



## **Analysis of macrobenthic community structure in relation to different environmental sources of contamination in two littoral ecosystems from the Gulf of Cádiz (SW Spain)**

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### **Abstract**

Selected chemicals in sediments and the abundances and biomasses of macrobenthic species were determined at seven stations from two littoral ecosystems in the Gulf of Cádiz. The marine macrobenthic communities were described in both ecosystems that are subjected to different sources and levels of contamination. A qualitative relationship between source of contamination and biological effects for each station is proposed and the results of the univariate and multivariate analysis used are discussed. Univariate analyses using conventional community descriptive parameters (number of species, abundance and richness, Shannon–Weaver and evenness indices) and the numerical contribution of major taxonomic groups (i.e., Polychaeta, Oligochaeta, Mollusca and Crustacea) could not be used as a means of classifying the sites as clean or polluted with respect to the contamination measured. The results showed that multivariate methods are much more sensitive than univariate techniques. Abundance/biomass comparison (ABC) plots classified the macrobenthic communities into different classes mainly related to organic contamination. However, this analysis did not reflect the alteration due to inorganic sources of contamination. This kind of analysis is valuable for the evaluation of alteration of effects but it cannot discriminate between two different phenomena: pollution (adverse effect associated with chemical concentrations) and alteration (adverse effect associated with chemical concentrations or with natural variations).

### **Introduction**

Measures of benthic community structure in marine ecosystems have been subject to a number of investigations to assessment of *in situ* alteration of residential community structure related to pollution-induced changes for the stated reasons that: (1) the organisms are sedentary, thus reflecting local conditions; (2) many species reside at the same sediment-water interface where many pollutants concentrate; (3) these communities are taxonomically diverse consisting of species that exhibit different tolerances to stress (and be classified into functional groups); (4) the lifespan of

many species allows community structure to integrate and reflect sources of stress over time (indicate and integrate water/sediment quality conditions); (5) are commercially important or are important food sources for economically or recreationally important species, and (6) have an important role in cycling nutrients and other chemicals between the sediments and the water column (i.e., Gray, 1980; Phillips & Segar, 1986; Gray et al., 1988; Weston, 1990).

Two major problems in interpretation of monitoring data are the identification and removal of sources of natural variation in space and time, and the identification of control sites (Holland et al., 1987). Complex

circulation patterns, tidal forcing and steep gradients in physical factors (e.g., salinity and sediment type) in coastal marine and estuarine sites often make the designation of control sites problematical.

Methodologies that do not require control comparisons have been employed (indicator species: Pearson & Rosenberg, 1978; nematode-copepod ratios: Raffaelli & Mason, 1981; lognormal species distribution: Gray, 1979). However, there are several reviews that have been critical of some these methods (i.e., Gray & Pearson, 1982; Warwick, 1988). An alternative to the simple dominance curves are the  $k$ -dominance curves, which are the cumulative ranked abundance and biomass plotted against species ranks (ABC method: Warwick, 1986). This method relies upon  $k$ -dominance curves as visual representations of the evenness of number the individuals and biomass distribution among species. The appeal of the ABC method is that it is easily interpreted, does not require reference to a control site, is presumably insensitive to natural variations in component populations, and may be applied without species-level taxonomic resolution (Warwick, 1986; Warwick et al., 1988).

Multivariate methods have been used in a sizeable number of published studies (e.g., Warwick et al., 1988; Addison & Clarke, 1990; Warwick et al., 1991). These methods are based on species abundances or derived variables at different stations (depending on the choice of similarity or distance measures can take one of many different forms). We used MDS (Multi-dimensional Scaling), one of the most used methods in benthic ecology studies (Clarke & Ainsworth, 1993). In this way, multivariate analysis appears to be an especially sensitive tool for detecting changes in the structure of the faunal community (Warwick & Clarke, 1991).

The scope of the present study was to examine the spatial development of the benthic assemblages by using univariate and multivariate methods. Our aim was to apply some of these techniques to evaluate the state of the marine benthic communities in two littoral ecosystems in the Gulf of Cádiz (SW Spain) in relation to various sources and degrees of contamination measured in those areas. In addition, we attempted to evaluate the different methods used in terms of their sensitivity to alteration effects. In this case, we accepted the postulate that selected chemicals measured in the sediments were suitable to allow identification of contamination sources in the area studied.

