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THE BENTHIC MACROFAUNA AT THE OUTFALLS OF THE UNDERWATER SEWAGE DISCHARGES IN THE GULF OF TRIESTE (NORTHERN ADRIATIC SEA, ITALY)

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ABSTRACT

The macrobenthic communities living at the outfalls of the five underwater sewage ducts servicing the Italian area of the Gulf of Trieste were sampled from 1990 to 1993 and analysed using uni- and multivariate techniques. 19,947 organisms from 217 taxa were identified. Polychaetes dominated the macrobenthic community, followed by molluscs; together they composed 92% of the total abundance and 80% of the number of species. Faunal composition differed between stations and was found to be influenced by sediment composition and depth rather than by discharges, even though the stations were located directly at the outfalls. Although the Biotic Index was low at all stations and no biological indicators of organic enrichment were found, the whole analysis indicated some degree of environmental unbalance, but it is similar to most soft bottom areas in the Gulf. These results indicate that the waste treatments have been effective in controlling the adverse effects of the urban discharges or at least have not negatively influenced the local benthic populations over the study period.

Key words: soft-bottoms macrobenthos, Biotic Index, sewage discharges, Gulf of Trieste, Adriatic Sea

LA MACROFAUNA BENTONICA DELLE CONDOTTE DI SCARICO SOTTOMARINE NEL GOLFO DI TRIESTE (ALTO ADRIATICO, ITALIA)

SINTESI

Le comunità macrobentoniche, insediate in vicinanza delle cinque condotte di scarico dei reflui nella parte italiana del Golfo di Trieste, sono state campionate dal 1990 al 1993 e sono state studiate con tecniche di analisi uni- e multivariata. Complessivamente sono stati identificati 19.947 individui appartenenti a 217 gruppi tassonomici. I policheti sono risultati il gruppo dominante, seguiti dai molluschi: i due gruppi insieme costituiscono il 92% dell'abbondanza totale e l'80% del numero di specie. La composizione faunistica di ciascuna stazione è maggiormente influenzata dalla tessitura del sedimento e dalla profondità, piuttosto che dalla vicinanza delle condotte di scarico. Sebbene i valori dell'indice biotico siano risultati bassi e non siano state ritrovate specie indicatrici di arricchimento organico, l'analisi complessiva dei popolamenti bentonici ha indicato un leggero grado di instabilità ambientale, il quale è piuttosto tipico delle comunità di fondo mobile del Golfo di Trieste. In base ai risultati ottenuti, il trattamento dei reflui scaricati dalle condotte sembra sia stato efficace e le comunità macrobentoniche non hanno mostrato evidenti alterazioni della loro struttura nel tempo, durante il periodo di studio.

Parole chiave: macrobenthos di fondo molle, indice biotico, condotte di scarico, Golfo di Trieste, Mare Adriatico

INTRODUCTION

Pollution of coastal waters has been one of the big ecological concerns of the 20th century; the fast development of coastal cities, ports and tourist localities worldwide with consequent increasing concentrations of human populations, has created, among others, serious problems of disposal of urban and industrial wastes. The effects of these discharges into the sea have long been known to be harmful to the coastal zone environment, threatening the well-being of people and the lucrative benefits derived from the development of activities such as tourism, fisheries or mariculture, to cite a few.

As an alternative to direct discharges to the littoral, offshore disposal has been adopted, as far away as possible from urban centres, sometimes as direct ocean discharges (or dumping), but increasingly through underwater ducts. Those who end up in deep waters tend to be increasingly used, where possible (Diener *et al.*, 1995; Koop & Hutchings, 1996; Gallagher & Keay, 1998; Zamouri-Langar *et al.*, 2001). Nowadays, sewage treatment prior to discharge is not yet the rule worldwide, but is already normative in developed countries, especially in Europe (Urban waste water treatment directive 91/271/EEC and directive 98/15/EEC amending the former).

The assessment of ecological impacts related to sewage disposal has been documented in several types of marine communities, from soft-bottoms benthos (Ghirardelli *et al.*, 1973; Smith *et al.*, 1973; Pearson & Rosenberg, 1978; Otway, 1995a), to hard bottoms benthos (Littler & Murray, 1975; Fairweather, 1990; Grigg, 1994; Koop & Hutchings, 1996), and fish (Puffer *et al.*, 1982; Grigg, 1994; Otway, 1995a, 1995b). The benthic communities are preferred as indicators of the health of marine environments, because of their main characteristics: reduced motility (*i.e.* incapacity of escaping polluting discharges, even if highly toxic or lethal), high diversity (*i.e.* selective response to environmental stress), and relatively long life cycles, which allow the observation of the short, medium, or long term effects of any discharged substance (Pearson & Rosenberg, 1978; Reish, 1980, 1986; Hily *et al.*, 1986; Dauer, 1993; Borja *et al.*, 2000).

The region of the Gulf of Trieste (northern Adriatic Sea), populated for at least the last 2000 years (Stevenson *et al.*, 1999), is now heavily anthropized. In some areas, pollution of coastal waters caused by the direct discharges to sea from the riverine urban and industrial centres had caused, in the past, growing problems to the marine environment and endangered the development of the tourist "industry" (bathing, beaches) which provides the first source of income for a large part of the population (Ghirardelli *et al.*, 1973, 1975). To solve this problem, five underwater ducts connected to the waste treatment plants were built between the 70s and 80s to serve the Italian coast of the Friuli-Venezia Giulia Region (Fig. 1).

To estimate the effects of those off-shore discharges on the environment, and in accordance with the (then) newly enforced ecological laws in the region, sampling of the macrobenthic communities was performed at the beginning of the 90s at the ducts' outfalls.

It is the purpose of this study to analyse these results trying to find the common components, which characterize the macrobenthic populations at the outfalls of the discharge ducts. In addition, these data will provide a baseline study of the conditions prevailing in the area for future reference.

MATERIAL AND METHODS

Study area description

Located at the north-eastern end of the Adriatic Sea, the Gulf of Trieste is a large and shallow embayment, with a coastline of 100 km, a surface close to 600 km^2 (Ogorelec *et al.*, 1991), and a maximum depth of 25 m (Fig. 1). Average bottom salinities range from 36 to 38.5 and annual temperatures ranges are 8 to 20 °C at the bottom (Cardin & Celio, 1997). Water circulation is from southeast to northwest. Sedimentation is controlled mainly by river inputs rather than by marine currents (Brambati & Venzo, 1967); the soft bottoms are not homogeneous in composition and can vary from sands with patches of beach rocks to muds, predominantly detrital (Brambati *et al.*, 1983). The other natural factors unique to this area that mainly influence the characteristics of the composition, evolution and persistence of its marine life are: 1) strong winds from the northeast that can provoke mixing of the waters down to the bottom; 2) thermal stratification of the water column (5–6 m at the beginning of spring until about 15 m at the end of summer) (Cardin & Celio, 1997) leading to occasional hypoxic and/or anoxic events (Aleffi *et al.*, 1992; Orel *et al.*, 1993; Malej & Malačič, 1995); 3) high sedimentation rates estimated at 1–2.5 mm y^{-1} (Ogorelec *et al.*, 1991; Covelli & Fontolan, 1997); and 4) occasional mucilage production (Degobbis *et al.*, 1995, 1999).

