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How mollusk assemblages respond to different urbanization levels: characterization of the malacofauna in subtropical Brazilian mangroves

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Abstract
This study aimed to describe the composition of mollusk assemblages in subtropical Brazilian mangroves with different levels of urbanization in their watersheds. Mangroves are important ecosystems, which are vanishing worldwide due to human impacts. The knowledge about the consequences of human pressure on the fauna of these ecosystems is still incipient. In addition, although Brazil is the country with the second largest mangrove area, there is a lack of studies on the mangrove fauna in this country. Mollusks are the second most abundant group of mangrove invertebrates and can be useful indicators of mangrove health. For this reason, mollusk species were assessed in two mangroves surrounded by a dense human population and in two mangroves away from urban centers. A total of 3820 individuals, representing 15 species, were sampled. The results revealed that the mollusk abundance, diversity estimators, and sediment organic matter content were not good indicators of the effects of urbanization on the mangroves studied. However, the species composition of mollusk assemblages differed according to the urbanization level. This survey of the mangrove malacofauna represents another step toward the effort to investigate and conserve the fauna of these important estuarine environments.

Keywords Diversity · Macrofauna · Estuary · Gastropod · Bivalve

Introduction

Mangrove forests are important ecosystems located at the interface between the land and the sea in tropical and subtropical coastlines (Spalding et al. 1997). These unique environments offer shoreline protection, by minimizing the effects of erosion, strong winds, tidal bores, and even tsunamis (Alongi 2008). Mangroves play a key role in carbon and nutrient cycling, showing the highest level of primary production and sediment carbon storage and export to coastal environments (Alongi 2014). Mangroves are economically important because numerous coastal human communities around the world depend on the extraction of food and wood from these forests, and because they are also explored for fishing and aquaculture. In addition, mangroves are important nursery areas, supporting high densities of juvenile fishes and invertebrates, which support the fishery supply (Lee et al. 2014). Moreover, extracts from different mangrove plants have been described to possess diverse medicinal properties, for example, inhibitors of HIV-1 reverse transcriptase, and anticancer and antiviral chemicals and substances with analgesic and anti-inflammatory activity (Bandaranayake 1998; Walters et al. 2008). Despite their importance, mangroves are vanishing worldwide by a rate of between 0.16 and 0.39% per year. However, critical region, as in Southeast Asia, presents a deforestation rate between 3.58 and 8.08% per year (Hamilton

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and Casey 2016). The exploration of their natural resources has been historically intense and the protection of this ecosystem is clearly insufficient (FAO 2007; Polidoro et al. 2010). Mangroves situated in areas under high anthropogenic influence are negatively affected (Pons and Fiselier 1991). Urban development, aquaculture, continuous exposure to pollutants, and hydrological alterations contribute to the degradation of mangroves and lead to a decrease in mangrove biodiversity (Valiela et al. 2001; Alongi 2002, 2015). The characterization and monitoring of the biodiversity in mangroves could be a valuable tool to measure natural and human-induced impacts (Duke et al. 2007; Gilman et al. 2008; Friess et al. 2012).

Mangroves host an exclusive set of associated fauna, which is able to tolerate salt and tidal disturbance (Nagelkerken et al. 2008). The fauna is composed of a variety of species, and mollusks are the second most abundant group of invertebrates, after arthropods (Cannicci et al. 2009; Kabir et al. 2014). Mollusks have great ecological importance to mangroves, since they affect the recycling processes of organic matter due to their burrowing, detritivorous, and filtering habits (Printrakoon et al. 2008). Furthermore, the abundance and diversity of mollusks can be directly impacted by changes in the structure and quality of mangroves, since many species are exclusive to these forests (Bodin et al. 2011; Salmo et al. 2017). Studies reporting the human impact consequences on the mangrove malacofauna have shown lower diversity and richness of mollusks and dominance of few species at highly disturbed forest sites compared to moderately disturbed sites (Amin et al. 2009; Cannicci et al. 2009; Kabir et al. 2014). In tropical mangroves from Colombia and Thailand, the abundance and diversity of mollusks are high, and these organisms show different responses to pollution and habitat loss due to forest devastation (Vilardy and Polanía 2002; Printrakoon et al. 2008; Blanco and Castaño 2012). Macintosh et al. (2002) proposed that members of the family Potamididae (gastropods) could be used as indicators of ecological change in mangroves, and Cannicci et al. (2009) indicated that domestic wastewater has detectable effects on mollusks from East African mangroves. Therefore, the attributes and sensitivity of the malacofauna suggest their usefulness as bioindicators of the health of mangroves. However, studies on the mangrove mollusk fauna are still scarce, and more knowledge needs to be generated to achieve a satisfactory level (Maia and Coutinho 2013). In addition, although Brazil is the country with the second largest mangrove area (11,940 km^2, Ferreira and Lacerda 2016), there are few studies on these invertebrates in Brazilian mangroves (Netto and Lana 1997; Koch and Wolff 2002; Boehs et al. 2004; Beasley et al. 2005; Barroso and Matthews-Cascon 2009; Melo et al. 2013). Some areas of the Brazilian coast are under significant urban pressures due to their high population densities, industries, petrochemical poles, and important ports (Cordeiro and Costa 2010; Pinheiro et al. 2012; Ferreira and Lacerda 2016). However, information about the fauna response to urban pressures is not sufficient (Bernardino et al. 2016). Thus, our present study aimed to characterize the diversity and composition of mollusk assemblages in subtropical mangroves with different levels of urbanization in their watersheds, in an attempt to identify patterns of the malacofauna that can be used as bioindicators of mangrove health. We expected to record lower abundance and diversity of mollusks and higher sediment organic matter content in mangroves with high level of urbanization in their watersheds than in mangroves with low level of urbanization. To accomplish this, we investigated and compared the mollusk assemblage of four mangroves: two located in areas of high population density and two located in areas of low population density. The results revealed that the mollusk abundance, diversity estimators, and sediment organic matter content were not good indicators of the effects of urbanization, since most of them were similar between the mangroves studied. However, we found that the species composition of mollusk assemblages differed according to the urbanization level.