## Material and methods

*Approach* The stations selected for this study are located in two littoral ecosystems in the Gulf of Cádiz. The first one is the Bay of Cádiz and the second one is the saltpond of the Barbate River, both shallow water temperate ecosystems (Figure 1). In both areas, approximately half of the area (20 km<sup>2</sup>) has a water depth of less than 3 m and is stagnant. The Bay of Cádiz (along its 41.2 km<sup>2</sup> coastline) supports an intensive marine aquaculture industry and also maintains other important industrial developments such as industries related to the manufacture of car and aircraft components, and seafood industries. The semi-closed Bay supports a human population of approximately 600,000 inhabitants. In addition, the climatic conditions in the Bay have given rise to increasing recreational activities. Five sites were selected in this region while the other two stations selected were in the area of the saltponds in the Barbate River. This river has been characterised as an almost clean area with an amount of urban sewage corresponding to a population of only 20,000. Also, the river presents a strong gradient in salinity (Cordón et al., 1986). Little or no research on the adverse effects of toxic chemicals has been performed on the sediments of these areas (DeValls et al., 1994, 1996, 1997, 1998b). Thus, the stations were chosen based on the best available information to represent presumably low, moderate and high levels of chemical contamination (Gomez-Parra et al., 1990). Stations chosen, in order of decreasing potential anthropogenic influences, were: CB2, CB3, CB5, CB4 and CB1 in the Bay of Cádiz and BR2 and BR1 in the Barbate River saltponds (Figure 1). These stations were sampled for benthic infaunal and sediment chemistry analyses. The following determinations were made: (1) detailed chemical analyses on composited surface sediments and (2) benthic infauna identification of biota retained on a 500  $\mu$ m screen.

*Sample collection.* Sediment samples at all stations were collected with a 0.025 m<sup>2</sup> Van Veen grab. Only grabs that had achieved adequate penetration (2/3 of total volume) to collect the superficial 5 cm of the sediment and that showed no evidence of leakage or surface disturbance were retained for the study. Water depths were constant (3–4 m) along the stations studied. For the benthic infaunal samples, the entire contents of the grab including the overlying water, were wet-sieved at the study site with a 0.5 mm stainless steel screen. Residues were gently washed, placed