The study area is composed of the stations located directly at the outfalls of the five ducts operative in the area (Fig. 1). Stations 1 and 3 correspond to the sewage outfalls of the urban zones of the tourist cities of Lignano and Grado, characterised by considerable annual population fluctuations; *e.g.* Grado varied, in 1985, from 9,000 inhabitants in the winter to 53,000 in the summer, and in 2000 from 9,000 to 80,000; Lignano's population varied more drastically: in 1985, from 5,500 to 140,000 and in 2000 from 6,500 to ca. 250,000 (F.V.G., 1985, 2000). Station 2 is located at the outfall of the duct serving the Friuli lowlands, an industrial area with corresponding human settlements, with a population of about

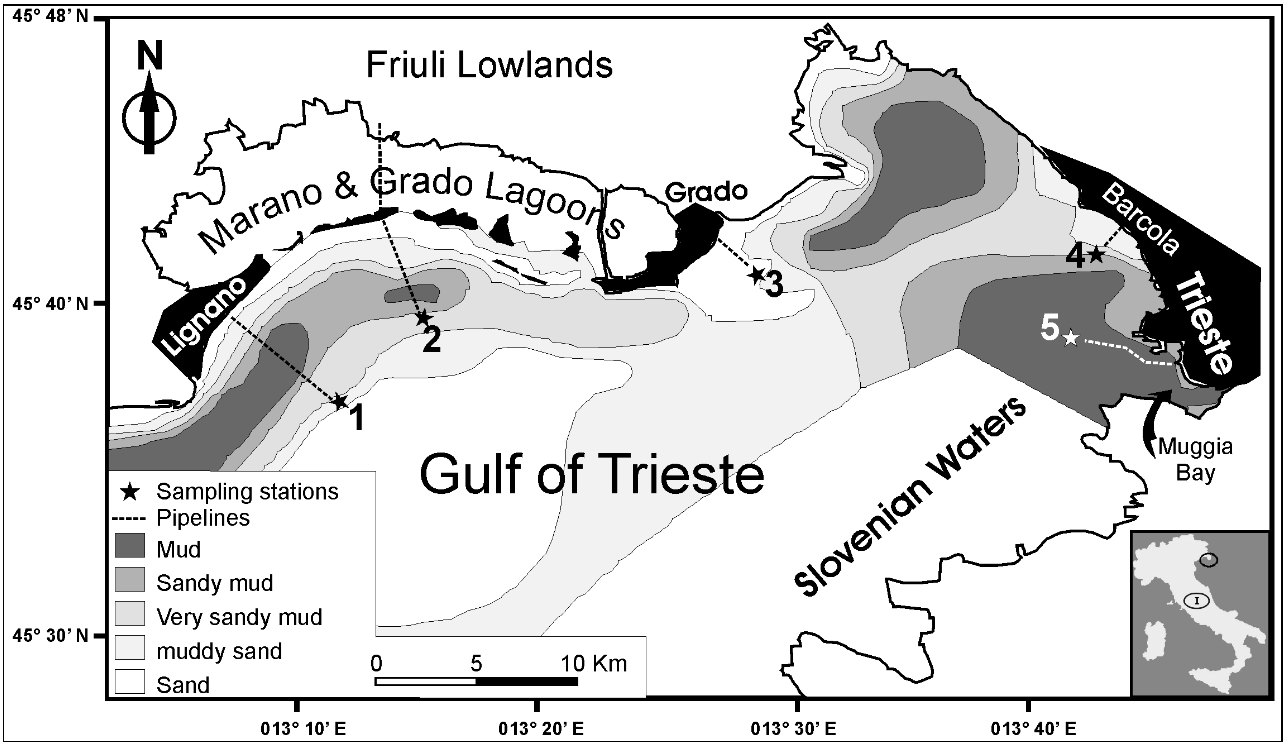

Fig. 1: Study area showing the outfalls position – sampling stations of the underwater sewage diffusers in the Italian part of the Gulf of Trieste.
Sl. 1: Raziskovano območje z vzorčevalnimi postajami ob podvodnih izlivih kanalizacijskih odplak v italijanski del Tržaškega zaliva.

Tab. 1: Main features of the five outfalls areas in the Gulf of Trieste (data from Novelli, 1996).
Tab. 1: Glavne značilnosti petih območij z vzorčevalnimi postajami ob podvodnih izpustih kanalizacijskih odplak v Tržaškem zalivu (podatki po Novelli, 1996).

Ducts	Treatment	Water depth (m)	Duct length (m)	Diffuser length (m)	Max flow (l sec^{-1})
Lignano (st. 1)	secondary	16	6000	1500	700
Porto Buso (st. 2)	secondary	15	6000	800+800	1780
Grado (st. 3)	primary (chemical)	10	4156	600	456
Barcola (st. 4)	primary	19	900	60	70
Trieste (st. 5)	primary (chemical)	23	6000/6500 (two parallel ducts)	500/1000	6000

375,000. Station 4 serves the beach area of the city of Trieste and a residential zone (Barcola) of about 12,000 inhabitants, whereas station 5 serves Trieste, a city of about 240,000 inhabitants, with well developed industrial, commercial and port activities. All the ducts operate with diffusers at their end, Y-shaped in the case of station 2.

In Table 1, all the ducts characteristics that could be gathered are reported. It is noteworthy that no reliable data are available for the daily average flow or the nitrogen or phosphorus discharges for any of the ducts in the Gulf.

Sampling

Sampling was performed with a 0.1 m^2 Van Veen grab (Aug-Sept 1990, 1991 and 1993, Nov-Dec 1990, 1991) (Fig. 1). At each station, three grabs (0.3 m^2) were taken. A small fraction of each grab was preserved for sedimentological analyses. Bottom salinities and temperatures as well as oxygen concentrations were measured, using an Idronaut Mod 401 multiparametric probe.

The sediment was sieved on a 1 mm mesh and fixed in formalin following standard methodology (Holme & McIntyre, 1984), then separated and the fauna identified to the lowest possible taxonomical level. For comparative purposes with other similar studies, the abundances were adjusted to a surface of 1 m^2. Unidentified species or groups (as in the case of the amphipods), which cannot be confused with any other identified species, were included, thus the total number of taxa reported represents the lowest number of species possible. The identified species are listed in Appendix 1.