Materials and methods

Characterization of the study area

The study was carried out on the coast of the state of São Paulo, Brazil, where mangroves represent 52% of the shore line, but only 8.8% of which are protected (Afonso 2006; Cantagallo et al. 2008). The south half of the São Paulo coast shows extensive mangroves, but also has a dense human population, with approximately two million inhabitants (IBGE 2010; Colpo et al. 2011). In this region of São Paulo, we studied the mollusk assemblages from four mangrove areas: Rio Branco mangrove, in São Vicente (23° 56′ 22″ S 46° 25′ 25″ W), Portinho mangrove, in Praia Grande (23° 59′ 20″ S 46° 24′ 25″ W), Guaratuba mangrove, in Bertioga (23° 45′ 09″ S 45° 53′ 33″ W), and Una mangrove, in the Juréia-Itatins Ecological Station, Peruíbe (24° 25′ 33″ S 47° 05′ 05″ W). The areas were selected following the criterion of urbanization levels in their watersheds. According to the last demographic census (IBGE 2010), São Vicente has a population density of 2232 inhabitants km^{-2}, Praia Grande of 1776 inhabitants km^{-2}, and Bertioga and Peruíbe of 490 and 192 inhabitants km^{-2}, respectively. In addition, Portinho and Rio Branco mangroves are surrounded by one of the largest industrial centers in Brazil and are impacted by several pollution sources (Cordeiro and Costa 2010; Pinheiro et al. 2012, 2013). These data, together with satellite images (Fig. 1), suggest that Rio Branco and Portinho mangroves are more affected by urbanization than Guaratuba and Una mangroves, which can be regarded as the most protected and less affected by human activities.

