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Effects of the Intertidal Burrowing Crab *Chasmagnathus granulatus* on Infaunal Zonation Patterns, Tidal Behavior, and Risk of Mortality

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ABSTRACT: Rhythmic movements in response to tidal cycles are characteristic of infaunal inhabitant of intertidal soft-bottoms, allowing them to remain in the area with best living conditions. The effect of bioturbators as modifier of local environmental conditions and thus of gradients in intertidal habitats, has not been investigated yet. The Atlantic estuarine intertidal areas are dominated by the burrowing crab *Chasmagnathus granulatus* that generates strong environmental heterogeneity by affecting the physical-chemical characteristics of the sediment. The comparison between intertidal areas with and without crab shows that sediments in the crab beds remain more humid, softer, and homogeneous across the intertidal and along the tidal cycle than areas without crabs. The densities of infauna were higher at high intertidal zones in crab beds than in similar areas without crabs. Infaunal organisms performed vertical movements into the sediment following the tidal cycle that were always of higher magnitude in habitats without crabs. Infaunal species tend to spend most of the time buried into the sediment in the crab bed. Migratory shorebirds use the Atlantic estuarine environments as stopover or wintering sites. They feed (mainly on polychaetes) in the low intertidal zones of both habitats (with and without crabs), but they also feed in the upper intertidal of the crab bed; polychaete per capita mortality rate is higher in the upper part of the crab bed. Environmental heterogeneity produced by crab disturbance has an effect on the infaunal behavior, risk of mortality, and the zonation pattern. This is another example of the ecosystem engineering ability of a burrowing intertidal species.

Introduction

Intertidal species are subject to a wide range of environmental variables that include changes in temperature, light, salinity, humidity, and action of waves (Chelazzi and Vanini 1988). Common responses of benthic organisms are rhythmic movements (vertical or horizontal), mainly related to the tidal cycle (Chelazzi and Vanini 1988). Vertical movements in response to tidal cycles are characteristic of this fauna, allowing them to remain in areas with good feeding conditions and minimizing the exposition to terrestrial and marine predators (Reise 1985; Brown and McLachlan 1990).

In soft-bottoms, where disturbance by wave action is almost absent, infauna build semi permanent burrows and moves up and down in the burrows as the tide rise and falls. These movements are common (e.g., harpacticoid copepods, Reise 1985; benthic foraminifers, Alve and Bernhard 1995) and they could be beneficial in terms of maximizing food acquisition and decreasing the infaunal mortality rate due to predation by shorebirds (Zwarts and Wanink 1991).

Shorebirds (Goss-Custard 1984), fish, and crabs (Quammen 1984) are important predators in soft-bottom intertidal areas. Their predation rate is often high, and depends on prey size, density (Goss-Custard 1984), substrate type, and climatic conditions (Pienkowski 1981). The consumption rate increases in soft-bottom areas (Goss-Custard 1984; Quammen 1984) and with low tidal levels (Pienkowski 1981; Goss-Custard 1984). Their effect on the infauna varies between habitats with different sediment characteristics that affect the bill penetrability (Botto et al. 1998) or due to infaunal vertical distribution (Zwarts and Wanink 1991). Burial offers protection to soft-bottom bivalve species such as *Donax* sp. and *Tellina* sp. (Brown and McLachlan 1990) and infaunal vertical movements has been shown to decrease the risk of mortality (Zwarts and Esselink 1989). Given that most sedimentary characteristics are altered by bioturbators (Botto and Iribarne 2000), it would be expected that their activities also modify the infaunal behavior and consequently their risk of mortality.

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The burrowing crab *Chasmagnathus granulatus* is one of the most abundant macroinvertebrates in the intertidal estuaries and bays located between southern Brazil and northern Patagonia (Spivak et al. 1994; Iribarne et al. 1997). These organisms remove large quantities of sediment (Iribarne et al. 1997; Botto and Iribarne 1999) increasing sediment water content (Botto and Iribarne 2000), redistributing sediment particles (Botto and Iribarne 1999, 2000), and generating environmental heterogeneity. The bioturbator activities affect infaunal polychaetes and nematodes (Botto et al. 1998; Botto and Iribarne 1999), indirectly affecting predators due to the alteration in prey density and availability (Botto et al. 2000). At ecosystem level, this crab may be considered as an ecosystem engineer (sensu Jones et al. 1994) because of their large impact on the sediment characteristics, which directly and indirectly affect resources for other species such as polychaetes, other crabs and shorebirds (Botto et al. 1998, 2000; Botto and Iribarne 1999, 2000). All these estuaries are also used by migratory shorebirds as stopover or wintering sites (i.e., Myers and Myers 1979; Morrison and Ross 1989; Botto et al. 1998, 2000) but most holarctic migrants avoid the crab beds while only *Charadrius falklandicus,* a species that only migrate within the southern hemisphere, use these areas (Botto et al. 2000). It is likely that this pattern of habitat use is the result of the indirect effect of crabs on polychaete by changing the sediment characteristics, which at the end affect their availability to shorebirds.

We expect that environmental heterogeneity generated by crabs will affect, directly or indirectly, the infaunal vertical movements, zonation pattern, and their risk of mortality along the intertidal. Based on a comparative study between nearby areas with and without crabs the objective of this work was to evaluate: sediment characteristic during the tidal cycle, variations across the intertidal in abundance and size distribution of infaunal organisms, vertical movements during the tidal cycle as function of infaunal sizes, and infaunal risk of mortality due to shorebirds during the tidal cycle due to the predators activities.

Materials and Methods

STUDY AREA

Field work was performed at the Mar Chiquita coastal lagoon, Argentina (37°32′S and 57°19′W; an UNESCO MAB Reserve). This is a 46 km^2 estuarine habitat affected by low amplitude tides (< 1 m) and characterized by soft-bottom intertidal areas dominated by the burrowing crab *C. granulatus* (see Iribarne et al. 1997; Botto and Iribarne 2000) that generate extensive areas with high densities of crabs (thereafter called crab bed).