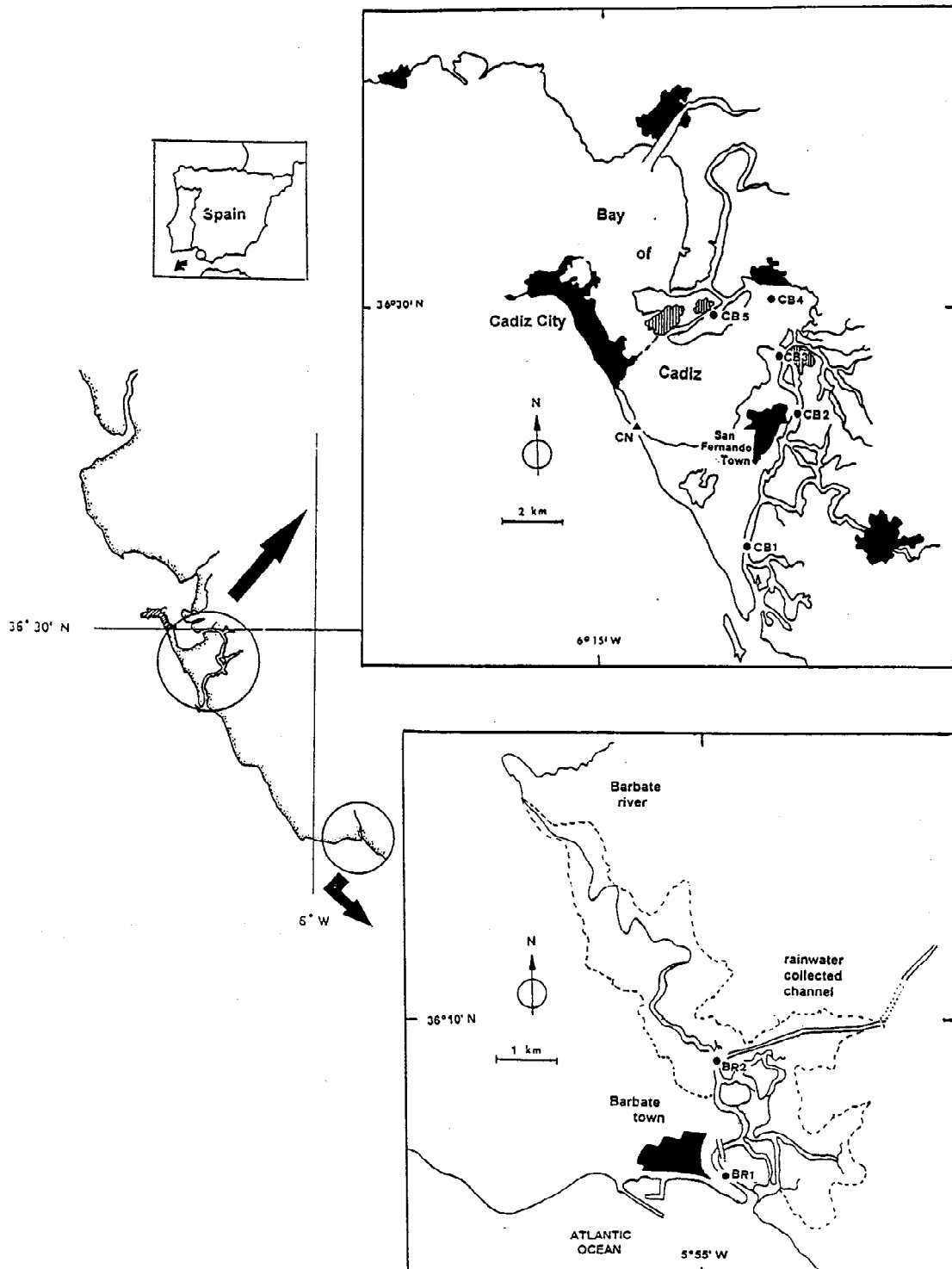


Figure 1. Map of the Gulf of Cádiz showing general areas sampled and locations of sampling stations.

in polyethylene bags, preserved with 10% buffered formalin and stained with Rose Bengal. Ten replicate grabs were collected for infauna at each station. Sediments for chemical analyses were collected and transferred to a cooler. When sufficient sediment had been collected from each station (at least 2L) the contents of the cooler were homogenised with a stainless steel spoon until no colour or textural differences could be detected. Then the coolers, chilled with ice, were transported to the laboratory. Samples were received at the laboratory 6–7 h after collection. Sediments used for analysis of grain size distribution were maintained in the dark at 4 °C prior to processing and analysis. The remaining sediments were dried at 60 °C in an oven.

*Chemical analyses.* Grain size distribution was determined in wet sediment samples by laser diffraction using a Analysette 22 particle size analyser following ultrasonic dispersal in distilled water (Loizeau et al., 1994). Organic carbon content was determined using El Rayis (1985) modification of the Gaudette et al. (1974) method. Elemental analysis was carried out using a CHN Carlo Erba (Model 1106). Surfactants (LAS) were measured using the procedure described by González-Mazo et al. (1997). For trace metal analysis, the sediment dissolution procedures were developed from the total decomposition method originally reported by Loring & Rantala (1992). Fe, Mn, Zn and Cu concentrations in the extracts were determined by Flame Atomic Absorption Spectrophotometry with a Perkin-Elmer 2100. Hg and As concentrations in the extracts were determined by using a Perkin-Elmer MHS-FIAS 400 coupled with a Perkin-Elmer, 4100 ZL spectrophotometer. The other trace metals were measured by graphite furnace atomic absorption spectrophotometry (Perkin-Elmer, 4100 ZL). Accuracy of the assays was assessed by reference to analyses of two certified reference materials (MESS-1 NRC and CRM 277 BCR).

*Benthic infaunal analyses.* Taxonomic analyses involved initially sorting each sample into major constituent taxa. Taxonomic identifications were then performed to the lowest possible level consistent with presently available references, counted, blotted to remove excess formalin and weighed (molluscs without shells). A variety of univariate, graphical/distributional and multivariate methods were employed in the analysis of the biological data set.

