Seasonal changes in the subtidal benthic macrofauna of a mangrove coast in northern Brazil

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ABSTRACT

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Mangrove coasts in northern Brazil are subject to seasonal discharges of freshwater input from rivers and terrestrial runoff. However, little is known about the effects of freshwater discharge on the subtidal benthos in the region. Thus, three sites, each representing a different level of seasonal discharge, Canela island (low), Ajuruteua beach (medium), and the Caeté Bay (high) were sampled in the dry (October) and wet (April/May) seasons of 2007 and 2008, respectively. Sampling was carried out using a Petersen grab and a bottom dredge (total n=74). Additional replicas (n=12) were obtained by grab for sediment analyses. Salinity and turbidity of the bottom waters, and percentage of silt/clay in the sediment were also measured. Benthic macrofauna and environmental data were analyzed using uni- and multivariate techniques. A total of 661 individuals were distributed among 47 taxa of which polychaetes and crustaceans dominated at all three sites. No significant differences in macrofaunal abundance were found among sites and dates. At all sites, in the wet season, the number of taxa was low and dominance was high, especially in the Caeté Bay. The polychaete *Nephtys fluviatilis* only occurred in the Caeté Bay in the wet season, and yet was the dominant taxon (22.7% of total abundance). The Ajuruteua beach and Canela island samples were composed of typically marine taxa. The polychaete *Armandia* sp., the gastropod *Olivella minuta,* mysiid and phoxocephalid crustaceans and the echinoderm family Mellitidae were most abundant in the dry season at Canela island. Salinity was the most important environmental factor associated with high numbers of taxa in the dry season. Silt and turbidity were associated with low numbers of taxa in the wet season. In conclusion, the subtidal benthic macrofauna at Canela island, Ajuruteua beach and the Caeté Bay is mainly composed of polychaetes and crustaceans. The higher freshwater discharge in the wet season, resulting in lower salinity, higher percentage silt and greater turbidity, is associated with lower abundance, fewer taxa and a different faunal composition at Canela and Ajuruteua, and strong dominance of an oligohaline polychaete in the Caeté Bay

ADDITIONAL INDEX WORDS: *Temporal variation, zoobenthic structure, freshwater discharge, tropical estuary.*

INTRODUCTION

Freshwater inflow affects salinity, turbidity, temperature and nutrient levels, among other factors (Turek *et al*., 1987; Baldó *et al*., 2005; Palmer *et al*., 2011; Pollack *et al*., 2011). Inflow may vary greatly over temporal (Xu and Wu, 2006; Miguel Pardo *et al*., 2011; Pollack *et al*., 2011; Tolley *et al*., 2012) and spatial scales (Palmer *et al*., 2011; Tolley *et al*., 2012) and in turn affect biological assemblages and processes in estuaries and coastal areas (Montagna and Kalke, 1992; Aller and Stupakoff, 1996; Palmer *et al*., 2011; Pollack *et al*., 2011; Rozas *et al*., 2005; Silva *et al*., 2012), although there is some doubt over the generality of such effects (Ardisson and Bourget, 1997).

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Benthic macrofaunal assemblages may alternate between being dominated by marine, brackish or freshwater taxa due to freshwater inflow (Kim and Montagna, 2009; Palmer *et al*., 2011; Silva *et al*., 2012; Correia *et al*., 2012). Marine macrofauna have economic and ecological importance due to their secondary production, and their roles in linking the benthos with the water column, filtration, bioturbation and nutrient cycling (Snelgrove, 1998). Thus, seasonal changes in freshwater input may alter the rate of certain ecological processes as the assemblage adjusts to differing conditions. For example, nutrient regeneration increases with freshwater inflow but may decrease if low salinity conditions are prolonged, due to mortality of the organisms responsible for the regeneration process (Kim and Montagna, 2009).

