

Diversity and spatial distribution patterns of soft-bottom macrofaunal communities inhabiting two Croatian recreational marinas

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ABSTRACT The effects of human pressure on benthic macrofaunal assemblages inhabiting touristic marinas have seldom been studied, especially in the eastern part of the Adriatic Sea. In July 2019, we investigated the macrofaunal communities in two Croatian marinas (Špinut and Strožanac) to evaluate the diversity and feeding structure influenced by the morphology of the basins and long-lasting anthropogenic activities. In both marinas, macrofaunal densities and species richness were observed to be higher at the main entrances than at the inner sites. In Špinut, a clear confinement gradient pattern was mirrored in the number of species. In Strožanac, the basin is more open to the sea compared to Špinut, thus the macrofaunal community was not directly influenced by a confinement gradient, despite the presence of boat careening and painting activities. In contrast, Špinut was characterised by a community impoverished in species number near the boathouse area. In both marinas, the variation of the feeding structure was likely driven by grain-size. Subsurface deposit feeders characterised the finer sediments whereas suspension feeders dominated at the sandy-bottom stations. Well-balanced feeding guilds characterised the station near the boathouse in Špinut. However, only a few species represented each trophic guild at this site, indicating possible higher community vulnerability.

Key words: feeding habits, β diversity, macrofauna, marina, eastern Adriatic Sea.

1. Introduction

Coastal tourism plays a major role in the economic development of many countries. Its recreational value is estimated to be the highest along the densely populated coastal areas (Ghermandi and Nunes, 2013). However, this increased recreational use of coastal waters has led to greater demands for boat-mooring facilities over the last decades. To meet this demand, the number of marinas has rapidly increased, and concern about their environmental impact is growing (Guerra-García and García-Gómez, 2005; Davenport and Davenport, 2006). Artificial constructions in coastal areas, such as marinas and touristic ports, can cause substantial habitat destruction and local environmental change (Bugnot *et al.*, 2021). The building of such infrastructures requires a considerable amount of modifications to the natural environment (i.e. seabed dredging, land reclamation, installation of seawalls, pilings, and pontoons) that change the physical-chemical characteristics of the nearby and adjacent marine areas. These alterations potentially influence the diversity and distribution of the resident biological communities (Rivero *et al.*, 2013; Cordell *et al.*, 2017). Ports can be the recipient and source of considerable

anthropogenic disturbances, both for marine and adjacent land habitats, since they centralise a range of environmental problems, such as air pollutant emissions, noise, sediment dredging and transport, industrial installations, jetty constructions, wastewater discharges, oil spill accidents, leaks of petroleum derivatives and antifouling coatings, storage and spillage of hazardous materials, as well as the introduction of invasive species (Darbra *et al.*, 2005; Chatzinikolaou *et al.*, 2018).

Furthermore, marina structures (e.g. piles, pontoons and walls) may also alter water circulation and decrease the current flow, thus increasing natural sedimentation rates (Mcgee *et al.*, 1995; Turner *et al.*, 1997). These structures are designed for smaller boats and are frequently semi-enclosed, with their innermost parts experiencing lower water renewal and, thus, anoxia with detrimental effects on the benthic communities (Guerra-García and Garcia-Gómez, 2005).

Benthic macroinvertebrates are sessile, semi-sessile, or confined to restricted bottom areas and spend their complete life cycle, or its greater part, in direct contact with bottom sediments. For all these reasons, they are most traditionally used as biological indicators of ecosystem health in marine environments (Borja *et al.*, 2003). Moreover, macrofaunal invertebrates have different feeding habits [e.g. suspension feeders (SF), deposit feeders (DF), and carnivores (C)], representing the higher trophic levels of the benthic food web (Gray and Elliott, 2009). The structure of benthic communities can, thus, be directly linked to disturbance exposure. Changes in macrofaunal diversity (species composition and feeding structures) can be mirrored in variations of the ecosystem functioning in marinas.

Overall, while little information is available on the diversity and species composition of soft-bottom benthic communities in small marinas (Mcgee *et al.*, 1995; Chatzinikolaou *et al.*, 2018; Dimitriou *et al.*, 2020), the feeding structures of the macrofaunal community inside the marinas are even less explored.

To support small local ports in the design and application of better environmental strategies aimed at a sustainable management of their maritime space, various initiatives have recently been set up among which the international European project ECOsustainable management of MARine and tourist Ports (ECOMAP). The ECOMAP project aims at improving the environmental quality conditions of nautical ports by promoting a coordinated development and implementation of environmentally friendly solutions and the exchange of knowledge and good practices between Italian and Croatian recreational ports. In this context, one of the project goals is to improve the environmental quality conditions of small marinas and touristic ports, influenced by anthropogenic activities. In this study, we investigated the variation in diversity, species composition, and feeding habits of macrofaunal communities inhabiting two Croatian marinas, namely Špinut and Strožanac, characterised by different morphology and anthropogenic impacts. To this end, we considered the macrofaunal α - and β -diversity, with the first referring to the biodiversity and its components (richness, diversity, and evenness) at each site, and the second focusing on two distinct processes: the replacement, and the loss or gain of a subset of species on a spatial scale. This study is one of the outcomes of the ECOMAP project, as it represents a first insight into the actual conditions of the soft-bottom macrozoobenthic community in the Špinut and Strožanac marinas. More precisely, we hypothesised that: i) the morphology of the basin and the long-lasting anthropogenic activities strongly affect the species composition and biodiversity of benthic invertebrates; ii) the environmental features of the marinas shape the feeding structures of the macrofaunal communities.

