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ECOSYSTEMS

The bivalves *Amarilladesma mactroides* and *Donax hanleyanus* as bioindicators of the impact of vehicles on Cassino Beach, southern Brazil

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Abstract: Sandy beaches are the main recreational ecosystems of the world, enabling high ecological impacts, especially on the benthic macrofauna, which inhabit the sandy matrix and have a low capacity of locomotion. Cassino Beach, located in southern Brazil, has intense vehicle traffic during the summer, so the purpose of this study was to evaluate the impact of vehicles on the key species *Amarilladesma mactroides* and *Donax hanleyanus*. For this purpose, samplings were performed in three sectors of this beach (High Impact, Moderate Impact and Control) during six periods of the year. The results showed lower densities of both bivalves in the High Impact sector than in to the other sectors in all periods, except in first summer sampling, and a predominance of recruits throughout the study. Thus, it suggests that the two species were influenced by the intense vehicle traffic, especially in the most impacted sector. In this way, we conclude that these bivalves could be used as good indicators of pulse disturbance by vehicle traffic on this beach and the results can support in management plans regarding the use of Cassino Beach, considering ecological aspects of this ecosystem in addition to economic and cultural demands.

Key words: Ecology, benthic macrofauna, anthropogenic impact, coastal management.

INTRODUCTION

Sandy beaches are widely used by humans for recreational and economic purposes, causing high disturbances in these ecosystems, mainly through the development of coastal areas (Afghan et al. 2020, McLachlan & Defeo 2017). These disturbances can differ by multiple orders of magnitude, depending on their intensity and duration (Defeo et al. 2009, Costa et al. 2020), are classified as pulse or press (Glasby & Underwood 1996). A pulse disturbance is characterized by be short-term, causing a sudden change in species numbers and recovery after the disturbance ends. In contrast, a press disturbance is characterized by being a continuous disturbance that causes the abundance or density of species to change permanently (Glasby & Underwood 1996).

The most common types of beach recreation are hiking, camping, fishing and vehicle traffic (Costa et al. 2020, McLachlan & Defeo 2017), which can cause severe damage to dune and macrobenthic populations (Farris et al. 2013, Hesp et al. 2010, Machado et al. 2017). According to Defeo et al. (2009), these beach uses are configured as pulse disturbances, with effects generally occurring for weeks or months at special scales of up to 10 km; thus, these pulse disturbances are emerging as significant environmental issues.

Vehicle traffic is possibly the most severe form of impact among recreational uses, altering the physical characteristics of the sediment (Bom & Colling 2020, Schlacher & Thompson 2008, Vieira et al. 2004) and disturbing the beach fauna, including vertebrates and invertebrates (McLachlan & Defeo 2017). The main studies concerning this type of impact are related to benthic macroinvertebrates (Schlacher et al. 2008a, Sheppard et al. 2009, Lucrezi & Schlacher 2010, Davies et al. 2016), since they occupy the intertidal sandy matrix, where the most vehicle traffic occurs (Schlacher & Thompson 2007, Schlacher et al. 2008a). In addition, benthic macroinvertebrates play a key role in the trophic chains of sandy beaches, serving as a link between primary producers and predators, such as fish and seabirds (McLachlan & Brown 2006, Pinotti et al. 2014). Therefore, the mortality of these invertebrates on sandy beaches by vehicles can cause effects at broad ecological levels, such as modifications of food webs, making this impact even more significant (Defeo et al. 2009).

On Cassino Beach, located in the southern Brazil, vehicle traffic occurs for approximately 20 km, mainly during the summer, when thousands of cars circulate daily (Figure 1), without any restriction or access control (Vieira et al. 2004). Bom & Colling (2020) identified the impact caused by vehicles on whole benthic macrofauna assemblages; however, further studies are needed regarding the behavior of each species. Thus, the present study aimed to evaluate the disturbance from vehicle traffic on two dominant species: the bivalves *Donax hanleyanus* Philippi, 1847 and *Amarilladesma mactroides* (Reeve 1854). These species were chosen because they have high densities and biomass on southern Brazilian beaches, representing an important link between phytoplankton and higher levels of the food chain (Pinotti et al. 2014). Furthermore, bivalves have been monitored in studies of recreational impacts on beaches, such as the effects of harvesting and trampling, and the vehicle traffic (Defeo & De Alava 1995, Laitano et al. 2019, Schlacher et al. 2008b, Vieira et al. 2012), since they have a wide distribution range, well-known biology and low mobility, and are

Figure 1. Different sampling sectors of Cassino Beach: (a) High Impact; (b) Moderate Impact; and (c) Control. (d) Bivalve *Donax hanleyanus* killed by direct vehicle action.

considered good bioindicators of impact in these ecosystems (Costa et al. 2020).