The Amazon estuary is one of the best known examples of a large freshwater input to a coastal area. Other rivers along the Amazonian mangrove macrotidal coast (Souza-Filho *et al*., 2009) of northern Brazil have significant seasonal discharges causing variation in salinity, nutrients and suspended sediment loads to coastal areas (Dominguez, 2009). These changes in turn cause responses in a variety of different organisms, including phytoplankton (Santos *et al*., 2008) and the benthic macrofauna (Aller and Stupakoff, 1996). Seasonal effects of freshwater runoff have been well documented for coastal macrofauna in diverse North and South American estuaries and lagoons (Montagna and Kalke, 1992; Aller and Stupakoff, 1996; Ieno and Batisda, 1998; Rozas *et al*., 2005) and the aim of the present study was to verify if differences in macrofaunal structure occur at three sites off a mangrove coast in northern Brazil, each one reflecting a different level of freshwater inflow during high and low seasonal freshwater discharge.

METHODS

Study Area

The Ajuruteua Peninsula is located on the highly indented macrotidal mangrove coast of Pará state, northern Brazil (Souza-Filho *et al*., 2009; Saint-Paul and Schneider, 2010). Semi-diurnal macrotides up to 6 m in height and current velocities up to 1.5 ms^{-1} occur during spring tides (Cohen *et al*., 1999). The Caeté river discharge varies from 180 to 0.3 $m3s^{-1}$ in the wet and dry season, respectively (Lara and Dittmar, 1999). Between 2007 and 2008, mean annual rainfall was 2688 mm with over 75% falling between January and May (INMET, www.inmet.gov.br). In 2007, precipitation was lowest in October (10.4 mm), whereas in 2008, highest rainfall was in March (650 mm and 29 days of rainfall). In April and May 2008, rainfall was 418 and 456 mm, respectively with 28 days of rainfall in each month. Almost the entire coast in the study area is mangrove, dominated by *Rhizophora mangle*, followed by *Avicennia germinans* and *Laguncularia racemosa* (Mehlig *et al*., 2010).

Field and laboratory work

Three locations, each representing a different level of freshwater discharge, Canela (low), Ajuruteua (medium) and the Caeté Bay (high) were sampled around the Ajuruteua Peninsula (Figure 1) for subtidal benthic macrofauna. Sampling was carried out twice at each site, in October 2007 and in April/May 2008, representing low and high levels of rainfall, respectively.

A Global Positioning System was used to locate random positions, along 3 parallel transects, roughly 20 m apart, at each site on each sampling date. Depth varied between 4 and 10 m at sites. Bottom sampling was carried out at each of the three sites on both sampling dates using a 3 L Petersen grab (n=12) and a 60 by 20 cm horizontal dredge towed for 3 minutes (n=3). The volume of sediment in each grab or tow was standardised on board to 3 L. A total of 90 replicate collections of sediment were made (72 by grab and 18 by dredge). Sediment was washed and sieved on board through a 300 µm mesh and sealed in labelled plastic bags with 10% MgCl over ice. An additional 18 grab replicas at three random points at each site on both sampling occasions were used to analyse sediment characteristics. Bottom water was collected using a 2 L Van Dorn bottle. Salinity was measured on board whereas turbidity was measured in the laboratory using a colorimeter. Sediment containing macrofauna was transferred to 5% formaldehyde for 12 hours. Later, the material was washed with water and sorted using a stereoscope. Animals were transferred to 70% alcohol with glycerine (1:1) for counting and identification to the lowest taxonomic level possible. Sediment collected for analysis was washed to remove salt, dried to a constant weight at 60ºC and 100 g sub-samples were sieved through 2000 and 62 µm meshes to separate gravel, sand and silt/clay. Percentage silt/clay (hereafter % silt) was calculated with SYSGRAN 3.0 (Camargo, 2006).