GENERAL DESIGN AND STATISTICAL CONSIDERATIONS

In this work, we used nearby intertidal habitats with (inside crab beds) and without (outside crab beds) crabs at the same tidal effect. These crab beds are solid beds of burrowing crabs that cover the intertidal completely (see Iribarne et al. 1997; Botto and Iribarne 2000). Based on the tidal level each habitat (inside and outside crab beds) was divided in 7 intertidal zones (upper = A to lower = G) parallel to the shore, each zone with tidal amplitude of 10 cm. The intertidal was divided in zones of similar exposition time. To study changes during the tidal cycle, we also defined the following tidal levels (measured as the water levels in relation to low tide) in both habitats: 1 = depth of water 20 cm with ebbing tide, 2 = depth of water 5 cm with ebbing tide, 3 = depth of water 1 cm with ebbing tide, 4 = humid sediment after the low tide, 5 = 60 minutes after the previous sample, 6 = depth of water 1 cm with flooding tide, 7 = depth of water 5 cm with flooding tide, 8 = depth of water 20 cm with flooding tide.

For all statistical analysis transformation were used when assumptions were not satisfied (following Underwood 1997; Zar 1999). In the case of counts of individuals in infaunal samples, when necessary we used transformations of the type root of (variable + constant), while for bird foraging rates we performed logarithmic transformations. Transformations were always reevaluated to test the efficiency of the procedure. In the case of obtaining significant differences with ANOVA a Least Significant Difference (LSD) test of multiple comparisons were used to identify differences (Day and Quinn 1989; Underwood 1997). We corrected all p-values of all LSD tests from Type I error bias (Day and Quinn 1989) applying the sequential Bonferroni technique (thereafter SBT; Rice 1989).

PARTICULAR OBSERVATIONS

Changes in Sediment Characteristic During the Tidal Cycle

Many activities (i.e., movements, feeding) of the benthic intertidal fauna depend directly on the substrate conditions, which in turn may influence the activity of intertidal predators. One of the objectives of this work was to evaluate some of these conditions during the tidal cycle. In both habitats (inside and outside crab beds) we obtained 10 randomly allocated samples of sediment for each intertidal zone (4 cm diameter and 5 cm depth). Wet weight was determined (WW; Precision 0.0001 gr) and, after drying (50°C) for 72 h, samples were weighed again (dry weight [DW]) to obtain the sediment water content (WW-DW). In both habitats and for each intertidal zone we also measured substrate penetrability (n = 15, random samples) using a hand penetrometer that determines the necessary pressure to penetrate the substrate surface and, the sediment torsion resistance (n = 10). A two-way ANOVA (Zar 1999) was performed to test the null hypotheses of no difference in the substrate conditions between intertidal zones (A to G) and habitats (inside and outside crab beds).

To evaluate how those parameters change with the tidal cycle, samples were obtained at all tidal levels in both habitats and at four different sediment depths (a = 0–3 cm; b = 3.1–6 cm; c = 6.1–9 cm; d = 9.1–12 cm) for the case of water content. Samples were analyzed as previously described. We also evaluated penetrability at all tidal levels. The sediment resistance to the torsion was only evaluated in the tidal levels 3, 4, 5, and 6 of the cycle due to the impossibility of introducing the instrument inside the water. A two-way ANOVA (Zar 1999) was used to assess differences in the sediment characteristics in relation to tidal levels (1 to 8) and habitats (inside and outside crab beds).

Infaunal Abundance and Size Frequency Distribution Across the Intertidal

Besides variations in infaunal abundance due to environmental heterogeneity produced by crabs (Botto and Iribarne 1999), variations may also exist between intertidal zones. To evaluate this hypothesis we employed similar procedures as previously described.

In each intertidal zone of both habitats, 10 macroinfaunal samples (10 cm diameter and 15 cm of depth) were obtained and sieved through a 500-μm mesh. The retained organisms were preserved in alcohol 70%, and they were identified and counted under 20× binocular microscope. Similar samples of meiofauna were randomly obtained (n = 10, corers of 2 cm diameter and 4 cm depth) that were then sieved through a 100-μm mesh. Organisms were preserved in formalin 5%, identified and counted under 40× binocular microscope. In both cases, a two-way ANOVA (Zar 1999) was performed to evaluate differences in the density of each group between intertidal zones (A to G) and habitats (inside and outside crab beds).

Individuals of the polychaete *Laeonereis acuta* were randomly sampled using corers of 10 cm of diameter and 15 cm of depth from all intertidal zones in both habitats to evaluate their size frequency distribution. When individuals were damaged, size was estimated using the relationship between the jaw size and the total length. For this purpose, a regression model between total length and jaw size was constructed for both habitats (Zar 1999). This model was then used to estimate the total length of damaged polychaetes using the jaw size. The null hypothesis of no difference in slope between habitats was analyzed with *t*-test (Zar 1999). A Kolmogorov-Smirnov test was used to evaluate the null hypothesis of no differences in size frequency distribution between intertidal zones and between habitats (Conover 1980).

Effect of Crab Burrows on the Available Area for Predation

Given that the surface area available for predators depends on the area occupied by crab burrows, the density and the diameter of crab burrows (sampling units were squares of 0.25 m^2; n = 20 for each intertidal zone) were sampled in the different intertidal zones defined inside crab beds. The available area without burrows was estimated from these samples, using the difference between total area and area occupied by crab burrows. ANOVA (Zar 1999) was used to evaluate the null hypothesis of no difference in the number of burrows and available area between intertidal zones (A to G).

INFAUNAL VERTICAL MOVEMENTS DURING TIDAL CYCLE

Macroinfaunal samples were obtained for each tidal level (1 to 8; n = 10 for each level) at both habitats. The samples were divided in situ in 5 layers of depth of 3 cm each one, and processed then as previously described. The null hypothesis of no difference in abundance of individuals between sediment depths, habitats, and during the tidal cycle (levels 1 to 8) was evaluated using a three-way ANOVA (Zar 1999) using sediment depth, habitat, and tidal level as fixed factors.

Macroinfaunal individuals obtained in the previously described sampling were measured to evaluate size distribution at each depth during the tidal cycle in both habitats. The null hypothesis of no difference in size frequency distribution during different tidal levels (1 to 8), and between habitats (inside and outside crab beds), for each sediment depth, was evaluated using a Kolmogorov-Smirnov test (Conover 1980).

RISK OF INFAUNA MORTALITY DURING THE TIDAL CYCLE

Benthic animals have a zonation pattern across the intertidal, which could be different inside and outside crab beds given the effect of crabs. If so, shorebirds may feed in different intertidal zones. To explore the use of habitat by the lesser yellowlegs *Tringa flavipes,* abundance of birds per unit of time was estimated at different intertidal zones, differentiating flooding and ebbing tide in both habitats. For this purpose, for each intertidal zone the time of observation was standardized to 1 h counting the number of birds that were foraging. The null hypothesis of no difference in the distribution of birds across the intertidal (zones A to G), inside and outside the crab beds, and with flooding or ebbing tide was evaluated using a Kolmogorov-Smirnov test (Conover 1980).