MATERIALS AND METHODS

Sampling design

The species *A. mactroides* and *D. hanleyanus* were evaluated among three sectors of Cassino Beach, located on the Brazilian southern coast: High Impact (-32.15 and -32.16); Moderate Impact (-32.25 and -32.26); and Control (-32.38 and -32.39), and the study area has a shoreline of approximately 30 km (Figure 2). These sectors were chosen based on the following: (a) High Impact: sector with a high number of vehicles in a long and continuous line of more than 10 km, reaching thousands of vehicles during the summer due to the proximity of the urbanized area (Balneário Cassino) (Esteves et al. 2003, Vieira et al. 2004); (b) Moderate Impact: sector in which vehicle traffic occurs but in smaller numbers due to the distance from the urbanized region and lack of infrastructure; and (c) Control: sector in which the passage of vehicles is hampered by the presence of streams, in addition to distance from the urbanized region.

For the spatiotemporal evaluation of bivalve densities each sector was represented by two points (distance of 2 km between them), comprising 6 sample points (Figure 2) and samplings were performed during six periods: pre-summer I (October/16), pre-summer II (November/16), summer I (February/17) summer II (March/2017), post-summer I (June/17) and post-summer II (July/17).

Biological and sediment samples

Biological samples were collected through a PVC-core, 20 cm in diameter (0.031 m^2), collecting 20 cm of the substrate. Each sampling point was represented by two beach levels of the intertidal zone (minimum and maximum of swash zone), with three replicates at each level (Figure 2), totaling 216 samples. The samples were previously sieved in 0.5 mm aperture meshes and fixed in 4% formalin. In the laboratory, the bivalves were identified with a stereoscopic microscope, quantified and preserved in 70% alcohol.

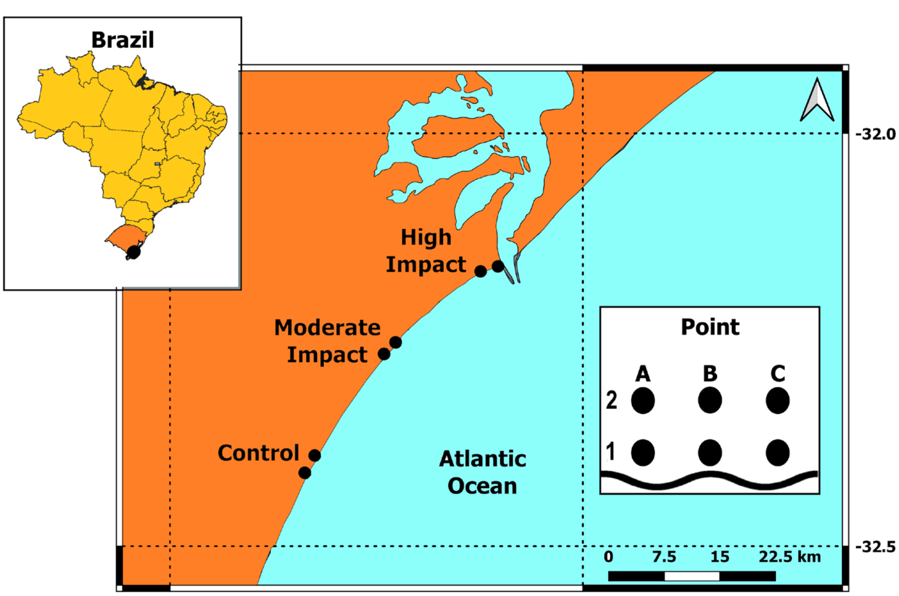

Figure 2. Location of the High Impact (-32.15 and -32.16); Moderate Impact (-32.25 and -32.26); and Control (-32.38 and -32.39) sectors on Cassino Beach, southern Brazil, each being sampled at two points. The sample design of each point is highlighted, comprising 6 biological samples at each point per sampling.

The lengths of the organisms were measured and classified into different size classes according to Defeo (1998): recruits (*A. mactroides*: <10.0 mm; > 5.0 mm), juveniles (*A. mactroides*: 10.1-42.9 mm; *D. hanleyanus*: 5.1-14.9 mm) and adults (*A. mactroides*: > 43.0 mm; *D. hanleyanus*: > 15.0 mm).