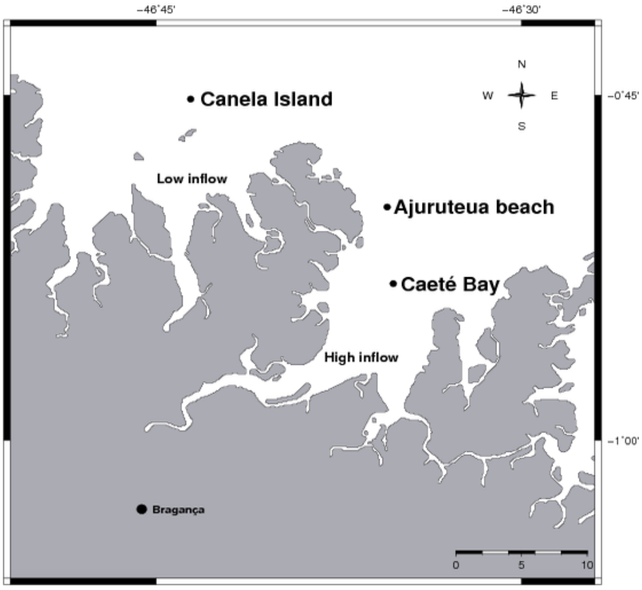

Figure 1.The Ajuruteua Peninsula, north of the city of Bragança, with sites at Canela island (low inflow), Ajuruteua beach (medium inflow and the Caeté Bay (high inflow) sampled for subtidal macrofauna in October 2007 and April/May 2008.

Statistical analysis

Density (number of individuals per replica of sediment), number of taxa and Berger-Parker dominance were calculated and differences in mean values among the three sites and between both sampling dates were tested for using two-way analysis of variance (ANOVA). Diagnostic graphics were used to evaluate if the assumptions of ANOVA were met. Tukey's Honestly Significant Difference (HSD) *post hoc* test was used to identify significant differences between pairs of samples following a significant ANOVA result.

Abundance data was fourth root transformed and a distance matrix was calculated using Czekanowski's index (Yoshioka, 2008). Zero sum rows and a single anomalous replica were removed leaving 57 and 17 grab and dredge replicas, respectively. Replicas were ordinated using non-metric multidimensional scaling on the macrobenthos distance matrix and differences in macrofaunal structure among sites and dates were tested for using permutational Multivariate Analysis of Variance (perMANOVA). The relationship between environmental data and macrofaunal structure was verified using BIOENV where 15 macrofaunal replicas were used that corresponded to the same coordinates as the environmental replicas. All analyses, unless otherwise indicated, were carried out with GNU R using the *vegan* and *sciplot* packages (R Project, 2012). An R script may be requested from the senior author.

RESULTS

A total of 661 individuals distributed among 47 different subtidal macrofaunal taxa were found. *Nepthys fluviatilis* dominated (22.7%) and was found exclusively in the Bay in April/May. No differences in mean macrofaunal density occurred among sites or between dates (Table 1). However, interaction between Site and Date was significant. In October, macrofaunal density was highest at Canela and Ajuruteua and lowest in the Bay, whereas in April/May, the opposite was the case (Figure 2a), due to the very high abundance of *N. fluviatilis* in the Bay. The number of taxa was significantly higher in October at all sites and was significantly lower in the Bay (Figure 2b, Table 1).

Table 1. F values from ANOVAs of density, number of taxa and Berger-Parker dominance of the subtidal macrofauna sampled at Canela island, Ajuruteua beach and the Caeté Bay in October 2007 and April/May 2008. SV=Source of variation, df=degrees of freedom, *=p<0.05, **=p<0.01, ***=p<0.001. Tukey's HSD: Number of taxa Bay<Ajuruteua=Canela, Berger-Parker dominance Bay>Ajuruteua=Canela.

SV	df	Density	Number of Taxa	Berger-Parker
Site (S)	2	0.30	7.06**	7.9***
Date (D)	1	2.69	23.60***	22.12***
S×D	2	3.91*	2.09	0.32
Error	68			

Berger-Parker dominance was significantly higher in April/May at all sites and was significantly higher in the Bay. (Figure 2c, Table 1). There was no interaction between factors for number of taxa and Berger-Parker dominance. Other abundant taxa were Mysidae (17.7%), Mellitidae (13.0%), Nepthys sp. (8.5%), Phoxocephalidae (5.9%) and *Olivella minuta* (5.6%). Abundances of all other taxa were below 5% of the total. Macrofaunal structure was significantly different on each sampling date and also between sites (Figure 3, Table 2).