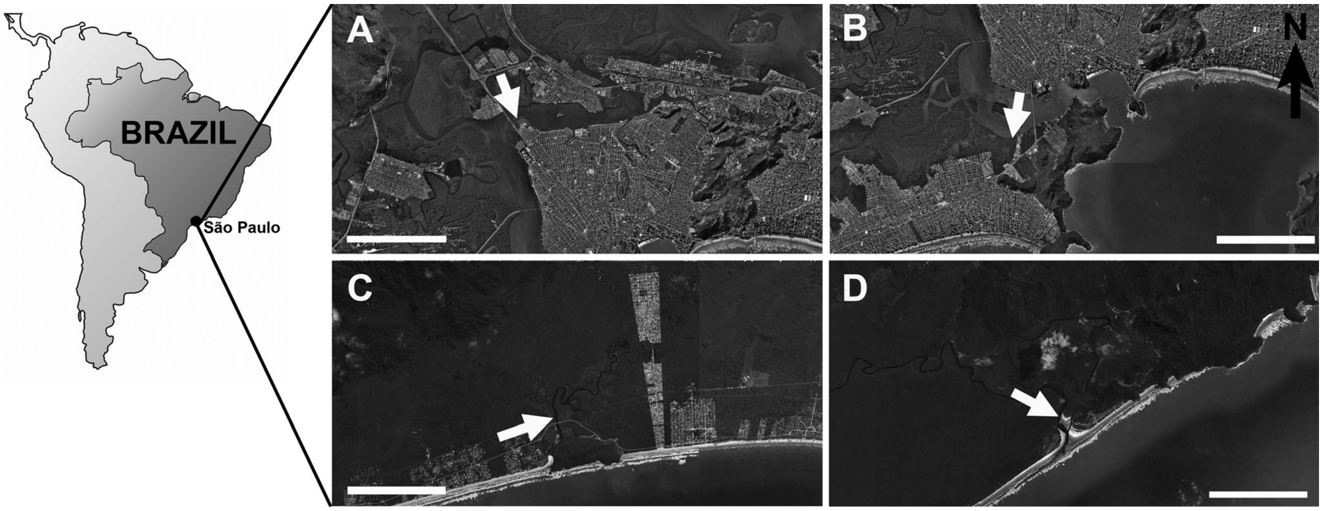

Fig. 1 Satellite images of the four mangroves studied in the coast of São Paulo, Brazil. The images show each mangroves surroundings. In (A) Rio Branco mangrove (23° 56′ 22″ S 46° 25′ 25″ W) and (B) Portinho mangrove (23° 59′ 20″ S 46° 24′ 25″ W); it is possible to observe the large population density located very close to the mangrove. In (C) Guaratuba mangrove (23° 45′ 09″ S 45° 53′ 33″ W) and (D) Una mangrove (24° 25′ 33″ S 47° 05′ 05″ W); reference areas, the mangrove are more isolated, affected less by population density. White arrows point the sampling location (modified from www.google.com). Scale bar = 3 km

Sampling

Samplings were performed during the low tide on four occasions for each mangrove, between July 2011 and April 2012. During the samplings, the mean air temperature in the region was 21.8 ± 3.8 °C, and the mean air relative humidity was 85.8 ± 11.6% (database available at the Center for Weather Forecasting and Climate Studies - CPTEC, São Paulo, Brazil). In average, during the low tide, the water salinity and temperature of the four estuaries were 22.0 ± 1.7‰ and 23 ± 0.6 °C, respectively, and no statistical differences were recorded among the mangroves.

Four samples of mollusk assemblages of each mangrove were collected at different times. The sampling unit was an area of 125 m^2, in each mangrove forest. In total, we covered an area of 2000 m^2 sampling of mollusks (500 m^2 in each mangrove). To access the species of mollusks that live associated with the sediment (infauna), 15 random substrate samples were collected in each sampling unit with a corer-type sampler of 10 cm in diameter and 5 cm deep. Thereafter, the visible mollusks that generally live associated with tree roots and trunks (epifauna) were collected manually or with a spatula, according to the method proposed by Printrakoon et al. (2008), with a capture effort of 100 min for each sampling unit, performed by a single collector. All mollusks collected were properly labeled, stored, and carried to the laboratory. Also, for each sampling unit, about 15 kg of fragments of tree trunk was separated, stored in labeled plastic bags, and taken to the laboratory to search for the presence of Teredinidae. The sediment was washed thoroughly in a 350-μm mesh sieve and then examined under a stereomicroscope. The mollusks found were removed and fixed in 70% ethanol. The teredinids found in the tree trunk fragments were carefully removed. All mollusks were identified to species level or to the lowest taxonomic level possible (see Supplementary Material).

The sediments of the mangroves studied were also characterized. In each sampling occasion, composite soil samples were collected from each mangrove. These samples were dried at 60 °C for 48 h or to constant weight. After that, they were sieved according to Wentworth's scale (1922) to determine the proportion of each granulometric fraction and the central tendency (Phi φ) of the grain size in the samples (Surguio 1973). The organic matter content (%) of the sediments was determined by the ash-free dry weight.

Analyses

The diversity of mollusk assemblages was characterized by the number of species (S), Shannon-Weiner Index (H′), and evenness (J). These diversity estimators and the abundance of mollusks were compared between mangroves by one-way ANOVA. The number of species and number of singletons found in each mangrove were evaluated. However, it is known that most surveys have species that are not recorded (Chao et al. 2000). For this reason, the completeness of the species inventory in the mangroves studied were analyzed and compared, considering the performance of three richness estimators (Chao 1, Jackknife 1, and Bootstrap). The richness estimators were calculated using Primer-E.6, performing 999 permutations in each analysis.

Rank-abundance curves are useful representations of the structure of assemblages. The number of points in the curves reveals the species richness, the order of these species in the curve indicates their relative abundance, the tail of the curve represents the rare species, and the slope of the curve shows the evenness (Negrete et al. 2014). Here, rank-abundance curves were made for the four mollusk assemblages studied. The relative abundance of each species, on a logarithmic scale (log_{10}), was plotted following the rank order of the species, from the most to the least abundant.

The mollusk assemblage composition between mangroves was compared using a non-metric multidimensional scaling (nMDS), based on the Bray-Curtis similarity coefficient (Gotelli and Ellison 2004). The analysis of similarities (ANOSIM) was used to complement the nMDS analysis. We used the ANOSIM R values as a valid approach to detect similarity patterns among mangroves (Clarke and Warwick 2001). Therefore, high R values (close to 1) indicate differences between mollusk assemblages, while small R values suggest little or no dissimilarity. In addition, the similarity percentage (SIMPER) analysis was carried out to identify the most important mollusk species to generate the patterns of dissimilarity observed between mangroves. We also used the Bray-Curtis coefficient as a measure of similarity in the SIMPER analysis (Clarke and Warwick 2001).

