



Article

# Characterization of Intertidal Macrofaunal Communities of Two Sandy Beaches under Different Anthropogenic Pressures

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Abstract: The macrofauna in the intertidal zone of sandy beaches provides the trophic connectivity between land and sea, by linking microbiome, meiofauna, and megafauna, representing a food source for several terrestrial animals, including shorebirds and mammals. However, the macrozoobenthos in urbanised beaches is subjected to intense disturbances, such as breakwater barriers and tourism, which limit or impede the energy transfer from the marine to the terrestrial habitats. Because the information about diversity and abundance of the macrozoobenthos of the intertidal zone on the Mediterranean sandy coasts is scant, the main objective of this study is to increase the knowledge on the macrofauna living in this habitat and to identify taxa sensitive to cumulative human-induced stresses. To achieve this purpose, the structure and dynamics of macrozoobenthic communities from (1) a highly frequented beach characterized by breakwater barriers and (2) a marine protected area (MPA) in the Adriatic Sea were compared. The hypotheses that macrofauna composition and abundance changed in the two sites and over time were tested. Results highlighted that the macrozoobenthos in the MPA is mainly dominated by juvenile bivalves, which peaked from autumn to winter, and to a lesser extent by ostracods and mysids. Conversely, ostracods and the bivalve Lentidium mediterraneum (O. G. Costa, 1830) are particularly abundant in the highly disturbed beach, while the gastropod Tritia neritea (Linnaeus, 1758) increased only during summer. A possible combined effect of breakwater barriers and intense trampling has been theorized to explain the main differences between the two sites especially in the summer.

Keywords: Adriatic Sea; macrozoobenthos; biodiversity; artificial barriers; marine protected areas



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# 1. Introduction

Benthic macrofauna living in soft substrates is a diverse and complex community of organisms, playing a pivotal role in the mixing, ventilation, oxygenation, and irrigation of sediments (phenomena commonly known as *bioturbation*) [1–4]. The bioturbation activity improves nutrients cycling [1,5], substrate permeability [6], redistribution of food resources [4], buffering against nutrient enrichment [7], and benthic–pelagic coupling [8–10]. The chemical reactions in general (e.g., redox) are positively influenced [1] and the depth of the oxic layer is extended over the anoxic one [1,11].

All these benefits can contribute to the increase or the maintenance of alpha and beta biodiversity [12] as well as influence the complex environment that these organisms inhabit [1]. Macrofauna can indeed act as an "ecosystem engineer" [4,13–15] that creates, modifies, and maintains the habitat and its complexity, being able to model the sediment structure by constructing tubes, or by digging channels and burrows [16].

The macrofauna of sandy coastal habitats is a key ring in the trophic chain, connecting microbiome, meiofauna and megafauna. Specifically, macrofauna in the intertidal zone represents a food source for several terrestrial species, including mammals [17] and shorebirds [18]. For example, the Kentish plover, *Charadrius alexandrinus* Linnaeus, 1758, feeding on small crustaceans, molluscs, and polychaetes [19,20], is a species included in the Annex I of the EU Birds Directive, the Annex II of the Bern Convention and considered as Threatened Species, LC (Least Concern), in the IUCN Red List.

The community structure of macro-, meio-, and microfauna hosted in beach environments is influenced by several natural physical and biological factors, acting synergistically or independently, such as sand granulometry, mineralogy, tides, beach exposure [21–25]; nutrients and food supply [26], organic matter content [27], hypoxia [28], local hydrodynamic conditions [29], sediments texture and heterogeneity [30]; competition for limited resources, predation and physical disturbance [31]. Thus, the alteration of natural beach dynamics due to human impacts, such as trampling, bait collection, and mechanical beach cleaning [32–34], could potentially affect the ecological traits of these organisms and the overall functioning of the beach ecosystem [35,36].

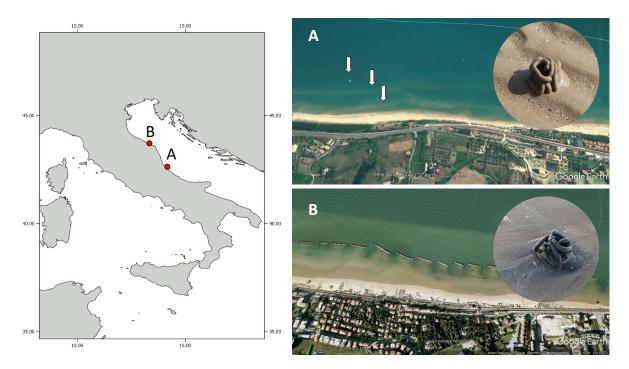
The ecological status of submerged sandy beaches can be assessed by analyzing the composition and abundance of benthic macrofauna as macrofauna living in the intertidal zone can be particularly vulnerable to beach activities and disturbances [37–40]. Even if the data on specific impacts influencing macrofauna are limited, the number of studies regarding the response of macrofaunal communities and populations towards physical disturbances has increased in recent decades [41–49]. Some of the studies are based on single indicator species, while others are focused on the overall macrofaunal community. Especially the organisms living buried in the sand, such as polychaetes, molluscs and crustaceans that are directly damaged by beach activities [39]. However, different taxa have different levels of sensitivity to disturbance, with certain species having been reported to be more vulnerable than others [42].