Table 2. PerMANOVA summary of differences in subtidal macrofaunal structure among sites and between sampling dates at Canela island, Ajuruteua beach and the Caeté Bay in October 2007 and April/May 2008. SV=Source of variation, df=degrees of freedom, MS=Mean Square, ***=p<0.001.

SV	df	MS	F	R^2
Site (S)	2	2.027	6.621***	0.135
Date (D)	1	2.546	8.318***	0.085
S×D	2	1.346	4.398***	0.089
Error	68	0.306	0.691	
Total	73	1.000		

Twenty nine taxa were found only in October whereas 9 were found only in April/May. Taxa with greatest abundance in October were Mysidae, Mellitidae, *Olivella minuta*, *Armandia* sp., *Scoloplos* sp., *Cylichna* sp., Sergestidae and Diogenidae whereas those in April/May were *N. fluviatilis*, *Tellina* sp., *Dispio* sp., *Notomastus* sp., Spionidae, *Scolelepis* sp. and Nemertea (Table 3). On both dates, the macrofauna from Ajuruteua and Canela appeared to be similar. In October, the Bay macrofauna was very variable, overlapping significantly with that of the other two sites. However, in April/May, the Bay macrofauna was very homogeneous and distinct from the other two sites.

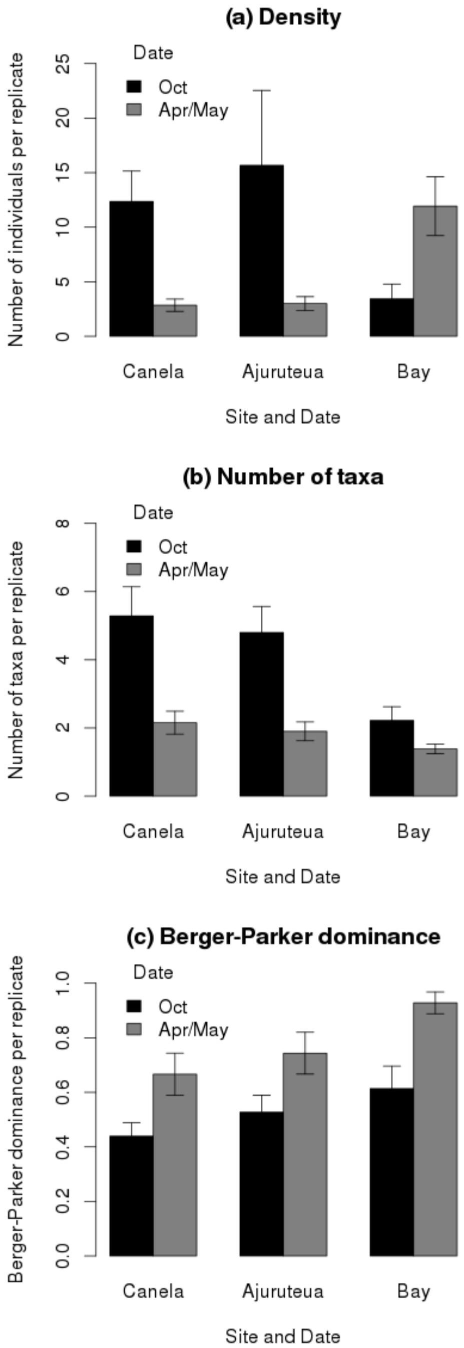

Figure 2. Density, number of taxa and Berger-Parker dominance of the subtidal macrofauna sampled at Canela island, Ajuruteua beach and the Caeté Bay in October 2007 and April/May 2008.

Table 3. Abundance (sum of replicas) of selected subtidal macrofaunal taxa at Canela island, Ajuruteua beach and the Caeté Bay sampled in October 2007 and April/May 2008.

