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DISSOLVED ORGANIC CARBON IN TWO BRAZILIAN COASTAL LAGOONS: SOURCES AND UTILIZATION FOR HETEROTROPHIC BACTERIA

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Resumo

Carbono Orgânico Dissolvido em duas lagoas costerias brasileiras: fontes e utilização por bactérias heterotróficas. Foi avaliado neste trabalho a utilização da matéria orgânica (MOD) originada da lixiviação de uma macrófita emergente, *Typha domingensis* Pers, dominante em duas lagoas costeiras de diferentes concentrações de carbono orgânico dissolvido (COD), no norte do estado do Rio de Janeiro. O lixiviado foi adicionado a microcosmos com água do meio pelágico da duas lagoas e inoculado com bactérias naturais aos ambientes. Experimentos controles foram realizados utilisando somente água filtrada e a adição do inóculo. A produção bacteriana, densidade e biomassa das bactérias foram quantificadas a cada 24 horas, durante 5 dias. A produção bacteriana e biomassa foram significativamente maiores nos experimentos com a adição do lixiviado, nas duas lagoas, em comparação aos controles. Estes resultados indicaram que a MOD da macrófita tem uma influência posistiva no crescimento bacteriano. Resultados normalizados indicam que isto não ocorreu somente devido a maior quantidade de MOD, mas também a qualidade do material originado das macrófitas.

Palavras-chave: bactéria, COD, macrófitas aquáticas, lixiviado, lagoas costerias.

Abstract

We evaluated the bacterial utilization of dissolved organic matter (DOM) of a leachate from emergent macrophyte, *Typha domingensis* Pers, dominant in two coastal lagoons of different dissolved organic carbon (DOC) concentrations, in the north of Rio de Janeiro State, Brazil. The leachate was added to microcosms with pelagic water from the two lagoons and inoculated with natural bacteria. Control experiments were performed using only pelagic filtered water added to inocculum. Bacterial production, density and biomass were measured every 24 hours, during five days. Bacterial production and biomass were significantly higher in experiments with leachate addition, in both lagoons, in comparison to the controls. Those results indicate that DOM from the macrophyte has a positive influence on bacterial growth. Normalized results also indicate it is not a matter of quantity, but the quality of DOM is better utilized than the DOM from pelagic region.

Key-words: bacteria, DOC, aquatic macrophytes, leachate, coastal lagoons.

Introduction

Coastal lagoons are among the most productive aquatic ecosystems in the world, with carbon production rates similar to upwelling regions and higher than offshore and shelf waters (Knoppers, 1994).

In many countries, such as in Brazil, those ecosystems are found close to dense urban centers, implying in a range of environmental problems. At same time, coastal lagoons have an important social and economical role, either as a food source or as tourist attraction.

Sand deposition during the Quaternary Period resulted from combined factors as littoral drift currents, changes in the sea level and natural traps to sediment retention, generating in the Brazilian Coast geological formations known as "Restingas" (Sugio & Tessler, 1984). The same processes also originated coastal lagoons, by the closure of bays or sedimentation in the rivers mouth (Esteves, 1998).

The Restinga de Jurubatiba is located in the North Coast of Rio de Janeiro State, where 14 different coastal lagoons are located within 14 km of coastline. Although the short distance among them is short and, in some cases similar vegetation on the drainage basin, those lagoons have different characteristics in the water chemistry. Among those characteristics, the most remarkable and ecologically important is the watercolor, ranging from clear to dark brown, which results mainly from differences in dissolved organic carbon (DOC) concentrations.

Morphological characteristics of coastal lagoons, such as small size and shallow depth, imply in important role of the littoral region on the pelagic metabolism (Wetzel, 1983). Panosso *et al.* (1998) described four coastal lagoons in the Jurubatiba region, Imboassica, Cabiúnas, Comprida and Carapebus, as shallows and high perimeter/area ratio water bodies. These four ecosystems have a developed community of emergent macrophytes on its marginal zone. Furtado (1994), studying Imboassica lagoon, concluded that detritus of two emergent macrophytes, *Typha domingensis* Pers. and *Eleocharis fistulosa*, are the main stock compartment for energy and nutrients, performing an important function on the ecosystem metabolism. In the same lagoon, Palma-Silva (1998) found high biomass and net primary productivity rates in stands of *T. domingensis*, identifying even the expansion of the specie distribution on the ecosystem.

