CHANGES IN BIOMASS, CHEMICAL COMPOSITION AND NUTRITIVE VALUE OF *Spartina alterniflora* DUE TO ORGANIC POLLUTION IN THE ITANHAÉM RIVER BASIN (SP, BRAZIL)

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(With 4 figures)

ABSTRACT

We compared the values of the biomass, chemical composition and nutritive value of the emergent aquatic macrophyte *S. alterniflora* in a river affected by the discharge of domestic sewage (Guaú River) and in an unpolluted river (Itanhaém River). *S. alterniflora*, water and sediment samples were obtained in the two rivers in November, 2001. The Guaú River presented the highest levels of Total-N and Total-P in the water (415 and 674 $\mu g.L^{-1}$, respectively) and in the sediment (0.25 e 0.20% of the Dry Mass, respectively), when compared to the water (TN = 105 $\mu g.L^{-1}$; TP = 20 $\mu g.L^{-1}$) and the sediment (NT = 0.12% DM; PT = 0.05% DM) of the Itanhaém River. Aerial (316 g $DM.m^{-2}$) and subterraneous (425 g $DM.m^{-2}$) biomass of *S. alterniflora* were significantly higher in the Guaú River than in the Itanhaém River (146 and 115 g $DM.m^{-2}$, respectively). In addition, the values of TN, protein, TP, lipids and soluble carbohydrates were significantly higher in *S. alterniflora* biomass from the Guaú River. On the other hand, the values of the polyphenols and the cell wall fraction were significantly higher in the biomass of *S. alterniflora* from the Itanhaém River. We concluded that domestic sewage discharge in water bodies may increase the biomass and change the chemical composition of *S. alterniflora*. The high N and P availability in the water of the Guaú River is probably the cause of the higher biomass, TN, TP, protein, lipids and soluble carbohydrates measured in *S. alterniflora* in this river.

Keywords: biomass, chemical composition, nutritive value, *Spartina alterniflora*.

RESUMO

Mudanças na biomassa, composição química e valor nutritivo de *Spartina alterniflora* devido à poluição orgânica na Bacia do Rio Itanhaém (SP, Brasil)

Foram comparados a biomassa, a composição química e o valor nutritivo da macrófita aquática emersa *S. alterniflora* em um rio impactado por descargas de efluentes domésticos (Rio Guaú) e em um rio bem conservado (Rio Itanhaém). Amostras de *S. alterniflora*, água e sedimento foram coletadas nos dois rios, em novembro de 2001. O rio Guaú apresentou as maiores concentrações de N-Total e P-Total na água (415 e 674 $\mu g.L^{-1}$, respectivamente) e sedimento (0,25 e 0,20% de Massa Seca, respectivamente), em relação a água (NT = 105 $\mu g.L^{-1}$; PT= 20 $\mu g.L^{-1}$) e sedimento (NT = 0,12% MS; PT = 0,05% MS) do rio Itanhaém. A biomassa aérea (316 g $MS.m^{-2}$) e subterrânea (425 g $MS.m^{-2}$) de *S. alterniflora* no rio Guaú foram significativamente maiores do que no rio Itanhaém (146 e 115 g $MS.m^{-2}$). Além disto, os valores de NT, proteínas, PT, lipídios e carboidratos solúveis foram significativamente maiores na biomassa de *S. alterniflora* no rio Guaú. Por outro lado, a fração de parede celular e os teores de polifenóis foram maiores na biomassa de *S. alterniflora* no rio Itanhaém. Concluiu-se que o lançamento de efluentes domésticos em corpos d'água pode aumentar a biomassa e alterar a composição química de *S. alterniflora*. A maior disponibilidade de N e P no rio Guaú, provavelmente, é a causa dos maiores valores de biomassa, NT, PT, lipídeos e carboidratos solúveis em *S. alterniflora* neste rio.

Palavras-chave: biomassa, composição química, valor nutritivo, *Spartina alterniflora*.