The main objective of this study is to define the structure and the dynamics of macro-zoobenthic communities of two sandy beaches characterized by different anthropic pressures. The hypotheses that there are no differences in macrofauna composition and abundance (1) among considered sites and (2) among different periods of the year (quarters, see ahead) were tested. The obtained results will help in characterizing the macrofaunal elements of the mentioned sites that could be used to set future assessments of the beach health status.

# 2. Materials and Methods

# 2.1. Study Areas

The two selected sites are Torre del Cerrano (thereinafter: Torre Cerrano, 42°35′5″ N, 14°5′26″ E) and Palombina (43°36′59″ N, 13°25′46″ E), both along the North-Central Adriatic Sea (Figure 1). Torre Cerrano is a small Marine Protected Area (Silvi-Pineto, Italy). divided into three different zones with increasing degree of protection (D, C, B) while no reserve zone A is present. The sampling area is in the B zone, whose backshore is characterized by the presence of scarcely developed sand dunes, and absence of beach facilities; however, the site is frequented from May to September by people walking along the beach or bathers. Vegetal detritus of marine and land origin (mainly brought by nearby rivers and sea current) is often observed along the emerged beach, while the submerged beach is characterized by nearshore sandbars [50].



**Figure 1.** Study area in the north-central Adriatic Sea (Italian coast). **(A)**. Torre Cerrano MPA. **(B)**. Palombina. Arrows indicate sandbars; the insets show casts of burrowers containing oxic and anoxic sediments in Torre Cerrano and Palombina, respectively. Background photos from Google Earth.

Palombina (Ancona, Italy, Figure 1B) is characterized by many touristic facilities, while its backshore is neighbored by a railway track. Originally, it was a dissipative beach [21], but breakwater barriers placed in the 1970s at about 150 m from the coast modified the waves' interaction with the shoreline [51]. Here, sediments are anoxic 4 cm  $\pm$  2 cm SD below the surface (Figure 1, insets). Most mechanical beach cleaning activities occur at the beginning of spring.

Local environmental agencies (EAs) classified the ecological status of the stretch of coast including Palombina as 'Sufficient' [52], and that of Torre Cerrano as 'Good' [53], while the quality of bathing waters was labelled as 'Excellent' at both sites [54].

# 2.2. Sampling Design and Fieldwork

Monthly samplings were carried out at low tide in Palombina and Torre Cerrano (Figure 1) from February to December 2019. The distance of sampling points from the shore was around 40 m, where the depth was about 50–70 cm. Each month, 15 samples of sediments were randomly collected at daytime along a 150 m transect parallel to the coast. Sediment samples were collected with PVC corer ( $\varnothing$ : 9.5 cm, h: 70 cm) equipped with a handle; the height of the sediment core was 15 cm. Therefore, the volume of each core was  $4.25 \times 10^{-3}$  m³. Once the samples of sediments were collected from the intertidal zone, they were gently sieved using a 500  $\mu$ m mesh for macrofauna. The collected sediment samples containing macrofaunal components were preserved in 95% alcohol and transported carefully to the laboratory for sorting.

The sieved sediments were sorted to obtain the living component, i.e., macrofaunal organisms alive at time of samplings, which were grouped in 13 main taxa (Gastropoda, Bivalves  $\leq 5$  mm, Bivalves > 5 mm, Lentidium mediterraneum (O. G. Costa, 1830), Polychaeta, Ostracoda, Mysida, Tanaidacea, Amphipoda, Isopoda, Cumacea, Decapoda, Ophiuroidea, Table 1) for data analysis.

**Table 1.** List of living-component items detected in the samples; PAL = Palombina; TC = Torre Cerrano. The last column indicates main taxa considered for data elaboration.

