Bacterial community structure in two sediments with different organic matter content of a tropical coastal lagoon (Brazil).

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ABSTRACT: Bacterial community structure in two sediments with different organic matter content of a tropical coastal lagoon (Brazil). The size structure of the bacterial benthic communities was studied in two distinct sediments with different organic matter contents (clay and sand) from a shallow eutrophic lagoon (Barra Lagoon), located in the southeastern of Brazil. Samples of sediments were taken from the pelagic and littoral zones from 1993 to 1995. The bacterial number and biomass were estimated per g DW (Dry Weight) of sediment. Total bacterial number (DAPIDC) showed an average of 3.0 - 4.0 x 10° bact/g, regardless of organic matter content. The average bacterial biomass was higher in the clay sediment (1.54 x 10<sup>3</sup> mgC/g) than in the sandy sediment (7.12 x 10<sup>2</sup> mgC/g). Total heterotrophic culturable bacteria were 3 to 4 orders of magnitude lower than DAPIDC and varied from 3.02 x 10<sup>4</sup> to 7.46 x 10<sup>5</sup> cfu/g. Proteolytic heterotrophic culturable bacteria were more abundantly found in the clay and lipolytics in the sandy sediments. Cocci forms exhibited the highest densities while filament forms accounted for most of the biomass. The density and biomass of benthic total and heterotrophic culturable bacteria, as well as average size biovolume of cells, were affected by organic matter content of the clay and sandy sediments.

Key-words: bacteria, community structure, sediment, tropical lagoon, Brazil.

RESUMO: Estrutura da comunidade bacteriana em dois sedimentos com diferentes concentrações de matéria orgânica de uma lagoa costeira tropical (Brasil). As bactérias bentônicas foram estudadas em dois tipos de sedimento (argiloso e arenoso), com diferentes concentrações de matéria orgânica, de uma lagoa eutrófica rasa (Lagoa da Barra), situada na região sudeste do Brasil (Região dos Lagos -RJ). As amostras foram coletadas nas zonas pelágica (sedimento argiloso) e litorânea (sedimento arenoso), no período de 1993 a 1995. Os resultados de densidade e biomassa bacterianas foram expressos em gramas de peso seco de sedimento. A média do número de bactérias totais (DAPIDC), independente da concentração de matéria orgânica, variou de 3,0 - 4,0 x 10º bact/g. Entretanto, a biomassa média das bactérias totais foi maior no sedimento argiloso (1,54 x 10<sup>3</sup> mgC/g) do que no sedimento arenoso (7,12 x10<sup>2</sup> mgC/g). O número total de bactérias heterotróficas cultiváveis (UFC) foi 3 a 4 ordens de magnitude menor que o número total estimado diretamente em microscopia (DAPIDC), variando de 3,02.10<sup>4</sup> a 7,46.10<sup>5</sup> ufc/g. As bactérias cultiváveis heterotróficas (NMP) proteolíticas foram dominantes no sedimento argiloso, enquanto as lipolíticas dominaram no sedimento arenoso. As formas cocóides apresentaram as maiores densidades, enquanto as formas filamentosas e bastonetes contribuíram com maior biomassa. Tanto a densidade como a biomassa das bactérias totais e das heterotróficas cultiváveis totais, bem como o biovolume celular médio, mostraram relação com as diferentes concentrações de matéria orgânica dos sedimentos argiloso e arenoso. Palavras-chave: bactérias, estrutura da comunidade, sedimento, lagoa tropical, Brazil.

# Introduction

The Brazilian Atlantic coast has many saline, freshwater and estuarine lagoons (Esteves, 1988). Despite the bacteria's key role for the productivity and transformation of the organic compounds in such aquatic systems, they have been little studied, particularly the benthic fraction. It is true especially in tropical regions like Brazil, where studies about microorganisms on freshwater and coastal lagoon date from the 1970s and 1980s respectively.

Bacteria can use very low concentrations of organic compounds, thus providing dissolved organic carbon for other organisms. This fact makes them the basis of the microbial food webs (Azam et al., 1983; Pomeroy, 1974; Sherr & Sherr, 1988). The aquatic food web is supported by the microbial loop which is regulated by nutrients and grazing (protozoans, microcrustaceans) resulting in a dynamic equilibrium of the system (Pace & Funke, 1991).