During the tidal cycle, the infaunal organisms may be at different depths into the substrate. If this were the case, their availability to shorebird predation should be different, and the predation efficiency of shorebirds should change. To evaluate this hypothesis we performed focal censuses of *T. flavipes* using a monocular telescope (18–36×). Observations were restricted to this species given that was the only abundant shorebird during the study period. This was made in all previously described intertidal zones (A to G) in both habitats (inside and outside crab beds), differentiating flooding and ebbing tide (each bird was observed for a period of 5–10 min). During the focal censuses, we registered the time that each bird spends in the following behaviors: walk, pause, peck, and capture of polychaetes (see Botto et al. 2000). The consumption rate was calculated as the amount of prey consumed per unit of time. The null hypothesis of no difference in consumption rates between intertidal zones within habitats was evaluated using the non parametric test of Kruskal-Wallis (Conover 1980) and the null hypothesis of no difference in consumption rate for each intertidal zone (A to G) comparing habitats was evaluated using a Mann-Whitney test (Conover 1980).

Using the data obtained from focal observations we calculated: consumption rate (a), habitat use (b) and infaunal density (c), data that was then used to estimate the per capita mortality rate of infauna for each intertidal zone and habitat and in the different tidal levels as

$$\text{Mortality rate (per capita)} = a * b/c,$$

where $a = \text{prey} * \text{minute}^{-1}$, $b = \text{minute} * \text{m}^{-2}$, and $c = \text{prey} * \text{m}^{-2}$.

For the calculation of mortality rate, we employed a resampling method (Manly 1998), taking data of each one of the three variables (with replacement) independently for each intertidal zone and habitat, and in the different tidal state. This process was repeated 5,000 times for each case mentioned previously (intertidal zones, habitats, and tidal states). A two-way ANOVA test (Zar 1999) was used to evaluate the null hypothesis of no differences in the risk of mortality between intertidal zones (A to G) and habitats (inside and outside crab beds). Given that rejection of the null hypothesis is directly related with the sample size, performing different number of iterations may produce different results. For this, following a conservative approximation and to be independent from the number of iterations, we used as degree of freedom the sum of the sample size of the three variables employed in the calculations of the mortality rate.

Results

CHANGES IN SEDIMENT CHARACTERISTIC DURING THE TIDAL CYCLE

Sediment water content was higher in the low intertidal zones in both habitats (Table 1 and Fig. 1), but in general was higher inside crab beds. During the tidal cycle, differences existed in the upper sediment layer (0–3 cm) outside crab beds and inside them (Table 2 and Fig. 2), the water content was higher in the flooding tide than ebbing tide at both habitats.

Sediment hardness decreases from the highest intertidal zones to the lowest zones for both habitats (Table 1 and Fig. 1). Differences were larger in habitats without crabs. Hardness was also higher in the habitats without crabs than in the crab beds, although at the lower intertidal zones the differences are smaller (Fig. 1). The values of hardness during the tide cycle differed in both habitats when the different tidal levels were compared (Table 2 and Fig. 2). Hardness of substrate was lower in the tidal levels where the substrate was flooded and it increased toward lowest tidal levels when the substrate was dry or slightly humid. Differences between habitats were significant for each tidal level; the substrate resistance was always smaller inside the crab beds than outside them (Fig. 2).

The sediment was more resistant to the torsion in high intertidal zones in both habitats (Table 1 and Fig. 1). This measure was always higher in habitats without crabs than inside crab beds. The difference between habitats decreases from high to low intertidal zones (Fig. 1). During the tidal cycle the substrate in habitats without crabs was always more resistant than inside crab bed (Fig. 2). In habitats without crabs, the resistance to the torsion decreases when the sediment was flooded (tidal levels 3 and 6; Table 2 and Fig. 2), but inside crab bed none differences were seen between tidal levels.

INFAUNAL ABUNDANCE AND SIZE FREQUENCY DISTRIBUTION ACROSS THE INTERTIDAL

Macroinfaunal densities differed significantly between intertidal zones (Table 1 and Fig. 3) and they were different for each intertidal zone comparing both habitats (Fig. 3). For all intertidal zones, the density was higher in the habitats without crabs, except in intertidal zone C where the

TABLE 1. Summary of two-way ANOVA evaluating sediment penetrability, sediment water content, sediment torsion resistance, densities of infauna, densities of nematodes, densities of copepods, densities of ostracods, per capita mortality rate of infauna during ebbing tide, and per capita mortality rate of infauna during flooding tide. All these variables were evaluated in relation to intertidal zones (Z) and habitats (H). *** Indicates $p < 0.001$; ** indicates $p < 0.01$; * indicates $p < 0.05$.

Source of Variation	df_{effect}	df_{error}	MS_{effect}	*F*
Sediment penetrability				
Zone (Z)	6	123	15.82	59.75***
Habitat (H)	1	123	142.04	536.29***
Z × H	6	123	1.38	5.21***
Sediment water content				
Zone (Z)	6	123	445.72	12.66***
Habitat (H)	1	123	2,748.22	78.07***
Z × H	6	123	52.88	1.50ns
Sediment torsion resistance				
Zone (Z)	6	123	14.85	10.23***
Habitat (H)	1	123	340.93	234.86***
Z × H	6	123	3.87	2.67*
Densities of infauna				
Zone (Z)	6	123	9.46	63.89***
Habitat (H)	1	123	16.07	108.51***
Z × H	6	123	0.38	2.55*
Densities of nematodes				
Zone (Z)	6	123	397.34	37.08***
Habitat (H)	1	123	1,836.06	171.33**
Z × H	6	123	33.10	3.09**
Densities of copepods				
Zone (Z)	6	123	34.12	16.06***
Habitat (H)	1	123	5.21	2.45ns
Z × H	6	123	5.62	2.65*
Densities of ostracods				
Zone (Z)	6	123	101.31	18.04***
Habitat (H)	1	123	146.06	26.01***
Z × H	6	123	12.90	2.30*
Per capita mortality rate of infauna during ebbing tide				
Zone (Z)	5	391	2.68	15.31**
Habitat (H)	1	391	2.87	16.38**
Z × H	5	391	0.90	5.15*
Per capita mortality rate of infauna during flooding tide				
Zone (Z)	5	386	2.10	13.61**
Habitat (H)	1	386	3.03	19.61**
Z × H	5	386	1.02	6.63*