Simultaneously to the biological sampling, a sample of 50 g of sediment was collected in all sectors and periods (one sample at each level) to perform granulometric analyses, following the techniques of Suguio (1973). Additionally, the morphodynamic characterization of each sector was based on the comparison of grain size averages evaluated by Pereira et al. (2010) and the results found in the present study.

Data analysis

Using RStudio software (RStudio Team 2020), three-way ANOVA tests were performed using the density of species to identify significant differences between sectors, periods, beach levels and their interactions. To comply with the prerequisites of normality, homogeneity of variance and independence, the original data were transformed by log(x+1). ANOVA tests were also performed on the sediment data to check for possible differences between sectors and periods.

RESULTS

Sediment characteristics

The High Impact sector was dominated by fine sands (76% by weight), followed by very fine sands (23%) and medium sands (0.5%). The Moderate Impact and Control sectors were also dominated by fine sand (81 and 83%, respectively), but the medium sand size was the second most abundant, representing more than 9% of the total sediment weight in these sectors. Significant differences were found between sectors, periods and their interaction (Table I), with average grain sizes (Φ) of 2.73, 2.50 and 2.49 phi for the High Impact, Moderate Impact, and Control sectors, respectively, characterizing them as dissipative (High Impact) and intermediate of low mobility (Moderate Impact and Control) beaches.

Biological data

During the study, a large number of samples did not contain *A. mactroides* and *D. hanleyanus* (36.6%; 79 samples). Of these, 45 samples were from the High Impact sector (63%), mainly from the pre-summer period. On the other hand, a lower number of samples from the Moderate Impact and Control sectors did not contain these species (33 and 14%, respectively).

A total of 7599 bivalves were recorded (*D. hanleyanus*: 3985 org; *A. mactroides*: 3614 org.) with greater abundance at level 1 for both species (77.7% and 80.7% of *D. hanleyanus* and *A. mactroides*, respectively). Regarding the spatiotemporal densities of the bivalves, it was observed that both species presented the lowest densities in the High Impact sector during all periods, except for a peak of density in the first summer sampling for *D. hanleyanus* (Figure 3). In the other sectors, significantly higher values were recorded mainly in summer, with densities greater than 3000 ind. m^{-2} for both species (Figure 3). It is also worth mentioning that the different density peaks between the sectors, with the highest values for both species occurring in the first summer sampling in the Moderate Impact sector, while in the Control sector the highest densities were observed during the second summer sampling. Three-way ANOVAs showed significant differences between the beach levels, periods, sectors, and their interactions, for both species (Table I).

Of the total *D. hanleyanus*, 3613 were classified as recruits (90.6%), 371 were classified

Table I. Summary of ANOVAs showing possible spatio-temporal significant differences for the densities of *D. hanleyanus*, *A. mactroides* and for the averages of grain size.

Coluna1	Df	Sum Sq	Mean Sq	F value	Pr(>F)	
Donax hanleyanus						
Period	5	676.9	135.37	52.910	< 2e-16	***
Sector	2	170.5	85.24	33.317	4.90e-13	***
Beach Level	1	91.7	91.67	35.828	1.14e-08	***
Period:Sector	10	311.6	31.16	12.180	5.45e-16	***
Period:Beach Level	5	98.5	19.71	7.702	1.38e-06	***
Sector:Beach Level	2	54.0	27.01	10.555	4.63e-05	***
Period:Sector:Beach Level	10	208.0	20.80	8.130	8.55e-11	***
Residuals	180	460.5	2.56			
Amarilladesma mactroides						
Period	5	175.4	35.09	10.207	1.26e-08	***
Sector	2	461.5	230.77	67.128	< 2e-16	***
Beach Level	1	245.1	245.12	71.304	9.96e-15	***
Period:Sector	10	144.3	14.43	4.197	3.05e-05	***
Period:Beach Level	5	112.6	22.51	6.549	1.27e-05	***
Sector:Beach Level	2	27.1	13.54	3.938	0.0212	*
Period:Sector:Beach Level	10	184.4	18.44	5.364	6.31e-07	***
Residuals	180	618.8	3.44			
Sediment						
Period	2	0.639	0.0319	3.681	0.0308	*
Sector	2	0.9216	0.4608	53.118	3.03e-14	***
Period:Sector	4	0.1057	0.0264	3.046	0.0233	*
Residuals	63	0.5465	0.0087			

Signif. codes: 0 '*' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1.**

as juveniles (9.3%), and only 1 organism was classified as adult (0.02%). Concerning *A. mactroides*, 3008 specimens were classified as recruits (83.3%), followed by juveniles (582 organisms, 16.1%) and adults (24 organisms, 0.66%). Regarding the spatiotemporal variability of size classes, it was possible to identify the predominance of recruits of *D. hanleyanus* and *A. macrtoides* in all sectors and periods of the year. The only exceptions occurred during the pre-summer period in the Moderate Impact and Control sectors, where higher percentages of juveniles were observed (Figure 4).