Bacteria make up a heterogeneous group in an aquatic ecosystem; most of them are heterotrophic and responsible for the degradation of organic matter (Amon & Benner, 1996; Chrzanowski & Hubbard, 1989; Rheinheimer, 1985; Sanders et al., 1989). Sediments favour the nutrients accumulation and provide a more homogeneous and stable environment for these organisms. Such particularity, besides the natural tendency to adhere to particles, result in a bacterial community much more numerous and active on sediment (Jones, 1980). Their cell number is usually proportional to the amount of organic matter available in sediments, where in the surface layers the most heterotrophic activity takes place and the bacterial biomass may account for about 2.5 to 7.8% of the total amount of organic matter (Drabkova, 1983; Dutka & Kwan, 1983; Freitas & Godinho-Orlandi, 1991).

Organic matter first reaches the sediment in particulate form, being mostly composed by macromolecules such as proteins, carbohydrates, lipids and nucleic acids. Hydrolytic activity is the first step in the decomposition process of detritus and dead cells (Jones, 1979).

Total number, biomass, cell morphology and species diversity of aquatic bacteria are greatly influenced by the trophic state of the environment. Many abiotic and biotic factors affect the bacterial community where many times one factor can define the type of bacterial population present (Konopka, 1993; Rheinheimer, 1985).

In the present work, the structure of number and biomass of the benthic bacterial community present in a natural tropical coastal ecosystem was studied.

# Study area

Barra Lagoon (22° 57° S, 42° 47° W) is located at 50 km north from the city of Rio de Janeiro, in the south eastern of Brazil (Fig. 1), where the climate is tropical. It is a shallow and eutrophic coastal lagoon belonging to the Maricá System arranged by four lagoons, Barra (area 6.2 km<sup>2</sup>, max. depth 1.3 m), Maricá (18.2 km<sup>2</sup>), Padre (3.1 km<sup>2</sup>) and Guarapina (8.6 km<sup>2</sup>). This system is inserted between a sandy beach ("restinga") and the coastal mountain ridge ("Serra do Mar"). The lagoon is surrounded by typical "restinga" vegetation with scattered macrophyte stands (*Typha*). Average densities and biomass of total planktonic bacteria (AODC) were similar at pelagic and littoral zones (9.5 x 10<sup>6</sup> bact/mL, 0.8 mgC/mL) during the same period studied at the present work, but total planktonic heterotrophic culturable bacteria was higher in the littoral zone (2 x 10<sup>4</sup> bact/mL) with dominance of proteolytics at two stations (> 64%) (Gomes et al., 1998). In 1991 spring-summer season a massive fish kill occurred, as a consequence of an anoxia, caused by a rapid growth of the toxin-producing cyanobacteria *Synecocystis aquatilis f. salina* that changed the predominant metabolism in the system from autotrophic to heterotrophic (Carmouze et al., 1993).



Figure 1: Upper: Location of Região dos Lagos, Rio de Janeiro State. Lower: map of the Maricá Lagoon System showing the location of Barra Lagoon and the sampling locations.

## Material and methods

Water sampling: morning samplings were carried out in September and December 1993, March, July and October 1994 and October 1995, at two sites, pelagic zone and littoral zone (Fig. 1), using two alcohol-sterilized PVC pipes with rubber stoppers (10 cm diameter, 30 cm and 1 m length for each station, respectively), and then transported to the laboratory at low temperature. Due the contaminations with fungi, bacterial densities and NMP were not measured in the last month.

Sample Treatment: sediment samples were homogenised in a blender with a solution of 1% NaPP (sodium pyrophosphate) plus 0.1% tween 80 and dilutions performed with a sterile saline solution (0.85% NaCl).

