

THE INFLUENCE OF SEWAGE POLLUTION ON POLYCHAETES ASSOCIATED WITH MUSSEL BEDS OF THE SOUTHERN ROMANIAN BLACK SEA COAST

VICTOR SURUGIU⁽¹⁾

⁽¹⁾ "Alexandru Ioan Cuza" University of Iași, Faculty of Biology, Bd. Carol I, 20A, 700507 Iași, Romania,
e-mail: vsurugiu@uaic.ro

Abstract: A study of the response of the polychaete community associated with the rocky mussel beds to a sewage discharge was carried out in the area of Eforie Sud during summer 2005 and summer 2006. Three stations and a control site were randomly sampled around the Eforie Sud effluent. In each station water quality variables were also measured. A total of 15 species were encountered. The most impacted station, situated at the effluent, had the lowest species richness and diversity. Here only *Alitta succinea* (Frey & Leuckart, 1847) and *Polydora websteri* Hartman, 1943 were present. These two opportunistic species peaked at the station nearest to the effluent, where low inter-specific competition for food and space and high organic matter input prevailed. The control station, situated 5 km from the effluent, had the highest number of species, diversity and evenness. In this site dominant species were *Salvatoria clavata* (Claparède, 1863) and *Platynereis dumerilii* (Audouin & Milne-Edwards, 1833). Uni- and multivariate methods were employed to assess the degree of sewage-induced disturbance on the rocky mussel community. The distribution pattern of the polychaete community was governed especially by pH and dissolved oxygen concentration.

Keywords: Polychaeta, mussel beds, sewage pollution, Black Sea, Romanian coast.

INTRODUCTION

Organic enrichment is probably the most common, and hence the most extensively studied, disturbance on marine environments (Pearson & Rosenberg, 1978). The discharge of domestic waste into the sea leads to a local increase in the quantity of the particulate and dissolved organic matter. This organic matter serves as food for many benthic surface or sub-surface deposit-feeders. If the quantity of domestic waste is large the bottom-dwelling organisms cannot assimilate this increase. As a result of bacterial decomposition of this unassimilated organic matter the concentration of dissolved oxygen diminishes to critical levels. Because oxygen deficiency is the main ecological factor causing severe stress to the macrobenthos, the number of species and the number of individuals will decrease until they disappear altogether.

The use of benthic organisms to assess the impact of human-mediated changes is favoured by the fact that they are relatively sedentary: they cannot avoid the pollution in the way that fishes can; they integrate environmental quality

conditions over longer periods of time compared to plankton; they comprise species with different tolerances to stress; and have an important role in the cycling of nutrients and other chemicals at the water-sediment interface (Reish, 1960, 1973; Gray & Pearson, 1982; Bellan, 1991). However, sorting and identifying of all benthic organisms is very laborious and time-consuming. Thus marine ecologists are searching for cost-effective methods to detect the impact of pollution without the necessity of identifying the whole benthic community.

Among bottom-dwelling organisms, polychaetes are considered one of the most useful indicators of the quality of the marine environment (Reish, 1960, 1963; Pocklington & Wells, 1992; Elias *et al.*, 2003, 2006; Surugiu, 2005). They have been used successfully as bioassay organisms, as monitors for toxic compounds and as pollution indicators at species, as well at population and community, levels (Pocklington & Wells, 1992). The use of polychaetes in biomonitoring studies is of special value because they are extremely responsive to changes in environmental conditions over time, even over a

wide geographic area. The presence or absence, but especially the mass proliferation of certain polychaete species, constitutes a good indicator of the health of benthic habitats. Thus, the species of the family Capitellidae (e.g., *Capitella capitata* complex) and Spionidae (e.g., *Polydora cornuta*, *Streblospio benedicti*, *Malacoceros fuliginosus* etc.) have been widely accepted as indicators of marine organic pollution (Reish, 1957, 1960, 1963; Grassle & Grassle, 1974; Pérès & Bellan, 1972; Pearson & Rosenberg, 1978; Anger, 1975; Losovskaya, 1977; Rygg, 1985; Tsustsumi, 1990; Tsustsumi *et al.*, 1990; Vallarino *et al.*, 2002). In many studies the establishment of the degree of organic pollution was based on the analysis of polychaete communities (Reish, 1955, 1957; Bellan, 1964, 1980; Cognetti & Talierico, 1969; Cognetti, 1972; Dauer & Conner, 1980; Losovskaya, 1983; Elías *et al.*, 2003). Bellan (1980) even proposed an Annelid Index of Pollution to assess the degree of pollution due to municipal sewage.

The Southern Romanian Black Sea coast, situated between Constanta and Mangalia, has high densities of people and industry. As a result, large volumes of polluted water are discharged into the sea in this zone. According to the objectives of the national strategy for the protection of the marine environment, certain actions are required for the preservation of biological diversity and sustainable use of its components. Rehabilitation and protection of the Romanian Black Sea coast and its adjacent marine waters can be achieved only by the reduction of pollution from point sources along the coast and off-shore. The Water Framework Directive (WFD 2000/60/EC) stipulates liabilities to assess biological effects along coastal waters, especially with respect to toxic pollutants. However, studies of the influence of pollution on the benthos in the Romanian littoral zone are very scarce. Țigănuș (1982) studied the composition and distribution of zoobenthos from soft-bottom substrates affected by harbour pollution. Later, she analysed the influence of industrial wastewater discharge on benthic populations and strengthened the importance of the knowledge of benthic communities' status in the assessment of marine pollution (Țigănuș, 1986, 1997).

Thus the aims of this study are: (a) to conduct a detailed analysis of the distribution pattern of polychaetes along an organic pollution gradient, and (b) to investigate the key environmental variables affecting the polychaete community structure.

MATERIAL AND METHODS

STUDY AREA

In order to assess the influence of sewage pollution on polychaete populations, the sampling area was located in the vicinity of the Eforie Sud seaside resort, where an outfall pipe of the local wastewater treatment plant is situated (Fig. 1). This treatment plant uses mechanical and biological processes and receives domestic sewage from the Eforie Nord, Eforie Sud, and Techirghiol towns (a total of about 20,000 inhabitants). Annually it discharges an average of $12.062 \cdot 10^9$ m³ of

sewage, including 178.5 tonnes of ammonia, 10.47 tonnes of nitrites, 62 tonnes of nitrates, 997.5 tonnes of suspended materials and 586 tonnes of Biochemical Oxygen Demand loading into the sea (Petran, 1997). The outlet of the drain-pipe, 70 cm in diameter, is situated at the level of the shoreline (44°01'15.7"N; 28°39'36.9"E) and discharges approximately 0.25 m³ effluent per second. The substrate around the sewage outfall is rocky, consisting of submerged, almost horizontal, Sarmatian limestone platforms.

SAMPLING PROCEDURE

Sampling strategy was of systematic type, having as the final objective the determination of changes in the polychaete community structure in relation to the degree of pollution (i.e., the distance of a station from the outfall). The quantitative sampling was carried out in August 2005, June 2006 and July 2006 at three locations, situated at different distances south of the outfall pipe: one at the effluent outlet (station 1), one 50 m from the sewage outlet (station 2), and one 100 m away (station 3). An additional control site (station C) was also sampled at Agigea, situated approximately 5 km north of the outfall (Fig. 1). In each site 3 replicate samples were taken from 1.5 m depth by removing the mussel epibiosis from an area of 400 cm² with a knife. In order to minimise the loss of organisms during their transportation ashore, the samples were placed *in situ* in polyethylene bags. Samples were immediately fixed in 4% formaldehyde (~10% formalin), rinsed in the laboratory with freshwater and then sieved through a 0.5 mm mesh. The retained polychaetes were sorted under the stereomicroscope, identified to the species level and counted. Sorted and identified animals were preserved in 80% ethyl alcohol.