Site	Canela		Ajuruteua		Bay	
Date Taxon	Oct	Apr/ May	Oct	Apr/ May	Oct	Apr/ May
Nephtys fluviatilis						150
Dispio sp.		5		3		
Tellina sp.	2	6	1	3		
Notomastus sp.		2				1
Spionidae		1		1		
Scoleleps sp.		1				
Nemertea		5		1		
Mysidae	7		104	1	5	
Mellitidae	43		41	1	1	
Olivella minuta	26		11			
Nephtys sp.	21	9	12	13	1	
Phoxocephalidae	11	3	11	7	6	1
Armandia sp.	9		13		2	
Sergestidae	1		9		3	
Diogenidae	5		6			
Cylichna sp.	11					1
Scoloplos sp.	6	2	2		2	

Salinity was significantly higher at all sites in October, mean 37.3, than in April/May, mean 10.2 (Figure 4a, Table 4). There was very little intra-site variation and all sites differed significantly, values being highest in Canela and lowest in the Bay. Interaction was significant due to the greater decline in salinity at Ajuruteua between sampling dates. In contrast, turbidity was significantly higher at all sites in April/May, mean 159.5 FAU, than in October, mean 64.9 FAU. There were no differences among sites nor was there significant interaction (Figure 4b, Table 4). Percentage silt was significantly lower in October, mean 0.5%, than in April/May, mean 15.5% (Figure 4c, Table 4) and, although there was no general effect of site, interaction was significant as the temporal difference was much greater in the Bay than in the other two sites. Tukey comparisons for % silt were close to significance for the Bay site (Canela:Ajuruteua P=0.98, Bay:Ajuruteua P=0.059, Bay:Canela P=0.08). Salinity and % silt were the two most important parameters associated with macrofaunal structure (BIOENV, r=0.362, p<0.05).

Table 4. F values from ANOVAs of salinity, turbidity and % silt at sites sampled for subtidal macrofauna at Canela island, Ajuruteua beach and the Caeté Bay in October 2007 and April/May 2008. SV=Source of variation, df=degrees of freedom, *=p<0.05, **=p<0.01, ***=p<0.001. Tukey's HSD: Salinity Bay<Ajuruteua=Canela.

SV	df	Salinity	Turbidity	% Silt
Site (S)	2	286.8***	1.48	4.13*
Date (D)	1	14884.0***	10.28**	5.53*
S×D	2	120.2***	0.52	4.39*
Error	12			

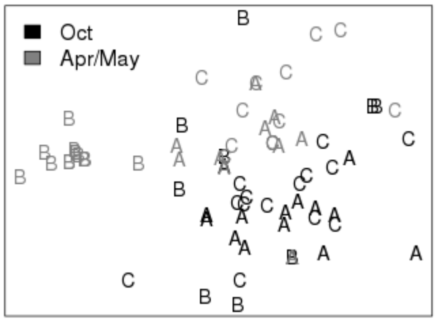

Figure 3. Ordination of plots based on fourth root transformed macrofaunal abundance at Canela island (C), Ajuruteua beach (A) and the Caeté Bay (B) in October 2007 and April/May 2008.

DISCUSSION

Freshwater inflow in April/May is associated with lower diversity and a different subtidal macrofauna around the Ajuruteua Peninsula. Around the mouth of the Amazon, Aller and Stupakoff (1996) found that abundances of lumbrinerid polychaetes, bivalves and gastropods were greatest during low inflow and increased with distance from the shore. Lower salinity due to a greater regulated freshwater inflow reduced abundance and biomass of macrofauna in the Guadalquivir estuary, southern Spain (Baldó *et al*., 2005). The shallow-water infaunal assemblage of the inner Doubtful Sound, New Zealand was less species rich and consistently different from other parts of this fjord complex due to a persistent freshwater input that reduced salinity in that area (Rutger and Wing, 2006). Many estuaries have two or more distinct macrofaunal assemblages dominated by either freshwater or marine taxa depending on freshwater inflow. In Texan estuaries, Palmer *et al*. (2011) found an oligo-mesohaline Chironomidae dominated assemblage and a polyhaline group with polychaetes and other typically marine taxa, whereas three assemblages with low (freshwater), medium (brackishwater) and high (marine) diversity were identified by Montagna and Kalke (1992). In southern Portugal, lower abundance of Oligochaetes, Spionidae and *Capitella capitata* is associated with a decrease in groundwater discharge into the estuary, especially at the end of the dry season (Silva *et al*., 2012). In years of high freshwater input in the Santo André lagoon, Southern Portugal, the macrofaunal assemblage is dominated by many insect species (Correia *et al*., 2012). Macrofaunal compositon at higher salinity sites (Canela and Ajurutuea) was different from that at the lower salinity site (Caeté Bay) and was associated with salinity and % silt. In the Caeté Bay, greatly increased silt values were found when freshwater runoff was high. Variation in macrofaunal structure among sites on different spatial scales may be attributed to local hydrodynamics, and its effect on sediment type, dissolved oxygen and other factors, and/or proximity to a freshwater/marine influence (Montagna and Kalke, 1992; Bachelet *et al*., 1996; Rozas *et al*., 2005; Rutger and Wing, 2006; Palmer *et al*., 2011; Correia *et al*., 2012; Silva *et al*., 2012). Along the Texas coast, Palmer *et al*. (2011) found that macrofaunal densities and species diversity increased with increasing salinity and that biomass and abundance were lower in river-dominated estuaries and high in lagoons, except for hypersaline ones.