Analyses

Uni- and multivariate techniques were used to analyse the communities' structure and included: abundance, number of species, diversity indexes (Shannon-Wiener diversity index (H') on $\log_e$ basis (Shannon & Weaver, 1949), Margalef's index (d) (Margalef, 1958) and Pielou's evenness index (J) (Pielou, 1966)). The feeding guilds' analysis was based on Fauchald & Jumars' (1979) and Bachelet's (1981) definitions. The Bray-Curtis similarity coefficient was calculated on square-root transformed data, using complete linkage; then, non-metric multidimensional scaling ordination (MDS) (PRIMER software package developed at the Plymouth Marine Laboratory) was used to evaluate the composition similarity among the stations. Since the stress factor was found to be greater than 0.1 and following Clarke & Warwick (2001) recommendations, hierarchical classification was applied. AMBI and Biotic Index (BI) were applied (Borja *et al.*, 2000; 2003) using the AMBI program – version 4.0 (AZTI Marine Biotic Index) (www.azti.es).

These indexes are based on the classification of the benthic species in five (I–V) ecological groups (EG), according to their tolerance to pollution (from EG–I = species very sensitive to organic enrichment, intolerant to pollution, EG–II = species indifferent to enrichment, EG-III = species tolerant to enrichment, slightly unbalanced environments, EG-IV = second-order opportunistic species, slight to pronounced unbalanced environments, to EG-V = first-order opportunistic species, pronounced unbalanced environment), then applying an algorithm to calculate the AMBI on a scale of increasing pollution (from 1 to 6) and obtaining the corresponding BI (0–1 = unpolluted sites, 2 = slightly polluted, 3 = moderately polluted, 4–5 = moderately to heavily polluted, 6 = heavily polluted, and 7 = extremely polluted, azoic state).

A recent multimetric index (M-AMBI) to assess the ecological quality status was applied, where the species richness and Shannon-Wiener diversity are also taken into consideration together with AMBI at the very same time (Muxika *et al.*, 2007). The AMBI program provides "Bad" and "High" reference conditions as default. As "Bad" conditions the values are always 6 for AMBI and 0 for diversity and richness. For "High" conditions the software selects the lowest AMBI value and the highest diversity and richness values. The user can modify these values (bad and high) if some reference conditions have been defined, within the Water Framework Directive, for the studied area (Muxika *et al.*, 2007). Waiting for the definition of reference conditions in Italian countries, the present paper adopted the default boundaries suggested by Borja *et al.* (2007).

RESULTS

Abiotic parameters

Bottom temperatures ranged from 9.34 °C (winter) to 23.55 °C (end of the summer). During the same campaign, differences among stations were negligible even if depth varied from 10 m (station 3) to 23 m (station 5) with the only exception of August 1991 when, at the deepest stations, temperature dropped of about 5 °C with respect to the shallower ones (Tab. 2). Bottom salinities varied from 36.26 to 38.20; oxygen concentrations (D.O.) values at the bottom were always around saturation, except for stations 4 and 5, at the end of the summer (Tab. 2).

Sediment composition varied from sands at station 3 to muds at station 4 and 5. At stations 1 and 2, mixed sediments were present, with higher sand content in station 1 than in station 2 (Tab. 2).

Faunal structure

19,947 organisms from 217 taxa (Appendix 1) were identified; the polychaetes were by far the dominant group with 12,223 organisms (61.3% of the total population) followed by the molluscs with 6,113 organisms (30.7%). Together, they constituted 92% of the total. Crustaceans with 883 (4.4%), echinoderms with 480 (2.4%) and "others" (the remainder of the usually scarce groups such as: ascidians, anthozoans, sipunculids, nemertines, phoronids and turbellarians) with 247 organisms (1.2%), complete the list; in this last group, the sipunculids constituted 47.3%. The polychaetes were also the richest group with 125 species (57.6%), followed by the molluscs with 48 species (22.1%); together they constituted almost 80% of the total. Crustaceans (15 taxa, 6.9%), echinoderms (14 taxa, 6.5%) and "others" (15 taxa, 6.9%) followed far behind.

The number of species and the mean of Margalef index decreased from station 1 to 5 (55 to 22, 7.9 to 3.2), while abundance and H' dropped from stations 1–2 to 3, and then to 4–5. Mean values of evenness were lowest at stations 4 and 5 (Tab. 3).

Cumulative percentages of the ten most abundant species were significantly lower at stations 1–3 (55.3–65.1%) than at stations 4–5 (83.9–88.8%). Dominance of the most abundant species was more pronounced at stations 1 and 2, where it represented 6 and 7 times the percentage of the next species (Tab. 4). *Corbula gibba* was strongly dominant at stations 2, 4 and 5, but was absent at stations 1 and 3.

Tab. 2: Abiotic parameters registered at the bottom in the five ducts' outfalls of the Gulf of Trieste.
Tab. 2: Abiotični parametri, ugotovljeni na dnu petih vzorčevalnih postaj ob podvodnih izpustih odplak v Tržaškem zalivu.

Station	Date	depth (m)	T (°C)	Sal	D.O.(cm^3 dm^{-3})	D.O.(%)	sand (%)	mud (%)
St. 1	4.9.1990	16.7	22.83	37.58	5.14	106.30	71.4	28.6
	13.11.1990	15.9	17.79	36.84	5.20	97.64	75.3	24.7
	29.8.1991	16.5	21.39	37.02	4.81	96.65	70.5	29.5
	18.12.1991	15.7	9.92	37.03	5.63	90.10	72.2	27.8
	1.9.1993	16.7	23.45	37.19	4.89	102.00	75.8	24.2
St. 2	4.9.1990	16.0	22.86	37.54	5.04	104.26	42.1	57.9
	13.11.1990	14.2	17.55	36.51	5.13	95.69	45.8	54.2
	29.8.1991	15.4	23.55	35.91	4.97	103.09	41.2	58.8
	18.12.1991	14.1	10.42	37.24	5.75	93.15	43.4	56.6
	1.9.1993	15.6	23.45	37.35	4.47	93.33	42.8	57.2
St. 3	4.9.1990	10.0	22.79	37.51	4.96	102.46	95.2	4.8
	13.11.1990	10.4	17.05	36.42	5.21	96.20	96.9	3.1
	29.8.1991	9.8	23.04	36.26	5.01	103.20	97.4	2.6
	18.12.1991	8.6	9.34	36.97	6.14	96.98	96.3	3.7
	1.9.1993	10.0	22.80	37.00	4.80	98.88	95.6	4.4
St. 4	5.9.1990	19.5	21.83	37.96	1.74	35.43	3.8	96.2
	14.11.1990	20.6	17.46	37.22	5.23	97.80	4.0	96.0
	30.8.1991	19.4	18.41	37.63	2.83	54.02	3.5	96.5
	19.12.1991	18.9	9.90	37.29	5.71	91.49	2.7	97.3
	31.8.1993	19.7	22.56	37.68	3.77	77.64	3.3	96.7
St. 5	5.9.1990	22.4	21.30	38.20	0.80	16.16	1.8	98.2
	14.11.1990	21.8	17.52	37.33	5.11	95.73	1.5	98.5
	30.8.1991	22.6	17.23	37.80	4.95	92.48	1.1	98.9
	18.12.1991	22.3	10.21	37.38	5.99	96.69	2.0	98.0
	31.8.1993	23.0	21.42	37.88	1.41	28.49	1.7	98.3

Tab. 3: Ecological parameters measured for this study: H' – Shannon-Wiener diversity index; d – Margalef's index; J – Pielou's evenness index.
Tab. 3: Ekološki parametri, ugotovljeni za pričujočo študijo: H' – Shannon-Wienerjev diverzitetni indeks; d – Margalefov indeks; J – Pieloujev indeks izenačenosti.