Some physical and chemical variables of the water column were also measured at each sampling site. The temperature, pH and dissolved oxygen were measured with a portable CONSORT Model C535 Water Quality Meter. The salinity was determined by titration of chloride with a special Aquamerck® kit and subsequent conversion of chlorinity to salinity by means of Almazov's formula (Bondar *et al.*, 1973). The concentration of nitrates, nitrites, ammonia and phosphates was determined spectrophotometrically with a Merck RQFlex Plus device, which uses special Reflectoquant® strips.

DATA ANALYSIS

The structure of the polychaete community was analysed in terms of species composition (*S*), population density (*A*), dominance (*D*), frequency (*F*) and diversity. Density was estimated from the average of the three replicates taken at each station. Dominance was calculated as the percentage of the total abundance of a given polychaete species relative to the total number of polychaetes. The diversity was calculated by the Shannon-Wiener diversity index (*H'*) on a log 2 base (Shannon & Weaver, 1963). The equitability (*J'*) was expressed as Pielou's evenness index (Pielou, 1966). The hierarchical agglomerative clustering (Bray-Curtis similarity coefficient, group-average linking) and the non-metric

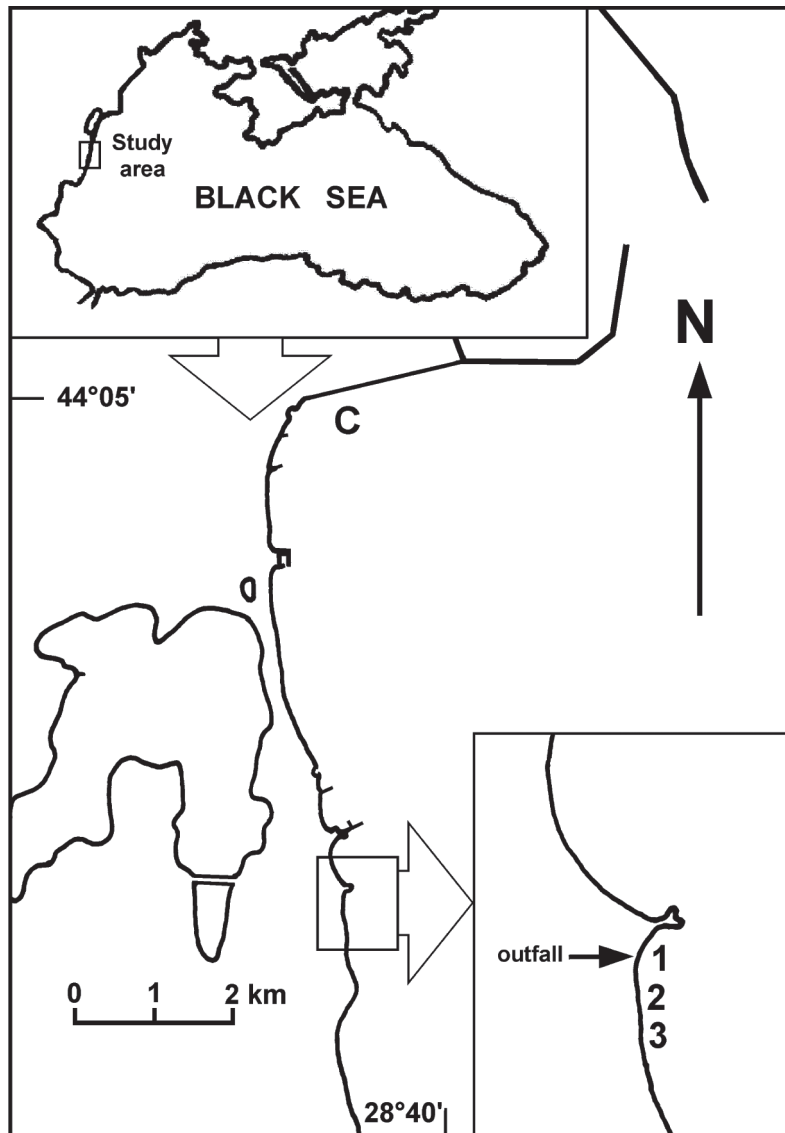


Fig. 1 Sampling area with the location of sampling sites around sewage effluent of Eforie Sud (44°01'15.7"N; 28°39'36.9"E)

Multi-Dimensional Scaling (MDS) were used to investigate faunal similarities among sampling sites (Clarke & Warwick, 1994). Square-root transformation was applied prior to calculating similarities in order to down weight the influence of dominant species.

The similarity breakdown, as proposed by Clarke & Warwick (1994) and implemented in the SIMPER routine ("similarity percentages") within the PRIMER program, was used to define indicator species for areas subjected to sewage pollution and those from unpolluted areas, by analysing contributions of each species to the average similarity (\bar{s}) within groups of sites exposed to different levels of organic enrichment. Discriminating species between unaffected and sewage-polluted sites were defined in the same way, by breaking down the average dissimilarity ($\bar{\delta}$) between different sites into the separate contributions from each species.

Spearman's harmonic rank correlation coefficient (ρ_W), as proposed by Clarke & Ainsworth (1993) and implemented within PRIMER's routine BIO-ENV (Clarke & Gorley, 2001), was used to determine which environmental parameters best correlated with the distribution pattern of the polychaete fauna

Clustering, ordination, similarity breakdown and the calculation of correlation coefficients were carried out using PRIMER v5.0 software package developed at the Plymouth Marine Laboratory.

RESULTS

ENVIRONMENTAL VARIABLES

The values of dissolved oxygen concentration, salinity and pH gradually increased with increasing distance from the effluent outlet (Fig. 2). The highest nutrient concentrations

(nitrates, nitrites, ammonia, and phosphates) were recorded at the effluent outlet (station 1), and the lowest values at the control site (Fig. 3). Differences in nutrient levels between station 1 and the other stations were highly significant ($p < 0.01$). The high variability (standard deviation) in environmental parameters in the area most affected by the sewage station is indicative of a severely fluctuating environment.

THE FAUNA

A total of 4,714 specimens belonging to 15 species were identified during this study. Among them, *Alitta succinea*, *Platynereis dumerilii* and *Polydora websteri* were common to all 4 sampling stations.

Within the study area the most abundant species were *Polydora websteri*, accounting for 45.27% of the total number of individuals, followed by *Salvatoria clavata* (18.16%), *Alitta succinea* (12.77%), *Polydora cornuta* (9.07%), and *Platynereis dumerilii* (8.55%). The remaining species accounted for 6.18% of the total number of specimens (Table 1).

The frequency index values showed that *Alitta succinea* (91.67%), *Polydora cornuta* (75%), *Polydora websteri* (75%), and *Platynereis dumerilii* (58.3%) were constant in the sampling area ($F \geq 50\%$), *Nereis zonata* (41.67%) and *Salvatoria clavata* (25%) were common ($25\% \leq F < 50\%$), and the remaining 9 species were rare ($F < 25\%$) (Table 1).