DAPI Direct Counting (DAPIDC): treated samples were fixed with filtered formaldehyde 2% (v/v), stored at 4°C, dyed with DAPI (1.0 mg/L) and filtered onto a black Millipore membrane (25 mm, 0.22 mm pore size). Three hundred cells were counted at 1250-x magnification under an epifluorescence microscope (Olympus BH-RFL-W) (Porter & Feig, 1980; Velji & Albright, 1993).

Total Bacterial Biomass: was estimated through biovolume of geometric cell forms (Rodina, 1972; Sorokin & Kadota, 1972). Twenty to fifty cells of each geometric form in each sample were measured under epifluorescence microscopy (DAPIDC). Dry weight (dw) and carbon content were estimated using the conversion factor  $1 \text{ mm}^3 = 308 \text{ fgC}$  (wet weight – ww) and the ratios dw/ww = 0.33 and carbon ww/dw = 0.50 (Bakken & Olsen, 1983; Fry, 1988; Watson et al., 1977). Data are presented in average and standard deviation.

Total Heterotrophic Culturable Bacteria: samples not fixed were diluted in saline solution and the number of colony forming unity (cfu) per gram (dw) was estimated by the spread plate technique using a modified Marine Agar medium (Difco) with salinity reduced to 20% using filtered seawater. Triplicates of plates were incubated for 5 days at 28 °C.

Amylolytic, Lipolytic and Proteolytic Heterotrophic Culturable Bacteria: the numbers were estimated in a mineral medium with 0.5% of starch, tween 80 or gelatine, respectively, using the most probable number (MPN) technique and incubated in a 5-tube series for 7 days at 28 °C (Greenberg et al., 1992).

Sediment Dry Weight: estimated as the weight after drying each sediment sample (triplicates) in a Pasteur oven for 2 hours at 100°C.

Physico-chemical sediment analysis: temperature (thermometer 0.1 °C precision), salinity (refractometer Shibuya Optical S-1) and dissolved oxygen (Winkler's Method; Golterman, 1969) was measured in sediment-water interface. Soil Laboratory of the Centro Nacional de Pesquisas do Solo (EMBRAPA, Rio de Janeiro) analyzed the chemical aspects. Data are presented in average and standard deviation.

Statistical Analysis: Spearman correlation coefficients (p < 0.05) and t-test for independent samples were obtained with Statistica 6.0<sup>b</sup> software program. Correlation was calculated using abiotic (temperature, salinity, pH, DO, chemical and physical composition) and biotic variables (number and biomass of total bacteria, number of heterotrophic culturable bacteria, amylolytic, lipolytic and proteolytic heterotrophic culturable bacteria, average size and biovolume of different bacterial forms). In order to assess if the biotic variables from the different sediments (clay and sand) were similar (H<sub>o</sub>), t-test was used at 5% significant level (8 to 10 degrees of freedom).

#### Results

The average depth of water column at pelagic zone was  $1.05 \pm 0.18$  m. Littoral zone was shallower, with an average depth of  $0.34 \pm 0.07$  m. There was no difference of temperature, dissolved oxygen and salinity among sampling sites (Tab. I). Sediment pH was close to neutral, being practically the same at the two stations, 6.4 (pelagic zone) and 6.9 (littoral zone). The sediment at pelagic zone was found to be basically of the clay-type (47% coarse sand, 31% clay, 12% silt), while that of littoral zone was of the sandy-type (88% coarse sand and 7% clay). Chemical analysis indicated that the former was richer in organic compounds (3.2%), nitrogen (0.26%) as well as in salts than the sediment from littoral zone.

 Table I: Physico-chemical analysis (average ± standard deviation) of sediment from pelagic and littoral zones.