INDRODUCTION

Domestic sewage discharges in water courses increase the concentrations of organic matter and nutrients that can affect the productivity of the system, increase particulate materials and reduce the amount of dissolved oxygen (Laws & Allen, 1996; Hall *et al.*, 1999). As a consequence, aquatic organism populations may change in quantity and quality (Ricklefs & Miller, 1999). According to Laws (1999), domestic sewage discharges affect the aquatic macrophyte communities in terms of growth and species occurrence and can promote species substitutions. Eutrophication, in general, causes the reduction or elimination of submerged aquatic macrophytes due to reduced light penetration caused by increased quantities of suspended matter in the water (Madsen & Adams, 1988). On the other hand, floating species are favoured by nutrient increases in water (Bini *et al.*, 1999). Emergent macrophytes, which are not in direct competition for nutrients or light with either algae or submerged plants, may show enhanced growth and biomass over a wide range of nutrient inputs (Harper, 1995).

Nutrient increases in water and sediment, as a consequence of eutrophication, can change the chemical composition of aquatic plants as there is a close relationship between the nutrient availability in the ecosystem and in the plant tissue (Tucker, 1981; Morris, 1982; Esteves, 1998). In fact, nutrient availability can directly affect the growth rate of some aquatic macrophyte species (Henry-Silva *et al.*, 2002). In addition, the chemical composition of the aquatic plants influences the use of biomass by herbivorous and detritivorous animals (Barbieri & Esteves, 1991).

In the estuarine region of the basin of the Itanhaém River (São Paulo South Coast), there are unpolluted rivers and some tidal creeks affected by domestic waste (Camargo *et al.*, 2002). In both aquatic environments the emergent aquatic macrophyte *Spartina alterniflora* is abundant and occurs close to the border of mangrove vegetation (Tonizza, 2002). The objective of the present study is to compare the values of biomass, chemical composition and nutrient value of *S. alterniflora* in a river affected by domestic sewage discharge (Guaú River) and in a non-affected river (Itanhaém River).

Study area

The Guaú and Itanhaém Rivers are located in the hydrographic basin of the Itanhaém River (Fig. 1). This hydrographic basin has a total area of 950 km^2 and is located on the South coast of São Paulo State, South-eastern Brazil (23° 50' and 24° 15' S; 46° 35' and 47° 00' W) (Suguiu & Martin, 1978). According to Laparelli & Moura (1998), the climate of the region presents little variation due to its latitude and proximity to the Atlantic Ocean. Rain is abundant (annual mean of 2183 mm), with more rainfall in March (279.9 mm) and less in August (84.7 mm). The atmospheric temperature in the region is high, with a minimum mean of 19.0 °C (July) and the maximum mean of 26.2 °C (February).

The Guaú River is a tidal creek that crosses the urban zone of Itanhaém municipality and receives domestic waste without previous treatment (Camargo *et al.*, 1996). In the lower course, the Guaú River flows through mangrove vegetation and some shore areas are colonized by monospecific stands of *S. alterniflora*.

The Itanhaém River is the final channel of the hydrographic basin and is located on a coastal plain. This river is formed by the confluence of the Preto and Branco rivers. These two rivers drain well preserved areas of tropical rain forest of the State Park of the Sea Range, preserved areas of restinga vegetation and some areas with banana farms (Camargo *et al.*, 1997). The Itanhaém River flows through mangrove vegetation with stands of *S. alterniflora*, *Crinum procerum* and *Scirpus californicus* on shore areas.

MATERIAL AND METHODS

Water, sediment and *S. alterniflora* samples were collected in the lower course of the Guaú