Phylum	Class	Superorder	Order	Family	Taxon	PAL	TC	Analysed as
Annelida	Polychaeta	-	-	Capitellidae	Capitella capitata (Fabricius, 1780)	Х		Polychaeta
Annelida	Polychaeta	-	-	Magelonidae	Magelona sp.		X	Polychaeta
Annelida	Polychaeta	-	Phyllodocida	Glyceridae	Glycera sp.	X	X	Polychaeta
Annelida	Polychaeta	-	Phyllodocida	Phyllodocidae	Eteone sp.	X		Polychaeta
Annelida	Polychaeta	-	Phyllodocida	Sigalionidae	Sigalion mathildae Audouin & Milne Edwards, 1832	X	X	Polychaeta
Annelida	Polychaeta	-	Spionida	Spionidae	Scolelepis sp.	X	X	Polychaeta
Annelida	Polychaeta	-	Spionida	Spionidae	Spionidae	X	X	Polychaeta
Annelida	Polychaeta	-	Terebellida	Ĉirratulidae	Cirratulidae	X		Polychaeta
Annelida	Polychaeta	-	Terebellida	Terebellidae	Terebellidae		X	Polychaeta
Arthropoda	Malacostraca	Eucarida	Decapoda	Callianassidae	Gilvossius candidus (Olivi, 1792)	X		Decapoda
Arthropoda	Malacostraca	Eucarida	Decapoda	Diogenidae	Diogenes pugilator (Roux, 1829)	X		Decapoda
Arthropoda	Malacostraca	Peracarida	Amphipoda	Bathyporeiidae	Bathyporeia guilliamsoniana (Spence Bate, 1857)	X	X	Amphipoda
Arthropoda	Malacostraca	Peracarida	Amphipoda	Caprellidae	Caprellidae	X		Amphipoda
Arthropoda	Malacostraca	Peracarida	Amphipoda	Gammaridae	Echinogammarus stocki G. Karaman, 1970	X	X	Amphipoda
Arthropoda	Malacostraca	Peracarida	Amphipoda	Oedicerotidae	Pontocrates altamarinus (Spence Bate & Westwood, 1862)	X	X	Amphipoda
Arthropoda	Malacostraca	Peracarida	Cumacea	Bodotriidae	Cumopsis sp.	X	X	Cumacea
Arthropoda	Malacostraca	Peracarida	Cumacea	Pseudocumatidae	Pseudocuma sp.	X	X	Cumacea
Arthropoda	Malacostraca	Peracarida	Isopoda	Cirolanidae	Eurydice spinigera Hansen, 1890	X	X	Isopoda
Arthropoda	Malacostraca	Peracarida	Isopoda	Idoteidae	Idotea pelagica Leach, 1816	X	X	Isopoda
Arthropoda	Malacostraca	Peracarida	Mysida	Mysidae	Paramysis (Longidentia) helleri (G.O. Sars, 1877)	X	X	Mysida
Arthropoda	Malacostraca	Peracarida	Tanaidacea	Apseudidae	Apseudopsis latreillii (Milne Edwards, 1828)	X	X	Tanaidacea
Arthropoda	Ostracoda	-	-	-	Ostracoda	X	X	Ostracoda
Echinodermata	Ophiuroidea	-	-	-	Ophiuroidea	X		Ophiuroidea
Echinodermata	Ophiuroidea	Ophintegrida	Ophiurida	Amphiuridae	Amphiura chiajei Forbes, 1843	X		Ophiuroidea
Mollusca	Bivalvia	-	-	-	Bivalvia ≤ 5 mm	X	X	Bivalvia $\leq 5$ mm
Mollusca	Bivalvia	Imparidentia	Cardiida	Donacidae	Donax trunculus Linnaeus, 1758	X	X	Bivalvia > 5 mm
Mollusca	Bivalvia	Imparidentia	Myida	Corbulidae	Lentidium mediterraneum (O. G. Costa, 1830)	X	Χ	Lentidium
Mollusca	Bivalvia	Imparidentia	Venerida	Veneridae	Chamelea gallina (Linnaeus, 1758)	X	X	Bivalvia > 5 mm
Mollusca	Gastropoda	-	Littorinimorpha	Naticidae	Neverita josephinia Risso, 1826		X	Gastropoda
Mollusca	Gastropoda	-	Neogastropoda	Nassariidae	Tritia neritea (Linnaeus, 1758)	X		Gastropoda

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In order to verify that the composition of the biogenic sand reflected that of the living component, the non-living fraction of the sediments (i.e., skeletal fragments of benthic organisms and vegetal detritus, Table 2) was sorted. Litter of anthropogenic origin was also quantified.

Table 2. Categories of non-living components.