| Sediment characteristics | Pelagic zone    | Littoral zone  |  |
|--------------------------|-----------------|----------------|--|
| T (°C)                   | 25.42 ± 3.09    | 26.25 ± 3.50   |  |
| S (g/L)                  | 10.00 ± 2.19    | 9.83 ± 2.40    |  |
| DO (mg/L)                | 7.45 ± 1.34     | 6.87 ± 1.27    |  |
| рН                       | 6.45 ± 0.32     | 6.88 ± 0.26    |  |
| C (%)                    | 3.17 ± 0.10     | 0.27 ± 0.03    |  |
| N (%)                    | 0.26 ± 0.03     | 0.10 ± 0.01    |  |
| Na (meq/100 ml)          | 3286.67 ± 11.11 | 608.33 ± 13.31 |  |
| Ca (meq/100 ml)          | 4.77 ± 0.19     | 0.59 ± 0.10    |  |
| Mg (meq/100 ml)          | 10.00 ± 1.55    | 0.92 ± 0.06    |  |
| P (ppt)                  | 86.67 ± 5.16    | 15.13 ± 1.77   |  |
| K (ppt)                  | 490.33 ± 35.34  | 77.83 ± 8.52   |  |
| Fine sand (%)            | 9.67 ± 1.32     | 3.07 ± 0.08    |  |
| Coarse sand (%)          | 47.05 ± 1.69    | 87.67 ± 1.86   |  |
| Silt (%)                 | 11.78 ± 1.27    | 2.03 ± 0.19    |  |
| Clay (%)                 | 31.50 ± 1.41    | 7.23 ± 1.75    |  |
|                          |                 |                |  |

Highest benthic total density and biomass  $(1.59 \times 10^{10} \text{ bact/g}; 5.17 \times 10^3 \text{ mgC/g})$ , as determined by DAPIDC, were found on September 1993 for pelagic zone, whereas at littoral zone, maximum density (6.84 x 10° bact/g) was found on October 1994 and maximum biomass (1.56 x 10<sup>3</sup> mgC/g) on October 1995. The lowest values of density and biomass were found on March 1994 (Fig. 2). On average the total bacterial density was similar at pelagic and littoral zones (4.34 x 10° bact/g and 3.27 x 10° bact/g, respectively) (t = 0.40). Despite that the average total biomass value was an order of magnitude higher at pelagic zone (1.54 x 10<sup>3</sup> mgC/g) than at littoral zone (7.12 x 10<sup>2</sup> mgC/g) the statistic analyzes showed similar values (t = 1.32).

Among the geometric cell forms observed, cocci were prevalent in numbers at both stations (@ 69%) (Fig. 3). Nevertheless, they were the least important group in terms of total biomass, accounting for just 22% (pelagic zone) and 21% (littoral zone). The opposite



Figure 2: Density (bact/g) and biomass (mgC/g) of total benthic bacteria (DAPIDC), estimated in dry weight of sediment in samples from pelagic and littoral zones, for the study period.



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was found for the filament forms, which were less numerous, 4% (pelagic zone) and 2% (littoral zone), but the most important in terms of biomass, accounting for 41% and 42% at stations pelagic and littoral zones respectively; rod cells exhibited intermediate values in terms of density (< 30%) and carbon biomass (@ 30%).

The number of total heterotrophic culturable bacteria (Fig. 4) reached a maximum on March 1994 at pelagic zone (2.91 x 10<sup>6</sup> bact/g) and in October 1994 at littoral zone (8.01 x 10<sup>5</sup> bact/g). Average densities were similar (t = 2.20) at pelagic zone (1.43 x 10<sup>6</sup> bact/g) and littoral zone (2.61 x 10<sup>5</sup> bact/g). Total heterotrophic culturable bacteria estimated by CFU method showed significant correlation with total bacterial densities analyzed by DAPIDC method only at littoral zone (r = 0.90, p < 0.05).



Figure 4: Density fluctuation of total heterotrophic culturable bacteria (UFC/g), estimated in dry weight of sediment in samples from pelagic and littoral zones, for the study period.