	Abundance (ind m^{-2})			Species richness (n°)			H'			d			J		
Station	min	max	mean	min	max	mean	min	max	mean	min	max	mean	min	max	mean
St. 1	710	1490	955	41	82	55	2.5	3.5	3.1	5.8	11.1	7.9	0.7	0.9	0.8
St. 2	170	1377	967	23	65	45	2.0	3.5	2.7	4.3	9.1	6.5	0.5	0.8	0.7
St. 3	343	1467	701	27	53	39	2.6	3.3	2.9	4.5	7.7	5.9	0.7	0.9	0.8
St. 4	363	1263	669	12	36	28	0.9	2.8	2.1	1.9	5.4	4.2	0.4	0.8	0.6
St. 5	490	847	698	7	41	22	0.9	2.4	1.6	0.9	6.0	3.2	0.5	0.7	0.5

The dendrogram resulting from the Bray-Curtis similarity matrix (Fig. 2a) shows that the stations were more closely related among themselves than with the others, a trend confirmed by the MDS ordination (Fig. 2b); three main groups were identified: the first is formed by two subgroups constituted by stations 4 and 5, the second is formed by station 3, and the third comprises stations 1 and 2 with the only exception of the sample of 11/1990 at station 3.

The value of Biotic Index was 1 in station 1 and 2 in remaining stations (Fig. 3), which correspond respectively to unpolluted and slightly polluted conditions for the "site pollution classification", and to impoverished and unbalanced situations regarding the "benthic com-

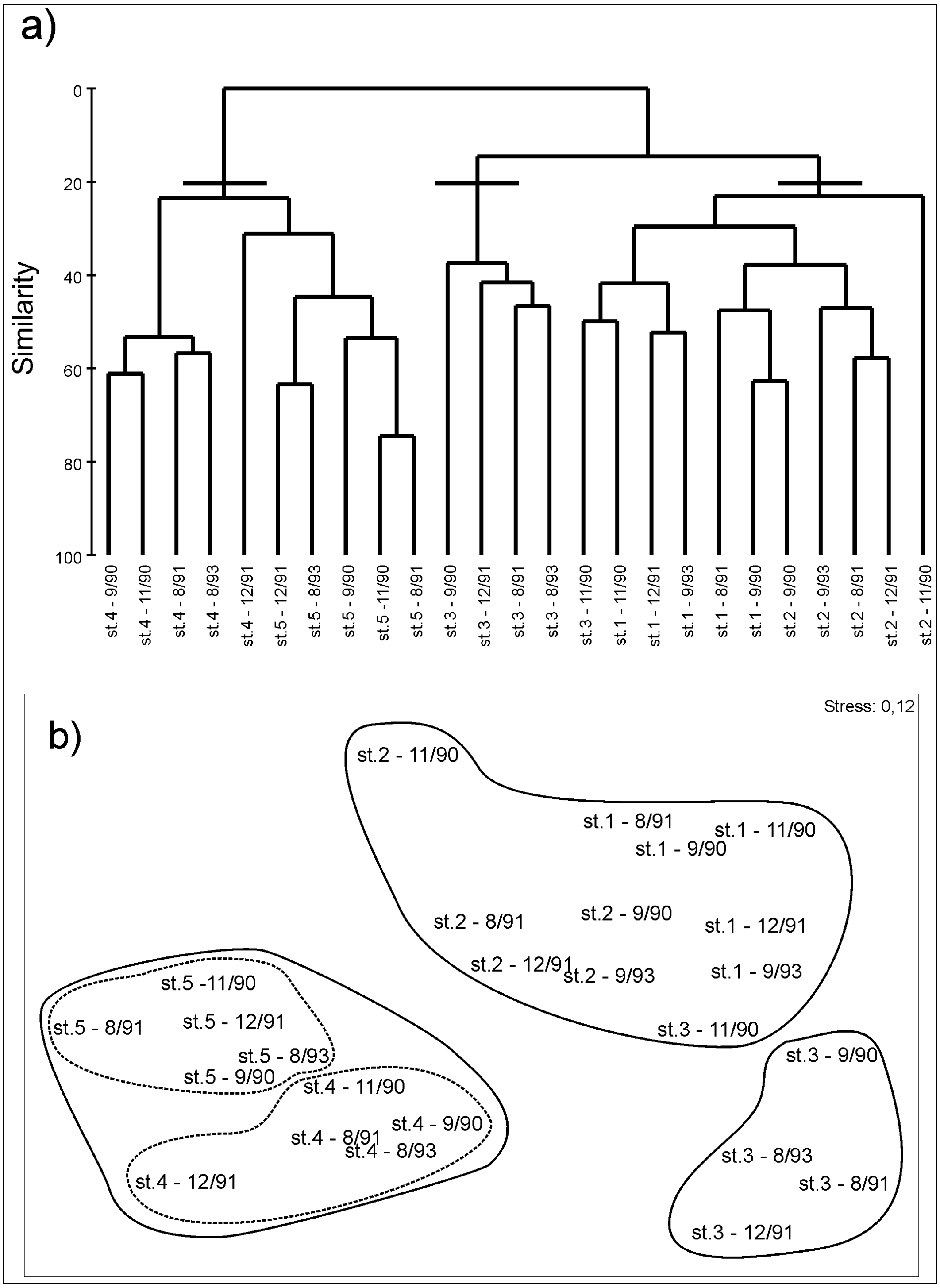

Fig. 2: (a) Hierarchical agglomerative clustering of square-root transformed macrobenthos data using complete linking on Bray-Curtis similarities (%); (b) multidimensional scaling ordination from square-root transformed macrobenthos data based on Bray-Curtis similarities.
Sl. 2: (a) Klasterski diagram na podlagi Bray-Curtisovega indeksa podobnosti; (b) večdimenzionalno skaliranje na podlagi Bray-Curtisovega indeksa podobnosti. Pri obeh analizah so bili makrobentoški podatki transformirani z uporabo kvadratnega korena.

munity health" (Borja *et al.*, 2000). M-AMBI index revealed a "Good" to "High" quality of benthic ecological status (Fig. 4).

About feeding guilds, most dominant species were surface deposit feeders, sub-surface deposit feeders or carnivores (Tab. 4), except for the absolute dominant, *C. gibba* (ca. 23% of the total), which is a suspension-feeder.