2. Material and methods

Špinut is a relatively large marina established in 1973, with 12 pontoons that can host approximately 780 boats, up to 25 m in length with a draft of 5 m. This marina is located to the north of the city of Split, under the Marjan Hill and next to its protected wood park. Špinut faces Kaštela Bay, which is characterised by several freshwater springs that influence the seawater temperature and salinity during the winter and spring seasons (Fritz and Bahun, 1997). The Strožanac marina, operational since 1975, hosts 330 small fishing and leisure boats (8 m), mainly owned by local people. The marina is located west of a 500-metre-long gravel beach along the coast (south of Podstrana municipality) and SE of the mouth of a small river named Žrnovnica.

In July 2019, sampling was carried out at five stations in each marina (Fig. 1). The sampling stations were chosen considering the confinement gradient (i.e. the distance from the main port entrance and the time required for the renewal of marine water) and the presence of anthropogenic activities like boat careening and painting (hereafter, named boathouse area) (Figs. 1a and 1b). Depths and geographical coordinates of all stations are presented in Supplementary Table 1.

At each sampling station, salinity and temperature of bottom seawater were registered by a multiparameter probe, YSI ECO2 EXP-7 2014.

Sediments for grain-size, contaminant and macrofauna analyses were sampled using a stainless steel Van Veen grab sampler (0.1 m²). For macrofauna investigation, three replicates per station were taken.

Sediment samples for grain-size analyses were sieved at 2 mm and pre-treated with 10%

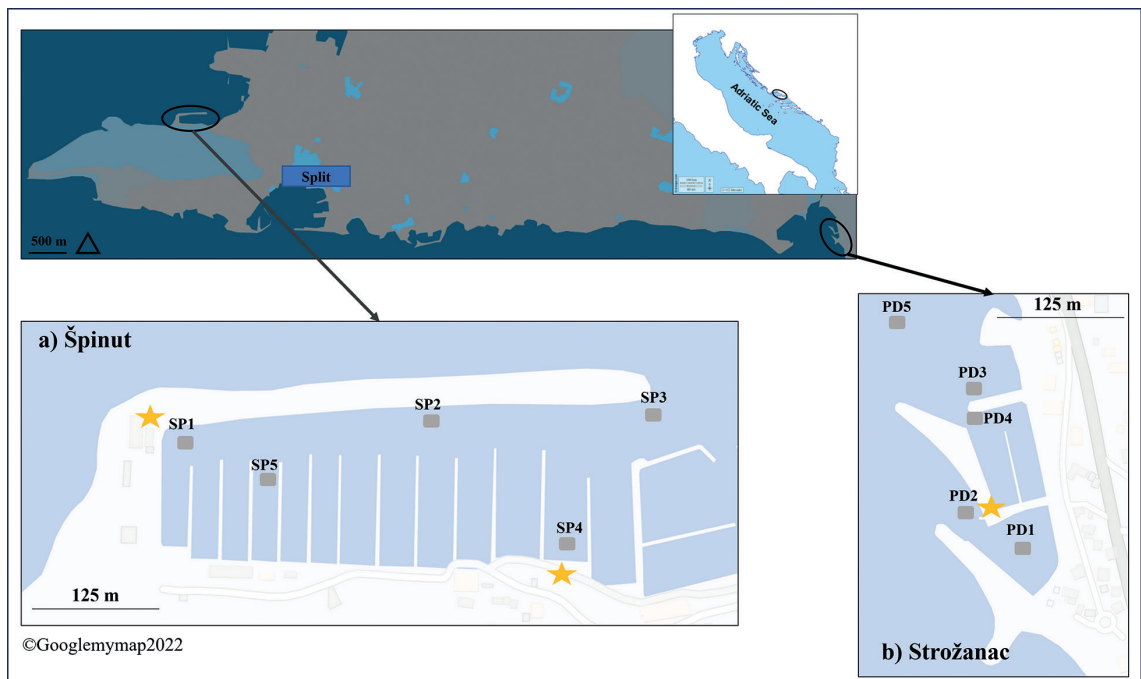


Fig. 1 - Sampling stations within the Špinut (SP) and Strožanac (PD) marinas. Asterisks indicate the stations near anthropogenic activities, such as careening and boat painting.

hydrogen peroxide before being analysed with a BECKMAN COULTER LS 13 320 laser diffraction particle size analyser. Data are expressed as percentages of sand, silt, and clay following the Udden-Wentworth grain-size classification (Wentworth, 1922).

A total of six metals [i.e. cadmium (Cd), chromium (Cr), copper (Cu), nickel (Ni), lead (Pb), and zinc (Zn); Singh and Turner (2009)], strictly linked to careening and boat painting activities, were analysed in the sediments of the marinas using inductively coupled plasma mass spectrometry (ICP-MS). For these analyses, powders (0.15 g) were totally digested with Suprapur® grade HF and HNO₃ (Merck, Darmstadt, Germany) on a hot plate. Dissolved samples were dried out and, then, re-dissolved in ultrapure water produced with a Millipore Milli-Q® Integral 10 water purification system. The analyses were carried out using an X Series Thermo-Scientific spectrometer. Data are expressed as mg/kg dry weight.