Among the groups of heterotrophic bacteria analysed, proteolytics and lipolytics were the most abundant at pelagic (86%) and littoral zones (57%), respectively (Fig. 5), demonstrating a more homogeneous distribution of the three groups at the latter. Maximum values of  $3.36 \times 10^5$  MPN bact/g at pelagic zone and  $7.09 \times 10^5$  MPN bact/g at littoral zone were found for the lipolytics bacteria on July and October 1994, respectively. The density of amylolytic benthic heterotrophic culturable bacteria was higher in average at pelagic zone (4.66  $\times 10^4$  MPN bact/g) and October 1994 for littoral zone (3.68  $\times 10^4$  MPN bact/g) and October 1994 for littoral zone (3.68  $\times 10^4$  MPN bact/g) and October 1994 for littoral zone (3.68  $\times 10^4$  MPN/g). Average densities of proteolytic benthic heterotrophic culturable bacteria were 8.05  $\times 10^5$  MPN/g at pelagic zone and 1.66  $\times 10^5$  MPN/g at littoral zone, with maximum values found on March and October 1994 at both stations. These averages of lipolytic (t = 0.84), amylolytic (t = 0.70) and proteolytic (t = 1.42) bacterial densities among pelagic and littoral zones were similar.

The different geometric cell forms and their dimensions found in this study are shown in Tab. II. Rod length varied from 1.28 to 2.38 mm (1.63  $\pm$  0.35 mm) for pelagic zone and from 1.50 to 2.34 mm (1.86  $\pm$  0.35 mm) for littoral zone. The maximum lengths were observed on September 1993 at both stations. These lengths were similar (t = 0.06). The average diameter of rods was greater for the clay samples (0.83  $\pm$  0.08 mm) than sandy samples (0.74  $\pm$  0.15 mm), but these averages were considered statistically similar (t = 0.99). Cocci cells also showed a very similar diameter (t = -0.12), ranging from 0.58 to 0.95 mm (0.75  $\pm$  0.12 mm) at pelagic zone and from 0.58 to 0.97 mm (0.76  $\pm$  0.13 mm) at littoral zone. Maximum diameters were found on September (pelagic zone) and December 1993 (littoral zone). Filament cells were frequent, varying in size and biovolume. Average



Figure 5: Density fluctuation of benthic heterotrophic culturable bacteria (amylolytic, lipolytic and proteolytic) (NMP/g), estimated in dry weight of sediment in samples from pelagic and littoral zones, for the study period. Note that scales of axis Y are different.

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|          |                 | PELAGIC ZONE  |             |             |               |  |  |  |
|----------|-----------------|---------------|-------------|-------------|---------------|--|--|--|
|          | Cocci           | F             | Rods        |             | Filaments     |  |  |  |
| DATE     | D               | D             | L           | D           | L             |  |  |  |
| Sep 1993 | 0.95 ± 0.21     | 0.92 ± 0.32   | 2.38 ± 0.63 | 0.83 ± 0.18 | 8.66 ± 4.60   |  |  |  |
| Dec      | 0.83 ± 0.17     | 0.93 ± 0.17   | 1.59 ± 0.38 | 0.74 ± 0.12 | 9.57 ± 7.46   |  |  |  |
| Mar 1994 | 0.79 ± 0.28     | 0.75 ± 0.24   | 1.45 ± 0.24 | 0.87 ± 0.87 | 11.55 ± 4.45  |  |  |  |
| Jul      | 0.74 ± 0.26     | 0.81 ± 0.17   | 1.56 ± 0.34 | 1.05 ± 0.25 | 12.60 ± 7.49  |  |  |  |
| Oct      | 0.62 ± 0.23     | 0.67 ± 0.18   | 1.28 ± 0.17 | 0.93 ± 0.18 | 16.10 ± 12.54 |  |  |  |
| Oct 1995 | 0.58 ± 0.25     | 0.77 ± 0.16   | 1.54 ± 0.19 | 1.05 ± 0.01 | 8.40 ± 5.05   |  |  |  |
|          |                 | LITTORAL ZONE |             |             |               |  |  |  |
|          | Cocci           | F             | Rods        |             | Filaments     |  |  |  |
| DATE     | D               | D             | L           | D           | L             |  |  |  |
| Sep 1993 | 0.84 ± 0.29     | 0.77 ± 0.14   | 2.34 ± 0.79 | 0.77 ± 0.16 | 6.30 ± 1.48   |  |  |  |
| Dec      | 0.97 ± 0.22     | 0.91 ± 0.21   | 2.27 ± 0.80 | NF          | NF            |  |  |  |
| Mar 1994 | 0.81 ± 0.28     | 0.82 ± 0.27   | 1.95 ± 0.69 | 0.96 ± 0.16 | 10.15 ± 3.94  |  |  |  |
| Jul      | 0.67 ± 0.26     | 0.68 ± 0.18   | 1.54 ± 0.40 | 1.24 ± 0.18 | 12.83 ± 8.64  |  |  |  |
| Oct      | $0.58 \pm 0.21$ | 0 80 + 0 26   | 1.50 ± 0.41 | 0.99 ± 0.21 | 14.00 ±7.92   |  |  |  |
|          | 0.30 ± 0.21     | 0.00 1 0.20   |             |             |               |  |  |  |

