuckey's test.

### Results and Discussion

Maximum denitrification rates under "real conditions" in sediment (Station 1) varied from 5.45.10<sup>-10</sup> to 2.66.10<sup>-9</sup> moles N<sub>2</sub>O/g/h, and in sediment (Station 2) from 4.35.10<sup>-10</sup> to 1.96.10<sup>-9</sup> moles N<sub>2</sub>O/g/h (Fig. 1). Under "potential conditions" rates varied in sediment (Station 1) from 3.77.10<sup>-10</sup> to 1.35.10<sup>-7</sup> moles N<sub>2</sub>O/g/h, and in sediment (Station 2) from 1.06.10<sup>-9</sup> to 5.39.10<sup>-8</sup> moles N<sub>2</sub>O/g/h. In the roots of *Scirpus cubensis* rates varied from 1.73.10<sup>-7</sup> to 1.08.10<sup>-6</sup> moles N<sub>2</sub>O/g/h (Fig. 2). Estimates for denitrifying bacteria in the roots varied from 2.2.10<sup>4</sup> to 1.5.10<sup>5</sup> bacteria / g (dry weight); in sediment (Station 1) from 6.4.10<sup>2</sup> to 3.0.10<sup>3</sup> bacteria / g (dry weight); and in sediment (Station 2) from 5.4.10<sup>2</sup> to 3.3.10<sup>3</sup> bacteria/g (dry weight) (Fig. 3).

The higher values (confidence limit 95%) were recorded during the rainy season, when the ecological conditions of the Infern o Lake undergo the greatest modification. It is at this time that direct contact is established between the Mogi-Gua u River and the lake along its banks by way of the flooded areas. When this occurs, allochthonous material is brought in from adjacent areas, either by rainwater or by the river itself. The entry of water during the rainy season (bringing with it organic matter and nutrients) results in a higher rate of oxygen consumption and in the establishment of more reductive conditions in the sediment (Gianotti, 1994). Thus, the ecological conditions prevailing in the lake during this period favour the denitrification displayed under both "real" and "potential" conditions. The presence of macrophytes had no differential effect on numbers or activities of denitrifying organisms in sediment, at either Station 1 or 2. Concentration of nitrites and nitrates in the water was very low (rates beneath the detection limit of the 5.10<sup>-3</sup> mg/l method) (Gianotti, 1994). In the interstitial water in the sediments, nitrate concentration varied from 10.10<sup>-3</sup> mg/l (rainy season) to 1.5 mg/l (dry season) (Feresin, 1991). Thus the low nitrate concentration had a clear effect on the denitrification process, principally in the roots of *Scirpus cubensis*, where it was not possible to estimate the rate under "real conditions" (without added nitrate). It was, however, under the "potential conditions" in the roots that the higher rates were recorded, showing this to be a suitable niche for denitrification. In the sediment, the process of nitrifi-

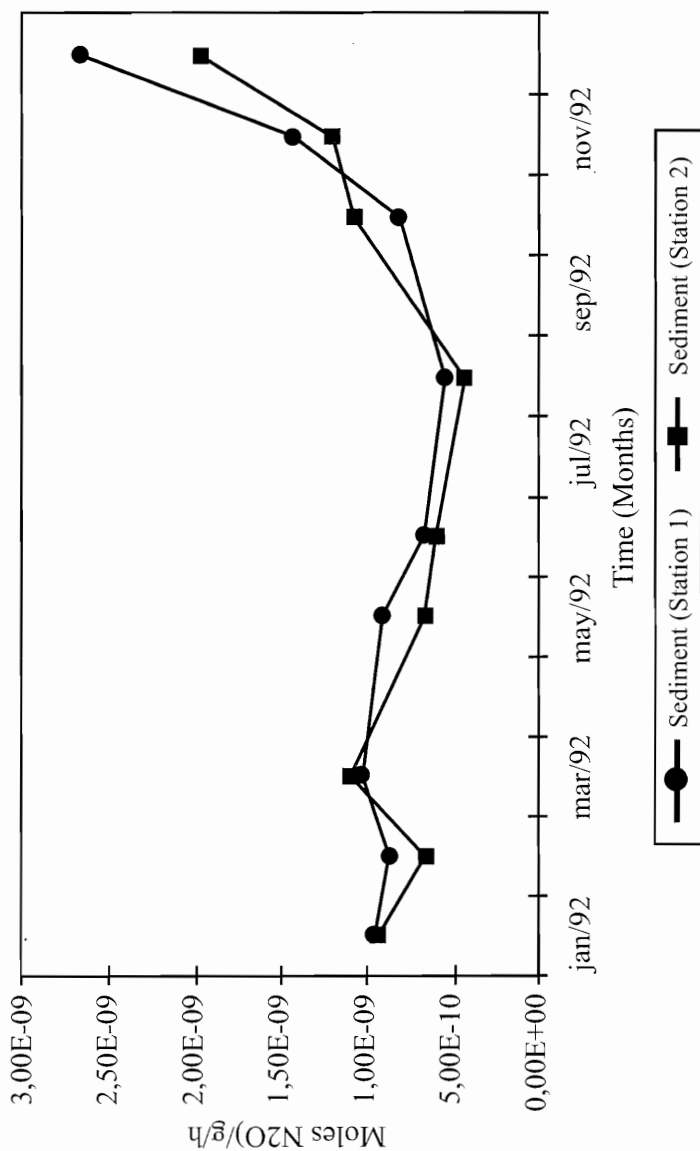


Figure 1. Estimated maximum rates for denitrification under “real conditions” (with endogenous nitrate) in sediment at Stations 1 and 2.