(1) Univariate measures; Benthic infauna data analyses were based on community descriptive parameters and abundance analysis which were calculated for each sample and then summarized for each station ( $n = 10$ ). Since they have been used extensively to evaluate pollution effects (cf., Pearson & Rosenberg, 1978, Chapman et al., 1987, Chapman et al., 1996), numerical contribution of major taxonomic groups (i.e., Polychaeta, Oligochaeta, Mollusca, Crustacea) were calculated as the proportions of the taxa abundance to total abundance for each of the 70 samples. Mean proportions, expressed as percentages, were also determined for each station. The descriptive indices used were: species richness (Margalef's R), Shannon–Wiener diversity ( $H'$ ) and evenness (Pielou's J). Numerical dominance, calculated as the complement of equitability (I–J) was related to the proportions of these major taxonomic groups. In order to determine whether there were differences among stations, a one-way ANOVA in each of these univariate measures was used followed by Scheffe multiple comparison test ( $P < 0.05$ ).

(2) Multivariate methods followed those outlined in Clarke (1993). Triangular matrices of similarities between all pairs of samples were computed using (a) the Bray Curtis coefficient for double square-root transformed species abundance data and (b) the euclidean distance dissimilarity coefficient for the environmental data. Clustering was by a hierarchical, agglomerative method using group average sorting, the results of which are displayed in a dendrogram. The species mainly responsible for the dissimilarity between sampling stations were determined using the computer program SIMPER (Clarke, 1993). Significance of differences among stations was tested using the randomisation/permutation test ANOSIM (Clarke & Green, 1988). The ordination analyses were carried out by means of an MDS ('non-metric multidimensional scaling') based on the similarity matrix among stations.

(3) Abundance-biomass plots (ABC) were produced for each station and collection data following the procedures of Warwick (1986) resulting in a total of six plots. The tripartite classification based on Warwick (1986) and Warwick et al. (1987) was used and the results of each station-collection graph were classified as: (a) unstressed, if the biomass curve was above the individuals curve for at least the first three species plotted; (b) highly stressed, if the individuals curve was above the biomass curve for at least the first three species plotted; or as (c) moderately stressed,

Table 1. Contaminants data and sediment physical characteristic (grain size) at different stations. BR1–BR2: stations from Barbate river. CB1–CB5: stations from the Bay of Cádiz

Chemicals	CB1	CB2	CB3	CB4	CB5	BR1	BR2
<b>Conventionals</b>							
TOC (%)	1.39	2.96	2.21	1.82	2.46	0.59	1.86
C/N	13.9	15.6	15.8	26.7	24.6	19.7	15.5
Sand (%)	1.0	1.0	1.1	0.5	0.6	7.1	0.9
Muddy (%)	21.3	22.0	19.2	18.2	21.1	43.2	18.6
Clay (%)	77.7	77.0	79.7	81.3	78.3	49.7	80.5
<b>Metals (ppm)</b>							
Fe	27734	33426	31872	33380	27759	12715	39820
Mn	333	278	332	452	272	262	295
Zn	82	157	163	73	105	34	140
Cu	51.4	69.6	66.4	34.8	49.6	37.4	73.7
Pb	30.5	84.6	64.4	24.4	51.1	66.8	30.0
Cd	0.51	0.67	0.75	0.99	0.81	1.10	0.68
Cr	49.6	77.1	53.0	41.1	283.9	42.5	101.2
Ag	0.48	1.34	1.20	0.78	1.06	0.75	0.61
Hg	0.11	0.25	0.46	0.25	0.57	0.06	0.15
V	79.1	106.5	77.0	80.7	83.3	17.9	147.5
Ni	24.9	35.5	27.9	34.4	32.5	8.2	42.8
Co	7.29	9.16	7.64	10.92	7.78	3.4	11.5
As	11.27	7.72	13.69	8.53	13.24	5.19	9.67
Sn	19.8	17.0	24.0	9.9	18.8	7.4	10.3
<b>Surfactans (ppm)</b>							
LAS	2.2	62.1	12.8	2.6	1.2	1.7	2.5

if the curves crossed for the first three species. To quantify the alteration, the indices SEP (Shannon–Wiener evenness proportion) and DAP (difference between abundance and biomass areas by percent) were used following those outlined by McManus & Pauli (1990).