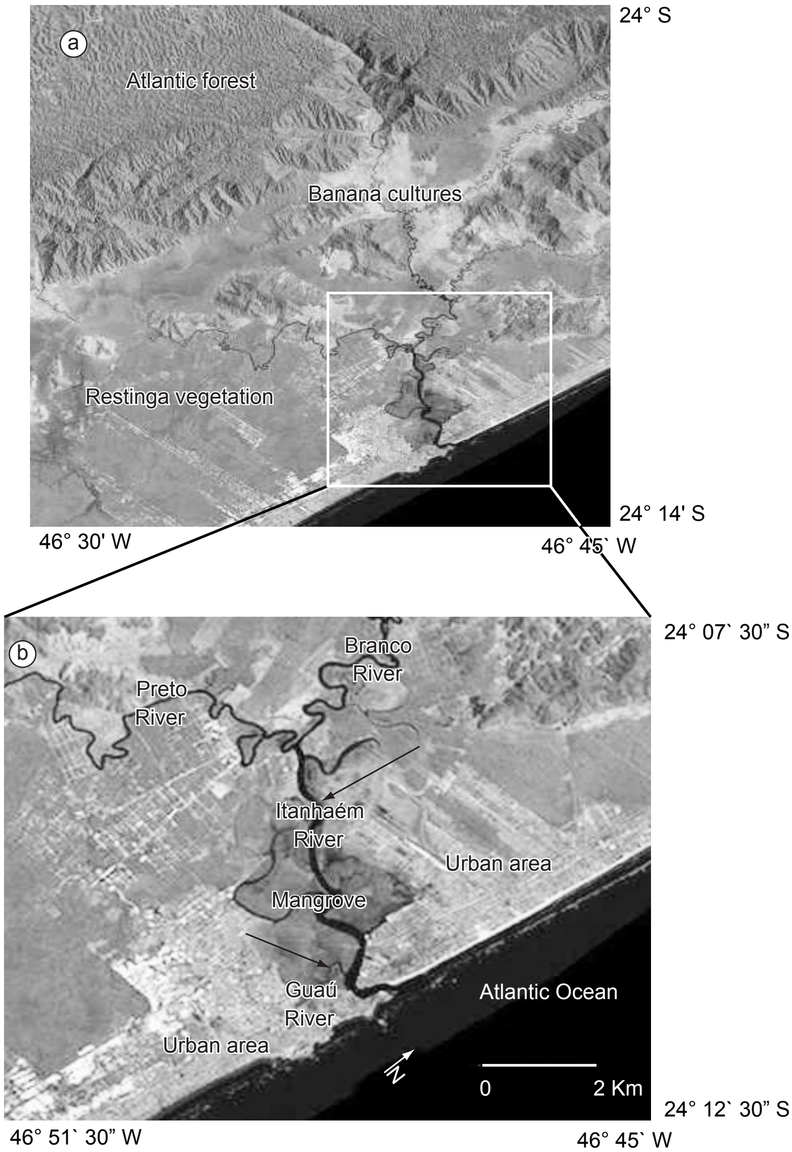

Fig. 1 — Satellite images: a) Itanhaém River Basin; and b) Estuarine region of the Itanhaém River basin. The arrows indicate the sample stations.

River and in the upper course of the Itanhaém River in November, 2001. Simultaneously, the pH values, salinity and turbidity were taken (in triplicate) using a Water Quality Checker, Horiba (model U10). Photosynthetic active radiation (PAR) was measured (in triplicate) at 0.0 m and 0.2 m, with the Light Meter LiCor (model 189) and the coefficient of the vertical attenuation (K) was calculated using the following equation: $K = (ln. Io - ln.I)/z$, where *ln* is the natural logarithm, *I* is the PAR at a determined depth (m), *Io* is the PAR at the surface and *z* is the depth (m) (Wetzel, 1983).

Water samples (in triplicate) were collected near stands of *S. alterniflora*. Afterwards, dissolved oxygen (dissolved O_2) was measured using Winkler`s method according to Golterman *et al.* (1978). The total nitrogen (TN), total dissolved nitrogen (TDN), nitrite (NO_2-N) and nitrate (NO_3 N) were obtained by the methods described in Mackreth *et al.* (1978). Ammoniacal nitrogen (Ammoniacal-N) was determined using the method proposed by Koroleff (1976). Total inorganic nitrogen (TIN) was calculated by the sum of NO_3 N, NO_2-N and Ammoniacal-N. Total phosphorus (TP), total dissolved phosphorus (TDP) and orthophosphates (PO_4-P) were measured through methods described in Golterman *et al.* (1978).

Sediment samples (in 5 replicates) were also collected near the stands of *S. alterniflora* to measure the total nitrogen (TN) using Kjeldahl`s method (Allen *et al.*, 1974), and the total phosphorus (TP) using the same methodology utilized by Esteves (1980).

S. alterniflora samples (in 5 replicates) were obtained within a square of 0.25 m^2. The plants were cleaned through successive washes in order to remove the periphyton, organic matter and associated inorganic particles. Thereafter, the plants were separated into the aerial parts (stalk and leaf) and subterraneous parts (root and rhizome), dried at 60 °C and weighed in order to determine the values of aerial and subterraneous biomass in grams of dry mass by square meter (g $DM.m^{-2}$). Afterwards, the samples were ground in a mill to determine the chemical composition.