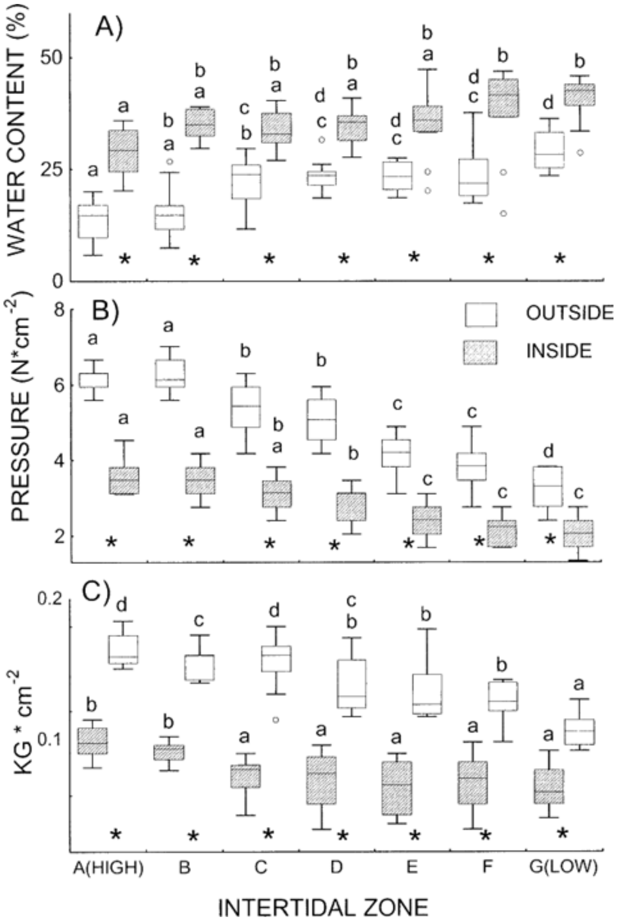

Fig. 1. Sediment characteristics across the intertidal (zones A to G) in both habitats (inside and outside crab beds). A) Sediment water content expressed as water percentage, B) necessary pressure to penetrate the sediment surface (penetrability) expressed as pressure, and C) torsion resistance of sediment expressed as $Kg \cdot cm^{-2}$. Here and thereafter box plots are constructed with limits of boxes being the 75th and 25th percentile, lines outside boxes represent 10th and 90th percentiles, lines inside boxes are medians, circles are outliers. Different letters on top of boxes represent significant differences between intertidal zones at the same habitat and, asterisks represent significant differences between habitats at the same intertidal zone (LSD test with $\alpha = 0.05$, after SBT).

density did not show differences between both habitats, although the trend is similar. The groups identified in the meiofaunal samples were nematodes, copepods and ostracods, with similar distribution patterns across intertidal in both habitats (Fig. 3). The nematodes density increased in both habitats toward low intertidal zones (Table 1 and Fig. 3). The copepods abundance was higher in the mid intertidal zones of both habitats (Table 1 and Fig. 3). The ostracods density increased toward the low zones of the intertidal in both habitats being the smallest densities in the high intertidal zones (Table 1 and Fig. 3). Few differences appear at each intertidal zone between habitats for the three groups, being these differences more frequent in nematodes (Fig. 3).

There was always a positive relationship between the jaw size (J) and total length (TL) for *L. acuta*

Outside:

$$TL = -8.37\ (\pm 2.06) + 92.76\ (\pm\ 4.15) * J$$

$$r^2 = 0.87,\ n = 79$$

Crab bed:

$$TL = -14.95\ (\pm 2.09) + 109.22\ (\pm\ 4.1) * J$$

$$r^2 = 0.90,\ n = 83$$

TABLE 2. Summary of two-way ANOVA evaluating sediment penetrability, sediment water content, and sediment torsion resistance; in relation to tidal levels (1 to 8, called "L") and habitat (H). *** Indicates $p < 0.001$; ** indicates $p < 0.01$.

Source of Variation	df_{effect}	df_{error}	MS_{effect}	F
Sediment penetrability				
Level (L)	7	144	7.26	38.07***
Habitat (H)	1	144	151.97	797.46***
L × H	7	144	0.79	4.13***
Sediment water content				
Level (L)	7	144	28.90	4.24***
Habitat (H)	1	144	2,353.21	345.99***
L × H	7	144	20.52	3.01**
Sediment torsion resistance				
Level (L)	3	72	15.53	38.71***
Habitat (H)	1	72	263.38	905.58***
L × H	3	72	4.87	12.13***

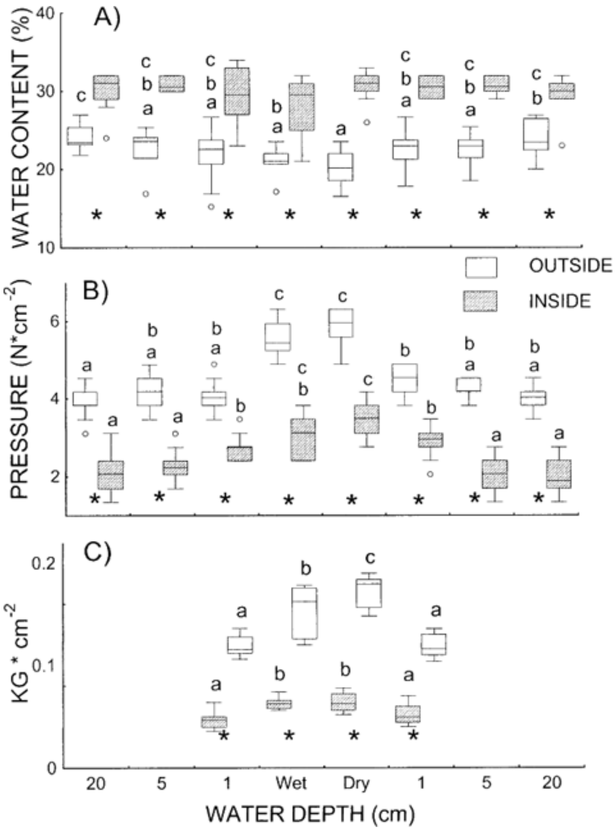

Fig. 2. Sediment characteristics during tidal cycle (represented by water depth) at both habitats (inside and outside crab beds). A) Sediment water content expressed as water percentage, B) necessary pressure to penetrate the sediment surface (penetrability) expressed as pressure, and C) torsion resistance of sediment expressed as Kg·cm^{-2}. Different letters above the boxes represent significant differences between tidal levels (water depth) at the same habitat and, asterisks represent differences between habitats at the same tidal level (LSD test with $\alpha = 0.05$, after SBT).