Table II: Dimensions (average ± standard deviation) of cellular forms (mm), found at pelagic and littoral zones, as determined by the epifluorescence microscopy (DAPIDC) method.

D: Diameter; L: Length; NF: Not found.

lengths varied from 8.40 to 16.10 mm, with an average value of 11.15 ± 2.68 mm for the clay sediment (pelagic zone), and from 6.30 to 15.05 mm, with an average value of 11.67 ± 3.14 mm for the sandy sediment (littoral zone). Maximum values were observed on October (1994 and 1995), coinciding with a hot and rainy period. Diameter varied from 0.74 to 1.05 mm (pelagic zone) and 0.77 to 1.24 mm (littoral zone) with averages of  $0.91 \pm 0.11 \ \text{mm}$  and  $1.03 \pm 0.17 \ \text{mm}$ , respectively. The maximum values were found in July 1994. These differences of size were not significant for length (t = 0.54) and diameter (t = 0.28).

Analysis of the different cell forms can also be analyzed through biovolume measurement (mm³) (Fig. 6). Maximum cocci biovolumes were found in September 1993



Figure 6: Average cell biovolume (mm3) of cocci, rods and filaments found in clay and sandy sediment samples (pelagic and littoral zones) in the study period.

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for the clay sediment and December 1993 for the sandy sediment, with similar averages of  $0.30 \pm 0.12 \text{ mm}^3$  and  $0.33 \pm 0.14 \text{ mm}^3$ , respectively (t = -0.24). A decreasing trend in cocci biovolume was observed along the studied period for both stations. The similar fluctuations were observed to the rods, average biovolume being higher (t = -0.67, p > 0.52) for littoral zone ( $1.09 \text{ mm}^3$ ) than for pelagic zone ( $0.94 \text{ mm}^3$ ), with maximum values in September 1993 and December 1993, respectively. At first view filament cells exhibited higher average biovolume in the sandy ( $10.75 \pm 5.27 \text{ mm}^3$ , littoral zone) than in the clay sediment ( $8.05 \pm 3.40 \text{ mm}^3$ , pelagic zone), with the maximum values of 12.71 mm^3 and 17.14 mm^3 observed in July-October 1994 (pelagic zone) and October 1995 (littoral zone). But the statistic analysis showed that the filament biovolumes were similar (t = -0.29).

Table III: Comparison of average biovolume as estimated by various authors in several environments using different methods.