Around the Ajuruteua Peninsula, no spatial or temporal differences in density occurred despite significant changes in the composition and numbers of taxa. High density in both the freshwater and marine end of an estuary may reflect different levels of connectivity with the coast (Montagna and Kalke, 1992) who found that macrofaunal density may increase with inflow as a result of the dominance of certain organisms that tolerate low salinity (such as *Nepthys fluviatilis* in the Caeté Bay in our study) making for a less diverse assemblage but diversity increases with greater marine influence, which fully agrees with our results. Organisms also need to be tolerant of silt and turbidity, which are often associated with freshwater runoff.

Sometimes no effect of freshwater inflow on benthic density and biomass is detected (McCarthy *et al*., 2000; Ardisson and Bourget, 1997). Controlled freshwater inflows reduced salinity, increased submerged aquatic vegetation and affected macrofaunal distributions (brown shrimp moved downstream, for example) and some species densities but not composition (Rozas *et al*., 2005).

Changes in macrofaunal structure with freshwater inflow are due to physical dislocation and/or level of physiological tolerance. Alongi (1989) concluded that, in general, tropical benthic habitats are subject to wide environmental variation and thus wide variation in benthic species richness and densities and smaller-bodied opportunistic species are expected to predominate that are subject to high levels of predation (see Aller and Stupakoff, 1996; Ieno and Batisda, 1998). Freshwater discharge causes high suspended solids loads, low salinity, physical stress, and may be associated with winds and waves that cause turbulence and rework sediments, especially in shallower waters (Aller and Stupakoff, 1996; Valiela *et al*., 2012). Along the Amazon coast, rapid erosion/deposition of the sediment may result in 2-4 fold decreases/increases in the mud fraction causing mortality by removal or burial of macrofauna and even zooplankton (Aller and Stupakoff, 1996). During low discharge, sediments are stable and animals are readily transported onshore by the North Brazil Current allowing recolonisation (Aller and Stupakoff, 1996).

Rates of export of fish and decapod crustacean larvae are higher with freshwater inflow and the duration of the planktonic stage, feeding and survival, as well as larval behaviour and movement may determine whether they are successful in settling or are lost to the outer coastal area (Tolley *et al*., 2012).

Physiological tolerance is also important. Richmond and Woodin (1999) found a direct effect of reduced salinity on oxygen consumption in larval invertebrates although tolerance varied with species. A negative effect of eutrophication on most of the macrofauna would be expected given that freshwater inflows are often high in nutrients and organic matter (Santos *et al*., 2008; Palmer *et al*., 2011) and mangroves may have even more importance (Dittmar and Lara, 2001).