COMMUNITY PATTERN

The total number of polychaete species gradually increased from 3 species near the sewage discharge to 12 species in the control site (Fig. 4a). The total average density of polychaetes peaked at the intermediate station 2 (50 m from the outfall) and decreased towards the control site (Fig. 4b). The lowest population density at the outfall pipe is due to the shortage of the dissolved oxygen concentration, to the sharp decrease of salinity and to the high concentration of toxic compounds resulting from the decomposition of organic matter (hydrogen sulphide and ammonium).

The Shannon-Wiener's diversity index increased from the sewage outfall to the control site (Fig. 4c). The equitability also increased with distance from the outfall, with the exception of the site situated in the immediate vicinity of the discharge (Fig. 4d), where only two species, each with approximately equal numbers of individuals, were present.

The distribution pattern of polychaete species shows different behaviour according to the distance from the effluent (Fig. 5). *Polydora websteri* and *Alitta succinea* were dominant near the outfall (station 2) with mean densities of 3937 ind. m^{-2} and 1169 ind. m^{-2} , respectively. Both species decreased in abundance towards the control site and were found at very low densities at the effluent (station 1). Thus, their occurrence shows a positive correlation with the sewage concentration. The populations of *Platynereis dumerilii* and *Nereis zonata* increased in abundance with the distance from the outfall and reached their maximum average densities in control station C

(500 ind. m^{-2} and 67 ind. m^{-2} , respectively). Thus, *Platynereis dumerilii* and *Nereis zonata* had a negative relationship with the level of organic enrichment.

Figure 6 shows the similarity dendrogram of the stations sampled for polychaetes. No polychaete species were recorded in station 1 in August 2005, and consequently this sample was not taken into consideration. Two main groups were formed: the first included the control site and the second included the stations more or less affected by sewage.

The MDS ordination plot indicates the existence of a pollution gradient along the horizontal axis, which corresponds to the increase in dissolved oxygen concentration and to the decrease in nutrient levels (Fig. 7). Thus, the samples from the unpolluted area (station C) are placed to the right of the ordination plot and those from polluted areas to the left. The secondary differences in the structure of polychaete assemblages along the vertical axis could be explained by the variation of other abiotic factors, such as the temperature, local hydrodynamics or rate of sedimentation.

The results of computing the contribution from each species to the average similarity \bar{S} within polluted areas are shown in Table 2. The average similarity within the group of stations subjected to sewage pollution is $\bar{S} = 59.08$. The most characteristic or indicator species, which is found at very consistent densities within mussel beds influenced by pollution, is *Polydora websteri*. This species contributes nearly 50% to the average similarity within polluted sites. *Alitta succinea* is the second indicator species for organically polluted areas, which contributes another 38% to the similarities within polluted zones.

Table 3 shows the similarity breakdown into separate contributions from each species to the average similarity within the control site. The total average similarity within this group is $\bar{S} = 65.69$. Over 90% of the contribution to the overall average similarity is accounted for by the first 5 species listed. However, the only species that could be considered characteristic for unpolluted areas were *Platynereis dumerilii*, *Nereis zonata* and *Salvatoria clavata*, as their $\bar{S}_i/SD(S_i)$ ratios were the highest.

The results of breaking down the dissimilarities between samples from sites influenced by sewage and those from the control site into species contributions are presented in Table 4. The total average dissimilarity between control and polluted sites is $\bar{\delta} = 71.92$. Eight species (53% of the total number) are responsible for most of the dissimilarities among stations affected and unaffected by sewage pollution. These species accounted for up to 90% of the observed dissimilarities. However, the only good discriminating species between polluted and unpolluted areas, which had a large $\bar{\delta}_i/SD(\delta_i)$ ratio, are *Salvatoria clavata*, *Platynereis dumerilii*, *Polydora websteri*, and *Nereis zonata*.

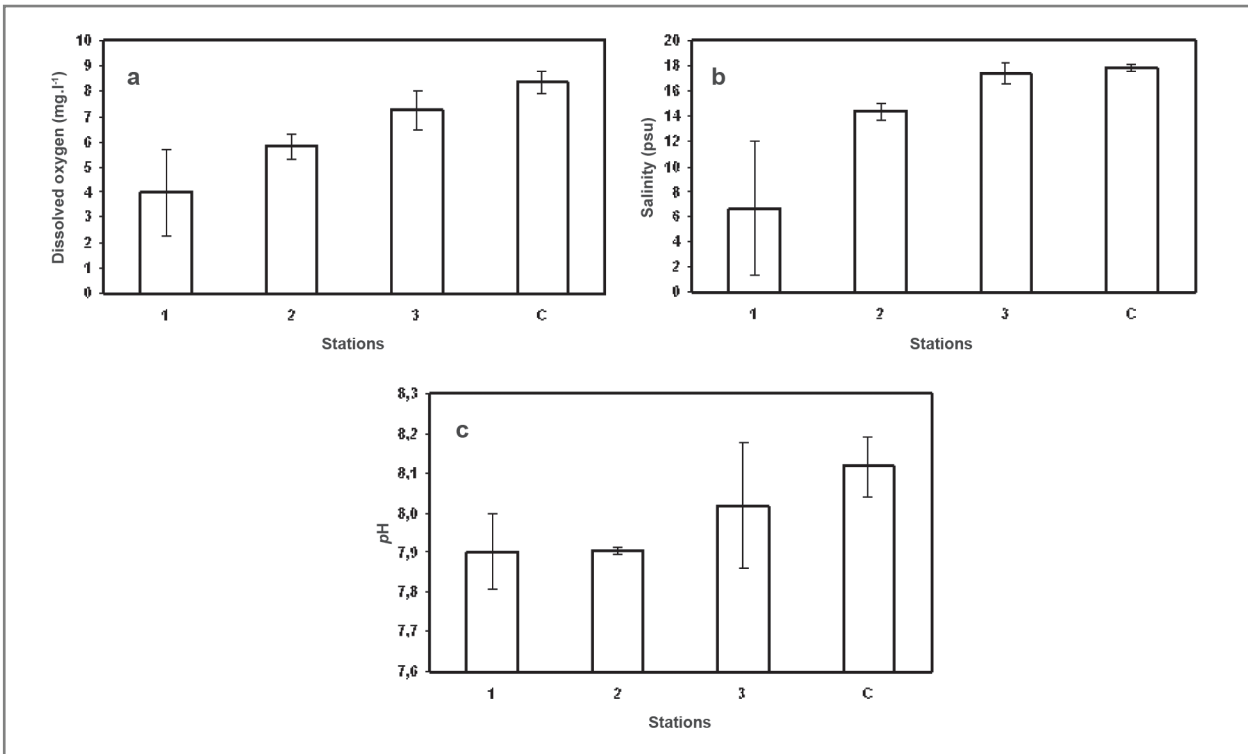


Fig. 2 Mean (\pm SD) of environmental variables in each sampling station: **(a)** dissolved oxygen (mg l^{-1}), **(b)** salinity (psu) and **(c)** pH. Station 1 is situated at the sewage discharge (0 m), station 2 at 50 m from the effluent and station 3 at 100 m. Station C (control) is located ~5 km north of the outfall