## Results and discussion

### *Environmental condition description*

Sediment chemical concentrations were determined for organic chemicals represented by the concentration of total organic carbon (TOC), the ratio of organic carbon and organic nitrogen (C/N), and the surfactant alkylbenzenesulphonates (LAS), and for inorganic chemicals represented by 14 heavy metals: Fe, Mn, Zn, Cu, Pb, Cd, Cr, Ag, Hg, V, Ni, Co, As and Sn. This set of chemicals represent the most typical sources of contamination in the studied areas (Gómez-

Parra et al., 1984; Blasco et al., 1996). Summarised results for the contaminant data and physical characteristics of the sediments are shown in Table 1. All stations had a similar texture composition except for BR1 sediments which present a higher percentage of sand (7.1%) due to the proximity of a sand-beach area. The ratio C/N was similar to those reported by other authors in the same area (Forja et al., 1994) ranging from 13.8 at station CB1 to 26.7 at station CB4. These values are higher than those reported by Redfield et al. (1963) associated to an organic matter with a seawater natural origin (6.6). Hence, the organic matter could have an antropoghenic origin, being higher at stations CB4 and CB5. Measured contaminants suggested the presence of various sources and degrees of contamination in each site and station. The presence of Sn and As at CB1 station could be related to recreational nautical activities (Bryan & Langston, 1992) as was reported by Blasco et al. (1996) and DelValls et al. (1998b) for the area studied; station CB2 is affected

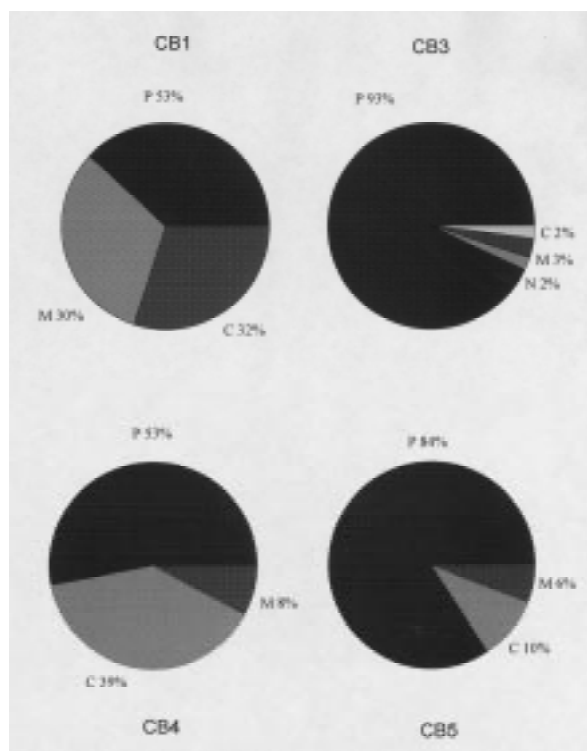


Figure 2. Dominance distribution of the main taxa in sediments from stations selected in the Bay of Cádiz. P = Polychaetes; C = Crustaceans; M = Molluscs; N = Nematodes.

by an urban-point disposal from San Fernando city, and characterised by high levels of TOC, LAS and those heavy metals which have been related to urban sources such as Cu, Pb and Ag (Alsenoy et al., 1993). Station CB3 has a miscellaneous sources of contamination including: high levels of heavy metals related to nautical activities (Hg, Sn and As), and intermediate levels of Pb, Ag, LAS and TOC which may be associated to the proximity of station CB2. Station CB4 has relatively low levels of contaminants and station CB5 was characterised by high levels of Cr, Hg and As, which could be associated with an old industrial disposal point (Gomez-Parra et al., 1990). The two stations in the saltpond of Barbate River (BR1 and BR2) showed, in general, low levels of contaminants, except for Sn at station BR2. More detail on the identification of sources of contamination in the studied areas are given by DelValls (1998a).

#### Community descriptive analysis

A total of 1045 individuals were identified, representing 70 different species. Polychaeta was the best represented

taxon with 35 species, followed by Crustacea with 22 species and Mollusca with 11. Among Crustacea, the order Amphipoda was the best represented and the most abundant while the order Cumacea was only represented by one occasional species. Bivalva was better represented and was more frequent than Gastropoda.