Items	Description				
Polychaetes soft tubes	Empty tubes made of cemented sand grains belonging to polychaetes				
Hard tubes	Carbonatic tubes of serpulids and tusk shells (Scaphopoda)				
Tube fragments	Other fragments of tubes of unidentified organisms				
Ophiuroid vertebrae	Ophiuroid skeletal elements (complete vertebrae)				
Sea urchin fragments	Fragments of tests > 5 mm				
Sea urchin spines	Only frgments including the basal part by which the spine is attached to the test.				
Gastropod shells	Entire, empty shells				
Anthropogenic litter	Every kind of rubbish				
Vegetal detritus	Vegetal detritus both of marine and land origin (Wood, seeds, plant and algal fragments > 5 mm)				

The macrofauna was identified to the lowest possible taxonomic level using a stere-omicroscope, therefore, the living and non-living items were counted to determine their abundance (n. of items  $m^{-3}$ ) in the examined volume of sediment. Successively, data from each site were averaged per quarter (Q1–Q4, Q1 = January, February, March; Q2 = April; May, June; Q3 = July August, September; Q4 = October, November, December), corresponding to the Italian winter, spring, summer, and autumn seasons. A dataset is available as supplementary material.

Additional sediment samples (3 per quarter) were collected with the same method described above to evaluate the granulometry. The sediment samples were dried in an oven and granulometric analysis was performed using a sieve shaker. The limits of the different granulometric classes have been fixed following the Udden-Wentworth scale [55].

Daily maximal wave heights (m) and sea temperature ( $^{\circ}$ C) from the Adriatic station nearest to the considered localities (i.e., Ancona), were retrieved from the National tide gauge network [56–58] (Creative Commons Attribution 4.0 International License) for the period 1 January 2019–31 December 2019. The frequency of calm/rough sea conditions was calculated for each quarter.

#### 2.3. Data Elaboration

The PRIMER (Plymouth Routines in Multivariate Ecological Research) version 7 package [59] and the Paleontological Statistics software [60] were used to analyse the collected data.

Two-way SIMPER analysis was used to identify taxa that mostly contributed to the similarity in the abundance among factors 'Sites' ((PAL = Palombina, TC = Torre Cerrano)) and 'Quarters' (Q1–Q4, Q1 = January, February, March; Q2 = April, May, June; Q3 = July August, September; Q4 = October, November, December).

Data on macrofauna from each site were processed by the function DIVERSE of PRIMER to measure diversity (number of taxa (S), total number of individuals (N), species richness (Margalef, d), Pielou's evenness (J'), and Shannon (H'  $\log_e$ )) indexes.

The hypothesis that the indexes' means would not change in space and time was tested by two-way ANOVA on normalised data across factors 'Sites' (PAL, TC) and 'Quarters' (Q1–Q4).

Non-parametric multivariate techniques were applied to abundance data of the living and the non-living components. Ranked lower triangular similarity matrices were constructed using the Bray–Curtis similarity measure on square root transformed, standardized (%) abundance data. The analysis of similarity (two-way ANOSIM) permutation test was

used to check the significance of differences between groups [61,62]. Bootstrap averages were computed as well to display clustering patterns among 'Quarters' in each site.

### 3. Results

## 3.1. Temporal Variations in Abundance of Living and Non-Living Components

A total of 28 and 21 taxa were found in Palombina and Torre Cerrano respectively with a few exclusive for each site (Table 1). At both sites and in all quarters the taxon bivalves  $\leq 5$  mm' dominated the assemblage contributing from 75% to 96% of the similarity (Figures 2 and S1 and Table S1). Taxa mostly contributing to dissimilarity among sites across quarters are ostracods (27.9%), Lentidium mediterraneum (17.22%), and bivalves  $\leq 5$  mm (12.6%), while those accounting for dissimilarity among quarters across sites are ostracods, Lentidium mediterraneum, Paramysis helleri, Scolelepis sp., Pseudocuma sp., Bathyporeia guilliamsoniana, and Tritia neritea (Table S1).

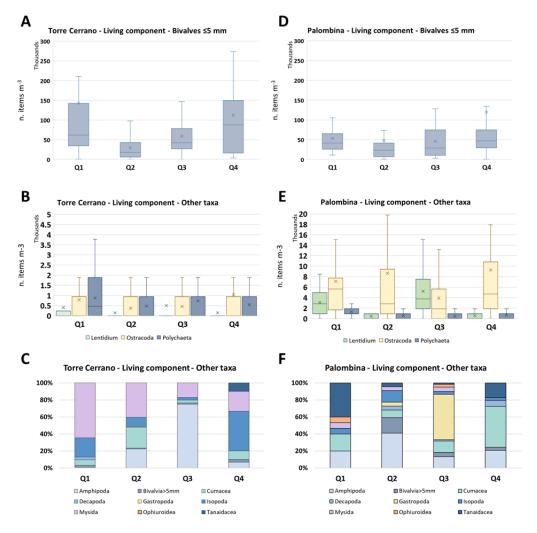


Figure 2. Temporal variations in abundance (n. items  $m^{-3}$ ) of the living component (macrofaunal taxa) in the sediments collected in Torre Cerrano MPA (A–C) and Palombina (D–F). Only Bivalves  $\leq 5$  mm were showed in (A,D), because their high abundance values would have overwhelmed the other taxa. (B,E) Abundance of ostracods, *Lentidium mediterraneum*, and polychaetes. (C,F) Percent composition of less represented taxa. Outliers not showed.