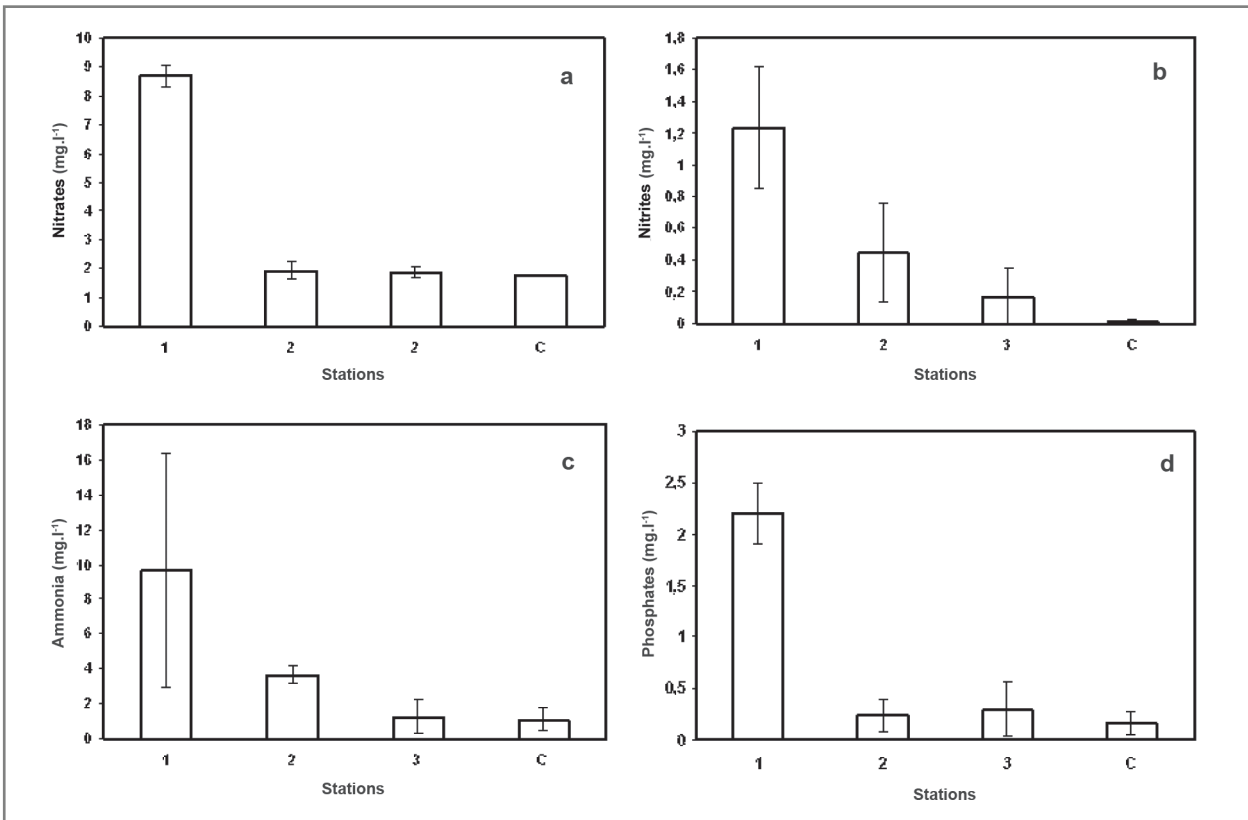


Fig. 3 Mean (\pm SD) of nutrient concentrations in impacted (1 to 3) and control stations (C): **(a)** nitrates (mg l^{-1}), **(b)** nitrites (mg l^{-1}), **(c)** ammonia (mg l^{-1}), and **(d)** phosphates (mg l^{-1}). Station 1 is the closest to the sewage discharge and station 3 is the farthest. Station C (control) is located ~5 km north of the outfall

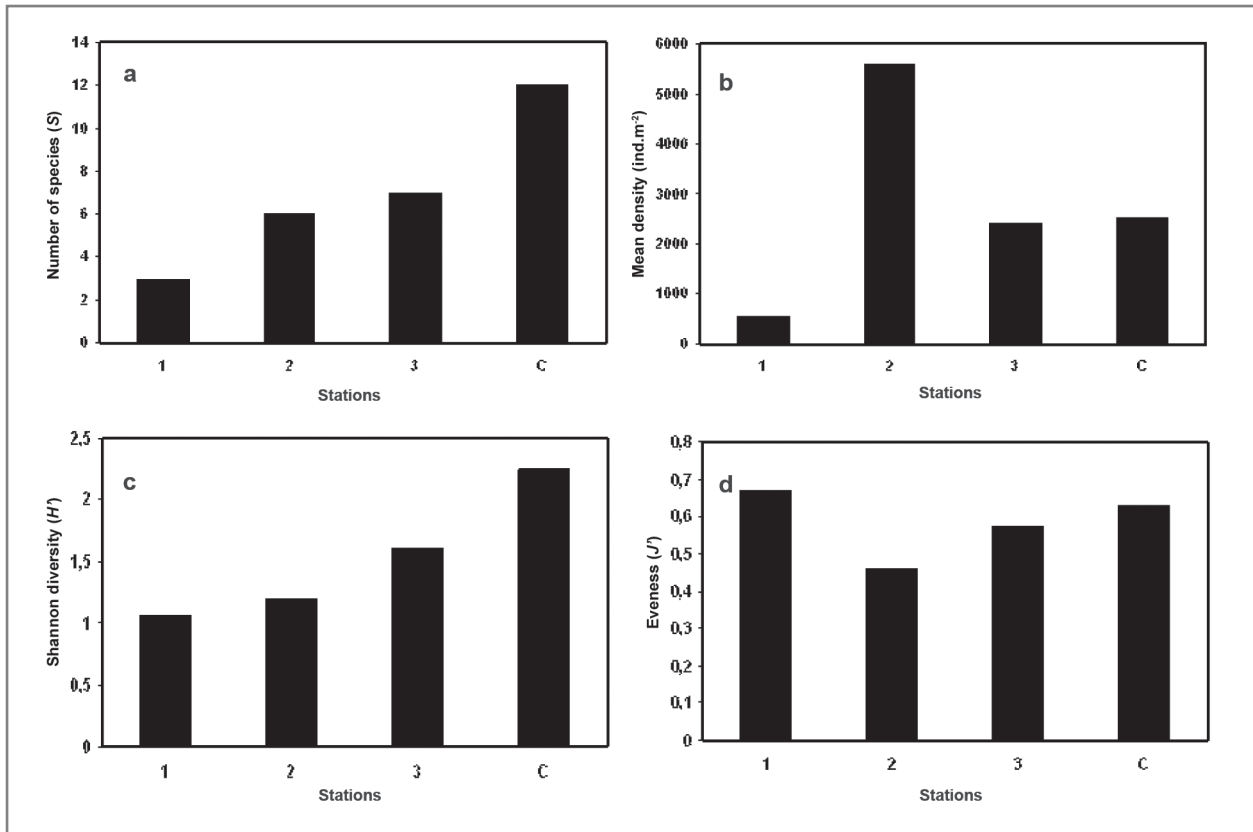


Fig. 4 Variation in univariate measures of the polychaete communities affected by domestic sewage outfall from the Eforie Sud treatment plant: **(a)** the total number of species (S), **(b)** the mean total density (ind. m⁻²), **(c)** the Shannon diversity index (H'), and **(d)** the Pielou's evenness index (J'). Organic enrichment decreases from Station 1 to 3. Station C is the control site