For sediment characterization, we compared the Phi (φ) and the organic matter content between the mangroves by one-way ANOVA and Kruskal-Wallis test, respectively, due to the homogeneity of variances (tested by Cochran's test). For all univariate analyses, the significance level used was 5% ($\alpha = 0.05$).

Results

A total of 3820 individuals, representing 15 mollusk species distributed in 13 genera of 11 families, were sampled. Although gastropods (92.4%) were more abundant than bivalves (7.6%), species richness of Bivalvia was higher (60%) than that of gastropods (40%) (Table 1). Most of the species (53.3%) were recorded associated with the sediment (infauna), 26.7% of mollusk species were found only on tree roots and trunks (epifauna), 13.3% were found in both habitats, and 6.7% were found inside tree trunks (Table 1). The diagnostic characterization, geographic distribution, and life habits of each species found are available as Supplementary Material.

The abundance of mollusks, Shannon-Weiner Index (H′), and evenness (J) were similar between the four mangroves (Table 1). The species richness (S) was different between mangroves. Portinho mangrove (high level of urbanization) and Una mangrove (low level of urbanization) showed the highest species richness. Meanwhile, Rio Branco mangrove (high level of urbanization) and Guaratuba mangrove (low level of urbanization) showed the lowest species richness (Table 1).

In Rio Branco, we found seven species, 42.8% of which were singletons, in Portinho, we found 11 species, 36.4% of which were singletons, in Guaratuba, we found eight species, 12.5% of which were singletons, and in Una, we found 11 species, 27.3% of which were singletons (Table 2). Among the richness estimators, Bootstrap was the most conservative; it estimated the lowest numbers of species, similar to that recorded at Rio Branco, Portinho, and Guaratuba (Table 2). Considering the mean of the three richness estimators, the inventory completeness of mollusk assemblages was about 10% lower in mangroves with high level of urbanization (Table 2).

The genus *Heleobia* was dominant in all mangroves, representing 80.7% of the mollusks sampled. We recorded two species of this genus: *Heleobia australis* and *Heleobia charruana*, which were found essentially in the sediments. The species *Littoraria angulifera*, *Crassostrea mangle*, *Neoteredo reynei*, and *Sayella hemphilli* showed intermediate abundances, and together represented 17.9% of the mollusks sampled. The remaining species were rare (Table 1, Fig. 2).

The nMDS analysis clustered the mollusk assemblages into two groups, with 53% similarity (Bray-Curtis coefficient). The first group was composed of Rio Branco and Portinho mangroves, while the second group was composed of Guaratuba and Una mangroves (Fig. 3). The ANOSIM R values confirmed the results of the nMDS analysis. The values of R (ANOSIM) and the average dissimilarities (SIMPER) were lower for comparisons within mangroves with the same level of urbanization than between the mangroves of different urbanization levels (Table 3). The SIPMER analysis indicated that *H. charruana* was the principal species that contributed to dissimilarity (more than 25%) between mangroves with different levels of urbanization (Table 3). In urbanized mangroves, *H. charruana* showed low contribution to dissimilarity (6.6%) since it was abundant in Rio Branco (46.8% of total individuals) and Portinho (67.1% of total individuals). In mangroves away from the urban centers, *H. charruana* also showed low contribution to dissimilarity (4.3%); however, this pattern was due to its low abundance in Guaratuba (0.9% of total individuals) and absence in Una. In these areas, *H. australis* was the dominant species (74.5% in Guaratuba, and 72.2% in Una) (Table 3, Fig. 3).

The spectrum of sediment particles ranged from very coarse sand to silt-clay, but fine sand, very fine sand, and silt-clay fractions predominated in all mangroves (Fig. 4). The values of central tendency of the grains (Phi φ) did not vary among mangroves (ANOVA: $F = 0.637$, $p = 0.608$). The organic matter content of sediments in the mangroves studied was similar (Kruskal-Wallis: $H = 7.147$, $p = 0.067$) (Fig. 4).

Discussion

Contrary to our expectations, the diversity estimators, abundance of mollusks, and organic matter content of sediments did not represent significant indicators of the level of urbanization in the mangroves studied. However, the species

Table 1 List of mollusks found in each mangrove accompanied by their abundance and habitat. The habitat is classified as infauna (I, found associated with the sediment), epifauna (E, found in tree roots and trunks) and wood (found inside tree trunks). The comparison of the abundance and diversity estimators of mollusk assemblages between the mangroves is shown at the foot of the table. Values with different letters indicate statistically significant differences ($p < 0.05$)