DISCUSSION

When analysing species richness in the Gulf of Trieste, we can see that the richest areas in number of species are the centre of the Gulf, around the discharges ducts of the urban centres, the zone to the east of the Isonzo estuary and the Barcola and Sistiana areas. The poorest areas were found to be in the northernmost zone, at the farther end of the Bay of Muggia and the deepest muddy areas, sometimes subjected to hypoxia events. Typical species indicating pollution conditions were found in some restricted coastal areas and in the Bay of Muggia, and generally they were in low density (Solis-Weiss *et al.*, 2001). The average Biotic Index in the Gulf is 2 and indicates a general not polluted and quite diversified condition of macrozoobenthos, excepting some area characterized by slight or moderate disturbance due to *C. gibba* dominance, where both natural and man induced stresses are involved (Solis-Weiss *et al.*, 2001; Rossin, 2005).

The Biotic Index calculated in the present study seemed to indicate a similar response of the macrofauna to the discharges, excepting the undisturbed condition revealed in station 1. However, this value is the average of very different proportions of the four ecological groups (EG) of species present at each station particularly evident at stations 1 and 5 (Fig. 3); at station 1, EG-I species clearly dominated (50%), while at station 5 there were practically no species of EG-II and III, and EG-IV species dominated, closely followed by EG-I species. In the other three stations, the proportions between ecological groups were more balanced, with a prevalence of EG-IV species in stations 2 and 4.

The application of M-AMBI (Muxika *et al.*, 2007) within the European Water Framework Directive (WFD 2000/60/EC) indicated an overall good ecological status of the study area.

The analysis of the species composition and abundance further emphasized the differences among the stations. Considering the ten most abundant species for each station (Tab. 4), *Aponuphis bilineata*, was among the first at stations 1, 2 and 3, being the dominant at station 1, while totally absent from stations 4 and 5; this species is mainly found in fine sands (Ameziane *et al.*, 1995; Desroy & Retiere, 2001). *Lucinella divaricata*, another sand dweller (Sardà *et al.*, 1999) and *Owenia fusiformis*, a species characteristic of fine sands (Pérès &

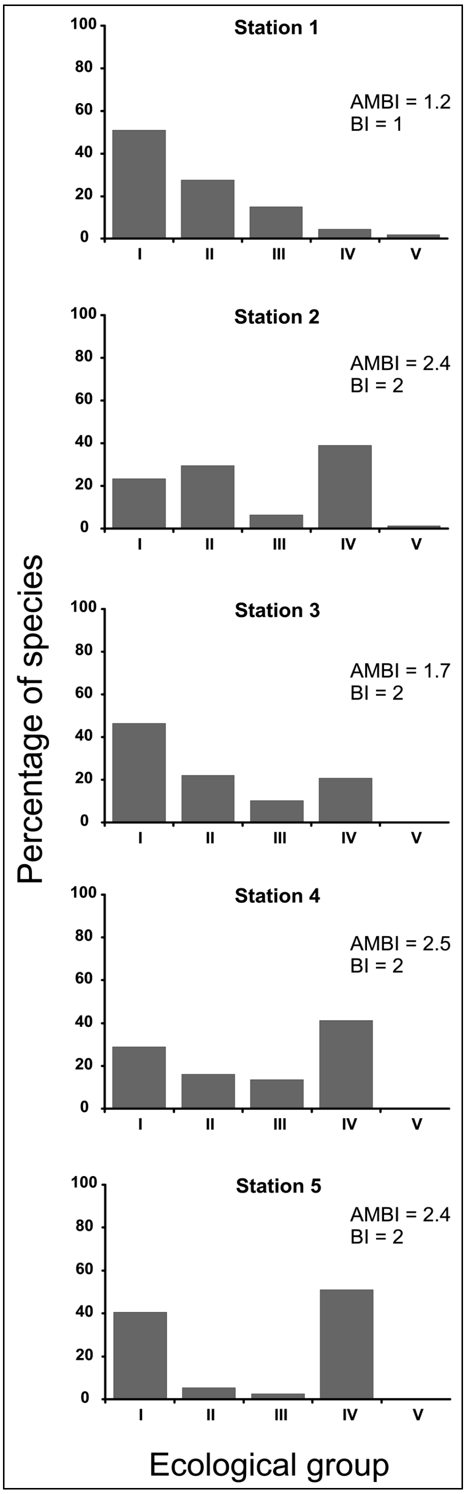

Fig. 3: Percentage of species of EG I to EG V per station with the corresponding AMBI and Biotic Index values, following Borja* et al. *(2003).
Sl. 3: Odstotek vrst EG I do EG V na posamezno vzorčevalno postajo z vrednostmi AMBI in biotskih indeksov po Borja* et al. *(2003).

Tab. 4: Cumulative percentage and feeding guilds of the first ten most abundant species in the study area: Tot. abund. – total abundance; SDF – surface deposit feeders; SSDF – sub-surface deposit feeders; C – carnivores; SF – suspension feeders.

Tab. 4: Skupni delež in prehranjevalni cehi prvih deset najštevilčnejših vrst v preučevanem območju: Tot. abund. – skupna abundanca; SDF – detritivori na površju morskega dna; SSDF – detritivori v morskem dnu; C – mesojedi organizmi; SF – filtratorji.

Station 1				
Species	%	Cum %	Tot. abund.	Feed. guilds
Aponuphis bilineata	26.8	26.8	1280	C
Notomastus sp.	4.5	31.3	217	SDF-SSDF
Chone duneri	4.3	35.7	207	SF
Clymene sp.	4.3	40.0	207	SSDF
Maldane glebifex	3.7	43.7	177	SSDF
Eunice vittata	2.8	46.5	133	C
Laonice cirrata	2.4	48.9	117	SDF
Pista cristata	2.4	51.3	113	SDF
Chone collaris	2.1	53.4	100	SF
Lumbrineris latreilli	2.0	55.3	93	C
Station 2				
Species	%	Cum %	Tot. abund.	Feed. guilds
Corbula gibba	35.2	35.2	1703	SF
Lumbrineris latreilli	5.1	40.3	247	C
Aponuphis bilineata	4.8	45.2	233	C
Amphiura chiajei	3.9	49.0	187	SDF
Lumbrineris gracilis	3.7	52.7	177	C
Maldane glebifex	2.7	55.4	130	SSDF
Clymene sp.	2.6	58.0	127	SSDF
Eunice vittata	2.6	60.6	123	C
Terebellides stroemi	2.4	63.0	117	SDF
Spiochaetopterus costarum	2.1	65.1	103	SF
Station 3				
Species	%	Cum %	Tot. abund.	Feed. guilds
Eunice vittata	12.2	12.2	427	C
Lucinella divaricata	11.2	23.4	393	SF
Clymene sp.	7.4	30.8	260	SSDF
Aponuphis bilineata	7.3	38.2	257	C
Prionospio malmgreni	5.8	44.0	203	SDF
Chaetozone setosa	5.1	49.1	180	SDF
Poecilochaetus serpens	4.8	53.9	167	SDF
Prionospio caspersi	2.9	56.8	103	SDF
Ancistrosyllis groenlandica	2.8	59.6	97	C
Owenia fusiformis	1.9	61.5	67	SDF-SF
Station 4				
Species	%	Cum %	Tot. abund.	Feed. guilds
Corbula gibba	37.6	37.6	1257	SF
Maldane glebifex	10.9	48.5	363	SSDF
Melinna palmata	9.7	58.1	323	SDF
Eunice vittata	8.5	66.6	283	C
Spiochaetopterus costarum	6.7	73.3	223	SF
Nucula nucleus	4.3	77.6	143	SDF
Pectinaria koreni	2.2	79.8	73	SSDF
Pomatoceros triqueter	2.0	81.8	67	SF
Paraonis sp.	1.2	83.0	40	SDF
Aricidea sp.	1.0	83.9	33	SDF
Station 5				
Species	%	Cum %	Tot. abund.	Feed. guilds
Corbula gibba	47.9	47.9	1670	SF
Maldane glebifex	28.7	76.5	1000	SSDF
Spiochaetopterus costarum	2.4	78.9	83	SF
Pomatoceros triqueter	2.2	81.1	77	SF
Pectinaria koreni	1.7	82.8	60	SSDF
Nucula nucleus	1.4	84.2	50	SDF
Processa sp.	1.3	85.6	47	C
Terebellides stroemi	1.1	86.7	40	SDF
Atrina pectinata	1.1	87.9	40	SF
Pectinaria auricoma	1.0	88.8	33	SSDF