The sediments collected for the macrofauna were sieved through a 500 µm mesh to retain the invertebrates and immediately fixed with 70% ethanol for further determination. In the laboratory, organisms were separated from the sediment remains, sorted, and identified to the species level, whenever possible. Species determination was carried out using traditional identification keys listed in Morri *et al.* (2004). Abundance was expressed as individuals per square metre (ind./m²).

To investigate the functional structure of the community, the feeding habits were considered as the functional trait of paramount importance. Six different feeding habits were assigned to all individuals: SF, DF divided into surface deposit feeders (SDF) and subsurface deposit feeders (SSDF), C, omnivores, and herbivores. These functional traits were assigned to each species based on the database by Faulwetter *et al.* (2014) and the literature (Jumars *et al.*, 2015). The values of feeding guild for each sampling station and area are expressed as their relative abundance (RA%).

We assessed the benthic community α -diversity and its components across space, calculating the i) species richness, ii) Pielou evenness (J'), and iii) Shannon–Weaver diversity index - $H' \log_2$. The H' diversity was coupled to the analyses of β -diversity. This diversity partitioning framework (Villéger *et al.*, 2013) was based on the Jaccard dissimilarity index (Baselga *et al.*, 2012). β -diversity equals zero when communities are identical and equals 1 when communities are maximally dissimilar along spatial scales [e.g. no species shared for taxonomic β -diversity; Baselga (2010)]. We determined whether the β -diversity between stations of each marina was mostly due to turnover (i.e. differences in species between stations due to a replacement) or to nestedness resultant processes (i.e. species between two stations representing a subset of those found at the other stations).

Furthermore, multivariate analyses were performed to assess the variation in species composition along spatial scales (one-way PERMANOVA with an unrestricted permutation of raw data and 9999 permutations). The following fixed factors were applied for each test: i) 'marina'; and ii) 'confinement gradient'. The latter factor was separately tested for each marina. In order to visualise any spatial patterns of macrofaunal species composition in the two marinas, a non-metric multidimensional scaling ordination (nMDS) was performed. The vectors of temperature, salinity, grain-size (sand, silt, and clay) and contaminants were overlaid.

The biotic matrix for the multivariate analyses was square-root transformed, and, then, the Bray-Curtis similarity was applied. Furthermore, to highlight differences between the two marinas, the macrofaunal biodiversity and relative abundance of feeding habits indices were tested by the Mann-Whitney U test, applying the 'marina' factor. This analysis was carried out using STATISTICA 7 software.

The univariate (i.e. H' and J') and multivariate analyses (PERMANOVA and nMDS) were performed using PRIMER 7 (PRIMER-E Ltd. Plymouth, UK) (Clarke *et al.*, 2014).

3. Results

The physical features of the water column (temperature and salinity) at the moment of sampling are reported in Supplementary Table 1.

The Špinut marina was characterised by muddy sediments in the inner part, and a higher sand percentage at the port entrance (SP3). A similar grain-size distribution pattern was noticed in Strožanac, where the highest percentage of sand was observed at the main entrance (PD2, Supplementary Table 2). Regarding the analysed metals (Supplementary Table 2), the Špinut marina was more contaminated compared to Strožanac, especially at its boathouse station where the concentrations of Cu, Pb, and Zn were up to two orders of magnitude higher than those at the other sampling sites.

In the Špinut marina, macrofaunal abundances varied from 76.6 ± 124.2 ind./m² (SP4) to 2760.0 ± 1459.3 (SP2) ind./m². Higher densities were noticed at stations near the main entrance (SP3 and SP2), whereas lower values were observed in the inner sites (SP1 and SP5, Fig. 2a). In the Strožanac marina, the lowest macrofaunal abundance was observed at PD3 (146.6 ± 136.1 ind./m²), whereas the highest value was recorded at PD2 (2573.3 ± 830.3 ind./m², Fig. 2c). Polychaetes generally dominated in Špinut, with relative abundances (RA) ranging from 61.3% at SP2 to 93.5% at SP1, except for SP3 and SP4, where molluscs accounted for the highest percentages (54.5% and 82.6%, respectively, Fig. 2b). A very similar pattern was observed in Strožanac, where polychaetes dominated at most stations, varying from 82.0% (PD5) to 85.5% (PD2 and PD4), except for PD1 and PD3 where molluscs represented 78.1% and 68.9% of the total macrofaunal community, respectively (Fig. 2d).