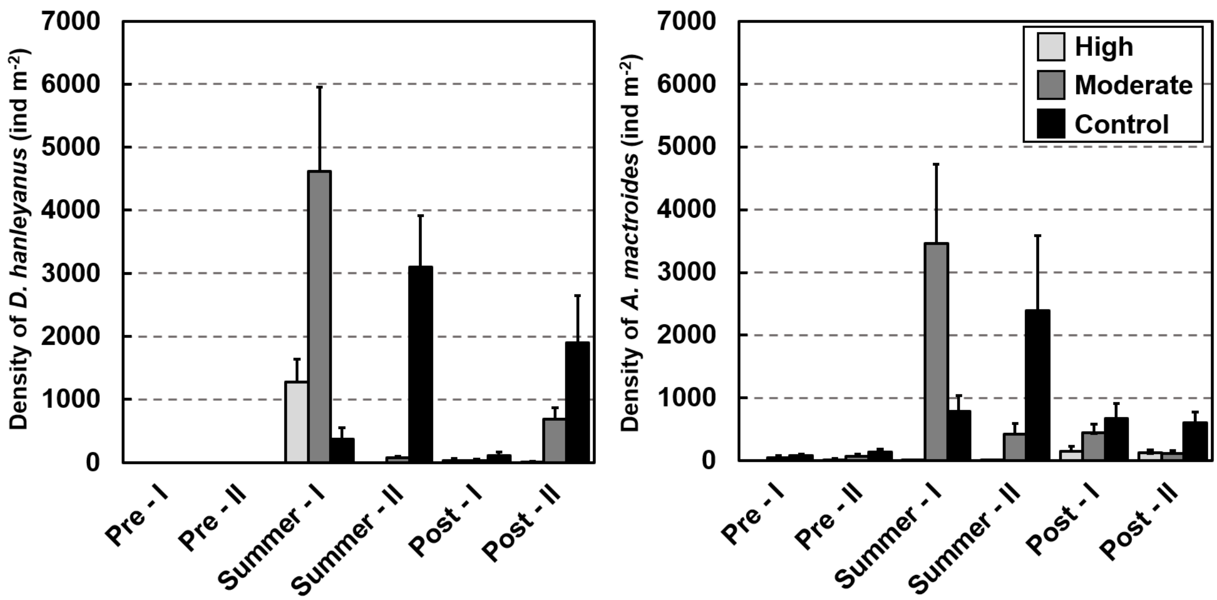

Figure 3. Mean densities + Standard Error of *D. hanleyanus* and *A. mactroides* in the distinct sectors for each sampling period. The color bars represent each sector: High Impact (light gray); Moderate Impact (dark gray); and Control (black).