Species	Rio Branco	Portinho	Guaratuba	Una	Habitat
Gastropoda					
Subclass Caenogastropoda					
Family Littorinidae					
Littoraria angulifera	1	85	160	70	E
Family Cochliopidae					
Heleobia australis	624	23	557	835	E/I
Heleobia charruana	574	463	7	0	I
Family Thiaridae					
Melanoides tuberculate	0	0	1	0	E
Subclass Heterobranchia					
Family Pyramidellidae					
Sayella hemphilli	18	46	13	29	I
Family Ellobiidae					
Melampus coffea	1	0	3	20	E
Bivalvia					
Order Pteriidae					
Family Isognomonidae					
Isognomon sp.	0	0	0	1	I
Order Ostreoidea					
Family Ostreidae					
Crassostrea mangle	1	55	4	97	E
Order Veneroidea					
Family Tellinidae					
Tellinidae sp1.	0	2	0	0	I
Tellinidae sp2.	0	1	0	1	I
Family Cyrenoididae					
Cyrenoida sp.	6	1	0	2	I
Order Myidae					
Family Myidae					
Sphenia sp.	0	1	0	1	I
Order Mytiloidea					
Family Mytilidae					
Mytella charruana	0	0	3	9	E/I
Dacrydium vitreum	0	1	0	0	I
Family Teredinidae					
Neoteredo reynei	0	12	0	92	Wood
Abundance (mean ± standard error)	306 ± 129 [a]	172 ± 54 [a]	187 ± 73 [a]	289 ± 107 [a]	
	ANOVA results:		F = 0.520	p = 0.677	
Species richness (S)	7 [b]	11 [a]	8 [b]	11 [a]	
	ANOVA results:		F = 4.301	p = 0.028	
Shannon-Weiner index of diversity (H′)	0.804 [a]	1.146 [a]	0.744 [a]	1.044 [a]	
	ANOVA results:		F = 1.067	p = 0.400	
Evenness (J)	0.319 [a]	0.286 [a]	0.263 [a]	0.258 [a]	
	ANOVA results:		F = 1.269	p = 0.329	

Table 2 Mollusk richness and singletons registered in each mangrove. Number of species expected and percentages of inventory completeness, according to different richness estimators

	Rio Branco		Portinho		Guaratuba		Una	
Number of species	7		11		8		11	
Number of Singletons	3		4		1		3	
Chao 1	11	64%	19	58%	10	80%	11	100%
Jackknife 1	10	78%	14	79%	11	73%	14	79%
Bootstrap	8	88%	12	92%	9	89%	12	92%
Average inventory completeness		76.6%		76.3%		80.6%		90.3%

composition of mollusk assemblages differed according to the urbanization level. Mangroves with higher urbanization level (Rio Branco and Portinho) shared similar mollusk assemblages but differed from mangroves where urbanization is less intense (Guaratuba and Una). The gastropod *Heleobia charruana* was the principal responsible for such differences. Rio Branco and Portinho shared high abundance of *H. charruana*, while in Guaratuba and Una, this species was rare.

Despite the extensive areas of Brazilian mangroves, studies about mangrove mollusks in Brazil are scarce (Spalding et al. 1997). There are a few works reporting mollusk species of tropical Brazilian mangroves (Koch and Wolff 2002; Beasley et al. 2005; Barroso and Matthews-Cascon 2009; Melo et al. 2013; Rodrigues et al. 2016). Our study is the first to assess the malacofauna in subtropical Brazilian mangroves, in which 15 species were recorded, with a completeness of inventory of about 81%, suggesting a suitable sampling effort. Most Brazilian mollusk mangrove surveys have also recorded low species richness (from 4 to 23 species of mollusks). In agreement with that found in tropical Brazilian mangroves (Koch and Wolff 2002; Beasley et al. 2005; Barroso and Matthews-Cascon 2009; Melo et al. 2013; Rodrigues et al. 2016), here we recorded the family Tellinidae, the genus *Mytella*, and the species *Crassostrea*, *Neoteredo reynei*, *Littoraria angulifera*, *Melanoides tuberculate*, and *Melampus coffeus*. We recorded important dominance of the genus *Heleobia* in the four mangroves studied, whereas Barroso and Matthews-Cascon (2009) and Rodrigues et al. (2016) reported low abundance of this genus. Probably, Koch and Wolff (2002), Beasley et al. (2005), and Melo et al. (2013) did not record these gastropods because their study areas are in the north edge of *Heleobia* geographical range (Cazzaniga 2011), and the population size tends to decrease toward the distribution boundaries (Gaston 2003; Hargreaves et al. 2014). There are other important works on mollusks in Brazilian estuarine systems, but they surveyed the malacofauna from other intertidal environments such as saltmarshes, lagoon estuarine systems, and muddy-sandy non-vegetated flats (Netto and Lana 1997; Boehs et al. 2004; Beasley et al. 2005; Fonseca and Netto 2006; Barroso et al. 2013). Furthermore, in general, these studies show a different set of species compared to mangroves, with only few species present in all environments (common taxa: Mellitidae,