Overall, 223 species were identified in both marinas. In the Špinut marina, the highest species number ($S = 85$) was obtained at the station near the main entrance (SP3), whereas decreasing numbers were noticed inside the marina (Table 1). The high species richness at SP3 was mirrored in the H' (Shannon-Weaver diversity) index ($H' = 4.75$). Although only 9 species were observed at SP4, the lowest value was obtained at SP1 ($H' = 2.12$; Table 1). This was probably due to the highest dominance of a single species, i.e. the polychaete *Pseudoleiocapitella fauveli* (RA = 65.0%). This was mirrored in the lowest value of J' observed at this station ($J' = 0.41$). Similarly, in Strožanac, the highest species number ($S = 79$) was observed at the station near the main entrance (PD2; Table 1). Although only 13 species were observed at PD3, the lowest values of H' and J' were obtained at PD1 ($H' = 3.00$ and $J' = 0.59$). This was due to the dominance of a single species, e.g. the bivalve *Loripes orbiculatus* (RA = 43.3%). The Mann-Whitney test did not highlight significant differences either in macrofaunal densities ($z = 0.52$; $p = 0.60$) or diversity values between the two marinas ($z = -0.94$; $p = 0.34$).

The spatial β -diversity analysis highlighted that a great variation in the assemblage composition was observed along the confinement gradient from the main entrance toward the inner stations of Špinut (Table 2). Despite the major contribution of turnover to the total β -diversity, the nestedness-resultant components between the stations were quite higher. In addition, the community in front of the boathouse was composed of species subsets also observed at the station near the entrance (nestedness-resultant SP3 compared to SP4: 0.56). On the contrary, the β -diversity analysed in Strožanac was characterised by lower nestedness-resultant components and was mainly ascribable to the turnover. The highest values of turnover were noticed between PD1 and PD5 (0.86; Table 2).

Both marinas were characterised by the same dominant species, as the polychaete *P. fauveli* and the molluscs *Abra alba* and *L. orbiculatus* (Table 3). Indeed, no significant variation in species composition was observed between Špinut and Strožanac (PERMANOVA test: pseudo $F = 1.50$;

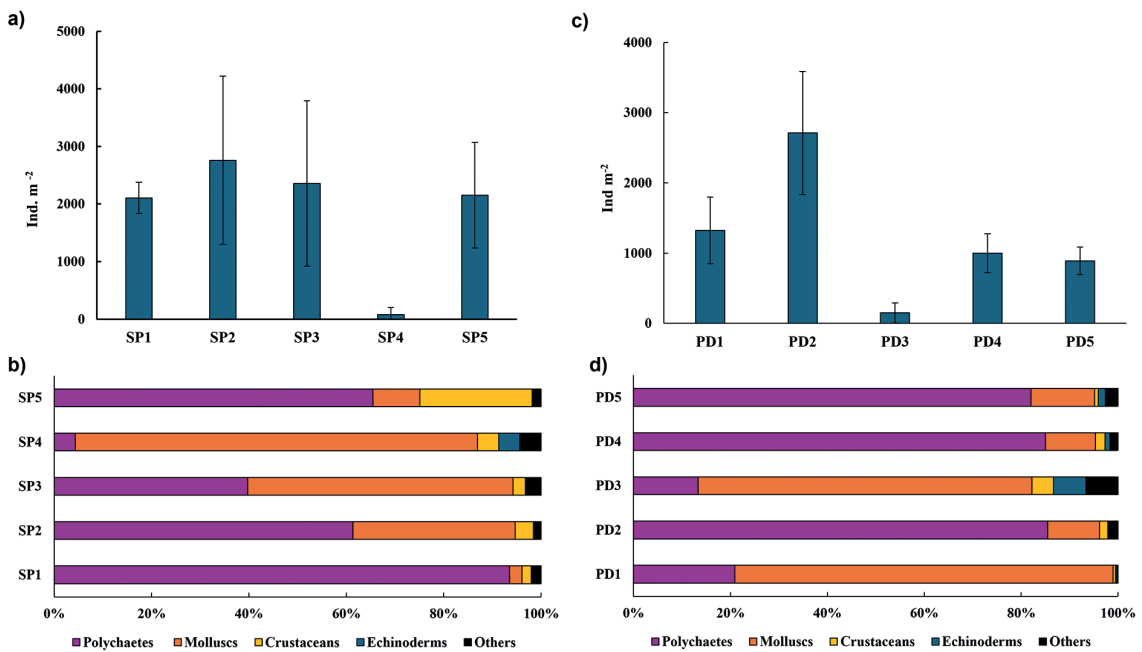


Fig. 2 - Total community abundance (ind./m²) and relative abundance (%) of the main macrofaunal taxa at the five sampling stations in the Špinut (a, b) and Strožanac (c, d) marinas. Others represent the sum of Anthozoa, Nemertea, Phoronida, and Sipuncula observed at each sampling site.

Table 1 - Number of species (S), Margalef richness index (d), Pielou evenness index (J'), and Shannon-Weaver biodiversity index - H' (Log2) at the five sampling stations in each marina.

Marina	Station	S	d	J'	H'
Špinut	SP1	37	6.73	0.42	2.19
	SP2	36	7.49	0.75	3.87
	SP3	90	16.30	0.75	4.87
	SP4	10	4.42	0.84	2.80
	SP5	45	8.19	0.57	3.11
Strožanac	PD1	37	7.37	0.59	3.07
	PD2	83	14.69	0.73	4.68
	PD3	16	5.54	0.88	3.50
	PD4	70	14.92	0.68	4.18
	PD5	53	11.58	0.84	4.81

$p = 0.08$). Moreover, in the Špinut marina, an alien species, the polychaete *Neopseudocapitella brasiliensis*, was identified at SP2 and SP3, with low abundance values of 6.6 ± 11.5 and 20.0 ± 17.3 ind./m², respectively.