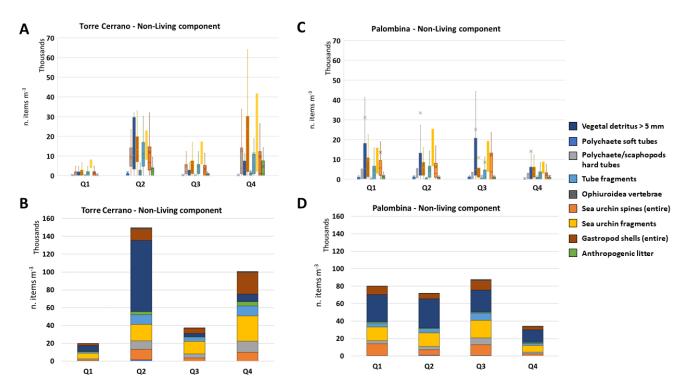
In Torre Cerrano (Figure 2A), bivalves  $\leq$  5 mm peaked in Q4 (about 112  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  111  $\times$  10<sup>3</sup> SD) and Q1 (142  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  205  $\times$  10<sup>3</sup> SD). Other taxa (Figures 2B,C and S2A) are almost 4 times less abundant than in Palombina. Ostracods were present

in all the quarters and more conspicuous in Q4 ( $10^3$  items m $^{-3} \pm 2.2 \times 10^3$  SD), while polychaetes peaked at Q1 (878 items m $^{-3} \pm 10^3$  SD). The mysid *Paramysis helleri* peaked in Q2 (940 items m $^{-3} \pm 2.4 \times 10^3$  SD; 16.7%), while *L. mediterraneum* increased in Q3 (500 items m $^{-3} \pm 3 \times 10^3$  SD). Amphipods decreased in Q4, were absent in Q1, and peaked in Q3, with *Bathyporeia guilliamsoniana* (Spence Bate, 1857) showing an average abundance of 919 items m $^{-3} \pm 1.9 \times 10^3$  SD.

In Palombina, bivalves  $\leq$  5 mm peaked in Q4 (Figure 2D) with values comparable to those in Torre Cerrano (120  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  371  $\times$  10<sup>3</sup> SD), while in Q1 their abundance was 2.6 times lower than in the MPA (54  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  39  $\times$  10<sup>3</sup> SD).

Regarding the other taxa (Figure 2E,F and Figure S2B), *L. mediterraneum* was particularly abundant in the urban beach in all quarters and reached the highest abundance in Q4 (9  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  11  $\times$  10<sup>3</sup> SD) and in Q3 (5.2  $\times$  10<sup>3</sup> items m<sup>-3</sup>  $\pm$  4.7  $\times$  10<sup>3</sup> SD), respectively. The gastropods, represented by *Tritia neritea* (Linnaeus, 1758), peaked in Q3 (334 items m<sup>-3</sup>  $\pm$  10<sup>3</sup> SD), in concomitance to the period with the highest beach frequentation (summer).

The non-living component in Torre Cerrano (Figure 3A,B) throve in Q2, dominated by vegetal detritus and sea urchin fragments, and in Q4, where the most representative items were again the fragments of sea urchins and gastropod shells. In Palombina, abundance values in Q1–Q3 are quite similar. Conversely, the non-living component was minimal in Q4. Vegetal detritus and gastropods dominated the assemblage even in Palombina.

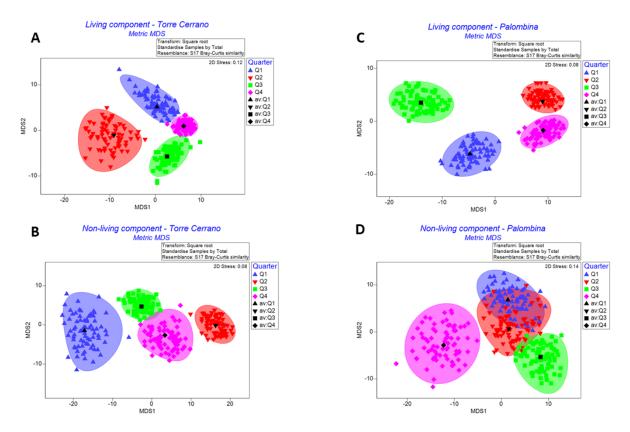


**Figure 3.** Temporal variations in abundance (n. items  $m^{-3}$ ) of the non-living component in the sediments collected in Torre Cerrano MPA (**A**,**B**) and Palombina (**C**,**D**). Outliers not shown.