The total nitrogen (TN) was determined by the Kjeldahl method according to Allen *et al.* (1974). Total phosphorus (TP) was determined by the method described by Esteves (1980). Soluble carbohydrates were obtained according to Esteves (1979). Lipids were obtained by the method described by Folch *et al.* (1957). The percentage of cell wall fraction was determined according to Van Soest & Wine (1967). Polyphenols were measured by the method described in King & Healt (1967). Protein was calculated by multiplying the results of total nitrogen by 6.25 (Boyd, 1970). Afterwards, the values of chemical composition in the total biomass were determined using the pondered mean of the respective values obtained in aerial and subterraneous biomass.

The t-test (Student) was applied to water and sediment data in order to check for significant differences ($p \leq 0.05$) between the Itanhaém and Guaú rivers. Prior to ANOVA (one way), we checked the homogeneity of variances in *S. alterniflora* data using Bartllet's Test. When significant differences ($p \leq 0.05$) occurred, Tukey's HSD test was used to identify differences in the values of biomass and chemical composition among total biomass, aerial biomass, subterraneous biomass of *S. alterniflora* in the two rivers (Zar, 1996).

RESULTS

Table 1 presents the mean and standard deviation of the physical and chemical variables measured in the water and sediment of the Itanhaém and Guaú rivers. The values of temperature, turbidity and K were significantly higher ($p \leq 0.05$) in the Guaú River, while dissolved O_2 was 0 $mg.L^{-1}$ in this river. The salinity and pH values of the two rivers were not significantly different. In relation to nutrients, the Guaú River presented the highest values of TN and TP in water and sediment. This river also presented the highest values of TDN, TIN, TDP and PO_4-P.

Fig. 2 shows the mean and standard deviation of the total, aerial and subterraneous biomass of *S. alternilfora*, in the Itanhaém and Guaú rivers. The total biomass *of S. alterniflora* in the Guaú River (732 g $DM.m^{-2}$) was significantly higher than in the Itanhaém River (262 g $DM.m^{-2}$). In the Guaú River, aerial (316 g $DM.m^{-2}$) and subterraneous (425 g $DM.m^{-2}$) biomass of *S. alterniflora* were also significantly higher when compared to aerial (146 g $DM.m^{-2}$) and subterraneous (115 g $DM.m^{-2}$) biomass of *S. alterniflora* in the Itanhaém River.

Fig. 3 and Fig. 4 present the mean and standard deviation of the chemical composition in total, aerial and subterraneous biomass of *S. alterniflora* in the Itanhaém and Guaú rivers.

When comparing the chemical composition of the total biomass of *S. alterniflora* in the two rivers, it can be observed that in the Guaú River there are significant higher values of TN, TP, protein, soluble carbohydrates and lipids. In the Itanhaém River, the values of polyphenols and cell wall fraction of the total biomass of *S. alterniflora* were significantly higher (Fig. 4). The same differences were observed for aerial and subterraneous biomass. In fact, both plant parts in the Guaú River showed significant higher values of TN, TP, protein, soluble carbohydrates and lipids. Aerial and subterraneous biomass of *S. alterniflora* in the Itanhaém River showed significantly higher values of polyphenols and cell wall fraction than in the Guaú River.

TABLE 1
Mean and standard deviation (SD) of physical and chemical variables of water and sediment from the Itanhaém and Guaú rivers.

222 Variables	Itanhaém River		Guaú River	
	Mean	SD	Mean	SD
pH	7.5	± 0.2	7.3	± 0.3
Salinity (‰)	9.8	± 0.6	10.3	± 0.2
Turbidity (NTU)*	15	± 7.5	68	± 8.8
Coefficient K (m^{-1})*	2.6	± 0.21	6.5	± 0.52
Dissolved O_2 ($mg.L^{-1}$)*	6.5	± 0.5	0	
Total Nitrogen ($\mu g.L^{-1}$)*	105	± 10	415	± 50
Total Dissolved N ($\mu g.L^{-1}$)*	67	± 10	320	± 20
Total Inorganic N ($\mu g.L^{-1}$)*	113.8	± 12.0	325.9	± 25.5
Total Phosphorus ($\mu g.L^{-1}$)*	20.7	± 2.1	674.1	± 72.1
Total Dissolved P ($\mu g.L^{-1}$)*	20	± 2.5	564	± 90.5
PO_4-P ($\mu g.L^{-1}$)*	12.2	± 0.6	320	± 20.5
Total N sediment (% DM)*	0.12	± 0.02	0.25	± 0.02
Total P sediment (% DM)*	0.05	± 0.02	0.20	± 0.05

The symbol * indicates significant difference ($p \leq 0.05$) between the rivers.