Picard, 1964), were practically found only at station 3 (respectively 97.5% and 96.5% of the specimens collected in this study). Station 3 differed from the other four stations in species composition: seven out of its ten most abundant species were found as dominant only at this station (Tab. 4) and the dominances were not marked. Moreover, in contrast with the other four stations, at station 3 no echinoderms were found.

At stations 4 and 5, *C. gibba* and *Maldane glebifex* were the absolute dominants, amounting together to almost 50% and 76% of the total, respectively (Tab. 4). In addition, six out of the ten most abundant species were common to those two stations. Both *C. gibba* and *M. glebifex* are prevalently found in muddy bottoms. *C. gibba* was also strongly dominant at station 2 (35% of the total), but the faunal composition of this station was very different from the former two (Tab. 4). The changes over time of *C. gibba*'s abundance were much more evident, being very high only in the 1991 samplings. The number of total taxa was also much larger at stations 1 and 2: 138 and 105, versus 73 and 63 at stations 4 and 5.

Since three different groups (1–2, 3, 4–5) emerged as a result of uni- and multivariate analyses, these populations cannot be typified as the macrofauna "characteristic of the outfalls areas" of the Gulf.

We found that the individuated groups share similar sediment and depth conditions. Sediment composition has been found to be a key element in structuring macrofauna community (Gray, 1974), also when related to sewage outfalls. Nicolaidou *et al.* (1993), for example, found that in areas influenced by organic discharges to Saronikos Gulf (Aegean Sea), diversity increased in areas of mixed sediments, rather than related to a gradient in organic enrichment, when conditions of pollution were not too severe, *i.e.* not directly at the outfalls.

Depth is also known to influence abundance and diversity because it is highly correlated to factors such as light, temperature, hydrodynamic properties and size of sediments' particles among others (Vio *et al.*, 1980; Orel *et al.*, 1987; Diener *et al.*, 1995): in this shallow bay, even a few meters make a difference for the local benthos (Aleffi *et al.*, 1995).

The structure of each station's populations appeared firstly related to local combinations of sediment composition and secondarily to depth. These two factors seem to act synergetically in the separation of the stations groups of Figure 2. At stations 1 and 2, shallow waters and mixed sediments led to the highest values of diversity and abundance; at station 3, the shallowest and with only sandy sediments, the populations' structure was different, with the highest dominance of sand dwellers (Tab. 2); at the deepest stations 4 and 5, where muds dominated, reduced abundance and diversity were recorded. The latter are also occasionally subjected to anoxia (Orel *et al.*, 1993).

On the other hand, even though the populations of the five stations cannot be typified as "characteristic of the outfalls", the very strong dominance of polychaetes and molluscs in their composition (92%), and the scarcity of echinoderms constitute an indication of environmental instability or anthropic impact common to all.

However, the impact does not seem to be too severe, as indicated by the Biotic Index (Fig. 3), the species composition, the predominant ecological groups (as defined by Borja *et al.*, 2003) and M-AMBI Index (Fig. 4). The dominant species, both in abundance and frequency (Appendix 1, Tab. 4) were *C. gibba*, *A. bilineata*, *Eunice vittata* and *M. glebifex*. *C. gibba* (EG-IV) is considered an indicator of instability (Orel *et al.*, 1987; Crema *et al.*, 1991; Elias, 1992; Aleffi & Bettoso, 2000; Zamouri-Langar *et al.*, 2001). It is able to withstand active sediment resuspension and even hypoxia (Diaz & Rosenberg, 1995), but it is never found in conditions of severe pollution (Solis-Weiss *et al.*, 2004). *A. bilineata* (EG-II) and *E. vittata* (EG-II) are found in unbalanced or moderately impacted bottoms, and *M. glebifex* (EG-I) is basically found in unpolluted muddy bottoms.

In addition, even though most abundant species were surface deposit feeders, sub-surface deposit feeders or carnivores (Tab. 4), able to withstand organically polluted conditions (Pearson & Rosenberg, 1978), no species indicating organic enrichment, or of EG-V (Borja *et al.*, 2000), such as *Capitella capitata*, *Malacoceros fuliginosus*, *Cirriformia tentaculata* or even *Neanthes caudata* (collected only once) (Pérès & Picard, 1964; Bellan, 1967; Grassle & Grassle, 1974; Bellan & Bourcier, 1990; Cardell *et al.*, 1999) were present. The high abundance of *C. gibba* at station 2 only recorded in 1991 certainly indicate a more unstable situation at that station in that period, but the source that triggered this response could not be identified.

The values reported for heavy metals related to anthropic activities (Pb, Cu, Mn, Ni, Zn, Cd and Cr) in the sediments of the study area (Rivetti, 1993; Covelli & Fontolan, 1997; Barbieri *et al.*, 1999; Covelli *et al.*, 2001) are difficult to evaluate as a measure of pollution, at least with reference to the local benthic populations. The fact that sediment composition importantly influences its heavy metals' concentrations (*i.e.* under similar conditions, in fine sediments the concentrations will always be higher) is not taken into account in Rivetti (1993) and Barbieri *et al.* (1999). In addition, the data published do not take into consideration the bioavailability of those metals; such studies have not been carried out so far in this area. It is very difficult to evaluate any environmental impact based only on abiotic data. Ideally, both abiotic and biotic parameters should be confronted, but we believe that the benthic populations are the best indicators of the global pollution "state" of an area, since it is their structure that most accurately reflects the synergetic effects of all substances to which they are subjected.