Differences in the occurrence of taxa among the stations of the Barbate River and those in the Bay of Cádiz were observed. The two stations selected in the Barbate River (BR) presented similar faunal assemblages, whereas those stations in the Bay of Cádiz were more different from each other (Figure 2). The communities of the Barbate River are clearly dominated by Oligochaeta (94.12% at BR1 and 81.3% at BR2), followed by Polychaeta (5.73% at BR1 and 18.24% at BR2) with *Prionospio cirrifera* as the most abundant species at both stations. Crustacea and Mollusca were almost absent in these stations (0.1% at BR1 and 0.4% at BR2, 0.04% at BR1 and 0.07% at BR2, respectively), with *Cyathura carinata* as the only crustacean species represented. Stations selected in the Bay of Cádiz sites had differing percentages in the major taxa (Figure 2). Thus, the abundance of Polychaeta varied among the stations studied. At CB1 this abundance was similar to those reached by Mollusca and Crustacea, at CB4 the percentage of Polychaeta increased while that of Crustacea decreased, at CB5 Polychaeta was the most frequent taxa and, at CB3 this group reached its maximum dominance (93%). The stations CB4 and CB1 had a higher percentage of taxa considered to be more sensitive to pollution (Crustacea and Mollusca) than stations CB5 and CB3 where those considered more tolerant (e.g., Polychaeta and Oligochaeta) were higher. With the exception of CB5 where *Hediste diversicolor* was the most frequent polychaete, the most abundant polychaete was *Cirriiformia tentaculata*. The most frequent crustacean species varied among stations. Thus, *Ampelisca spinifer* was the most abundant at CB1, the amphipod *Melita palmata* and the isopod *Cyathura carinata* were at CB3 and *Cyathura carinata* was the most frequent at CB4 and CB5. *Corbula gibba*, *Cerastoderma edule*, *Calyptera chinensis* and *Abra alba* were the most abundant molluscan species at CB1, CB4, CB3 and CB5, respectively. With regard to population parameters, species richness ranged from 0.32 at BR1 to 2.19 at CB1. Both stations, CB1 and BR1 presented the highest and lowest values in diversity, respectively (2.76 and 0.35). Evenness varied from 0.18 at BR1 to 0.78 at CB5 (Figure 3).