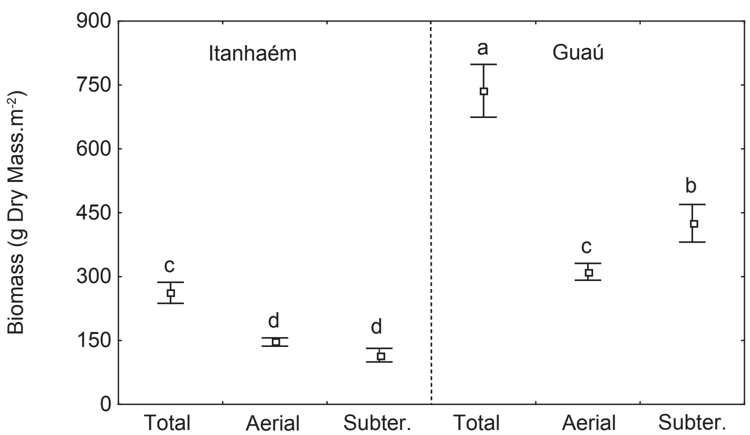

Fig. 2 — Mean and standard deviations (bars) of the total, aerial and subterraneous (subter.) biomass of *S. alterniflora*, in the Itanhaém and Guaú rivers. Different letters indicate significant differences ($p \leq 0.05$).

In both rivers the concentrations of TN, TP, proteins, lipids and soluble carbohydrates were higher in the aerial biomass of *S. alterniflora* (Figs. 3 and 4), while the percentage of cell wall fraction was higher in the subterraneous biomass (Fig. 4c).

DISCUSSION

The results of physical and chemical variables of water and sediment demonstrated that the Guaú River has higher levels of organic pollution than the Itanhaém River (Table 1). Other authors (Camargo *et al.*, 1995; Sanches & Camargo, 1995; Henry-Silva & Camargo, 2000) studying others rivers in São Paulo State submitted to urban wastewater discharge, also observed high nutrient concentrations (TN, TIN, TP and PO_4-P) and low dissolved O_2 levels as well as water transparency.

Probably, the highest concentrations of N and P in the water and sediment of the Guaú River are responsible for the higher values of biomass,