The link between the carbon from littoral and pelagic metabolism zone involves the export of particulate detritus or in a dissolved form, from the excretion of photosynthates (Wetzel, 1969) or from the leaching of the plant material (Findlay *et al.*, 1992).

Higher plants found in the drainage basin or in the littoral region are the main source of organic carbon in shallow and small lakes (Wetzel, 1992). The inflow of this material from plants occupying the littoral region occurs through the export of particulate detritus or in a dissolved form, from the excretion of photosynthates (Wetzel, 1969) or from the leaching of the plant material (Findlay *et al.*, 1992).

The main inlet of the dissolved organic carbon in the trophic chain of aquatic ecosystems is the incorporation by bacteria. (Pomeroy, 1974; Paerl, 1978; Azam *et* *al.*, 1983). The availability of the dissolved organic carbon to heterotrophic bacteria is dependent mainly of its biochemical composition and molecular size, concentration of inorganic nutrients, and other environmental factors, such as temperature (Amon & Berner, 1996). The growth efficiency of bacteria from this material is of utmost importance to estimate the role of this community in the transfer of carbon to other trophic levels (Coffin *et al.*, 1993).

In the north of the State of Rio de Janeiro, Brazil, a series of shallow and small coastal lakes are found, with a thriving community of emergent macrophytes in its littoral region, *Typha domingensis* Pers. being the dominant species in these ecosystems. The goal of this research is to evaluate, from microcosms, the bacterial utilization of the leachate of leaves of this macrophyte in samples of water from two lakes with different DOC concentration.

Study Area

Imboassica and Comprida lagoons are shallow coastal lakes located near Macaé, a city of State of Rio de Janeiro, Brazil. These shallow lakes present a high surface/volume ratio, which grant a high importance to the littoral zone. *T. domingensis* is the dominant macrophyte in the littoral zone of both lakes.

The urban development in its littoral zone and the discharge of domestic effluents are the main anthropogenic impacts over Imboassica Lagoon. Once a year, a man-made channel is opened in the sandy barrier, increasing the ocean influence in its ecosystem. Comprida lagoon can be considered an undisturbed system, without major anthropogenic influences. It is an acid and black-colored lake due to high concentrations of humic compounds.

Material and Methods

Pieces of naturally dry leaves of *T. domingensis* collected from the littoral zone of Imboassica and Comprida lakes were kept in glass flasks with filtered (0.2 μm) water sampled at the central body of each lagoon for 24 hours, at a proportion of 6.5g/l. The samples were again filtered (0.2 μm) and 180 ml of the leachate was added to 20 ml of GFF filtered respective lake water (inoculums), and incubated in the dark at 25 °C in capped (without bubbles) sterile glass flasks. Before the incubation, inorganic carbon (DIC) and DOC were measured. Control experiments were performed for each lake using 0.2 μm filtered pelagic water added to GFF filtered lake water. So, there were four different treatments in total: Imboassica leachate and control, Comprida leachate and control.

During 5 days, every 24 hours, 2 flasks (replicates) were picked out and used for IC, DOC, bacterial number, bacterial biomass and bacterial productivity measurements.

IC and DOC were determined by high temperature oxidation, Pt-catalyzed, using a Shimadzu 5000 TOC analyzer. Bacterioplankton productivity (BP) was evaluated by ^{3}H-thymidine incorporation method (Fuhrman & Azam, 1980). Empirical factors for conversion of the results of ^{3}H-thymidine incorporation to carbon production were obtained through the integration method (Kirchman, 1985). The conversion factor value obtained for Imboacica lagoon was 5.86 10^{10} μg C mol thymidine^{-1} (Thomaz, 1995) and for Comprida lagoon, 1.076 10^{10} μg C mol thymidine^{-1}.

Bacterial number was evaluated using epifluorescence microscopy after staining with acridine orange (Hobbie *et al.*, 1977). Bacterial biomass was estimated from biovolume measurements (Fry, 1990) of 30 bacteria/slide, using the conversion factor proposed by Bjørnsen (1986), 1μm^3 = 3.5 10^{-13} g C.