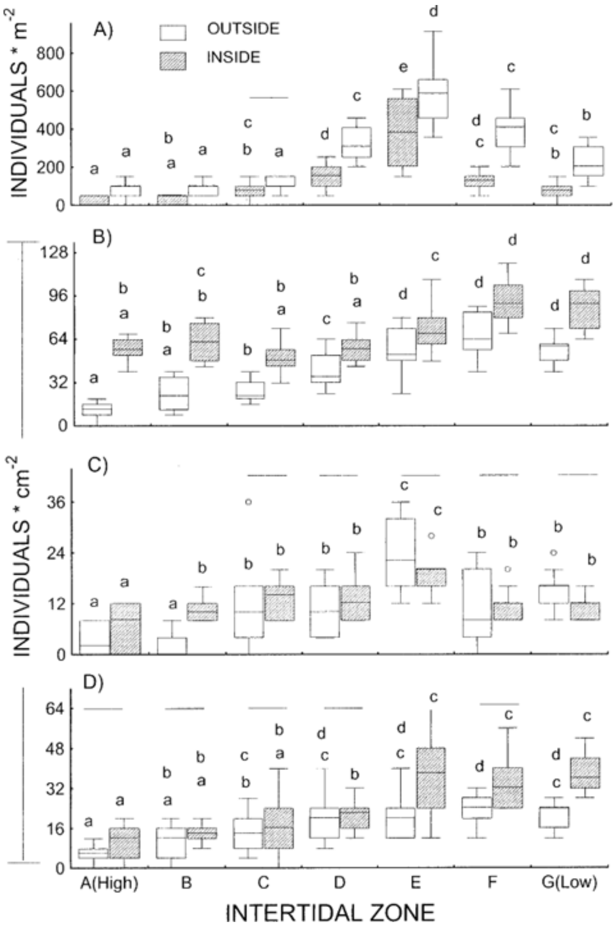

Fig. 3. Abundance of infauna across the intertidal zones (A to G) at both habitats (inside and outside crab beds), expressed as individuals/unit of area. A) Polychaetes, B) nematodes, C) copepods, and D) ostracods. Different letters above the boxes represent significant differences between intertidal zones at the same habitat and, lines represent no significant differences between habitats at the same intertidal zone (LSD test with $\alpha = 0.05$, after SBT).

the slopes of both straight lines were different ($t = 7.77$, $df = 158$, $p < 0.05$).

The above results plus data of non-damaged polychaetes were used to evaluate the size frequency distribution of polychaetes in each habitat. These size frequency distributions did not differ between crab beds and habitats without crabs in any zone except zone E. The mean size was larger at the high intertidal zones (A and B) than in the other zones (C to F) for both habitats (K-S test, $p < 0.05$; Fig. 4).

Effect of Crab Burrows on the Available Area for Predation

The highest burrow density of *C. granulatus* was present in the high intertidal zones decreasing toward the lower zones (ANOVA: $df = 6, 63$, $F =$

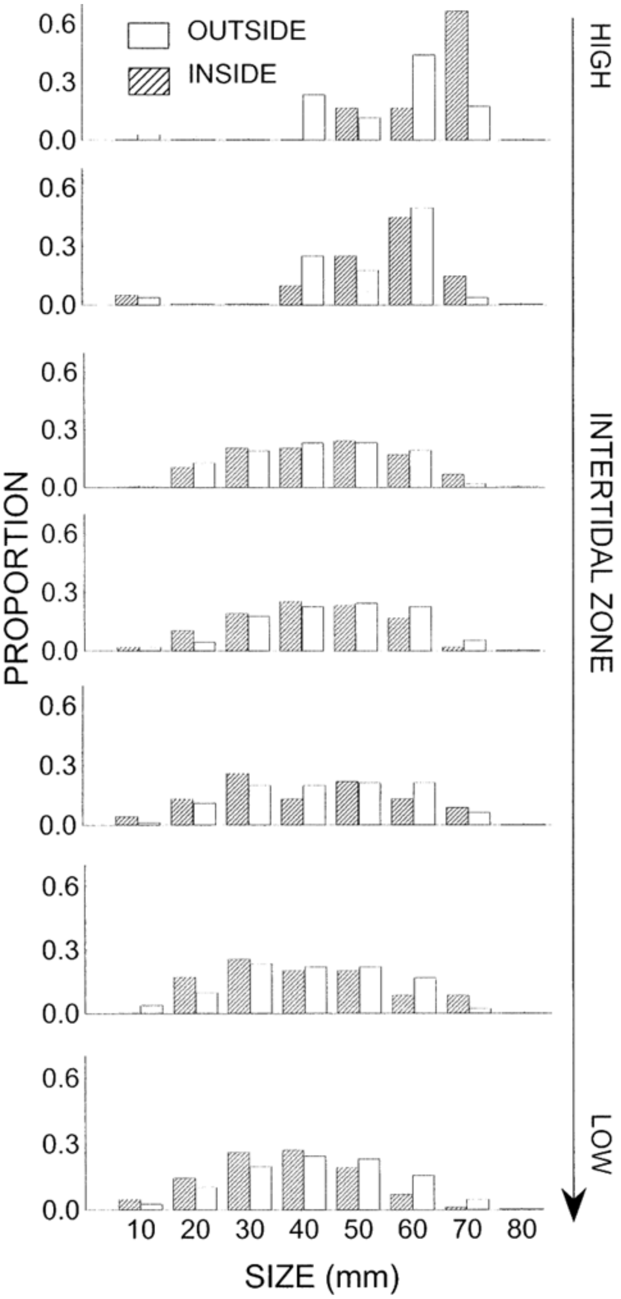

Fig. 4. Sizes distribution of polychaetes expressed as proportion from high to low intertidal zones (A to G) at both habitats (inside and outside crab beds). Significant differences existed inside and outside crab beds between intertidal zones (K–S test, being $p < 0.05$ for both habitats).

63.9, $p < 0.001$). The available area to predation (total area − area occupied by burrows) decrease considerably in the high intertidal zones inside crab beds (ANOVA: df = 6, 63, F = 76.67, $p < 0.001$), where the available area was approximately 70% of the total area (Fig. 5).