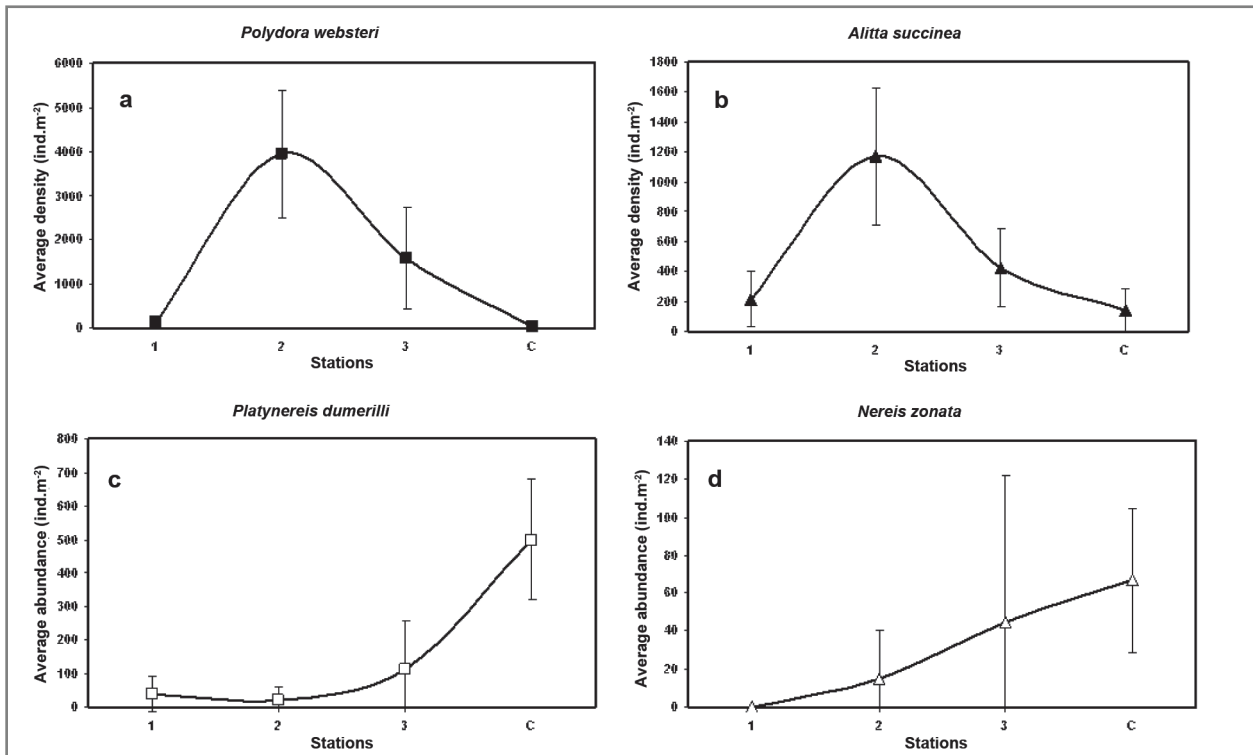


Fig. 5 Average densities of main polychaete species (ind. m⁻²) associated with the intertidal *Mytilus galloprovincialis* beds influenced by domestic sewage according to distance from the outfall: **(a)** *Polydora websteri*, **(b)** *Alitta succinea*, **(c)** *Platynereis dumerilii*, and **(d)** *Nereis zonata* (1 = 0 m, 2 = 50 m, 3 = 100 m, C = control)

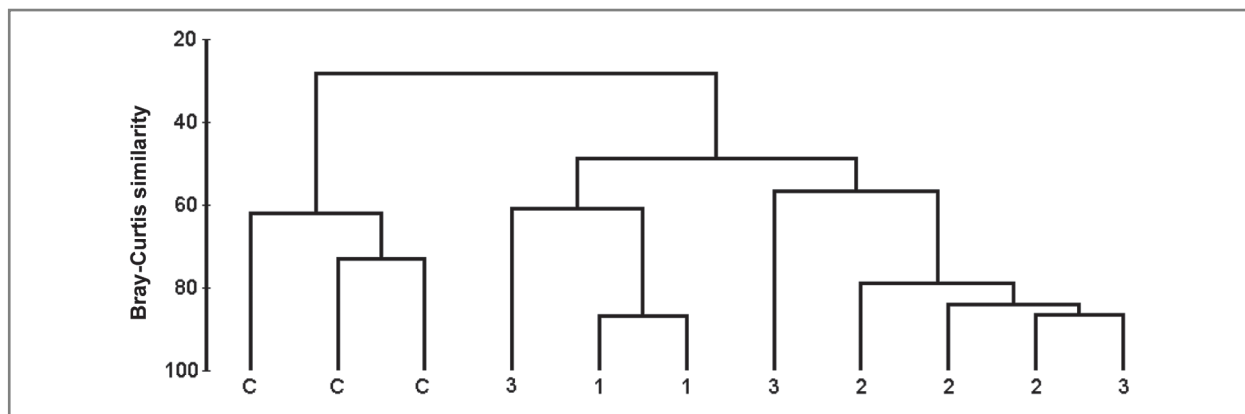


Fig. 6 The dendrogram for hierarchical clustering of the samples taken at different distances from the sewage effluent (1 = 0 m, 2 = 50 m, 3 = 100 m, C = control), based on Bray-Curtis similarities from root-transformed densities (one sample from station 1 not included in the analysis because of lack of polychaetes)

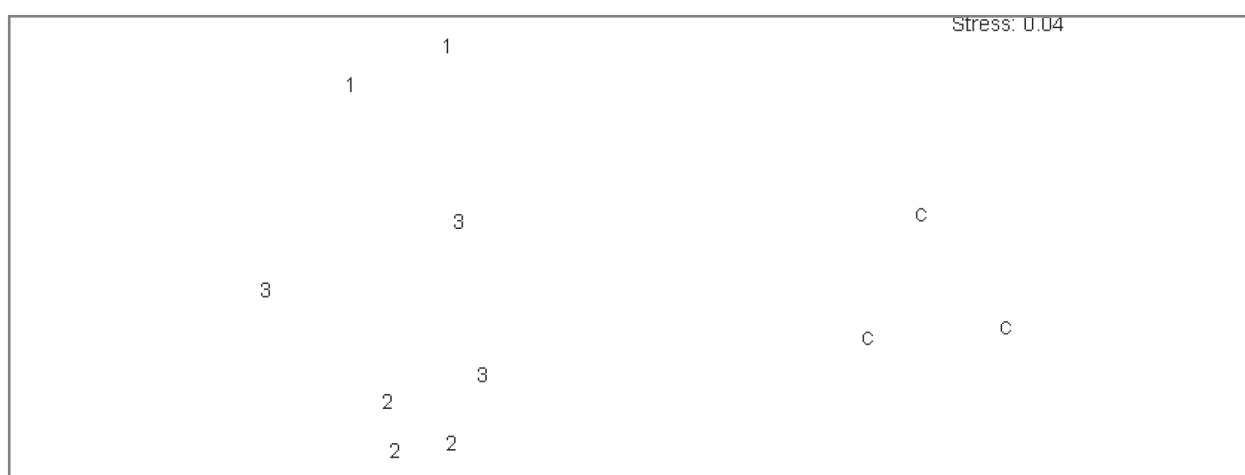


Fig. 7 The MDS ordination plot of the 11 samples taken at different distances from the sewage effluent (except station 1 where one sample contained no polychaetes), based on Bray-Curtis similarities from root-transformed densities (1 = 0 m, 2 = 50 m, 3 = 100 m, C = control)

Table 1 Dominance (*D*) and frequency (*F*) of polychaete species indentified in the *Mytilus galloprovincialis* bed community around Eforie Sud outfall area.