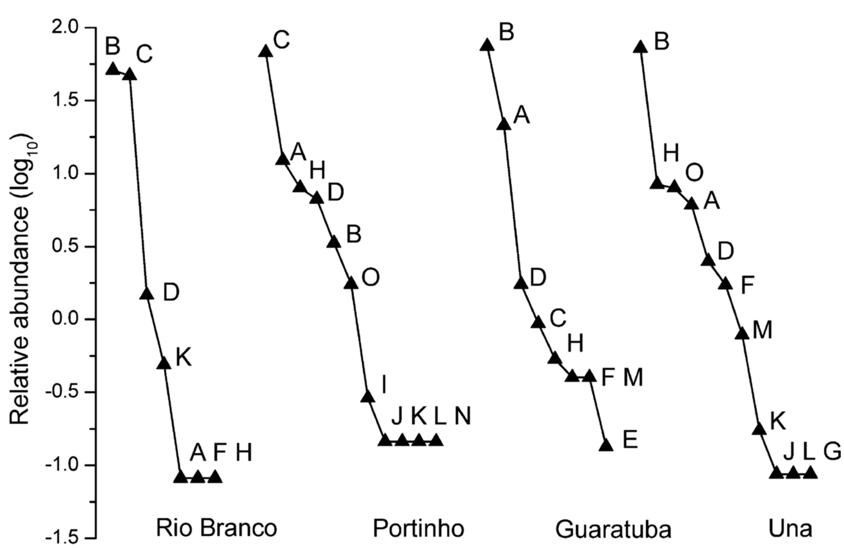

Fig. 2 Rank-abundance curves of mollusk assemblages in each mangrove. *Heleobia charruana* (C) was the most abundant species in Rio Branco and Portinho mangroves, while *Heleobia australis* (B) predominated in Guaratuba and Una mangroves. A, *Littoraria angulifera*; B, *Heleobia australis*; C, *Heleobia charruana*; D, *Melanoides tuberculate*; E, *Sayella hemphilli*; F, *Melampus coffea*; G, *Isognomon* sp.; H, *Crassostrea mangle*; I, *Tellinidae* sp1; J, *Tellinidae* sp2; K, *Cyrenoida* sp.; L, *Sphenia* sp.; M, *Mytella charruana*, N, *Dacrydium vitreum*; O, *Neoteredo reynei*

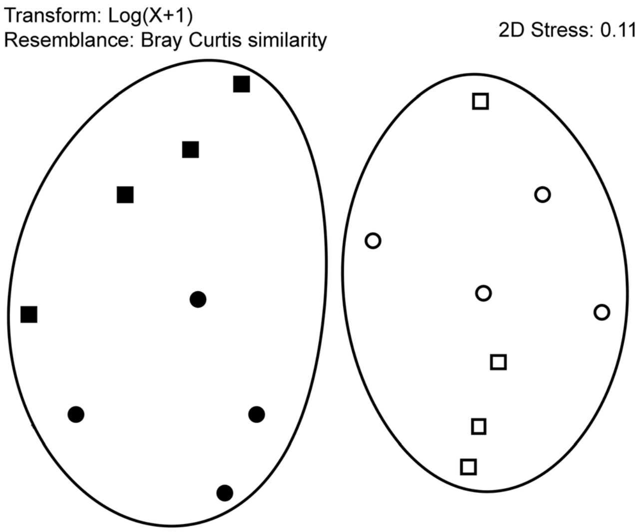

Fig. 3 Non-metric multidimensional scaling (nMDS) based on the mollusk assemblage composition of each mangrove. Two groups were determined with 53% similarity (Bray-Curtis coefficient). The first group included Rio Branco (closed squares) and Portinho mangroves (closed circles), while the second group included Guaratuba (open circles) and Una River mangroves (open squares)

Heleobia, *Crassostrea*, *Littoraria angulifera*, *Mytella charruana*, *Neoteredo reynei*, *Melampus coffeus*). Therefore, it is important to improve the recognized level of mollusk diversity and the distribution patterns in different estuarine environments (Beasley et al. 2005; Bernardino et al. 2016).

The sedimentological features of the mangroves studied were similar. These results were unexpected, because, despite the similar granulometry, we expected to find greater organic matter contents in sediments of highly urbanized mangroves, considering that urbanization processes usually can cause an increase in organic matter in these environments, leading to a high concentration of hydrocarbons (Gao et al. 2008; Muniz et al. 2015). Anthropogenic organic matter (e.g., contamination by fecal matter, hydrocarbons, and fatty acids) derived from land are carried by rivers and are buried in marine coastal areas, leading to increases in this parameter (Grilo et al. 2013). Sedimentological features can be reflected on the distribution of mollusks in estuarine ecosystems (Netto and Gallucci 2003; Kabir et al. 2014). However, in our study, the differences recorded in the mollusk species compositions between the mangroves with different levels of urbanization cannot be explained by the sediment parameters evaluated.