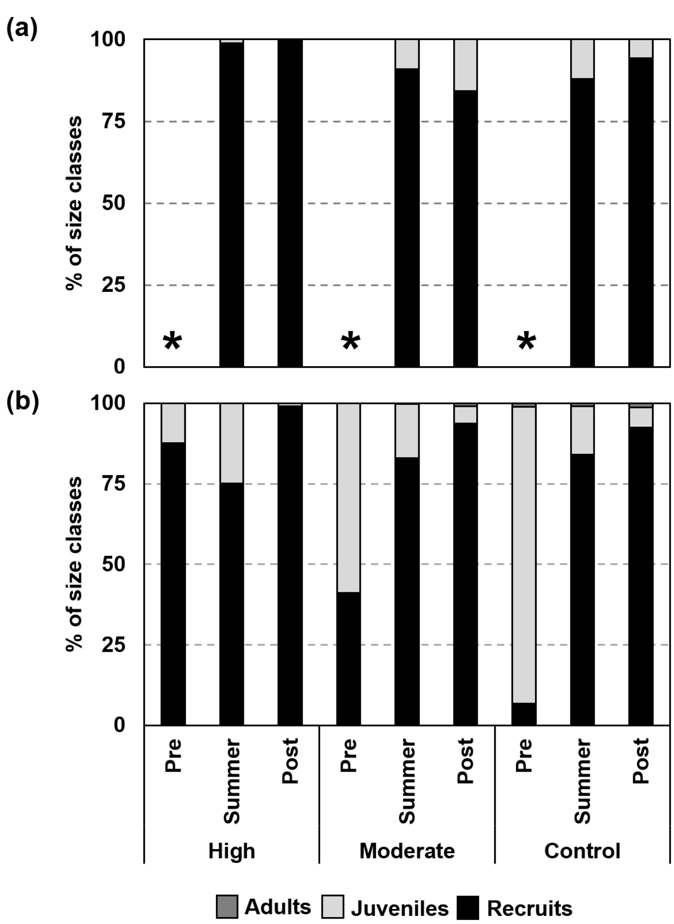

Figure 4. Percentage of each size class of *D. hanleyanus* (a) and *A. mactroides* (b) in the different sampling periods for the High Impact, Moderate Impact and Control sectors. Colors represent each size class: black (recruits); light gray (juveniles); and dark gray (adults). Asterisks represent periods in which the presence of bivalves was not observed.

DISCUSSION

Our results showed spatiotemporal differences in the densities of *D. hanleyanus* and *A. mactroides* with lower densities of both species and high number of samples without organisms in the High Impact sector. In addition, the interaction between the factors used in the ANOVA tests (Period, Sector and Beach Level) was also identified, which demonstrates the dependence between them, mainly due to the higher densities in the summer samplings in the Moderate Impact and Control sectors, especially in the beach level 1. Thus, it suggests that both bivalve species were influenced by the pulse disturbance caused by vehicles, especially in the High Impact sector, where significant traffic occurs and greater sediment compaction was already observed (Bom & Colling 2020).

The grain size, in turn, suggests that this was not a main factor that influenced the biological results in relation to the spatial scale, since the macrofauna densities were generally higher on dissipative beaches, and decreased on intermediate and reflective beaches (McLachlan & Defeo 2013), a pattern opposite to that found in the present study. Despite this, the different granulometric patterns found between the sectors cannot be disregarded and future studies should take them into account. For this, areas of vehicle restriction can be created in the High Impact sector to observe the population behavior of the two species.

Evaluations of other recreational uses on beaches in Brazil and other countries of the world also demonstrated impacts on macrofaunal organisms (Table II). For example, ghost crabs, sandhoppers, cirolanids, polychaetas, nematodes and insects showed lower densities on urbanized beaches (Bessa et al. 2014, Costa & Zalmon 2019, Santos et al. 2021, Vieira et al. 2012). Additionally, the abundance of bivalves decreased on urbanized beaches on the Argentine and Brazilian coastlines (Herrmann et al. 2009, Laitano et al. 2019, Vieira et al. 2012).

There are also studies that have evaluated the possible impact of vehicle traffic on bivalves (Table II). Donacid species have shown tolerance to the passage of vehicles on South African and American beaches (van der Merwe & van der Merwe 1991, Wolcott & Wolcott 1984). These authors suggest that these results were due to low vehicle traffic in the zone where these organisms live. On Cassino Beach, in contrast, intense traffic also occurs in the lower zone of the beach (Bom & Colling 2020), allowing a direct impact on these organisms. Experimental approaches have shown negative effects on bivalves caused by the passage of vehicles, significantly impairing the burial and body condition (Sheppard et al. 2009) or causing mortality of these organisms (Schlacher et al. 2008b).

Regarding the size classes, it was possible to identify a predominance of recruits of both bivalves at all beach levels, periods and sectors. This result can be explained by the spatial segregation by size, with a higher concentration of recruits in the shallow subtidal zone and a predominance of adults in the subtidal zone, as identified for *A. mactroides* (Bergonci & Thomé 2008) and donacid species (Ansell & Lagardère 1980, McLachlan & Hanekom 1979). In addition, the highest densities of recruits occurred during the summer, suggesting recruitment peaks, a similar process observed in Uruguayan and Brazilian beaches for both species (Defeo et al. 1992, Defeo & De Alava 1995, Silva et al. 2008).