Results and Discussion

The concentration of DOC, after all filtration steps, was 27.0 mg/l for Comprida lagoon and 13.0 mg/l for Imboassica lagoon. After the addition of the leachate, the concentrations rose to 42.5 mg/l and 20.2 mg/l respectively. So, DOC concentration increased 1.5 fold for Comprida and 1.6 for Imboassica with the leachate addition.

Similar trends in the bacterial activity could be observed in both lagoons, in the experiments with added carbon from *T. domingensis*. After 24 hours, there were significant increases in both the bacterial biomass (from 2.16 10^{-8} g C l^{-1} to 3.37 10^{-7}g C l^{-1} at Comprida and from 1.91 10^{-8}g C l^{-1} to 1.41$10^{-7}$g C l^{-1} at Imboassica) (Fig. 1) and in the bacterial production (from 0.83 μg C l^{-1} h^{-1} to 8.12 μg C l^{-1} h^{-1} at Comprida and from 2.86 μg C l^{-1} h^{-1} to 14.64 μg C l^{-1} h^{-1} at Imboassica) (Fig. 2). These values decreased 24 or 48 hours later, in both cases, evidencing a quick assimilation of this substrate. In the controls, no significant variations were observed in these variables, with the exception of the bacterial production at Comprida, with a slight increase up to 72 hours.

The trends observed in this experiment coincide with those obtained by other authors in similar experiments, using leaves of *Typha latifolia* and *Juncus effusus* (Mann & Wetzel, 1992) and detritus of *Typha angustifolia* and *Vallisneria americana* (Findlay *et al.*, 1992). This suggest the fast assimilation of the more labile material, stabilizing later when the material more resistant to degradation is used. This more labile material would be composed mainly by molecules of smaller size, which, even being a small fraction of the total pool of DOC, sustains most of the bacterial activity in aquatic ecosystems (Sundh, 1992; Coffin *et al.*, 1993).

Unfortunately, due to the sensibility of the employed methods, it was not possible to detect variations in IC or DOC, preventing any estimate of the bacterial growth efficiency.

The average of the results of bacterial production and biomass in the experiments in which leachate from *T. domingensis* was added was significantly higher when compared to those obtained in the controls (Mann-Whitney test, $p<0,005$), indicating that dissolved organic matter is a positive stimulus for the bacterial activity. The variation was further marked when we observe the bacterial biomass (959% at Comprida and 892% at Imboassica), but was also relevant for the production (215% at Comprida and 91% at Imboassica). This leads to the conclusion that these macrophytes could potentially sustain a considerable fraction of the bacterial metabolism in these ecosystems.

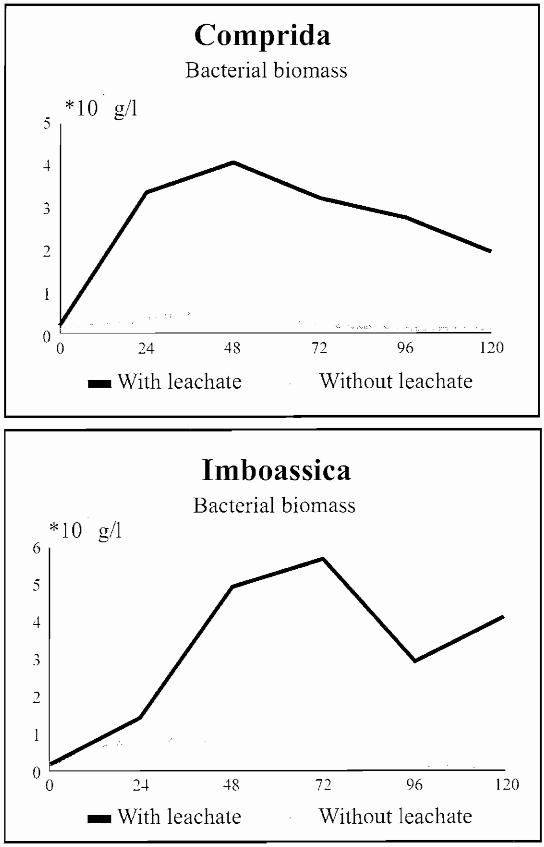
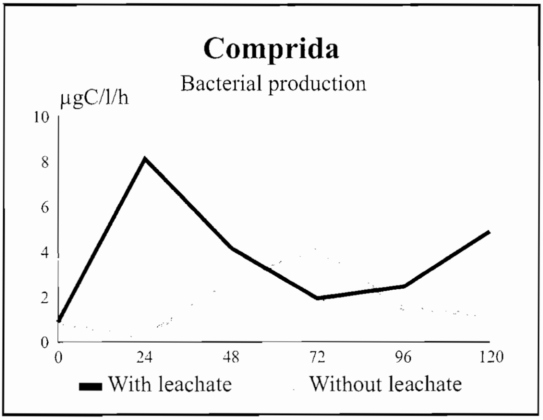