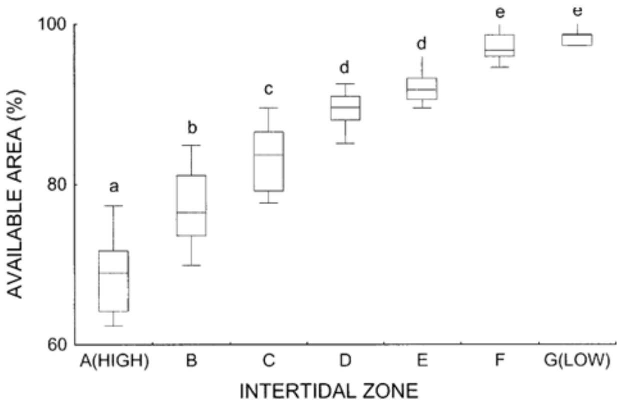

Fig. 5. Available area for predation expressed as m^2 across intertidal zones (A to G) inside crab beds. Different letters represent significant differences between intertidal zones (LSD test with $\alpha = 0.05$).

INFAUNAL VERTICAL MOVEMENTS DURING THE TIDAL CYCLE

Vertical movements were detected in both habitats, but these movements were of higher magnitude outside crab beds than inside. Differences in the abundance of macroinfaunal organisms were registered outside crab beds in the sediment depths A, C, and D, but no differences were seen for the sediment depth B during the tidal cycle (Table 3 and Fig. 6). The density decreased in depth A when the tide was ebbing, and increased in the depths C–D (deepest levels). The pattern is similar in the crab beds but without statistical differences between sediment depths, except for depth D with dried sediment, during the lowest tidal level (Fig. 6).

The size frequency distributions were different in the surface (0 to 3 cm, K–S test, $p < 0.05$; Fig. 7) at both habitats during the tidal cycle, although these differences were larger outside the crab beds (K–S test, $p < 0.01$; Fig. 7). There were no significant differences at the other sediment depths at any moment of the tidal cycle. The larger sizes

TABLE 3. Movements of infauna. Summary of three-way ANOVA evaluating densities of infauna in relation to tidal levels (1 to 8, called "L"), sediment depth (a to d, called "D"); and habitats (called "H"). (**) $p < 0.01$, (*) $p < 0.05$, (ns) nonsignificant differences.

Source of Variation	df_{effect}	df_{error}	MS_{effect}	F
H	1	576	2.04	16.21
L	7	576	0.15	1.22
D	3	576	11.15	88.59
H × L	7	576	0.05	0.42
H × D	3	576	0.09	0.75
L × D	21	576	0.98	7.79
H × L × D	21	576	0.21	1.66
H × L	7	576	0.05	0.42

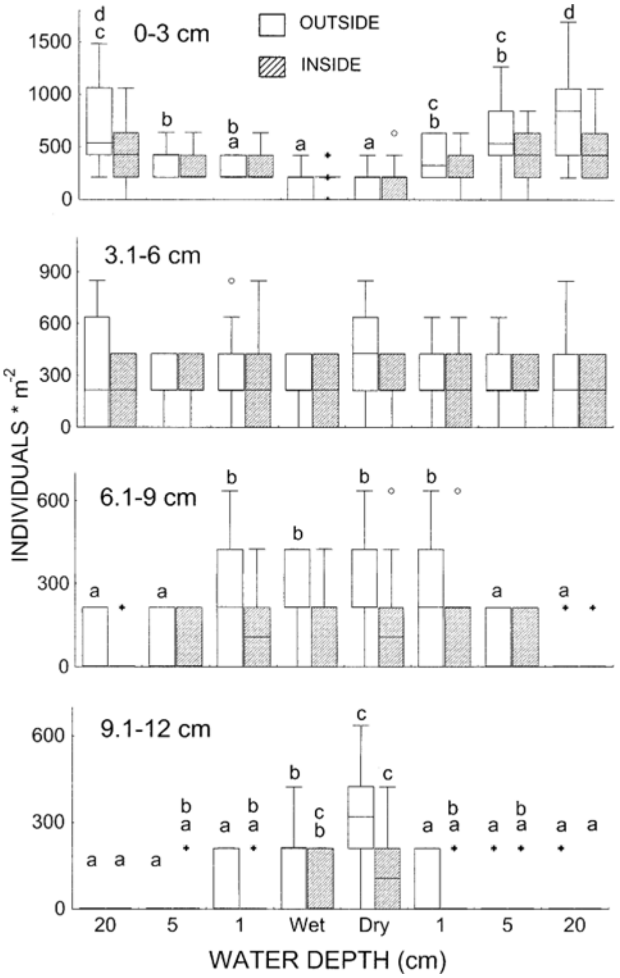

Fig. 6. Vertical movements (expressed as densities at different depths) of macroinfauna during tidal cycle (correspond to water depth) in both habitats (inside and outside crab beds). Different letters represent significant differences between intertidal zones at the same habitat (LSD test, $\alpha = 0.05$, after SBT).

were always at the deepest sediment depth. There were only two cases with differences between habitats at different tidal levels and depth. Size always increases with depth (K–S test, $p < 0.05$).

Risk of Infaunal Mortality During the Tidal Cycle

Habitat use by birds also differed between habitats with and without crabs during the ebbing tide. The shorebirds were distributed more homogeneously across the intertidal in the crab bed, while they were restricted to the lower intertidal outside the crab bed. There were no differences during the flooding tide, although the pattern is similar to the ebbing tide (Kolmogorov-Smirnov: ebbing, $p < 0.05$; flooding, $p = 0.06$; Fig. 8).