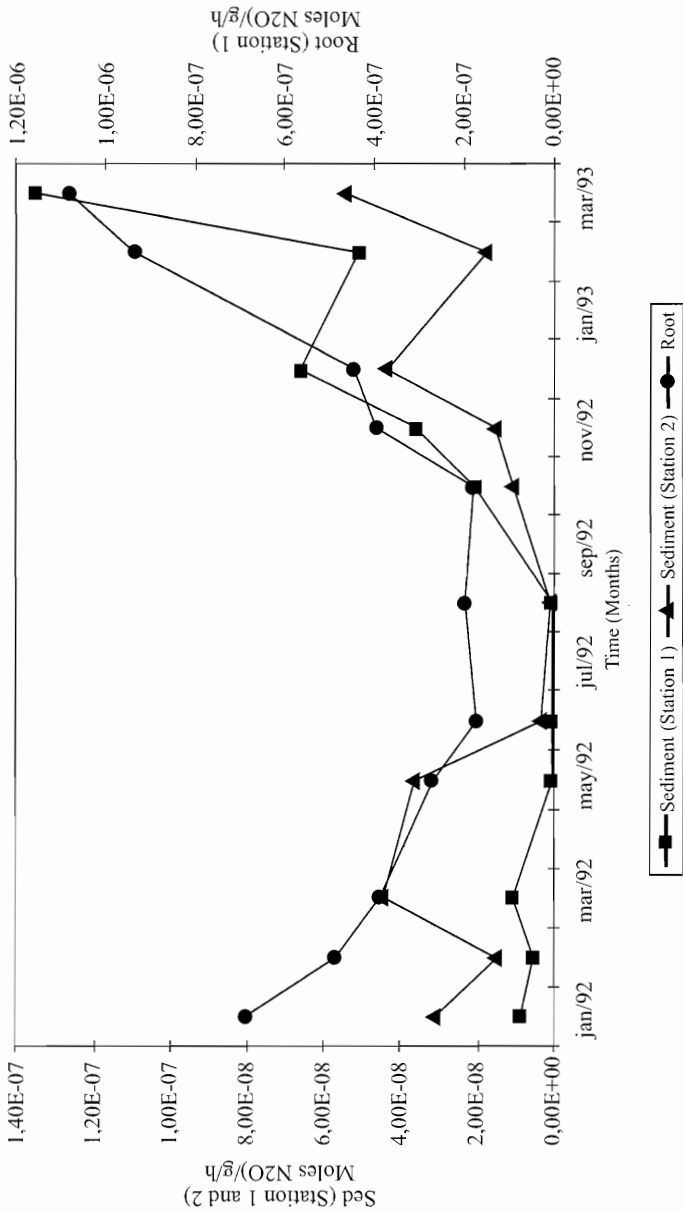


Figure. 2. Estimated maximum rates for denitrification under “potential conditions” (with supplementary nitrate) in sediment at Stations 1 and 2, and in roots at Station 1.

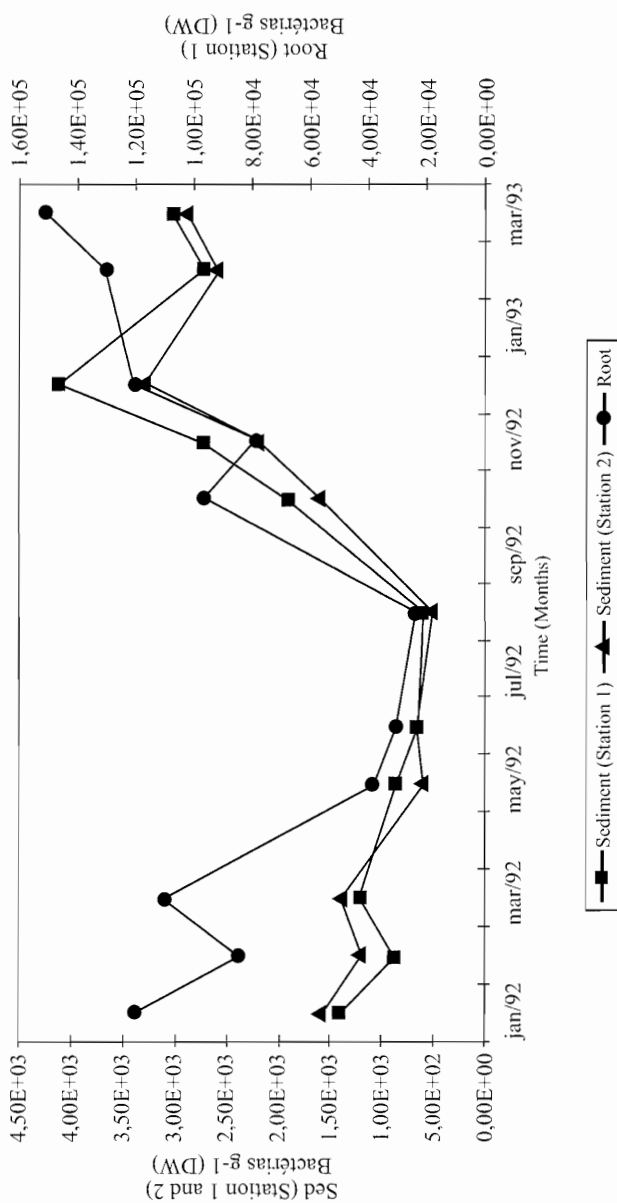


Figure. 3. Estimated numbers of denitrifying bacteria in sediment at Stations 1 and 2 and among roots of *Scirpus cubensis* at Station 1.

cation provided the electron acceptor for denitrification (Howard-Williams *et al.*, 1989); the two processes were represented by low generation rates for  $\text{NO}_3^-$  (Feresin, 1991) and  $\text{N}_2\text{O}$ , respectively. These considerations support the secondary ecological importance of both nitrification and denitrification processes to the aquatic system studied.

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