Figure 1: Changes on bacterial biomass (gC l^{-1}) on leachate and control experiments during 5 days, in Comprida and Imboassica Lagoons.

In an attempt to normalize production and biomass data related to the supply of dissolved organic carbon, ratios between these variables and the concentration of DOC were calculated in both lagoons and in the different treatments. The ratios between bacterial biomass and DOC were approximately 6 times greater in treatments with added leachate, notwithstanding the fact ratios of production did not exhibit such pronounced differences. This indicates that not only the quantitative increase of dissolved organic matter originated from the macrophytes is a stimulus for the bacterial growth, but also that the quality of the supplied material is an important factor. We can visualize two explanations for this fact: first, dissolved organic matter present in the pelagic region was already submitted to bacterial degradation in place of its origin, becoming more refractory as it penetrates into the lagoon. Another explanation would be that, compared with other sources of organic matter, such as plants located in the drainage basin, rich in complex molecules characteristic of its structural compounds, the matter originated



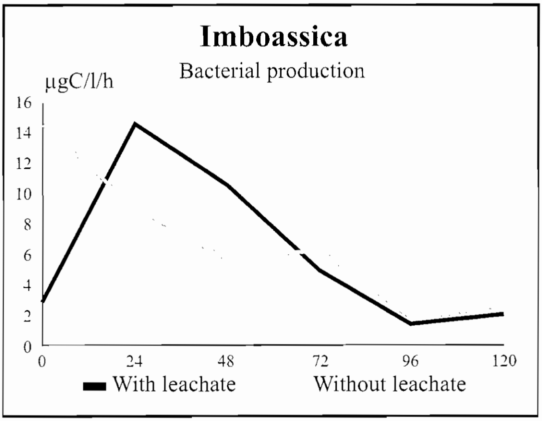


Figure 2: Changes on bacterial production (µgC l^{-1} h^{-1}) on leachate and control experiments during 5 days, in Comprida and Imboassica Lagoons.

from macrophytes has characteristics that make it easier to incorporate by the bacterial community. However, for a more in depth knowledge on these claims, other experiments are necessary involving other sources of organic matter for these ecosystems.

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References

AMON, R.M.W. & R. BENNER 1996. Bacterial utilization of different size classes of dissolved organic matter. *Limnol. Oceanogr.*, **41**(1): 41-51.

AZAM, F.A.; T. FENCHEL; J.G. FIELD; J.S. GRAY; L.A. MEYER-REIL & F. THINGSTAD 1983. The ecological role of water-column microbes in the sea. *Mar. Ecol. Prog. Ser.*, **10**: 257-263.

BANO, N.; N. MEHR-UN; N. KHAN; M. SALEEM; P.J. HARRISON; S.I. AHMED & F. AZAM 1997. Significance of bacteria in the flux of organic matter in the tidal creeks of the mangrove ecosystem of the Indus River delta, Pakistan. *Mar. Ecol. Prog. Ser.* **157**: 1-12.

BILLEN, G.; P. SERVAIS & S. BECQUEVORT 1990. Dynamics of bacterioplankton in a oligotrophic and eutrophic aquatic environments; bottom-up or top-down control. *Hydrobiologia*, **207**: 37-42.

BJØRSSEN, P.K. 1986. Automatic determinations of bacterioplankton biomass by means of image analyses. *Appl. Envir. Microbiol.* **51**: 1199-1204.