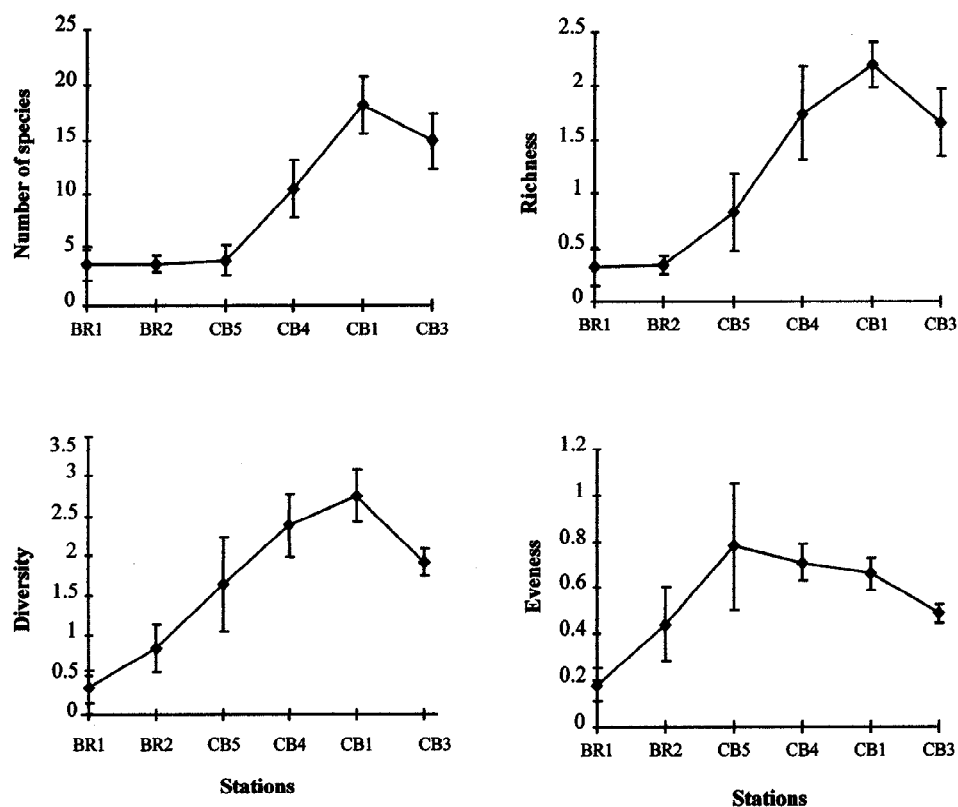


Figure 3. Mean values of the species richness, diversity and evenness indices along the stations. Error bars represent  $\pm$  SD of mean, based on 10 replicates at each station. BR1–BR2, Barbate stations; CB1–CB5, stations from the Bay of Cádiz.

The result of the different ANOVAS and the Scheffe multiple comparison tests ( $P < 0.05$ ) for each univariate measures in each of the stations studied are shown in Figure 4. In order to perform this analysis and integrate the data, we have assumed that clean sites typically have a high number of species, taxa richness and diversity, and the species are equally represented. Additionally, we have assumed that Crustacea and Mollusca are taxa which are sensitive to pollution while Polychaeta and Oligochaeta are less so. This analysis showed the Barbate River stations to be more stressed than those stations at the Bay of Cádiz since the former stations generally had lower values for number of species, richness, diversity and evenness indices. It is important to note in any case that the strong salinity gradient affecting this area could have importance enough to explain this differences (Cordón et al., 1986). With the exception of CB2 station, the Bay of Cadiz stations had higher values for these parameters than those in Barbate River. Furthermore, BR# stations had a higher percentage of Polychaeta, with Crustacea and Mollusca almost ab-

sent. Due to the high variability among stations in the Bay of Cádiz, little information could be obtained and used to classify the different stations of this Bay according to its degree of stress. In general, CB2 which was excluded from the analysis due to an absence of macrobenthic fauna, was the most stressed station. CB3 and CB5 were less stressed than CB2 ( $CB2 > CB3 > CB5$ ) while CB1 and CB4 were considered un-stressed. Indeed, Yong Cao et al (1996) found the diversity indices least informative than the multivariate analyses to point out the change of community structure along the pollution gradient.

#### Faunistic ordination

Figure 5 shows the results of clustering the stations using the Bray–Curtis index for similarities (CB2 station was excluded to prevent modification of patterns and misinterpretation). Cluster analysis distinguished two main groups of sites, which were considered to represent different communities on the basis of their geometric means. The first group (G1) includes

**DECREASED ALTERATION**

➔

	M.S.	BR1	BR2	CB5	CB4	CB1	CB3
<b>S</b>	4.29	3.8	3.8	4.1	10.6	18.2	14.9
<b>A</b>	20.20	13	51	249	429	342	2.
<b>R</b>	1.47	0.32	0.34	0.83	1.66	1.74	2.19
<b>H'</b>	1.54	0.35	0.83	1.64	1.92	2.38	2.76
<b>J</b>	0.21	0.18	0.44	0.49	0.66	0.71	0.78
<b>%P</b>	10.4	92	84	54	52	39	6
<b>%O</b>	34.93	84	82	0	0	0	0
<b>%M</b>	17.74	0.04	0.07	3.42	5.85	11.6	33.7
<b>%C</b>	20.94	0.1	0.4	1.6	6.9	12	27

Figure 4. Summary of ANOVA results for each univariate measure (mean values) ordered in decreasing alteration. Treatments not underlined by the same line are significantly different at  $p < 0.05$  (Scheffe's  $F$  tests). MS, mean squares; S, number of species; A, abundance; R, species richness;  $H'$ , Shannon diversity; J, evenness; %P, percentage of Polychaeta; %O, percentage of Oligochaeta; %M, percentage of Mollusca; %C, percentage of Crustacea; BR1–BR2, Barbate stations; CB1–CB5, stations from the Bay of Cádiz.

three stations (CB3, CB1 and CB4) in the Bay of Cádiz, with CB1 and CB3 the most similar. The most-abundant species found at these stations (the polychaetes *Cirriiformia tentaculata*, *Prionospio Cirrifera*, *Polydora ciliata*, the bivalve *Corbula gibba*, and the amphipod *Ampelisca spinifer*) showed a community similar to that described by Cabioch (1968) which is characteristic of fine sediments. In the second group (G2), the sites correspond with stations in Barbate River and one in the Bay of Cádiz (CB5). Within this group, the strongest similarity (70%) occurs between the two stations in Barbate River. The most frequent