In general, estuaries with different levels of urbanization in their watersheds show dissimilarities in macrofauna estimators, and usually, the abundance and diversity of the species decrease as the urbanization and pollution increase (Pagliosa and Barbosa 2006; Venturini et al. 2012; Souza et al. 2013; Pérez-Reyes et al. 2016). The urbanization levels stablished in this study showed no significant differences in these estimators between the mangroves studied. However, only the presence of human population not necessarily implies a disturbance per se; it is important to evaluate the real effect of urbanization to understand their influence on the fauna composition. The specific impacts of urbanization on species richness and abundance may vary depending on low, moderate, or high levels of human disturbance; geographic location of the city; presence of pollutants in the sediment; dynamics of nutrients; and spatial scale modifications (McKinney 2008; Cavalcante et al. 2009; Lee et al. 2014). Small-scale modifications on the physical structure of mangroves can also affect the diversity and abundance of macrobenthic organisms in these habitats (Skilleter and Warren 2000; Kabir et al. 2014). The complex architectures and hydrodynamics of estuaries and mangroves provide spatio-temporal variations in the availability of food, temperature, organic matter, water and sediment conditions, tidal inundation, larval recruitment, predation, competition, and mortality, generating several microhabitats, and affecting the distribution, density, abundance, and richness of macrofauna (Ashton et al. 2003; Reiss and Kröncke 2005). Therefore, the difference found in the richness

Table 3 *R* values of ANOSIM analysis and average dissimilarity of SIMPER analysis, showing that mollusk assemblages from mangroves with the same level of urbanization are more similar than those from mangroves with different levels of urbanization. Contribution (%) of *Heleobia charruana* to the dissimilarity among assemblages of the mangroves studied. Bray-Curtis coefficient was used as a measure of similarity

	ANOSIM	SIMPER	
Pairwise comparisons	*R* value	Average dissimilarity (%)	Contribution (%) *H. charruana*
Rio Branco vs. Portinho	0.583	45.9	6.6
Rio Branco vs. Guaratuba	0.906	62.4	32.5
Portinho vs. Guaratuba	0.958	57.1	28.6
Rio Branco vs. Una	0.969	66.1	26.5
Portinho vs. Una	0.76	57.8	25.2
Guaratuba vs. Una	0.344	39.8	4.3

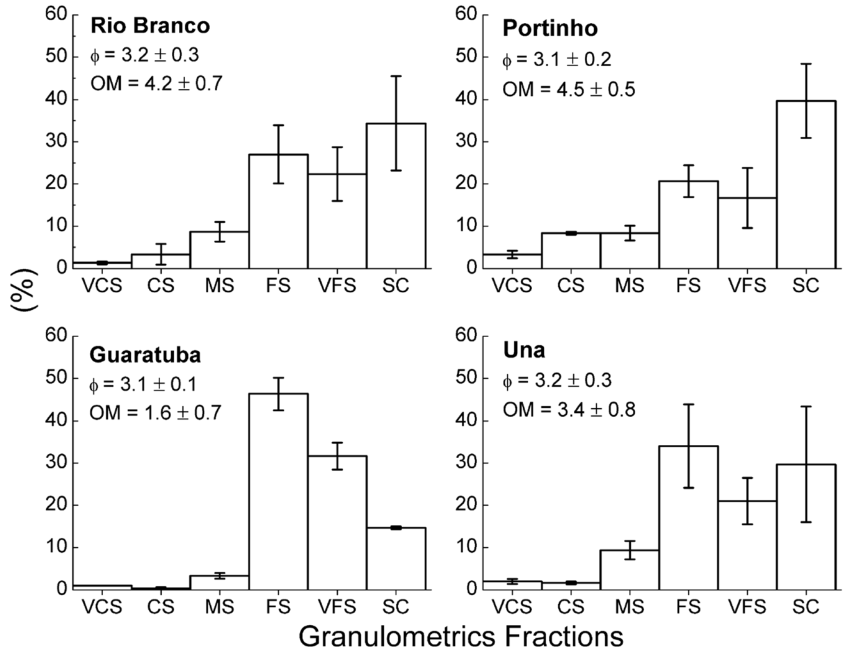

Fig. 4 Sediment characterization of the mangroves studied. Mean (± standard error) of percentage of each particle size fraction, of central tendency of the grain Phi (φ), and of percentage of organic matter content (OM). VCS very coarse sand, CS coarse sand, MS median sand, FS fine sand, VFS very fine sand, SC silt-clay

of mollusks between the mangroves can be due to the spatio-temporal variations affecting each environment rather than to the consequences of the levels of urbanization evaluated.