#### 3.2. Data elaboration

Regarding the living component (macrofauna), the two-way crossed ANOSIM (details in Table S1), testing for differences between unordered 'Quarter' groups across 'Sites', gave a low R value (0.067). This indicates that there are no significant differences among Quarters. A certain significance is found in testing for differences between unordered 'Site' groups (R = 0.428) with a 0.1% significance level of sample statistics. Similarly, there are no significant differences among Quarters (R = 0.064) or Sites (R = 0.123) for the non-living components.

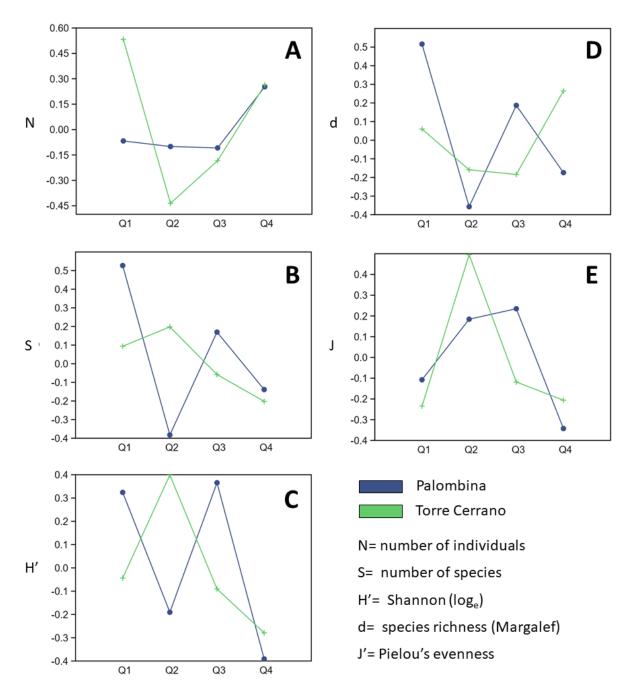
The 2D-metric multidimensional scaling ordination plots based on bootstrap averages of the living (Figure 4A,C) and non-living components (Figure 4B,D) among the quarters (Q1–Q4) display that, for the living component, the Q2 pattern differs from those of the other quarters in Torre Cerrano, while in Palombina Q1 and Q3 are the most diverse. Regarding the non-living component, Q1 and Q2 are clearly discernible in Torre Cerrano as Q4 is in Palombina.



**Figure 4.** 2D-metric multidimensional scaling ordination plots based on bootstrap averages visualise patterns in the living  $(\mathbf{A}, \mathbf{C})$  and non-living components  $(\mathbf{B}, \mathbf{D})$  among the quarters (Q1-Q4) in the sites Palombina and Torre Cerrano). Group averages (black symbols) and approximate 95% confidence intervals (colored ellipses) are provided.

Regarding diversity metrics (Figure 5A–E, Table S2), in Torre Cerrano, the highest diversity (H′ = 0.39) and evenness (J′ = 0.49) occurred in Q2 (April, May, June), when the number of individuals (N) and the species richness had the lowest values. In Palombina, the number of individuals (N) was constant in Q1, Q2, and Q3 and increased in the most uneven quarter, Q4. The number of species, as well as species richness (d = 0.51), peaked in Q1. Diversity (H′ = 0.36) and evenness (J′ = 0.23) were higher in Q3.

Two-way ANOVA (Table S2) highlighted significant differences in the values across 'Quarters' but not across 'Sites' for all the considered indices.