Species	D (%)	F (%)
<i>Polydora websteri</i> (Hartman, 1943)	45.27	75.00
<i>Salvatoria clavata</i> (Claparède, 1863)	18.16	25.00
<i>Alitta succinea</i> (Frey & Leuckart, 1847)	12.77	91.67
<i>Polydora cornuta</i> (Bosc, 1802)	9.07	75.00
<i>Platynereis dumerilii</i> (Audouin & Milne-Edwards, 1833)	8.55	58.33
<i>Nereis zonata</i> (Malmgren, 1867)	4.20	41.67
<i>Capitella minima</i> (Langerhans, 1880)	1.44	8.33
<i>Syllis gracilis</i> (Grube, 1840)	0.15	16.67
<i>Eteone picta</i> (Quatrefages, 1866)	0.11	16.67
<i>Capitella "capitata"</i> (Fabricius, 1780)	0.08	8.33
<i>Syllis hyalina</i> (Grube, 1863)	0.06	16.67
<i>Namanereis littoralis</i> (Grube, 1872)	0.04	16.67
<i>Eulalia viridis</i> (Linné, 1767)	0.04	0.26
<i>Harmothoe impar</i> (Johnston, 1839)	0.02	8.33
<i>Pterocirrus macroceros</i> (Grube, 1860)	0.02	8.33

Table 2 Breakdown of average similarity, within areas subjected to environmental stress due to sewage pollution, into contributions from each species (\bar{S}_i): \bar{A} = average density (ind. m⁻²); $SD(S_i)$ = standard deviation; $\bar{S}_i\%$ = % of the total similarity; $\Sigma\bar{S}_i\%$ = cumulative % of the total similarity (species are ordered by their average contribution \bar{S}_i to the total average similarity $\bar{S} = \Sigma\bar{S}_i$; typical species are indicated by an asterisk).

Species	\bar{A} (ind. m ⁻²)	\bar{S}_i	$\bar{S}_i/SD(S_i)$	$\bar{S}_i\%$	$\Sigma\bar{S}_i\%$
<i>Polydora websteri</i>	2114	29.48	2.77*	49.89	49.89
<i>Alitta succinea</i>	677	22.56	2.95*	38.18	88.08
<i>Polydora cornuta</i>	236	5.18	0.84	8.78	96.86

Table 3 Breakdown of average similarity, within areas unaffected by sewage pollution, into contributions from each species (\bar{S}_i): \bar{A} = average density (ind. m⁻²); $SD(S_i)$ = standard deviation; $\bar{S}_i\%$ = % of the total similarity; $\Sigma\bar{S}_i\%$ = cumulative % of the total similarity (species are ordered in decreasing contribution; typical species are indicated by an asterisk).

Species	\bar{A} (ind. m ⁻²)	\bar{S}_i	$\bar{S}_i/SD(S_i)$	$\bar{S}_i\%$	$\Sigma\bar{S}_i\%$
<i>Salvatoria clavata</i>	1150	19.35	2.35*	29.46	29.46
<i>Platynereis dumerilii</i>	500	16.64	21.85*	25.33	54.79
<i>Polydora cornuta</i>	483	12.47	1.85	18.98	73.77
<i>Alitta succinea</i>	142	5.80	1.98	8.83	82.60
<i>Nereis zonata</i>	67	5.25	3.57*	7.98	90.59

Table 4 Breakdown of average dissimilarity, between samples taken at impacted stations and the station unaffected by sewage pollution, into contributions from each species ($\bar{\delta}_i$): \bar{A} = average density (ind. m⁻²); $SD(\delta_i)$ = standard deviation; $\bar{\delta}_i\%$ = % of the total dissimilarity; $\Sigma\bar{\delta}_i\%$ = cumulative % of the total dissimilarity (species are ordered by their average contribution to the total average dissimilarity, good discriminating species are indicated by an asterisk).

Species	\bar{A} (ind. m ⁻²)		$\bar{\delta}_i$	$\bar{\delta}_i/SD(\delta_i)$	$\bar{\delta}_i\%$	$\Sigma\bar{\delta}_i\%$
	Impacted	Control				
<i>Polydora websteri</i>	2114	17	17.59	2.20*	24.45	24.45
<i>Salvatoria clavata</i>	0	1150	15.99	2.49*	22.23	46.68
<i>Platynereis dumerilii</i>	52	500	9.00	2.33*	12.52	59.20
<i>Polydora cornuta</i>	236	483	6.87	1.15	9.56	68.75
<i>Alitta succinea</i>	677	142	6.43	1.81	8.94	77.69
<i>Nereis zonata</i>	22	67	3.40	2.06*	4.73	82.42
<i>Syllis gracilis</i>	0	58	3.02	1.28	4.20	86.62
<i>Eteone picta</i>	0	42	2.68	1.11	3.73	90.35

RELATIONSHIP BETWEEN ENVIRONMENTAL FACTORS AND POLYCHAETE DISTRIBUTION PATTERN

The matching between the multivariate pattern of polychaete density and environmental variables were assessed by the calculation of the Spearman non-parametric correlation coefficient (ρ_W). The single abiotic variable best correlated with the quantitative distribution pattern of polychaetes is the pH ($\rho_W = 0.600$), followed by dissolved oxygen, nitrites and ammonia ($\rho_W = 0.314$ each). A negative correlation was observed for temperature ($\rho_W = -0.143$), salinity ($\rho_W = -0.143$), phosphates ($\rho_W = -0.200$) and nitrates ($\rho_W = -0.206$). The best 2-variable combination also involves pH but adds dissolved oxygen concentration ($\rho_W = 0.600$). The best 3-variable combination gives the overall optimum value for ρ_W of 0.486. The MDS ordinations based on pH values, and those based on pH and dissolved oxygen values, will group the sites in approximately the same manner as the polychaete community pattern. Thus, the structure of polychaete communities in organically enriched environments is highly consistent with pH and oxygen concentration.

DISCUSSION

Environmental data on water quality show a pattern consistent with an organic enrichment gradient from the effluent to the control site due to the dilution of sewage by marine waters (Reish, 1960; Cognetti & Talierico, 1969; Anger, 1975; Elías *et al.*, 2006). However, this pattern may be greatly altered by local circulation. Due to the exposed character of the Romanian coastline, the north-south long-shore littoral current and strong winter storms (Bondar *et al.*, 1973), there is generally a rapid dispersal of waste discharges.

At the outfall (station 1) the water was black in colour and smelled strongly of hydrogen sulphide. Large strands of mussel shells and decomposing algae were present on the shore. In this grossly polluted area only *Alitta succinea* and *Polydora websteri* were present. These species are very resistant to increased concentrations of toxic compounds and to depletion of dissolved oxygen. But even these very tolerant organisms were suffocated by waste. No polychaetes were recorded in this station in August 2005 due to strong depletion of oxygen and significant freshwater input. Gray *et al.* (2002) indicates that mortality of benthic organisms usually occurs at dissolved oxygen concentrations below 2.0 to 0.5 mg l⁻¹, with polychaetes being less sensitive to hypoxia than crustaceans, but more sensitive than molluscs.