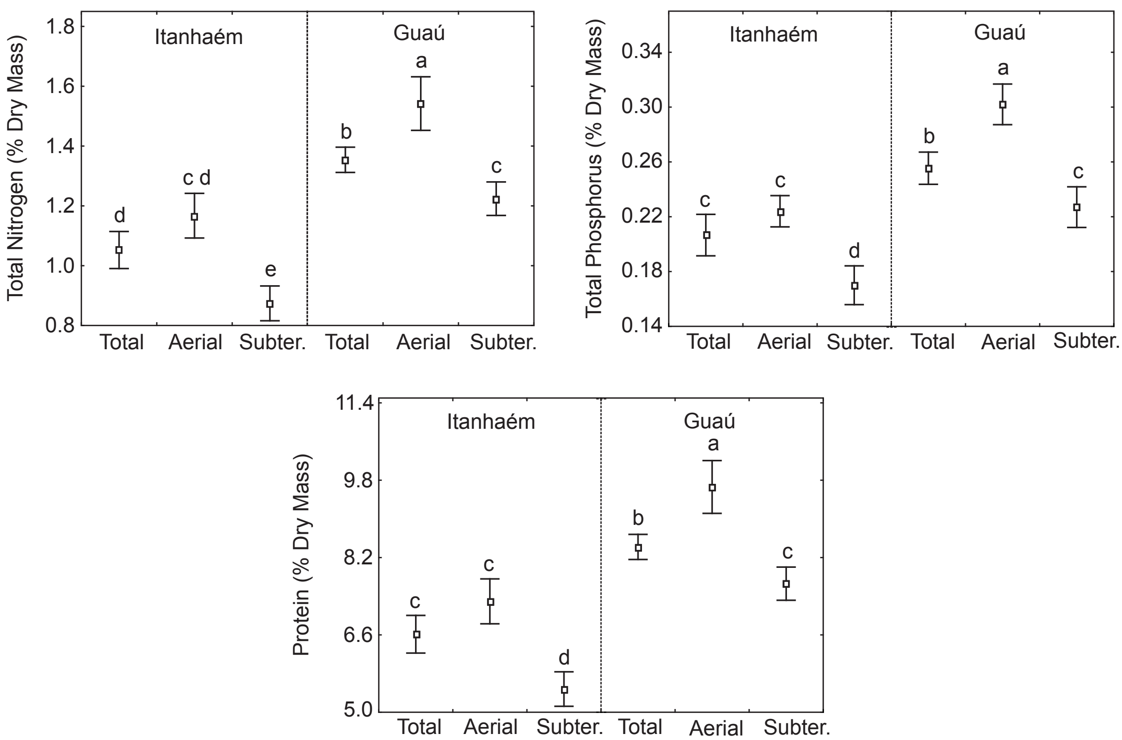

Fig. 3 — Mean and standard deviations (bars) of total nitrogen (a), total phosphorus (b), protein (c) in total, aerial and subterraneous (subter.) biomass of *S. alterniflora*, in the Itanhaém and Guaú rivers. Different letters indicate significant differences ($p \leq 0.05$).

TN and TP of *S. alterniflora* from this river. In fact, the environmental availability of N and P control the growth of freshwater plants (Ricklefs & Miller, 1999). Therefore, the absorption of these two nutrients by plants, in general, is higher when the environmental availability of N and P is higher (Larcher, 2000). On the other hand, anoxia and low transparency probably did not limit the growth of *S. alterniflora* in the Guaú River as emergent aquatic macrophytes make gaseous changes and use direct light from the atmosphere (Esteves, 1998; Camargo *et al.*, 2003). Morris (1982) in a laboratory experiment also observed higher biomass of *S. alterniflora* in higher concentrations of nitrogen. In addition, Pierce (1983) measuring biomass of *Spartina maritima* in Algoa Bay (South Africa) obtained higher values for the areas with higher concentrations of nitrogen and phosphorous (Table 2).

The highest concentrations of protein, soluble carbohydrates and lipids in the total biomass of *S. alterniflora* in the Guaú River are probably related to the higher availability of nitrogen and phosphorus in this river. These nutrients contribute to the biosynthesis of protein, soluble carbohydrates and lipids and are part of enzymes and cofactors (ATP, NADP) involved in these process (Salisbury & Ross, 1992). Barbieri & Esteves (1991) also observed a positive relationship between N and P availability in the water column and the protein concentration, soluble carbohydrates and lipids in the freshwater aquatic macrophytes *Pontederia cordata* and *Utricularia breviscapa*.

The differences in the chemical composition between aerial and subterraneous biomass of *S. alterniflora* are probably related to the different functions of the plant structures. According to Esteves (1998), in general, the plant part with a high metabolic rate has a high concentration of nutrients and according to Barbieri & Esteves (1991) it also has higher values of soluble carbohydrates, lipids and polyphenols. Therefore, the higher