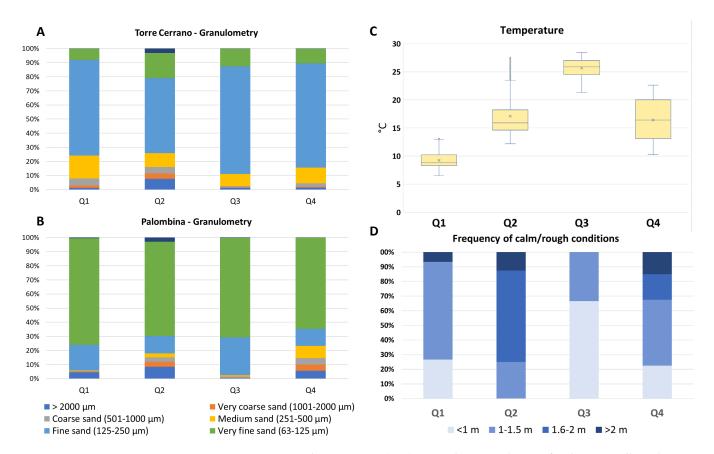


**Figure 5.** Diversity measured in Torre Cerrano and Palombina, PAST elaboration. In Torre Cerrano (green line), the number of individuals (N) (**A**), the number of species (S) (**B**) and species richness (d) (**D**) had the lowest values in Q2 (April, May, June), while diversity (H' = 0.39) (**C**) and evenness (J' = 0.49) (**E**) peaked in the same quarter. In Palombina (blue line), the number of individuals (N) (**A**) was constant in Q1, Q2 and Q3 and increased in the most uneven quarter, the Q4. The number of species (**B**) peaked in Q1, as well as species richness (d = 0.51) (**D**). Diversity (H' = 0.36) (**C**) and evenness (J' = 0.23) (**E**) were higher in Q3.

# 3.3. Environmental Parameters

Concerning the granulometry of the two sites (Figure 6A,B), the most represented fraction in all the quarters in Torre Cerrano is the 'Fine sand (125–250  $\mu$ m)' class (Figure 6A), while Palombina is mainly characterized by the 'Very fine sand (63–125  $\mu$ m)' (Figure 6B). The fraction 'Medium sand (251–500  $\mu$ m)' is more consistent in Torre Cerrano (ranging

from about 8.5% to 16%) than in Palombina (up to 8.6%), above all during Q1. In Palombina, the collected sediments were dark grey at 4 cm below the surface.



**Figure 6.** Environmental parameters. (**A**,**B**). Granulometric classes of sediments collected in Torre Cerrano MPA (**A**) and Palombina (**B**). (**C**). Temporal variations in superficial sea temperature. (**D**). Frequency of calm/rough sea conditions in the Adriatic Sea on the base of maximal wave height (m) recorded for each quarter (data source: [56–58]).

In the considered basin, sea water temperature sharply varies according to seasonal variations [63] with the lowest average value in Q1 (about 9  $^{\circ}$ C) and the highest in Q3 (25.6  $^{\circ}$ C).

Regarding the sea conditions, in 2019, Q3 included the most prolongated period with calm waters (wave height < 1 m, 67% of the events), while rough sea conditions were mainly recorded in Q2 (wave height 1.6-2 m, 62%; wave height > 2 m, 12.5%) and Q4 (wave height 1.6-2 m, 17.5%; wave height > 2 m, 15%). Wave incidence, due to the prevailing winds, Bora and Scirocco, is considered comparable [50].

# 4. Discussion

# 4.1. Overview of the Characteristics of the Two Sites

The composition and the abundance of macrozoobenthos from two sandy beaches along the western coast of the Adriatic Sea were analyzed. The two sampling sites are subjected to comparable morphodynamic conditions [64] and to different levels of anthropic pressure. At both sites, storm events are more frequent in winter (Q1), spring (Q2), and autumn (Q4), while the longest period with calm waters is summer (Q3). Both the macrofauna and the non-living component have a more irregular trend in Torre Cerrano than in Palombina (Figure S1), probably because storms have a stronger impact in the natural site due to the lack of the breakwater barriers. Diversity in Torre Cerrano in Q1 and Q4 is low, while the number of individuals is high, suggesting that in these quarters the assemblage is dominated by the species adapted to withstand rough sea conditions.

In Torre Cerrano MPA, the major (mechanical) anthropic stress for the macrofauna is represented by people walking on the intertidal zone, which may be responsible, at least in part, for the lower diversity in Q3.

Summer trampling in Palombina is much more intense than in Torre Cerrano with the presence of rows of sun umbrellas on the backshore reducing the width of the beach and forcing people to walk very close to the shoreline or into the water. Breakwater barriers in Palombina defuse the wave effects, limit water exchange, and favor fine sand accumulation [65–68], as confirmed by the results of the granulometric analysis. It is also likely that water quality is low in this site because of short but numerous episodes of discharge of untreated wastewater due to overflow following heavy rains [69]. These considerations suggest that species surviving in the intertidal zone in Palombina should be very tolerant to cumulative anthropic stresses.

## 4.2. Living and Non-Living Components in the Collected Sediments

Notwithstanding the strong differences in types and intensity of stresses in the two study sites in the considered periods, the statistical analysis did not highlight significant differences among quarters while differences among sites were slight. However, it was possible to discriminate the two sites by interpreting the temporal variations in abundance and diversity indexes.