COFFIN, R.B.; J.P. CONNOLLY & P.S. HARRIS 1993. Availability of dissolved organic carbon to bacterioplankton examined by oxigen utilization. *Mar. Ecol. Prog. Ser.*, **191**: 9-22.

FINDLAY, S.; M.L. PACE; D. LINTS; J.J. COLE; N.F. CARACO & B. PEIERLS 1991. Weak coupling of bacterial and algal production in a heterotrophic ecosystem: The Hudson River estuary. *Limnol. Oceanogr.*, **36**(2): 268-271.

FINDLAY, S.; M.L. PACE; D. LINTS & K. HOWE 1992. Bacterial metabolism of organic carbon in the tidal freshwater Hudson Estuary. *Mar. Ecol. Prog. Ser.* **89**: 147-153.

FRY, J.C. 1990. Direct methods and biomass estimation. *Met. Microbiol.* **22**: 41-85.

FUHRMAN, J.A. & F. AZAM 1980. Bacterioplankton secondary production estimates for coastal waters of British Columbia, Antarctica and California. *Appl. Envir. Microbiol.* **39**: 1085-1095.

FURTADO, A. 1994. *Contribuição das macrófitas aquáticas Typha domingensis Pers (TYPHACEAE) e Eleocharis cf. fistulosa (CYPERACEAE) para estoques de nutrientes e energia da Lagoa Imboassica (Macaé, RJ).* MsC dissertation, PPGE-UFRJ, Rio de Janeiro, 138p.

HOBBIE, J.E.; R.J. DALEY & S. JASPAR 1977. Use of nuclepore filters for counting bacteria by fluorescent microscopy. *Appl. Envir. Microbiol.* **33**: 1225-1228.

JOHANSSON, J. 1983. Seasonal development of bacterioplankton in two forest lakes in central Sweden. *Hydrobiologia*, **101**: 71-88.

MANN, C.J & R.G. WETZEL 1996. Loading and utilization of dissolved organic carbon from emergent macrophytes. *Aquat. Bot.*, **53**: 61-72.

PAERL, H.W. 1978. Microbial organic carbon recovery in aquatic ecosystems. *Limnol. Oceanogr.* **23**(5): 927-935.

PALMA-SILVA, C. 1999. *Ecologia de macrófitas aquáticas em uma lagoa costeira sujeita a impactos antrópicos (Lagoa Imboassica, Macaé, RJ).* PhD thesis, PPGE-UFRJ, Rio de Janeiro, 153p.

PANOSSO, R.F. 1998. *Remineralização do fósforo e dinâmica do plâncton em duas lagoas costeiras fluminenses, e o papel da matéria orgânica dissolvida para nutrição do fitoplâncton.* PhD thesis, IBCCF-UFRJ, Rio de Janeiro, 200p.

POMEROY, L.R. 1974. The ocean's foodweb, a changing paradigm. *Bioscience,* **24**: 499-504.

SIMOM, M.; B.C. CHO & F. AZAM 1992. Significance of bacterial biomass in lakes and the ocean: comparison to phytoplankton biomass and biogeochemical implications. *Mar. Ecol. Prog. Ser.,* **86**: 103-110.

SØNDEGAARD, M. 1997. Bacteria and dissolved organic carbon in lakes. pp:.138-161. In: SAND-JENSEN, K & O. PEDERSEN (eds.). *Freshwater Biology: Priorities and development in Danish Research.*

SUNDH, I. 1992. Biochemical composition of dissolved organic carbon derived from phytoplankton and used by heterotrophic bacteria. *Appl. Environ. Microbiol.,* **58**: 2938-2947.

THOMAZ, S.M. & F.A. ESTEVES 1997. Secondary productivity (^{3}H-leucine and ^{3}H-thymidine incorporation), abundance and biomass of the epiphytic bacteria attached to detritus of *Typha domingensis pers.* in a tropical coastal lagoon. *Hydrobiologia,* **357**: 17-26.

WETZEL, R.G. 1969. Excretion of dissolved organic compounds by aquatic macrophytes. *BioScience,* **19**(6): 539-540

WETZEL, R.G. 1992. Gradient-dominated ecosystems: sources and regulatory functions of dissolved organic matter in freshwater ecosystems. *Hydrobiologia,* **229**: 181-198.

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