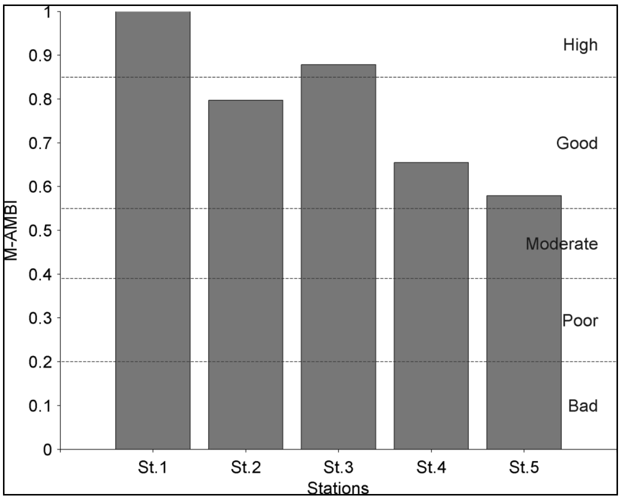

Fig. 4: M-AMBI Index per station, following Borja** et al. **(2007).
Sl. 4: Indeks M-AMBI na posamezno vzorčevalno postajo po Borja** et al. **(2007).

In this study, the values of dissolved oxygen measured at the ducts outfalls (Tab. 2) were almost always around saturation levels, *i.e.* better than expected in discharge areas.

Species composition and diversity values indicated conditions that can be qualified as "slightly polluted" or "unbalanced" (Borja *et al.*, 2000, 2003) and on the whole "Good" (Muxika *et al.*, 2007). However, in most of the other soft bottom areas of this large Gulf, similar qualifications can be applied, so that the outfalls areas come out as not particularly impacted with respect to the neighbouring zones (Solis-Weiss *et al.*, 2001).

This could mean that there is an environmental generalised impact or stress in the whole Gulf. It has been reported that the northern Adriatic benthic macrofauna has a lower number of species but higher values of abundance and biomass than the southern region (Gamulin-Brida, 1967; Šimunović, 1997) thus reflecting "harsher" environmental conditions. The Gulf of Trieste represents the most extreme conditions since it is located at the northeastern end of the Adriatic and under strong natural stress factors already outlined in the study area section, to which the anthropic pressure is added. Consequently "unbalanced" conditions are generalized throughout the area, with the influence of the natural stress factors prevailing over the anthropic pressure. It is noteworthy that at all stations diversity was higher than expected at an outfall zone (Bellan & Bourcier, 1990; Tsutsumi, 1990; Méndez, 1993; Taylor *et al.*, 1998), particularly at stations 1 and 2 where benthic macrofauna is among the richest and more diversified in the entire Gulf (Solis-Weiss *et al.*, 2001).

One possible explanation that should be verified could be that in these oligotrophic-mesotrophic waters of the northern Adriatic (Fonda Umani, 1996), some organic matter input could favour the local benthic populations. Several authors (Pearson & Rosenberg, 1978; Wilkinson, 1999) have indicated that in conditions of moderate organic enrichment, an increase in the number of species can occur, before an eventual decline.

CONCLUSIONS

Since little was known about the soft bottoms macrobenthos surrounding the sewage discharge areas in the Gulf of Trieste, the analysis of these first samplings open new insights on the real effect of the local discharges on the benthic fauna, although at this point they are to be considered as preliminary results and the baseline for further studies. The macrobenthic populations' structure differed between stations and was found to be more influenced by local environmental parameters, such as sediment composition and depth, than by the selected common denominator: *i.e.* the sewage discharges.

It is imperative to continue these studies and establish a monitoring programme to evaluate accurately the ecological importance of the impacts in the medium and long range, and (hopefully) confirm these trends over the following years. Within this framework, the heavy metals' bio-availability should be assessed, in order to interpret correctly the source and the effects of the impacts on the local marine fauna, since so far, these results potentially indicate that the waste treatments have been effective in controlling the negative effects of urban discharges on those bottoms or at least have not negatively influenced their populations.

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BENTOŠKA MAKROFAVNA PRI PODVODNIH IZPUSTIH KANALIZACIJSKIH ODPLAK V TRŽAŠKI ZALIV (SEVERNO JADRANSKO MORJE, ITALIJA)

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POVZETEK

Med letoma 1990 in 1993 so avtorji vzorčevali in z uporabo uni- in multivariatnih metod analizirali makrobentoške združbe, živeče pri izlivih odplak, ki se po petih podvodnih kanalizacijskih ceveh stekajo v Tržaški zaliv. Ugotovljenih je bilo 19.947 organizmov, pripadajočih 217 taksonom. V makrobentoški združbi so prevladovali mnogoščetinci in mehkužci, saj so sestavljali 92% celotne abundance in 80% števila vseh zabeleženih vrst. Ugotovljeno je bilo, da obstajajo razlike v favnistični sestavi na posameznih vzorčnih postajah in da nanje vplivata bolj sestava usedlin in globina kot pa odplake same, četudi je vzorčenje potekalo neposredno ob njihovih izlivih v Tržaški zaliv. Čeprav je bil biotski indeks nizek na vseh postajah in avtorji niso našli nobenih kazalcev organske obogatitve, je celotna analiza pokazala na določeno mero okoljskega neravnotežja, ki je sicer podobno neravnotežju v večini območij Tržaškega zaliva z mehkim dnom. Te ugotovitve kažejo, da je bilo čiščenje odplak učinkovito pri nadzorovanju škodljivih vplivov urbanih odpadnih voda ali pa da te vsaj niso negativno vplivale na lokalne bentoške populacije med preučevanim obdobjem.

Ključne besede: makrobentos mehkega dna, biotski indeks, kanalizacijske odplake, Tržaški zaliv, Jadransko morje

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APPENDIX / DODATEK

List of the macrofauna species found in the study area with total abundance and frequency as presences of species in the 25 collected samples.
Seznam makrofavnističnih vrst, odkritih v preučevanem območju s skupno številčnostjo in frekvenco pojavljanja vrst v petindvajsetih nabranih vzorcih.