|   |          |          | BIOVOLUME (mmr 3) |           |                        |  |
|---|----------|----------|-------------------|-----------|------------------------|--|
|   |          | Cocci    | Rods              | Filaments | Total                  |  |
| Present Work <sup>2,C</sup>                       | Sediment | 0.3      | 0.9-1.1           | 8.0-10.7  | -                      |  |
| Gomes et al. (1998) <sup>1,C</sup> (Barra Lagoon) | Water    | 0.2      | 0.6               |           |                        |  |
| Watson et al. (1977) <sup>6,M</sup>               | Water    | 0.1      | 0.07-0.4          |           |                        |  |
| Bakken & Olsen (1983) <sup>1,</sup> s             | Water    | 0.2-0.5  | 0.4-1.6           |           |                        |  |
| Bratbak (1985) <sup>E</sup>                       | Water    |          |                   |           | 0.1-0.3 <sup>1</sup>   |  |
|   |          |          |                   |           | 0.1-0.3 5              |  |
| Sieracki et al. (1985) <sup>2, **,M</sup>         | Water    | 0.1-0.3  | 0.08-0.2          |           |                        |  |
| Bjornsen (1986) <sup>1,**</sup>                   | Water    |          |                   |           | 0.2-0.3 <sup>L</sup>   |  |
|   |          |          |                   |           | 0.08-01 <sup>F</sup>   |  |
| Lewis et al. (1986) <sup>3,L</sup>                | Water    | 0.07-0.5 | 0.2-1.5           |           |                        |  |
| Lee & Fuhrman (1987) <sup>1,M</sup>               | Water    |          |                   |           | 0.03-0.07              |  |
| Gonzalez et al. (1990) <sup>4,M</sup>             | Water    |          |                   |           | 0.03-0.08              |  |
| Gomes & Godinho (in press) <sup>1,L</sup>         | Water    | 0.05     | 0.80              |           |                        |  |
| Lind & Lind (1991) <sup>1,L</sup>                 | Water    |          |                   |           | 0.05-0.07              |  |
| Abreu et al. (1992) <sup>1,*,E</sup>              | Water    |          |                   |           | 0.04-0.3               |  |
| Ritzrau \$ Graf (1992) <sup>1,M</sup>             | Sediment |          |                   |           | 0.15-0.54              |  |
| Suzuki et al. (1993) <sup>E</sup>                 | Water    |          |                   |           | 0.11-0.17 <sup>1</sup> |  |
|   |          |          |                   |           | 0.07-0.09 <sup>2</sup> |  |
| Robarts et al. (1996) <sup>2,M</sup>              | Water    | 0.05     | 0.11              |           |                        |  |
| Furtado et al. (2001) <sup>2,C</sup>              | Water    |          |                   |           | 0.35                   |  |
| Starink et al. (1996) <sup>2,L</sup>              | Sediment |          |                   |           | 0.1-0.2                |  |

1: AO epifluorescence microscopy; 2: DAPI epifluorescence microscopy; 3: FITC epifluorescence microscopy; 4: fluoresceine epifluorescence microscopy; 5: scanning eletronic microscopy; 6: transmission eletronic microscopy; \*: estimated value based on the data presented; \*\*: image analysing; C: coastal environment; E: estuarine environment; F: fjord; L: lake environment; M: marine environment; S: soil.

### Discussion

The small depth of Barra Lagoon with constant circulations without a determined stratification pattern and the winds promote a frequent oxygen distribution from surface to bottom layers of water column (Carmouze et al., 1993).

Similar total bacterial numbers (10<sup>7</sup>-10<sup>9</sup> bact/g) were observed at the sediment from the natural oxbow lagoon Lagoa do Infernão, of a Brazilian tropical river, Rio Mogi-Guaçu (Freitas & Godinho-Orlandi, 1991). Total density of bacteria (DAPIDC) was 2-4 orders of magnitude higher than total heterotrophic bacteria (plate counts). The methods for bacterial enumeration in natural environments are inefficient and need improvement. Many bacteria are viable in the environment but cannot grow in culture medium because it is simpler in composition when compared to the natural ecosystem where the bacteria use a wide range of survival strategies (Roszak & Colwell, 1987; Kogure et al., 1987; Güde, 1993; Oliver et al., 1995). It can explain the absence of correlation among total bacterial densities (DAPIDC method) and heterotrophic culturable bacteria (CFU method) for data from pelagic zone.

The average number found of heterotrophic culturable bacteria (plate counts) is very similar to that obtained in sediment samples from Sepetiba Bay (Rio de Janeiro State), a coastal Brazilian ecosystem (Pagnocca et al., 1991), but it was lower than the observed at the sediment from Lagoa do Infernão, the natural oxbow freshwater lagoon of a Brazilian tropical river (Freitas & Godinho-Orlandi, 1991), maybe because the physical conditions are diverse. The significant correlations (p < 0.05) among heterotrophic bacteria and salinity (r = 0.95), pH (r = 0.97) and nitrogen content (r = 0.90) found in pelagic zone, but not in littoral zone demonstrate diverse conditions in Barra Lagoon.