The nMDS analysis (Fig. 3) gathered the stations placed near the open part of the marinas (i.e. SP3, PD2, PD4, and PD5) on the left side of the plot, along with the vectors of salinity and sand. Instead, the inner stations of Strožanac were plotted on the lower and right side

Table 2 - Taxonomic β -diversity comparisons between stations in each marina. Values show β -diversity, its two components (turnover and nestedness-resultant), and relative contribution of turnover to β -diversity (%). The asterisks indicate the stations near anthropogenic activities.

Špinut					Strožanac				
<i>Turnover</i>	SP1	SP2	SP3	SP4*	<i>Turnover</i>	PD1	PD2*	PD3	PD4
SP2	0.48				PD2*	0.71			
SP3	0.60	0.62			PD3	0.56	0.70		
SP4*	0.62	0.36	0.36		PD4	0.80	0.71	0.70	
SP5	0.68	0.26	0.56	0.71	PD5	0.86	0.36	0.82	0.70
<i>Nestedness-resultant</i>	SP1	SP2	SP3	SP4*	<i>Nestedness-resultant</i>	PD1	PD2*	PD3	PD4
SP2	0.25				PD2*	0.14			
SP3	0.20	0.02			PD3	0.23	0.23		
SP4*	0.26	0.55	0.56		PD4	0.08	0.03	0.22	
SP5	0.03	0.33	0.19	0.20	PD5	0.03	0.21	0.11	0.05
<i>β-diversity</i>	SP1	SP2	SP3	SP4*	<i>β-diversity</i>	PD1	PD2*	PD3	PD4
SP2	0.73				PD2*	0.85			
SP3	0.80	0.64			PD3	0.79	0.93		
SP4*	0.87	0.91	0.92		PD4	0.88	0.74	0.92	
SP5	0.71	0.58	0.75	0.91	PD5	0.89	0.57	0.93	0.75
<i>Turnover/ β-diversity (%)</i>	SP1	SP2	SP3	SP4*	<i>Turnover/ β-diversity (%)</i>	PD1	PD2*	PD3	PD4
SP2	65.48				PD2*	83.50			
SP3	75.00	96.54			PD3	70.37	75.25		
SP4*	70.59	39.85	39.55		PD4	91.43	95.50	76.27	
SP5	95.42	43.77	74.61	78.23	PD5	96.50	63.65	87.77	93.17

of the plot, near the silt, clay, temperature, and Ni and Cr vectors. In addition, the station near the boathouse (SP4) in the Špinut marina was plotted separately from the others in the nMDS, driven by the higher Pb, Zn, Cd, and Cu concentrations in the sediments. The feeding habits taken into account in this study confirmed the nMDS outputs. The variation of feeding guilds could be directly linked to the position of stations inside the basins (Fig. 4). In the Špinut marina, SSDF dominated the community at the inner stations (i.e. SP1 and SP2), with RA values ranging from 82.7% to 84.5%, respectively. SDF reached high values at the outer ones (i.e. SP3 and SP4) with a RA of 49.1% and 45.5%, respectively, followed by C (24.5% at SP3 and 27.3% at SP4). An intermediate situation was displayed at SP5, with a high percentage of both SSDF (60.4%) and SDF (27.8%). As for Špinut, the position of the stations inside the Strožanac marina reflected a clear difference in the macrofaunal feeding structure. The inner stations (PD1 and PD3) were dominated by SF (50.4% and 26.2%, respectively) and SDF, with RA values ranging from 24.7% (PD1) to 33.3% (PD3). The outer stations (PD2, PD4 and PD5) showed a community dominated by SSDF (29.3%, 58.5%, and 44.5%, respectively), followed by SDF (38.4%, 15.3%, 21.3%, respectively).

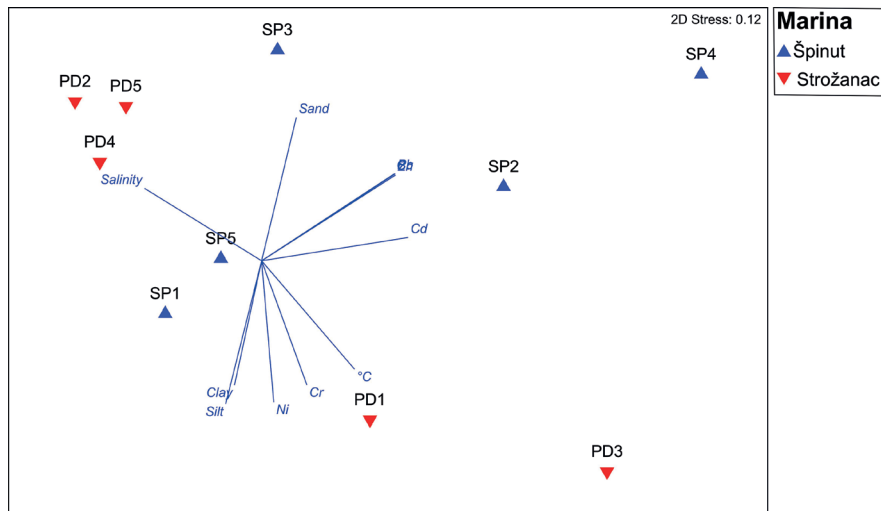


Fig. 3 - Non-metric multidimensional scaling (nMDS) ordination plot of the macrofaunal community in the study areas. The vectors of salinity, temperature, sand, silt, clay percentages, and heavy metals [cadmium (Cd), chromium (Cr), copper (Cu), nickel (Ni), lead (Pb), and zinc (Zn)] are overlaid. Zn, Pb and Cu are overlapped in the plot.