These two factors combined show that recruits are possibly the size class most impacted by vehicular traffic, especially in the High Impact sector, where the disturbance was more evident. A similar result was found by Laitano et al. (2019) on the Argentine coast, showing that places of

Table II. **Summary of studies that verified recreational impacts on the benthic fauna of sandy beaches.**

Species/group of organisms	Location	Type of Impact	Main Conclusion	References
Cylindera nivea (Insecta)	Brazil	Proximity to urbanization, beach cleaning, solid waste, vehicle traffic, visitors and buildings on the beachfront	*C. nivea* was negatively related to the urbanization level, and no individuals were found on the high-impacted beaches	Costa & Zalmon (2019)
Meiofauna and free nematodes	Brazil	Trampling and vehicles	Recreational activities reduced the richness and abundance of the community	Santos et al. (2021)
Atlantorchestoidea brasiliensis, Excirolana braziliensis and Ocypode quadrata (Crustacea)	Brazil	Proximity to urbanization, beach cleaning, solid waste, vehicle traffic, visitors and buildings on the beachfront	Abundance of organisms was negatively correlated with solid waste, which is affected by the number of visitors	Suciu et al. (2018)
Macrofauna assemblage	Brazil	Recreational activities	The abundance of some species has decreased due to recreational activities during the summer	Vieira et al. (2012)
Amarilladesma mactroides and *Donax hanleyanus* (Mollusca)	Argentina	Proximity to urbanization, beach cleaning, solid waste, vehicle traffic, visitors and buildings on the beachfront	Urbanization reduced the abundance of *A. mactroides* recruits and juveniles, while the opposite occurred for *D. hanleyanus* juveniles	Laitano et al. (2019)
Donax hanleyanus (Mollusca)	Argentina	Trampling	Human trampling caused a decrease in the abundance of *D. hanleyanus* during the holiday season	Herrmann et al. (2009)
Donax deltoides (Mollusca)	Australia	Vehicles	More than half of the organisms were killed with the increase in vehicular traffic	Schlacher et al. (2008b)
Donax deltoides (Mollusca)	Australia	Vehicles	Vehicle traffic significantly hampered shellfish burrowing performance, intensifying predation and desiccation risks	Sheppard et al. (2009)
Macrofauna assemblage	China	Trampling, camping, sunbathing, sand bathing, and collecting beach fauna	Macrobenthos were seriously disturbed by recreational activities	Wu et al. (2020)
Ocypode quadrata (Crustacea)	Cuba	Proximity to urbanization, beach cleaning, solid waste, vehicle traffic, visitors and buildings on the beachfront	A negative relationship was identified between the densities of *O. quadrata* burrows and the number of visitors, showing significant differences between the beaches analyzed.	Ocaña & de Jesús-Navarrete (2021)

Table II. Continuation.

Macrofauna assemblage	Portugal	Visitors	The increase of visitors in the most urbanized beach impacted the abundance of *Talitrus saltator* and *Tylos europaeus*	Bessa et al. (2014)
Bullia rhodostoma, Donax serra and *Donax sordidus* (Mollusca); *Gastrosaccus psammodytes* and *Tylos capensis* (Crustacea)	South Africa	Vehicles	The species *B. rhodostoma*, *D. serra* and *D. sordidus* showed high tolerance to the passage of vehicles, while the isopod *T. capensis* was highly impacted.	Van der Merwe & Van der Merwe (1991)
Emerita talpoida and *Ocypode quadrata* (Crustacea); *Donax variabilis* (Mollusca)	USA	Vehicles	Vehicle circulation is light and essentially does not occur on the foreshore, not affecting the species analyzed.	Wolcott & Wolcott (1984)
Ocypode quadrata (Crustacea)	USA	Visitors and vehicles	There was a change in density, individual size and excavation behavior due to the greater degree of urbanization	Gül & Griffen (2018)

greater urbanization reduced the densities of recruits and juveniles of *A. mactroides*. However, the variations in size classes and densities in the other sectors (Moderate and Control) may have been caused by natural aspects, such as recruitment peaks and mortality of organisms, as already observed in studies of Cassino Beach (Neves et al. 2007, Silva et al. 2008). Furthermore, the presence of juveniles, even at low densities, in the post-summer period in these sectors may indicate the relative success in the development of these species in less impacted sectors.

The results reported in the present study showed that the two species are impacted by vehicles, especially in the High Impact sector, with low densities in all sampling periods, except for the first summer sampling. In this way, the bivalves *D. hanelyanus* and *A. mactroides* can be considered good bioindicators of the pulse disturbance of vehicular traffic, since they are species with a low locomotion capacity and because they occupy the zone where vehicular traffic is present. In addition, it is essential to expand the temporal component of this research, to verify a possible long-term effect on these organisms, providing a greater understanding of the ecosystems of Cassino Beach, and promoting the management plans of this coastal region.

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