At 50 m from the effluent (station 2) the rocky seabed was covered by dense mats of *Polydora websteri* tubes, muddy and dark in their distal part and sandy at their bases. From place to place the stones were covered by mussels, many of them dead, and by whitish colonies of the filamentous bacteria *Sphaerotilus natans*. Due to the high sedimentation of suspended matter, the *Polydora websteri* tubes covered 95% of the available surface area. This species appears to be favoured by sewage. Generally, species of the genus *Polydora*,

when found in large numbers to the exclusion of others, have been accepted as indicators of organically enriched sediments (Pearson & Rosenberg, 1978). Rygg (1985) considers *Polydora* species to be very tolerant to pollution as they are present in large number in areas characterised by low diversity.

Alitta succinea is another opportunistic species (*sensu* Grassle & Grassle, 1974) enhanced by sewage discharge, and is found in or around organically polluted areas (Pearson & Rosenberg, 1978). However, Elías *et al.* (2006) consider this species as an indicator of moderately enriched sediments.

Besides these two opportunistic species, two more indicator species were present – *Polydora cornuta* and *Capitella capitata*. *Polydora cornuta* (as *Polydora ciliata*) has often been recorded as a dominant species in very polluted environments (Cognetti & Talierico, 1969; Cognetti, 1972; Pérès & Bellan, 1972; Grassle & Grassle, 1974; Anger, 1975). Pearson & Rosenberg (1978) rank *Polydora cornuta* (= *Polydora ligni*) as the second species in a sequence of settlement of azoic areas following an oil spill. *Capitella capitata* is an indicator of the most polluted areas (Reish, 1955, 1963, 1970, 1973; Bellan, 1964, 1967, 1980; Cognetti & Talierico, 1969; Cognetti, 1972; Anger, 1975; Rygg, 1985; Grassle & Grassle, 1974, 1976; Tsustsumi, 1990; Tsustsumi *et al.*, 1990; Elías *et al.*, 2003). This species is very tolerant to anoxic conditions, being capable of surviving prolonged periods of exposure to high concentrations of hydrogen sulphide (Jakubova & Malm, 1930). According to Pearson & Rosenberg (1978) *Capitella capitata* is the first species in the successional sequence of recovery after pollution abatement. Similarly, Losovskaya (1977) considered this species as indicative of the most polluted beaches of Odessa.

At 100 m from the effluent (station 3) the most abundant species were also *Polydora websteri*, *Alitta succinea* and *Polydora cornuta*, but with much reduced densities. This is due to greater pressure from less tolerant but competitively superior species.

In the control site (station C) which is unaffected by organic pollution, the numerically dominant species was *Salvatoria clavata* (1150 ind. m⁻²). This most abundant species was accompanied by *Platynereis dumerilii*. The latter species, despite being abundant in polluted areas, could not be considered an indicator of organically enriched environments because it is also common in undisturbed areas (Bellan, 1980). Grassle & Grassle (1974) consider this species a secondary coloniser as it is very sensitive to anaerobiosis and to the presence of hydrogen sulphide (Jakubova & Malm, 1930; Losovskaya, 1962). Species characteristic of polluted areas were also present, but at low levels of abundance. Conversely, the number and the abundance of sensitive species were considerably higher. Furthermore, Bellan (1980) showed that the absence of sensitive species such as *Nereis zonata* and *Syllis gracilis* is usually indicative of detrimental environmental conditions.

The decrease of the species richness with the increase of pollution intensity was indicated by several authors (e.g. Anger, 1975; Bellan, 1980). For example, Reish (1960) reported 7 polychaete species in areas unaffected by pollution in Los Angeles and Long Beach harbours, 5 species in slightly polluted areas, 3 species in polluted areas, whereas highly polluted areas were characterized by the total absence of polychaetes.

Consequently, the decrease of diversity and evenness indicate an increasing level of environmental stress due to pollution (Clarke & Warwick, 1994; Rygg, 1985). However, the intermediate disturbance hypothesis of Connell (1978) suggests that species diversity rises at intermediate levels of disturbance and falls drastically at still higher levels of disturbance.

CONCLUSIONS

Environmental variables show a spatial gradient of organic enrichment related to increasing distance from the outfall. Poor environmental conditions near the outfall were indicated by low pH and dissolved oxygen values. In the same way, nutrient levels showed highest values near the outfall, indicating high organic matter input in the area.

The dominance of specific polychaetes provided a good indication of the condition or health of a benthic environment. Only species very tolerant to polluted environments, such as *Polydora websteri* and *Alitta succinea*, were present near the outfall. These two opportunistic species attained

highest densities in areas where competitive pressure from other species was reduced.

Salvatoria clavata, *Platynereis dumerilii* and *Nereis zonata* proved to be sensitive to organic pollution, as they occurred in high densities only in the control site.

The community structure of polychaetes from areas affected by domestic sewage appears to be best correlated to dissolved oxygen concentration and to pH.

The assessment of the status of marine environments subjected to organic enrichment, based on the study of the alteration of the community or population units' structure under the influence of a pollution factor, in combination with the physical and chemical analyses, seems to be a very efficient tool in marine monitoring programmes. The use of indicator species allows not only for the estimation of the pollution intensity, but also for the judgement of putative trends of community structure in a given marine area, as a function of the predictable increase in land based pollution.

ACKNOWLEDGEMENTS

Special thanks to Dr. Carol Simon, lecturer at the University of Stellenbosch (South Africa), for correcting the English and for valuable comments on the manuscript. This study was supported by the research grant No. 164/2005-2006 from the National University Research Council (CNCSIS).

REFERENCES

- ANGER, K., 1975. On the influence of sewage pollution on inshore benthic communities in the South of Kiel Bay. Part I. Quantitative studies on indicator species and communities. *Merentutkimuslait. Julk./Havsforskningsinst. Skr.*, **239**: 116-122.
- BELLAN, G., 1964. Influence de la pollution sur la faune annélienne des substrats meubles. *Comm. int. Explor. sci. Mer Médit., Symp. Pollut. mar. Par Microorgan. Prod. pétrol.*, Monaco, 123-126.
- BELLAN, G., 1967. Pollution et peuplements benthiques sur substrat meuble dans la région de Marseille. *Rev. Intern. Oceanogr. Médit.*, **6/7**: 53-87.
- BELLAN, G., 1980. Annélides Polychètes des substrats solides de trois milieux pollués sur les côtes de Provence (France): Cortiou, Golfe de Fos, Vieux Port de Marseille. *Tethys*, **9**(3): 267-278.
- BELLAN, G., 1991. Characteristic, indicative and sentinel species: from the conception to the utilization. In: *Terrestrial and Aquatic ecosystems Perturbation and Recovery*, Ellis Horwood Limited, 95-100.
- BONDAR, C., ROVENȚA, V., STATE, I. (1973) *Marea Neagră în zona litoralului românesc. Monografie hidrologică*. Institutul de meteorologie și hidrologie, București. 516 pp.
- CLARKE, K.R. & AINSWORTH, M., 1993. A method of linking multivariate community structure to environmental variables. *Marine Ecology Progress Series*, **46**: 205-219.
- CLARKE, K.R. & WARWICK, R.M., 1994. *Change in marine communities: an approach to statistical analysis and interpretation*. Plymouth Marine Laboratory, Natural Environment Research Council, 144 pp.
- CLARKE, K.R. & GORLEY, R.N., 2001. *PRIMER v5: User Manual/Tutorial*. PRIMER-E, Plymouth, 91 pp.
- COGNETTI, G., 1972. Distribution of polychaeta in polluted waters. *Rev. Intern. Océanogr. Méd.*, **25**: 23-34.
- COGNETTI, G., TALIERICO, P., 1969. Policheti indicatori dell'inquinamento delle acque. *Pubbl. Staz. Zool. Napoli*, **37**(2 suppl.): 149-154.
- CONNELL, J.H., 1978. Diversity in tropical rain forests and coral reefs. *Science N.Y.*, **199**: 1302-1310.
- DAUER, D.M, CONNER, W.G., 1980. Effects of moderate sewage input on benthic polychaete populations. *Estuarine and Marine Science*, **10**: 335-346.
- DIRECTIVE 2000/60/EC of the European parliament and of the council of 23 October 2000 establishing a framework for Community ac-