Species	ind. m^{-2}	freq.
Annelida		
Amage adspersa	3	1
Ampharete acutifrons	17	4
Amphicteis gunneri	10	1
Amphitrite variabilis	7	2
Ancistrosyllis groenlandica	117	7
Aonides oxycephala	33	3
Aponuphis bilineata	1773	16
Arabella geniculata	33	5
Aricidea sp.	190	16
Capitellidae indet.	150	8
Ceratonereis costae	7	2
Chaetopterus variopedatus	10	2
Chaetozone setosa	193	5
Chone acustica	43	4
Chone collaris	177	5
Chone duneri	253	9
Cirratulidae indet.	297	17
Cirratulus filiformis	7	1
Cirriformia tentaculata	3	1
Clymene sp.	593	15
Clymenura clypeata	13	2
Dasybranchus caducus	23	3
Dorvillea sp.	3	1
Drilonereis filum	57	6
Euchone rosea	17	2
Euclymene lumbricoides	10	3
Euclymene oerstedi	60	6
Euclymene palermitana	103	5
Eulalia sp.	13	2
Eunereis longissima	3	1
Eunice vittata	993	24
Eupolymnia nebulosa	10	3
Eupolymnia nesidensis	10	3
Glycera alba	3	1
Glycera capitata	3	1
Glycera rouxii	127	13
Glycera sp.	30	4
Glycera unicornis	40	8
Goniada maculata	73	6
Harmothoe extenuata	3	1
Harmothoe sp.	10	2
Jasmineira caudata	3	1
Jasmineira elegans	60	4
Laonice cirrata	197	12
Lumbrineris gracilis	340	15
Lumbrineris latreilli	397	19
Lumbrineris tetraura	20	3
Magelona alleni	13	4
Malacoceros sp.	7	2
Maldane glebifex	1673	19
Marphysa bellii	60	10
Marphysa sanguinea	20	6
Megalomma vesiculosum	30	5
Melinna palmata	380	12
Monticellina dorsobranchialis	43	6
Myriochele heeri	70	1
Myriochele oculata	67	6
Mysta picta	53	9
Myxicola infundibulum	3	1
Nainereis laevigata	3	1
Neanthes caudata	10	1
Nematonereis unicornis	3	1
Nephthys hombergi	3	1
Nephthys hystricis	3	1
Nereis lamellosa	77	11
Nereis rava	23	3
Nereis sp.	63	8
Nicomache sp.	3	1
Nothria conchylega	27	4
Notomastus latericeus	30	3
Notomastus sp.	347	14
Ophiodromus flexuosus	7	2
Orbinia cuvieri	7	1
Owenia fusiformis	193	13

Paraonis sp.	53	7
Pectinaria auricoma	60	7
Pectinaria koreni	170	11
Petaloproctus terricola	23	6
Pherusa monilifera	10	3
Pherusa plumosa	7	2
Phyllodoce laminosa	17	4
Phyllodoce lineata	40	7
Phyllodoce mucosa	3	1
Phyllodoce sp.	23	3
Phyllodocidae indet.	13	1
Phylo foetida	10	3
Pilargis verrucosa	3	1
Piromis eruca	7	1
Pista cristata	183	4
Platynereis dumerilii	3	1
Poecilochaetus serpens	233	16
Polydora ciliata	27	4
Polydora flava	37	7
Polynoidae indet.	27	6
Pomatoceros triqueter	173	10
Prionospio caspersi	103	1
Prionospio cirrifera	27	3
Prionospio malmgreni	207	4
Pseudopolydora antennata	23	5
Pseudopotamilla reniformis	7	2
Sabellidae indet.	23	4
Sabellides octocirrata	3	1
Scalibregma inflatum	10	1
Schistomeringos rudolphii	7	2
Scolaricia typica	10	1
Scolelepis cantabra	3	1
Scolelepis tridentata	7	2
Scoloplos armiger	7	2
Serpula concharum	3	1
Serpula vermicularis	20	2
Sosane sulcata	7	2
Spio filicornis	50	6
Spiochaetopterus costarum	457	17
Spionidae indet.	13	1
Spiophanes kroyeri	33	7
Sternaspis scutata	10	3
Sthenelais boa	43	8
Sthenelais minor	3	1
Sthenolepis hyleni	10	2
Streblosoma bairdi	10	2
Syllis cornuta	13	3
Syllis sp.	3	1
Terebella lapidaria	7	1
Terebellidae indet.	60	3
Terebellides stroemi	160	6
Mollusca		
Abra alba	33	6

Abra prismatica	13	1
Acanthocardia aculeata	7	2
Acanthocardia paucicostata	3	1
Acanthochitona aenea	7	2
Anodontia fragilis	77	11
Anomia ephippium	7	1
Atrina pectinata	40	4
Azorinus chamasolen	10	3
Callista chione	3	1
Calyptraea chinensis	7	2
Chamelea gallina	3	1
Chlamys varia	7	1
Corbula gibba	4667	19
Dentalium inaequicostatum	20	4
Diodora gibberula	3	1
Diplodonta rotundata	37	4
Dosinia lupinus	7	2
Eulima glabra	3	1
Euspira guillemini	3	1
Euspira nitida	10	3
Glycymeris insubrica	3	1
Hiatella arctica	17	4
Lima exilis	3	1
Limea loscombi	3	1
Loripes lacteus	30	6
Lucinella divaricata	403	7
Modiolarca subpicta	7	2
Mysia undata	3	1
Nucula nucleus	227	12
Ostrea edulis	7	2
Paphia aurea	37	6
Pecten jacobaeus	3	1
Phaxas adriaticus	60	10
Philine aperta	20	3
Pitar rudis	53	7
Plagiocardium papillosum	7	1
Proteopecten glaber	3	1
Psammobia fervensis	20	5
Scapharca inaequivalvis	3	1
Solecurtus strigilatus	3	1
Spisula subtruncata	30	3
Striarca lactea	3	1
Tellina distorta	113	12
Tellina serrata	7	1
Thracia convexa	3	1
Thracia pubescens	23	4
Thyasira flexuosa	53	6
Crustacea		
Amphipoda indet.	493	16
Corystes cassivelaunus	10	3
Ethusa mascarone	27	5
Galathea intermedia	3	1
Isopoda indet.	120	11

Macropipus vernalis	3	1
Ostracoda indet.	3	1
Paguridea indet.	10	2
Paguristes eremita	10	2
Pilumnus hirtellus	7	2
Pisidia bluteli	43	8
Pisidia sp.	3	1
Processa edulis	10	2
Processa parva	17	1
Processa sp.	123	13
Echinodermata		
Amphiura chiajei	250	11
Astropecten aranciacus	10	3
Astropecten sp.	7	2
Cucumaria planci	3	1
Holoturidea indet.	3	1
Ophiothrix quinquemaculata	23	4
Ophiura albida	67	9
Ophiura grubei	13	1
Ophiura texturata	20	5
Paracentrotus lividus	10	2
Psammechinus microtuberculatus	17	5
Schizaster canaliferus	27	5
Thyone fusus	27	2
Trachythyone elongata	3	1

Others		
Anthozoa indet.	10	3
Ascidiacea indet.	3	1
Ascidiella aspersa	7	1
Aspidosiphon muelleri	7	1
Branchiostoma lanceolatum	40	4
Calliactis parasitica	3	1
Edwardsia claparedi	3	1
Golfingia sp.	7	1
Golfingia vulgare	13	1
Nemertea indet.	13	3
Phascolosoma sp.	7	1
Phoronida indet.	40	4
Sipuncula indet.	53	7
Sipunculus nudus	30	4
Turbellaria indet.	10	3
TOTAL ABUNDANCE	**19947**	
TOTAL TAXA	**217**	