Differences in consumption rates were obtained

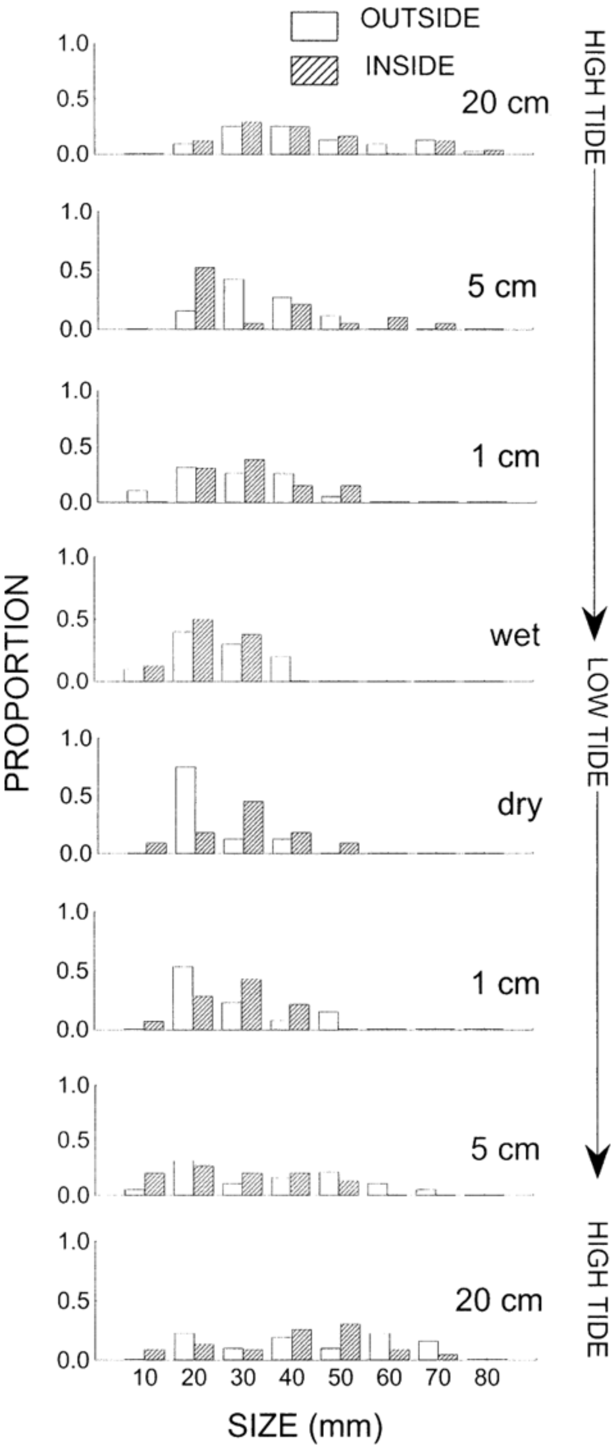

Fig. 7. Polychaetes size frequency distribution expressed as proportion during tidal cycle (correspond to water depth) in the upper sediment layer (0–3 cm) of both habitats (inside and outside crab beds).

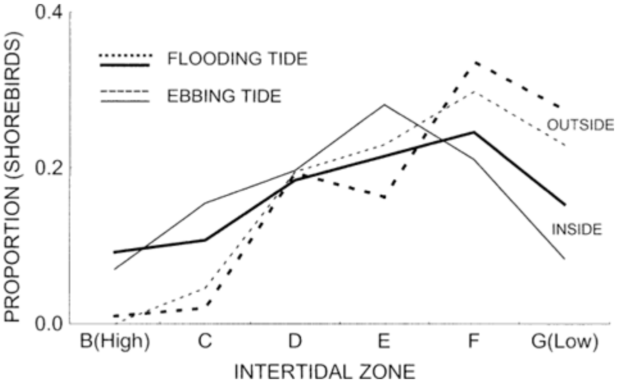

Fig. 8. Habitat use of *Tringa flavipes* as proportion of birds in the different intertidal zones of both habitats (inside crab beds: continue line, outside crab beds: dotted line) at different tidal states (ebbing–flooding).

in the comparison between habitats in some intertidal zones. Consumption rates of birds differ when ebbing and flooding tide were compared. Differences between intertidal zones were only in habitats without crabs during ebbing tide (Kruskal-Wallis: Inside: $H_{(5,\ n\ =\ 68)} = 5.8$, $p > 0.05$; Outside: $H_{(5,\ n\ =\ 83)} = 19.5$, $p < 0.01$). During the flooding tide there were differences between intertidal zones only in crab beds (Kruskal-Wallis: Inside, $H_{(5,\ n\ =\ 62)} = 12.3$, $p < 0.05$; Outside, $H_{(5,\ n\ =\ 104)} = 9.9$, $p > 0.05$). The low statistical power associated to these data in the multiple comparisons after Kruskal-Wallis does not allow to identify where the differences occurs.

The per capita mortality rate (risk of predation) differed between habitats in some intertidal zones with ebbing and flooding tide (Table 1 and Fig. 9). At the high intertidal, the mortality rate was higher in the crab beds than outside them during both tidal states, and in the low intertidal the mortality rates do not varied between habitats. During ebbing tide, the mortality rate was different between zones in both habitats (Table 1 and Fig. 9) and during flooding tide there were differences only outside crab beds (Fig. 9).

Discussion

Our results shows that the physical characteristics of the sediment (humidity, sediment softness, and torsion resistance), movement and zonation patterns of infauna, and habitat use by shorebirds and their consumption rate varies depending of the presence of beds of the burrowing crab *C. granulatus*. All evidence suggests that these differences are the indirect results of their sediment reworking activity.

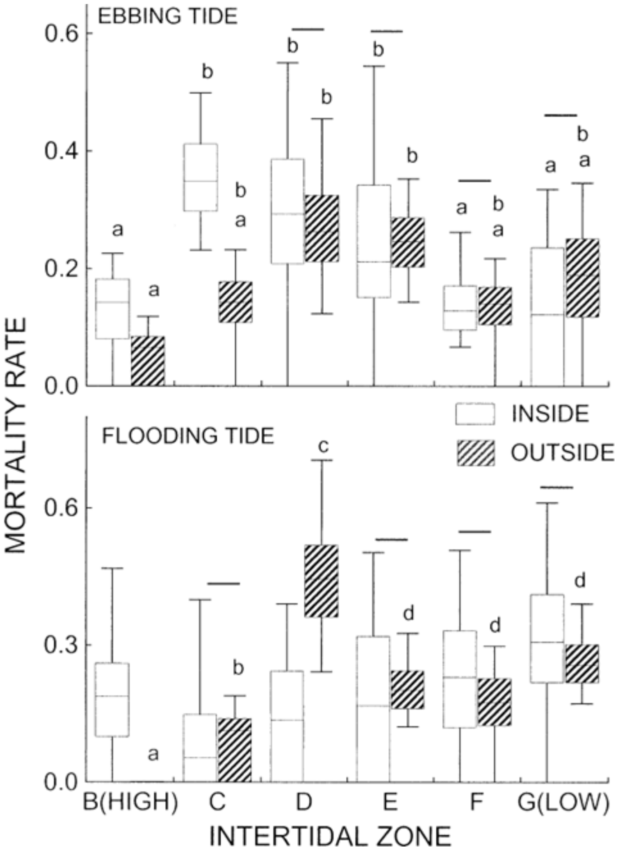

Fig. 9. Per capita mortality rate in the different intertidal zones of both habitats (inside and outside crab beds) at different tidal states (ebbing–flooding). Different letters represent significant differences between intertidal zones at the same habitat (LSD test with $\alpha = 0.05$, after SBT). Lines connect no significant differences between habitats at the same intertidal zone.