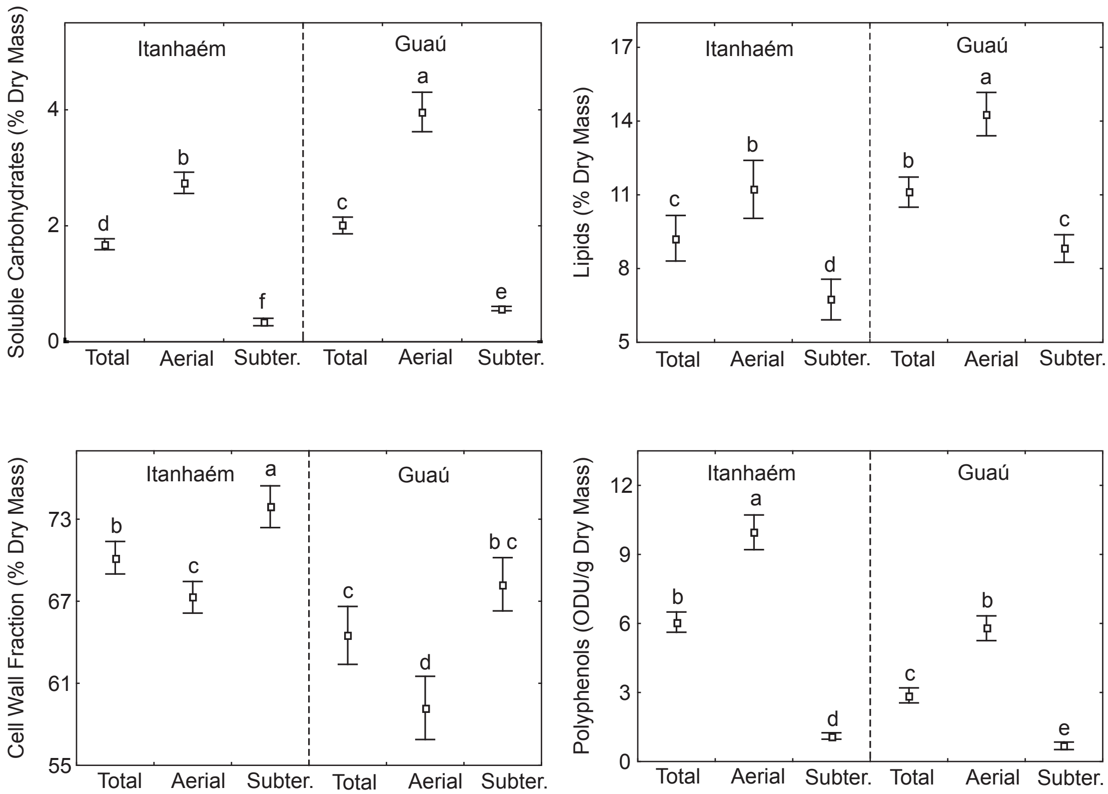

Fig. 4 — Means and standard deviations of soluble carbohydrates (a), lipids (b), cell wall fraction (c) and polyphenols (d) in total, aerial and subterraneous (subter.) biomass of *S. alterniflora*. Different letters indicate significant differences ($p \leq 0.05$).

TABLE 2
Total biomass of *Spartina alterniflora* and *Spartina maritima* in different concentrations of total nitrogen (TN) and total phosphorus (TP).

Species	Total Biomass ($g\ DM/m^2$)	Water		Sediment		Place	Source
		TN ($\mu g.L^{-1}$)	TP ($\mu g.L^{-1}$)	TN (% DM)	TP (% DM)		
Spartina alterniflora	850	350	-	0.5	-	Laboratory	Morris (1982)
	305	152	-	0.1	-	Laboratory	
Spartina alterniflora	732	415	674	0.25	0.20	River Guaú	This work
	262	105	20	0.12	0.05	River Itanhaém	
Spartina maritima	780	525	860	0.33	0.32	Algoa Bay	Pierce (1983)
	523	228	220	0.23	0.18	Algoa Bay	

values of TN, TP, soluble carbohydrates, lipids and polyphenols in aerial biomass of *S. alterniflora* are probably due to the high enzymatic activity of photosynthetic structures.

A whole analysis of the chemical composition of aquatic macrophytes provides information about the nutritive value of the biomass (Barbieri & Esteves, 1991). In fact, proteins, lipids and carbohydrates are essential in animal feeding as they are structural components and an energy source (Randall *et al.*, 1997). According to some studies (Onuf *et al.*, 1977; Kraft & Denno, 1982;

Coley, 1983), leaf protein content has been shown to play a central role in determining herbivore feeding patterns in several aquatic and terrestrial communities. On the other hand, high concentrations of polyphenols (antocians, tanins, flavonols, metil-propans and fenil-propans) and high percentages of cell wall fraction (cellulose, lignin and nitrogeneted lignificat substances of hard digestion) decrease the consumption of biomass by aquatic animals (Barbieri & Esteves, 1991; Henry-Silva *et al.*, 2001). Therefore, *S. alterniflora* from the Guaú River has a better nutritive value due to the higher values of protein, lipids, carbohydrates and lower values of polyphenols and cell wall fraction in the biomass. These results suggest that *S. alterniflora* is a food source more available for the herbivores food chain in the Guaú River because generalists' herbivores will first consume food with a high nutritive value (Begon *et al.,* 1996). Furthermore, the higher values of *S. alterniflora* biomass in the Guaú River will probably increase its availability as a food source even more.

We conclude that the discharge of domestic sewage in water bodies may increase the biomass and change the chemical composition of *S. alterniflora*. Among the limnological characteristics changed by the discharge of domestic sewage, the higher N and P availability, is probably the cause of the higher biomass, TN, TP, protein, lipids and soluble carbohydrates in *S. alterniflora* from the Guaú River.

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