Although the abundance of mollusks and diversity estimators were not useful tools to indicate the effects of large human population density, we detected differences in species composition between mangroves with different urbanization levels. *Heleobia charruana* was the species that most contributed to this dissimilarity, since it was abundant in the mangroves with high urbanization level (Rio Branco and Portinho) and rare in the less urbanized mangroves (Guaratuba and Una). The genus *Heleobia* (represented here by *H. charruana* and *H. australis*) represented 80% of our mollusk survey. *Heleobia* is widely distributed in estuaries of the eastern and western margins of South America (Aguirre and Urrutia 2002; Figueiredo-Barros et al. 2006) and has been used as a paleobiological and paleoenvironmental indicator (Aguirre et al. 2002; de Francesco and Isla 2004). Several studies have demonstrated that the hydrobiid gastropod population can attain high density and is the main constituent of the benthic community (Netto and Lana 1997; Muniz and Venturini 2005; Magnone et al. 2015). Alvarez et al. (2013) concluded that *Heleobia* snails modify the benthic assemblage by changing the relative species composition and abundances. The high densities and high percentage of biomass, in relation to other groups of benthic macroinvertebrates, as well as their habit of consuming organic detritus, suggest that this hydrobiid gastropod plays an important role in the nutrient cycling of estuarine ecosystems (Wells 1984; Figueiredo-Barros et al. 2006; Magnone et al. 2015).

Furthermore, Egres et al. (2012) found an increase in the density of *Heleobia* after an oil spill, suggesting that this species may be a resilient species and able to quickly recolonize oil-disturbed sites. Muniz and Venturini (2005) and Muniz et al. (2011) noted that *Heleobia* is resistant to organic-enriched and oxygen-depleted environments. In addition, it is known that some *Heleobia* species show high dominance in impacted environments and are used as sentinels to evaluate the presence of mutagenic agents in estuarine environments (Cannicci et al. 2009; Villar et al. 2011). Based on these studies, we can assume that *Heleobia* species can resist several anthropological impacts, and as opportunistic species, they occupy niches that are available after other species were not able to resist a particular impact. This can explain the high dominance of *H. charruana* in the most urbanized mangroves studied in the present study. Nevertheless, Souza et al. (2013) studied flats subjected to different levels of contamination from urban effluents and found that *Heleobia* was also abundant in the non-contaminated area. Therefore, the high abundance of *H. australis* in the mangroves with low urbanization level was not an unexpected result. However, the reason *H. charruana* was abundant in the most urbanized mangroves and rare in the least urbanized ones is an inquisitive question.

Since they are transitional areas between marine and freshwater systems, estuaries are naturally characterized by extreme environmental changes and constantly affected by marked differences in salinity, hydrodynamic energy, and tidal currents (Dauvin 2007). Although our nested approach between mangroves showed some consequences of human population density, the influence of those environmental factors seems to be the main source of variability that affected the malacofauna in the mangroves studied. Studies that found faunal differences between urbanized and non-urbanized mangroves related their results to effects of sewage or other products accumulated in the sediment (Morrisey et al. 2003; Pagliosa and Barbosa 2006;

Venturini et al. 2012). Although no pollutant was evaluated in the present work, previous studies in the region have shown the presence of contaminants (heavy metal and plastic residues) in the sediments of urbanized mangroves (Cordeiro and Costa 2010; Pinheiro et al. 2013; Banci et al. 2017). Therefore, we can assume that these pollutants may be affecting the mollusk assemblages, especially the infauna. However, specific studies are required to explore the pollutants present in these sediments and their concentrations, as well as to evaluate the tolerance levels of different species, to point out biomarkers indicative of sublethal contamination and understand the real effect of urbanization.

The pattern of occurrence of *H. charruana* in the mangroves studied caught our attention. Nevertheless, there are few studies reporting the distribution patterns, ecological response, and tolerance ranges of different *Heleobia* species (Canepuccia et al. 2007; Etchegoin and Merlo 2011; Fiori and Carcedo 2011) that may help us to explain these results. Moreover, the taxonomy of this group is complex and current works are scarce (de Francesco and Isla 2004). We suggest that *Heleobia* species, which are abundant in the mangrove sediments, could be a target for future researches evaluating their potential as bioindicators of health for this important ecosystem. However, an advanced taxonomic review of this genus and new studies about environmental tolerances, using biomarkers, are necessary to define the use of *Heleobia* species as bioindicators. Furthermore, there is a lack of studies about mollusks in subtropical Brazilian mangroves, since previous studies have mainly characterized the tropical mangroves or other estuarine environments (Bernardino et al. 2016). The findings of the present study are important to fill the gap in the knowledge of the Brazilian malacofauna. The knowledge about the consequences of human pressure on ecological processes and assemblage patterns in mangroves is still incipient to generate potential predictors of risks, declines, and losses of this ecosystem (Daru et al. 2013). Our study represents another step toward the conservation and integrity of this important estuarine environment.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

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