In Q3, in concomitance with the peak of presence on the beach, diversity and evenness were higher in Palombina, where the assemblage was dominated by *Lentidium mediterraneum*. This bivalve is one of the most commonly species found in the Northern-Central Adriatic beaches comprising up to 95% of the community [64]. It is likely that it tolerates the disturbance linked to the touristic season (i.e., trampling, sediment resuspension) probably because of its ability to burrow rapidly [70].

High abundance of *Tritia neritea* in Palombina in Q3 is due first to seasonality since they were observed at juvenile stages in early summer and mature towards the end of summer. This gastropod shows a flat, thick shell very resistant to mechanical disturbances (personal observation). Moreover, *T. neritea* is tolerant to sudden changes in temperature, high salinity, and dissolved hydrogen sulphide, and it can avoid the anoxic layer close to the substrate thanks to its ability to extend its inhalant siphon beyond the surface of sediments [71]. The high tolerance of this opportunistic species to extreme environmental conditions explains why this is so common in Palombina at summertime.

Ostracods from Palombina are quite abundant all year round and have a considerable decrease in Q3. Ostracods are largely used as an indicator of anthropic impact and hypoxia [45,72,73]. However, a shift towards opportunistic and more tolerant assemblages has been documented in the North Adriatic Sea over the last 20 years [73]. Even if further investigations are needed to establish whether natural drivers or anthropic pressures caused the decline of these crustaceans during summertime, it is possible that they are mechanically damaged by trampling [74].

At both sites, the non-living component is characterized by exogenous material dominated by vegetal detritus. In Torre Cerrano, the macrofauna diversity increased in Q2 likely as a consequence of the presence of vegetal detritus which shapes biodiversity and initiates trophic chains [75].

In the MPA natural predators in the intertidal zone may contribute to the decline in polychaete's abundance observed in spring (Q2), such as the Kentish plover, whose diet includes polychaetes [19]. It is likely that *Charadrius alexandrinus* Linnaeus, 1758 and its chicks, common in the protected area, feed on macrofauna above all during reproductive and hatching season [76] as confirmed by several shots from amateur photographers (Figure S3). Long-term studies demonstrated that invertebrate prey depletion is one of main factors causing declines in abundance and delays in migration in shorebirds [18], since the birds need to assimilate enough high-quality food to build up food reserves prior to departure.

These considerations highlight the importance of preserving the intertidal zone being the macrofauna diversity positively related to secondary production [77], while allochthonous inputs plays a crucial role in biodiversity maintenance [78]. However, main EU Directives implemented in MPAs [79–81] include monitoring programs for the dune system and the sandbank, while there are no specific protocols to assess the quality status of the intertidal zone, nor plans to improve its conservation.

# 5. Conclusions and Future Perspectives

This work highlights the importance of characterizing macrofauna assemblages at local scale and to identify main human pressures influencing the biodiversity on sandy beaches. The collected data could be used to define protocols to assess human impact, to regulate the access to the intertidal zone, and to establish the socio-ecological carrying capacity of a beach [82].

Assessing multiple stressors is crucial to mitigate the effects of human disturbance on benthic invertebrates [83]. Armoring, dune suppression, trampling, and beach cleaning are among the most destructive impacts on supralittoral invertebrates, such as Ocypodidae, Hippidae (decapods), and Talitridae (amphipods) [41,84]. However, beachgoers also walk in shallow waters, influencing the dynamics of species living in the submerged beach, such as mollusks and polychaetes [85]. Conversely, the lower beach is less studied [63], notwithstanding its relevance for the survival of other species.

In the considered study area, mysids, cumaceans, and likely ostracods could be used as an indicator of trampling, while the bivalve *Lentidium mediterraneum* and the gastropod *Tritia neritea* are more tolerant taxa to both the mechanical impact and the low-quality environmental conditions occurring during summertime in the urbanized site.

Our observations suggest that the influence of breakwaters should be further investigated as they create a sheltered and impoverished habitat where opportunistic species may develop, which may be unsuitable for bathers [86], and where the economic, social and ecological value of beaches are affected [87].

Future studies should focus on the behavior, trophic relationships, and life histories of less abundant taxa, to better understand their functional role in the intertidal communities and their relationship with the non-living component.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jmse10121976/s1, 1. a Word document: Figure S1: Monthly variations in the macrofaunal assemblage (living component); Figure S2: Trimestral variations in the macrofaunal assemblage (living component); Figure S3: Possible relationship between macrofauna and higher trophic levels; Section S1: Results of SIMPER and ANOSIM; Section S2: Diversity Indexes; 2. a spreadsheet containing the data presented in this study.

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