Among the heterotrophic culturable bacteria groups, the proteolytics were predominant in the clay sediment, while the amylolytics were in the sandy sediment. At the plankton compartment (water column) proteolytics were also dominants (Gomes et al., 1998). Generally the heterotrophic bacterial number is proportional to organic compounds content present in the water as well as in the sediment (Drabkova, 1983; Dutka & Kwan, 1983). Nevertheless we didn't find significant correlation among any kind of heterotrophic culturable bacteria and total carbon content of sediments. But a higher number of significant correlations among proteolytic bacteria in pelagic zone (pH r = 0.97, salinity r = 0.95, nitrogen content r = 0.90, sodium content r = 0.90) suggest that protein molecules are present in higher concentrations in the clay sediment.

There is little information about bacterial biomass in aquatic system, particularly from benthic compartment. Despite of diverse conditional of different ecosystems, benthic bacterial biomass found at Barra Lagoon was higher when compared with other environments, such as Kiel Bay (0.006-0.03 mg/mL) or Tyrrheniano Sea (7.0-76.8 mgC/g dry wt) (Fabiano & Danovaro, 1994; Ritzrau & Graf, 1992), probably because the higher temperatures in tropical region favor the growth of bacteria and other organisms. Our benthic bacterial biomass data and phyto- and zooplankton biomass of Barra Lagoon obtained during the same period by Domingos et al. (1994) and Arcifa et al. (1994) suggest the high productivity of this tropical coastal lagoon, such is indicated by Barnes (1980).

The contribution of geometric forms was similar in both sediments; cocci were more abundant whereas filaments showed higher biomass. Accumulation of nutrients in the sediments provides a more homogeneous, stable and diverse environment when compared with the water column (Jones, 1980). Bacterioplankton of Barra Lagoon was less diverse in forms, and cocci cells, including coccobacilli (very small rods), dominated the cellular density and biomass (Gomes et al., 1998).

Studies of the size structure of aquatic bacterial community started in the end of 1980's (Güde, 1989; Gasol et al., 1991 and 1997). Most of bacteria are very small (< 0.05 mm<sup>3</sup>) and coccoids in the water bodies, but they are able to assimilate labelled nutrients, therefore they are active metabolically (Roszak & Colwell, 1987; Morita, 1988; Güde, 1993).

The average sizes of the cell forms (cocci, rods, filaments) were similar in both clay and sandy sediments, and they were slightly greater than those found for bacterioplankton community at the same lagoon and period (Gomes et al., 1998). The morphology seems poor under a microscope analysis, but many bacteria exhibit morphological flexibility genetically determined how an adapting answer to environmental conditions (biotic and abiotic), such as the cell capacity to concentrate nutrient inside the cell ("in-take pressure"), nutrient availability ("bottom up pressure") and grazing by other organisms ("top down pressure") (Letarte & Pinel-Alloul, 1991; Ducklow, 1992; Gaedke, 1992; Jost et al., 1992; Güde, 1993; Starink et al., 1996).

Because of the scarcity of the data from similar environments we compared our biovolume data with biovolume found by different methods and environments (Tab. III). According Bratbak (1985) low differences can be due the different methods, but they are not significant. In Barra Lagoon, the average biovolumes of cocci and rods were higher in the sediment samples compared to water samples, contributing to higher biomass (Gomes et al., 1998). Among all of the form classes the filaments showed the highest biovolumes, so in general they are not considered in other papers. The biovolume values

obtained for Barra Lagoon were superior to the others, except that obtained from soil samples (Bakken & Olsen, 1983). Most environments showed very low average biovolumes (< 0.2 mm<sup>3</sup>) and the higher values were found in eutrophic environments (Bakken & Olsen, 1983; Lewis et al., 1986; Furtado et al., 2001; Gomes & Godinho, 2003).

We conclude that bacteria attributes (density, biomass, size cell, composition metabolic activity) answer to different trophic conditions, but not in clay and sand sediments of Barra Lagoon, a small and shallow lagoon where the circulation promote a efficient mixture and interactions of water and sediment. But this study is preliminary in microbial benthic system in tropical region and additional investigations about contribution of bacteria in the functioning of aquatic system are necessary.

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