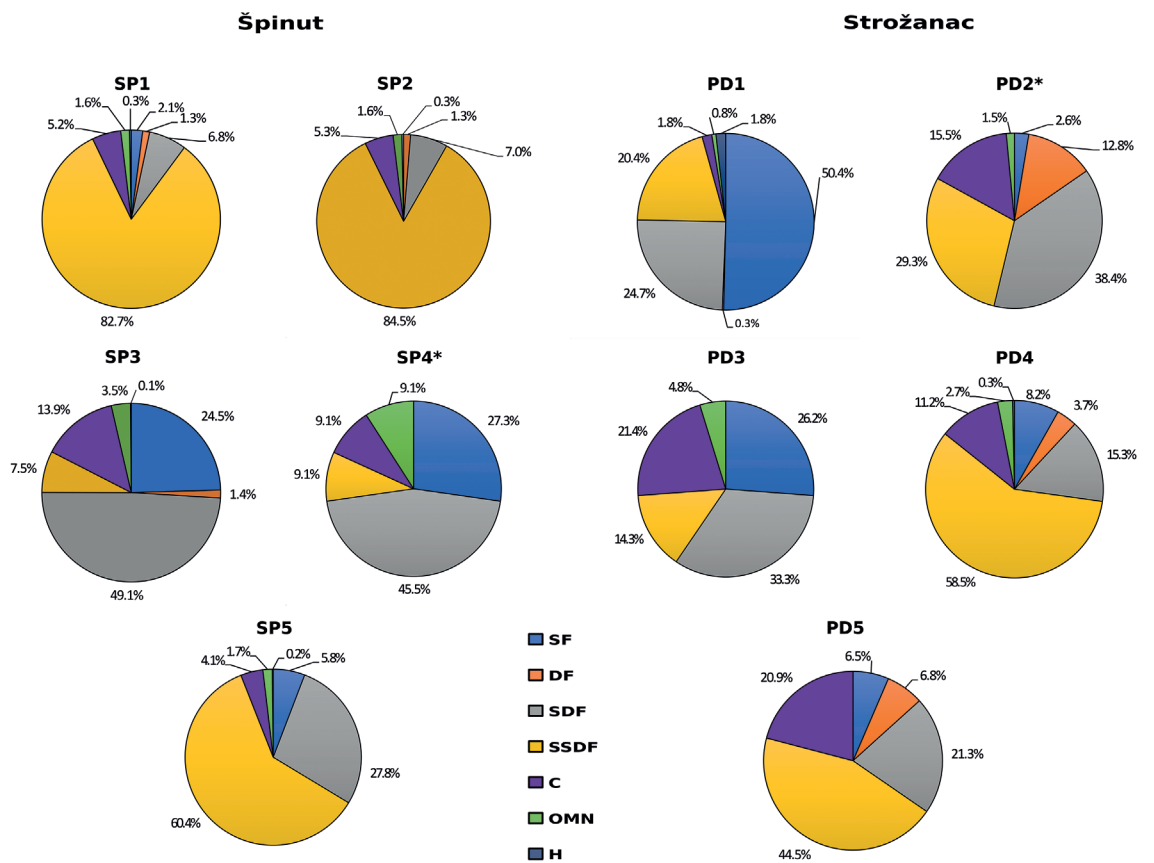


Fig. 4 - Relative abundance, expressed as a percentage, of the macrofauna feeding habits at the sampling sites. The asterisks indicate stations near anthropogenic activities. SF: suspension feeders; DF: deposit feeders; SDF: surface deposit feeder; SSDF: subsurface deposit feeders; C: carnivores; OMN: omnivores; H: herbivores.

Table 3 - Relative abundance, expressed as a percentage, of macrofaunal taxa in the Špinut and Strožanac marinas (cut-off < 0.3%).