- tion in the field of water policy. *Official Journal of the European Communities*, 22.12.2000, **L 327**: 1-72.
- ELÍAS, R., RIVERO, M.S., VALLARINO, E.A., 2003. Sewage impact on the composition and distribution of polychaeta associated to intertidal mussel beds of the Mar del Plata rocky shore, Argentina. *Iheringia, Sér. Zool, Porto Alegre*, **93**(3): 309-318.
- ELÍAS, R., RIVERO, M.S., PALACIOS, J.R., VALLARINO, E.A., 2006. Sewage-induced disturbance on polychaetes inhabiting intertidal mussel beds of *Brachidontes rodriguezii* off Mar del Plata (SW Atlantic, Argentina). In: R. Sardá, G. San Martín, E. López, D. Martín & D. George (eds.) *Scientific Advances in Polychaete Research, Scientia Marina*, **70**(3): 187-196.
- GRASSLE, J.F., GRASSLE, J.P., 1974. Opportunistic life histories and genetic systems in marine benthic polychaetes. *Journal of Marine Research*, **32**(2): 253-284.
- GRASSLE, J.P., GRASSLE, J.F., 1976. Sibling species in the marine pollution indicator *Capitella* (Polychaeta). *Science*, **192**: 567-569.
- GRAY, J.S., PEARSON, T.H., 1982. Objective selection of sensitive species indicative of pollution induced change in benthic communities. I. Comparative methodology, *Marine Ecology Progress Series*, **9**: 111-119.
- GRAY, J.S., SHIU-SUN WU, R., YING OR, Y., 2002. Effects of hypoxia and organic enrichment on the coastal marine environment. *Marine Ecology Progress Series*, **238**: 249-279.
- JAKUBOVA, L.I., MALM, E.N., 1930. Les phénomènes de l'anaérobiose temporaire chez quelques représentants du benthos de la mer Noire. *Doklady Akad. Nauk SSSR*, **14**: 363-366. [in Russian]
- LOSOVSKAYA, G.V., 1962. Otnoşenie nekotoraĥ cernomorskih polihet k izmeneniu solenosti, gazovogo rejima i grunta. *Voprosy ekologii*, **5**: 115-117. [in Russian]
- LOSOVSKAYA, G.V., 1977. *The ecology of polychaetes of the Black Sea*. Naukova Dumka, Kiev, 92 pp. [in Russian]
- LOSOVSKAYA, G.V., 1983. On significance of polychaetes as possible indicators of the Black Sea environment quality. *Ekologiya morya*, **12**: 73-78. [in Russian]
- PETRAN, A., 1997. *Black Sea Biological Diversity. România*. Black Sea Environmental Series, Vol. **4**, UN Publications, 314 pp.
- PEARSON, T.H., ROSENBERG, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *Oceanogr. Mar. Biol. Ann. Rev.*, **16**: 229-311.
- PÉRÉS, J.-M., BELLAN, G., 1972. Aperçu sur l'Influence des Pollutions sur les Peuplements Benthiques. In: M. Ruivo (ed.) *Marine Pollution and Sea Life*, Fishing News, W. Byfleet, Surrey, 3-14.
- PIELOU, E.C., 1966. The measurement of diversity in different types of biological collections. *Journal of Theoretical Biology*, **13**: 131-144.
- POCKLINGTON, P. & WELLS, P.G., 1992. Polychaetes - Key taxa for marine environmental quality monitoring. *Marine Pollution Bulletin*, **24**: 593-598.
- REISH, D.J., 1955. The Relation of Polychaetous Annelids to Harbour Pollution. *Unites States Public Health Reports*, **70**(12): 1168-1174.
- REISH, D.J., 1957. Effect of Pollution on Marine Life. *Industrial Wastes*, **2**: 114-118.
- REISH, D.J., 1960. The use of marine invertebrates as indicators of water quality. In: E.A. Pearson (ed.) *Proceedings of First International Conference on Waste Disposal in the marine Environment*, Pergamon Press, New York, 92-103.
- REISH, D.J., 1963. The Effects of Ocean Water Quality on Bottom-Dwelling Animals. *Division of Water and Waste Chemistry American Chemical Society*, New York, 74-81.
- REISH, D.J., 1970. The effects of varying concentrations of nutrients, chlorinity, and dissolved oxygen on polychaetous annelids. *Water Research*, **4**: 721-735.
- REISH, D.J., 1973. The use of benthic animals in monitoring the marine environment. *Journal of Environmental Planning and Pollution Control*, **1**(3): 32-38.
- RYGG, B., 1985. Distribution of Species along Pollution-induced Diversity Gradients in Benthic Communities in Norwegian Fjords. *Marine Pollution Bulletin*, **16**(12): 469-474.
- SHANNON, C.E. & WEAVER, W., 1963, *The Mathematical Theory of Communication*. University of Illinois Press. Urbana, 125 p.
- SURUGIU, V., 2005. The use of polychaetes as indicators of eutrophication and organic enrichment of coastal waters: A study case – Romanian Black Sea coast. *Analele Ştiinţifice ale Universităţii "Al. I. Cuza" Iaşi, s. Biologie animală*, **51**: 55-62.
- ȚIGĂNUŞ, V., 1982. Données préliminaires sur le zoobenthos du substrat meuble de la zone portuaire Constanţa. *Cercetări marine*, **15**: 107-114.
- ȚIGĂNUŞ, V., 1986. Influenţa deversării de ape reziduale industriale asupra populaţiilor bentale din Zona Năvodari, *Pontus Euxinus. Studii şi cercetări*. Constanţa, **3**: 75-82.
- ȚIGĂNUŞ, V., 1997. Importanţa cunoaşterii stării comunităţilor bentale în aprecierea gradului de poluare a unei zone marine. *Analele Universităţii "Ovidius" Constanţa, Seria Biologie-Ecologie*, **1**(1): 99-104.
- TSUTSUMI, H., 1990. Population persistence of *Capitella* sp. (Polychaeta; Capitellidae) on a mud flat subject to environmental disturbance by organic enrichment. *Marine Ecology Progress Series*, **63**: 147-156.
- TSUTSUMI, H., FUKUNAGA, S., FUJITA, N., SUMIDA, M., 1990. Relationship between growth of *Capitella* sp. and organic enrichment of the sediment. *Marine Ecology Progress Series*, **63**: 147-156.
- VALLARINO, E.A., RIVERO, S., GRAVINA, C., ELÍAS, R., 2002. The community-level response to sewage impact in intertidal mytilid beds of the Southwestern Atlantic, and the use of the Shannon index to assess pollution. *Revista de Biología Marina y Oceanografía*, **37**(1): 25-33.