Species may tolerate low salinity but behavioural adaptations such as burial in sediment with high salinity interstitial water may be critical for survival in habitats where large and rapid decreases in salinity occur as a result of sudden rainfall and increased freshwater inflow (Miguel Pardo *et al*., 2011). Others are unable to cope with changes in salinity and simply die out. Polyp prevalence of a hydrozoan oyster symbiont in Estero Bay, Florida, where sudden changes in salinity occur, was positively related to

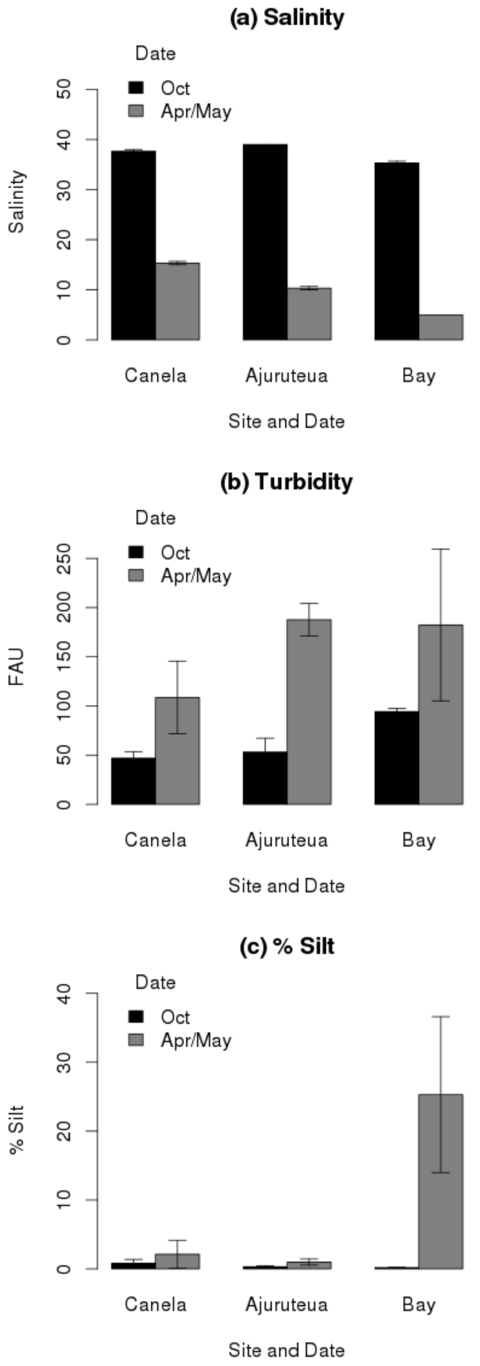

Figure 4. Salinity, turbidity and percentage silt at sites sampled for subtidal macrofauna at Canela island, Ajuruteua beach and the Caeté Bay in October 2007 and April/May 2008.

salinity and disappear where freshwater input is high (Tolley *et al*., 2010).

Nutrient enrichment and hydrodynamics due to increasing seasonal rainfall and freshwater inflow may cause significant changes in biogeochemical cycles and dynamics of coastal food webs (Valiela *et al*., 2012). Long term data from Louisiana rivers entering the Gulf of Mexico show increases in inflow rates, related with increased precipitation but also greater urbanization and the greater inflow may increase nutrient input to the already eutrophic Gulf waters (Xu and Wu, 2006). Similarly, long term declines in salinity, related to river inflow, which was variable over the years and associated with climatic phenomena, were associated with declines in benthic macrofaunal abundance, biomass and diversity in Lavaca Bay and Matagorda Bay, Texas (Pollack *et al*., 2011).

Although the subtidal macrofaunal around the Ajuruteua Peninsula mangrove coast is mainly composed of polychaetes and crustaceans, there is remarkable spatial and temporal variation in the composition of the fauna. Seasonal freshwater runoff and associated decreases in salinity and increases in silt (and turbidity) are associated with a reduction in the number of individuals and taxa with the disappearance of typically marine taxons at Ajuruteua beach and Canela island. Numbers of individuals and taxa are much lower in the Caeté Bay, where there is a constant influence of freshwater runoff. However, when discharge is highest in April/May, the Bay macrofauna becomes strongly dominated by the polychaete *Nepthys fluviatilis*, which appears to tolerate well low salinity, and silty and turbid conditions.

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