Špinut	SP1	SP2	SP3	SP4	SP5
<i>Abra alba</i>	0.00	11.28	0.00	34.78	0.00
<i>Antalis inaequicostata</i>	0.00	0.00	0.00	4.35	0.00
<i>Aricidea (Strelzovia) claudiae</i>	0.00	0.00	0.00	4.35	0.00
<i>Caecum trachea</i>	0.00	0.00	30.68	0.00	0.00
<i>Caprella rapax</i>	0.00	0.00	0.00	4.35	0.00
<i>Cirrophorus branchiatus</i>	0.00	4.30	0.00	0.00	0.00
<i>Cirrophorus nikebianchii</i>	14.24	21.96	0.00	0.00	20.59
<i>Fustiaria rubescens</i>	0.00	0.00	0.00	8.70	0.00
<i>Kirkegaardia dorsobranchialis</i>	3.16	0.00	0.00	0.00	0.00
<i>Loripes orbiculatus</i>	0.00	0.00	3.88	0.00	0.00
<i>Lucinella divaricata</i>	0.00	0.00	5.73	0.00	0.00
<i>Nemertea</i>	0.00	0.00	0.00	4.35	0.00
<i>Nucula nucleus</i>	0.00	0.00	0.00	8.70	0.00
<i>Oestergrenia digitata</i>	0.00	0.00	0.00	4.35	0.00
<i>Papillicardium papillosum</i>	0.00	0.00	4.44	4.35	0.00
<i>Polititapes aureus</i>	0.00	0.00	3.88	0.00	0.00
<i>Protodorvillea kefersteini</i>	0.00	0.00	10.72	0.00	0.00
<i>Pseudoleiocardia fauveli</i>	65.03	22.85	3.70	0.00	33.90
<i>Pseudolirius kroyeri</i>	0.00	3.26	0.00	0.00	20.90
<i>Syllis hyaline</i>	0.00	0.00	8.87	0.00	0.00
<i>Varicorbula gibba</i>	0.00	9.94	9.43	21.74	4.18
Strožanac	PD1	PD2	PD3	PD4	PD5
<i>Abra alba</i>	19.14	0.00	24.44	0.00	0.00
<i>Aonides oxycephala</i>	0.00	0.00	0.00	5.00	0.00
<i>Aricidea cfr. acmira meridionalis</i>	0.00	0.00	0.00	0.00	4.12
<i>Bivalvia</i>	0.00	0.00	6.67	0.00	0.00
<i>Cirrophorus nikebianchii</i>	8.82	0.00	0.00	0.00	0.00
<i>Cossura soyeri</i>	0.00	11.32	0.00	0.00	14.23
<i>Diogenes pugilator</i>	0.00	0.00	4.44	0.00	0.00
<i>Heteromastus filiformis</i>	3.27	0.00	4.44	0.00	4.12
<i>Paraleptopentacta elongata</i>	0.00	0.00	6.67	0.00	0.00
<i>Levinsenia gracilis</i>	0.00	22.26	0.00	0.00	9.36

4. Discussion

The species and diversity variation patterns indicate that the macrofaunal diversity was highly influenced by the renewal time of marine water, showing low biodiversity and strong dominance of few species (Guelorget and Perthuisot, 1983) in areas where the renewal time was high (see Table 1). In fact, from a confinement gradient point of view, small marinas can be compared to brackish environments, where high diversity is commonly observed toward the open part of the basin and, instead, lower species numbers with few dominant taxa are present in its innermost part (Tagliapietra *et al.*, 2009).

Our data clearly show that a high confinement was established in the inner part of the basins (i.e. SP1, SP4, PD1, and PD3). Moreover, the diverse sediment grain-size could be due to the intrinsic nature of the marinas that, similarly to any other area sheltered from waves and marine currents, are prone to siltation (Winterwep, 2005). In fact, a higher percentage of sand was found at stations placed near the main port entrance, whereas finer sediments were present at the inner stations.

Overall, regarding the different morphology of the Strožanac and Špinut marinas, Strožanac is undoubtedly a more open system compared to Špinut. This aspect was mirrored in higher nestendess-resultant values of β -diversity in Špinut that evidenced the presence of a community characterised by species that were a subset of those inhabiting the nearby stations (Baselga *et al.*, 2012). Conversely, the higher turnover values observed in the Strožanac marina indicated the presence of a higher variation in species composition between the stations. The occurrence of seagrass coverage [*Zostera* spp., Nasi (pers. comm.)] enhanced this species difference between the stations, particularly at PD1 and PD3. In the Špinut marina, the differences between the stations were mainly due to diverse sediment grain-size and anthropogenic influence in the basin (i.e. the boathouse area) (see Fig. 3). The highest diversity observed at SP3 was ascribable to mixed environmental conditions (i.e. major seawater renewal and higher sand percentage). In fact, at the latter station, common marine species, rare species (the polychaete *Goniadopsis* sp.), and alien taxa (the polychaete *Neopseudocapitella brasiliensis*) were all co-occurring. It is well-documented that alien species occur in ports principally as a result of intense marine traffic and discharge of ballast waters. However, the presence of alien species has been documented also in small touristic ports. The results of this study are in accordance with Travizi *et al.* (2019), who found non-indigenous species in a recreational marina (i.e. Pula, Croatia).

Despite the differences between the stations in each marina, we observed no significant variations in species composition between Strožanac and Špinut (PERMANOVA test). A few dominant species, such as the polychaete *Pseudoleiocapitella fauveli* and the mollusc *Abra alba*, probably masked the variation of less abundant species in both marinas. However, these two macrofaunal invertebrates could be considered ubiquitous species in costal environments, reaching the highest dominance in muddy sands, such as those found in the inner stations of both marinas (Dauvin, 1998; D'Alessandro *et al.*, 2016).