The burrowing crab *C. granulatus* is an important bioturbator of estuarine intertidal flats reworking up to 5 kg m^{-1} day^{-1} (Iribarne et al. 1997; Botto and Iribarne 2000). Their activity increases sediment softness, organic matter, and water content. Their large burrows (up to 1 m deep and entrance up to 20 cm diameter; Iribarne et al. 1997) are always open, which effectively trap clay and silt particles rich in organic matter (Botto and Iribarne 2000). Our results also show that all those characteristics remain across the intertidal, making the intertidal area more homogeneous in the crab bed than outside. The differences between habitats are higher at the high intertidal zones, being the intertidal environmental gradient more pronounced outside the crab beds. Crabs also remove very fine and cohesive sediments that stabilize the sediment decreasing bedload transport (Botto and Iribarne 2000). This fine sediment may also serve as a shield against evaporation during the low tide, maintaining higher humidity and homogeneity across sediment depth in the crab bed. The variation during the tidal cycle in the sediment water content was significantly higher outside the crab bed, being the sediment very homogeneous inside them. In terms of sediment humidity the activities of crabs decrease the tidal effect.

The pattern of infaunal zonation was very similar in both habitats. The density of infaunal organisms (i.e., polychaetes, nematodes, copepods, and ostracods) was higher at the low and medium intertidal zones. Density of meiofauna increases at the high intertidal zones in the crab bed. This pattern may be the results of the higher sediment water and organic matter retained in these sediments (see Botto and Iribarne 2000). The habitats without crabs showed much faster desiccation when the tide ebbs. This environment becomes harsher for meiofaunal organisms that live in the upper few centimeters of the sediment (see Coull 1992). In this way the crabs are moderating the physical-chemical conditions of sediment, consequently having a positive effect (sensu Bertness 1999) on some infaunal species.

As a general mechanism of intertidal zonation (see Bertness 1999), the upper distribution limit is determined by physical factors (i.e., temperature, desiccation) while the lower limit are established by biological processes (see Rafaelli and Hawkins 1996). We have no evidence that *C. granulatus* changes this mechanism, but their activity ameliorates the physical harshness of the upper intertidal allowing some organisms to extend their distribution to upper levels.

Individuals of *L. acuta* migrated into the sediment to a deeper position during low tide and return to the upper sediment layers when the sediment is flooded. The largest individuals move toward deeper layers during the tidal cycle. As a result, the mean size of individuals in the upper sediment layers largely change with the tidal cycle. This pattern may be to avoid predation (Wanink and Zwarts 1993) or intraspecific competition (Zwarts and Wanink 1991) between individuals of different sizes. The individuals that remain most exposed to predation during low tide are the smaller. Their vertical movements showed higher amplitude outside the crab beds. This pattern is similar to changes in sediment water content, and may be the reason of this behavior. One of the causes recognized for vertical movements are to avoid desiccation, which is a critical factor in intertidal areas (see Reise 1985; Brown and McLachlan 1990). Maximizing feeding has also been described as a cause of vertical movements (Reise 1985; Brown and McLachlan 1990). Sediment nutritious quality is higher and more homogeneously distributed across the sediment inside crab beds, largely due to the trapping of fine sediments into the crab burrows, which are rich in organic matter (Botto and Iribarne 2000). Occurrence of feeding on the surface by this polychaete is higher in habitats without crabs than in areas with crabs (Palomo and Iribarne 2001). This evidence suggests that there is a positive interaction of crabs on polychaetes.

Our result also showed that the smallest polychaete do not migrate toward deeper sediments remaining in the superficial layers during low tide. Given the length of shorebird bills, those individuals should be the most exposed to mortality by predation. Experiments in the same area have shown that in this area the small size-polychaetes were the most affected by shorebird predation (Botto et al. 1998).

Habitat use by migratory shorebirds is also affected by the crab bed (Botto et al. 2000). The two-banded plover (*C. falklandicus*) forage more frequently inside while the white-rumped sandpiper (*C. fuscicollis*), and the yellowlegs (*T. flavipes* and *T. melanoleuca*) forage mainly outside crab beds. This may be in part because the available area for predation, mainly on the high intertidal zones, inside crab beds decreases considerably due to increases in the density of burrows. Recent results show that shorebirds that tend to avoid the crab beds, when feeding inside use foraging paths that maximize the distance to burrows (Bruschetti and Iribarne 2001) which actually reduce the foraging area. Focal observations on the feeding behavior of the white-rumped sandpiper (*Calidris fuscicollis*) and the two-banded plover (*Charadrius falklandicus*) inside and outside crab beds showed that the plover was a visual searcher and captured more prey inside crab beds, and the white-rumped sandpiper was a tactile feeder. Although consumption rates (prey min^{-1}) did not differ between sites, their efficiency (prey probe^{-1}) inside crab beds was lower (Botto et al. 2000). These differences were probably related to changes in sediment characteristics and prey behavior, which vary with crab activity.

The only common shorebird species found in our study site was the lesser yellowleg *T. flavipes*. During low tide, they use all intertidal zones inside crab beds but foraging was restricted to lowest intertidal zones where the sediment is less compact in habitats without crabs. This observation may be the result of sediments that remain humid and softer for longer time at the high intertidal zones inside the crab beds when compared with similar zones outside. Although it is expected that higher sediment penetrability generated by crab activity should increase the risk of infaunal mortality (Myers et al. 1980; Pienkowski 1981; Goss-Custard 1984; Quammen 1984; Mouritsen and Jensen 1992), the foraging rates of *T. flavipes* did not show differences between habitats with and without crabs. This may be because of the low activity of polychaete in the surface inside the crab beds (Palomo and Iribarne 2001). Shorebirds use the crab beds more time and cover larger areas and thus total predation is higher. There is a positive indirect effect of crabs on shorebirds.

The changes in the sediment promoted by bioturbators have direct and indirect effects in the infaunal organisms that consequently modify their interactions with other species. In this work, we presented further evidences that bioturbator organisms such as *C. granulatus* promote changes in sediment characteristics that direct and indirectly influence the behavior and survival of sub-superficial organisms and in the activity of predators. This has not been evaluated in other systems, but it is likely that other bioturbators as the burrowing crustaceans *Callianassa* and *Upogebia* (see Posey 1986; Posey et al. 1991) have similar effects. This is another example of the ecosystem engineering (sensu Jones et al. 1994) capability of bioturbator species. They largely modify the availability of resources to other organisms and modify the interaction between species.

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