The findings of this study indicate that the presence of anthropogenic activities strongly affects the species composition at the inner station, where the lowest number of species was observed (see Fig. 3 and Fig. 4). In fact, an impoverished community was observed near the boathouse area in Špinut (SP4), characterised by a higher concentration of heavy metals deriving from boat painting activities [i.e. Cd, Cu, Pb, and Zn; Singh and Turner (2009)]. The results obtained agree with those of Chatzinikolaou *et al.* (2018) and Dimitriou *et al.* (2020), who reported a reduction of macrofaunal density and species in touristic port areas of the Mediterranean Sea directly influenced by human activity. Further, Chatzinikolaou *et al.* (2018) emphasised that the size of the harbour and the length and type of boats play a crucial role for the biodiversity of the port areas. The authors reported that, in a small marina hosting mostly local boats, both for leisure and fishery, the sediments were characterised by low contamination and higher biodiversity compared to those of larger touristic ports, hosting a higher number of and bigger boats for nautical tourism (e.g. charter expedition). Even if careening and boat painting activities were present both in Špinut and Strožanac, the effects of this human impact on the macrofaunal community were different. In Strožanac, hosting fewer boats principally belonging to local people and fishermen, we observed a lower impact compared to Špinut. At the station near the boathouse area (PD2), the macrofaunal community was not as strongly affected by the presence

of this activity, also due to its position at the open side of the marina, where greater depths and seawater flushing possibly enhanced the dilution of contaminants. As indicated by Mcgee *et al.* (1995), the features and design of marinas limit flushing and contaminant export, localising the impact in a small area of the basin. This situation is consistent with the case of Špinut, in which, the most contaminated site near the boathouse was located in the narrow part of the basin and far from the main entrance.

In both marinas, the variation of the feeding structure, as well as the species composition, could be driven by the grain-size pattern. Sediment composition is a key element in structuring the macrofaunal community and the distribution of dominant species, also when related to organic enrichment (Hermund *et al.*, 2008). Deposit-feeding invertebrates are reported to be numerically dominant in sandy-mud or muddy sediments (Nasi *et al.*, 2017 and reference therein). Much of the detrital food for these invertebrates is associated with a high proportion of particles within the silt-clay range. The high surface-area-to-volume ratio of small particles provides large expanse for the attachment and growth of microbial populations that produce mucopolysaccharide exudates, which are very palatable for these invertebrates (Donald and Larry, 1982).

Many SSDF are considered able bioturbators (Kristensen *et al.*, 2012). Bioturbator invertebrates are organisms whose activity produces constant and random local sediment biomixing, over a short distance, resulting in particle transport (Queirós *et al.*, 2015). According to Queirós *et al.* (2015), the high bioturbation activity at stations with a higher presence of SSDF, by intensifying oxygen fluxes in sediments, plays a major role in the reoxidation and detoxification of highly reduced sediments. This could also occur at the stations taken into consideration in this study, in particular towards the inner part of the marinas. Furthermore, the occurrence of SDF and SF near the main entrance could be linked to the higher percentage of sand. Similarly, Vesal *et al.* (2021) reported the high occurrence of SF as being related to high sand contents. Indeed, many bivalves (27.9% of the total community) were observed in SP3, the station characterised by a high sand content. In contrast, a large number of SF was detected in muddy sites at Strožanac (i.e. PD1 and PD3, Fig. 4). The presence of SF at the latter stations was mostly due to the dominance of *L. orbiculatus*. The occurrence of this species was reported in close association with seagrass *Zostera* spp. (El-Hacen *et al.*, 2018 and reference therein). Indeed, this bivalve was observed solely at PD1 and PD2, the only stations in the Strožanac marina characterised by finer sediments and the presence of seagrass.

Finally, SP4, the station near the boathouse in Špinut, was characterised by well-balanced feeding guilds. Despite low species richness, the community was constituted by different feeding habits. This result was not surprising, since the macrofaunal community is able to adapt their structure when in the presence of long-lasting pressure, as in the case of boat cleaning operations. However, the occurrence of few species for each trophic guild (e.g. carnivorous) could indicate higher vulnerability of the community. In turn, the vulnerability induces the reduction of long-term resilience of biological communities to cope with other environmental stressors, both of natural and anthropogenic origin (Naeem *et al.*, 2012).

5. Conclusions

This study confirmed the importance of the renewal time of marine water (confinement) for the dilution and dispersion rates of contaminants related to the nautical world. The Strožanac marina is characterised by seagrass coverage at the bottom, and the anthropogenic activities did not seem to influence the structure of the investigated macrofaunal community. This does

not hold true for Špinut, where long-lasting anthropic activities deeply modified the sediment characteristics and evidenced major macrofaunal adaptation to contamination at the main impacted site. Furthermore, these results indicate that the inclusion of macrofaunal community features in monitoring plans could help local port and marina managers design site-specific environmental interventions to mitigate anthropic disturbances.

Thanks to the ECOMAP project, over the past years the municipalities have implemented numerous activities in order to improve the ecological status of their marinas (Špinut and Strožanac). Among these, chemical and physical wastewater treatment plants were installed in order to improve wastewater treatment. In addition, two mobile recycling yards were procured, to reduce the amount of unclassified wastes in the nearby coastal area. Furthermore, as a future perspective, to find a lasting solution to contamination from boat cleaning, a first step could be to move this activity to larger and less confined areas, and, in parallel, to install holding tanks at the edge of the docks to collect the waste produced and dispose of it properly and directly on site.

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