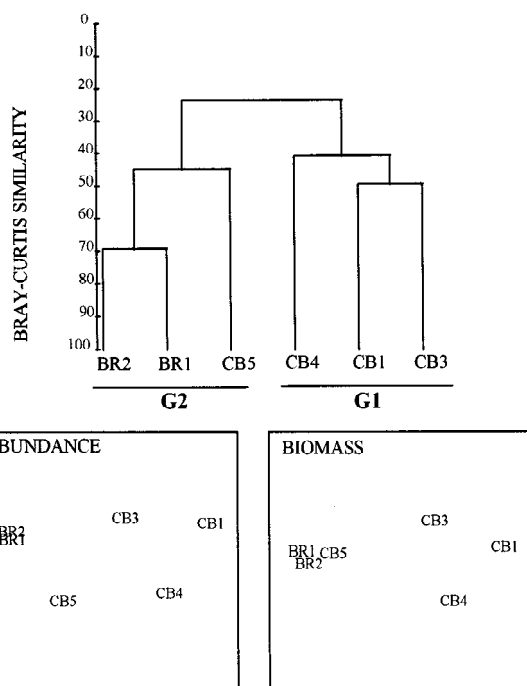


Figure 5. Dendrogram showing classification of six sampling station and two-dimensional non-metric multi-dimensional scaling (MDS) plot of stations based on a similarity matrix abundance species and biomass using Bray–Curtis index. BR1–BR2, Barbate stations; CB1–CB5, stations from Bay of Cádiz.

taxa in G2 group were Oligochaeta, the polychaetes *Prionospio cirrifera* and *Hediste diversicolor* and the isopod *Cyathura carinata*. These species pointed out the presence of a similar community to the one described as 'Reduced Macoma' by Thorson (1957) and characteristic of estuarine areas or sites with salinity stress. In fact, Barbate River is an area strongly influenced by continuous changes in salinity (Cordón et al., 1986) associated with the tidal regime, and because of the extreme narrowness of the estuarine channel (not more than 3 m wide). However, there are differences between those stations of Barbate River, and CB5 since the latter presented species such as *Abra alba*, which has a high tolerance to sediment modifications and quickly adapts to change in the environment (Hily, 1984; Dauvin & Gentil, 1989). This species indicates a transient community between 'Reduced Macoma' and the one called '*Abra alba*' by Thorson (1957). The dendrogram for the species biomass data was virtually identical to the species abundance cluster and is, therefore, not given here.

In terms of abundance, twenty taxa were responsible for more than 70% of the similarity between



Table 2. Comparison of species abundances between groups G1 and G2.  $\delta_i$ : contribution of the  $i$ th species to the average Bray-Curtis dissimilarity ( $\delta_i$ ) between the groups,  $\Sigma\delta_i$  (%): cumulative percentage. Species are listed in decreasing order of importance in contributions to  $\delta_i$ , with a cut-off at 72% of  $\delta_i$ . Average dissimilarity between groups = 76.38 (SD = 7.99)

Species	G2	G1	$\delta_i$	$\delta_i/SD(\delta_i)$	$\Sigma\delta_i$ (%)
Oligochaeta	2341.67	4.67	5.38	1.52	7.05
<i>Cirriiformia tentaculata</i>	6.33	950.67	5.18	3.19	13.83
<i>Corbula gibba</i>	0.00	226.00	2.92	1.65	17.64
<i>Corophium multisetosum</i>	0.00	11.33	2.13	4.54	20.43
<i>Paphia aurea</i>	0.00	12.67	2.07	10.01	23.15
<i>Ampelisca diadema</i>	0.00	15.33	1.98	3.67	28.35
<i>Melinna palmata</i>	1.33	38.67	1.93	1.69	30.88
<i>Prionospio cirrifera</i>	280.00	234.00	1.87	1.32	33.32
<i>Polydora ciliata</i>	0.33	129.67	1.83	0.81	35.72
<i>Paradoneis lyra</i>	0.00	24.00	1.76	1.33	38.03
<i>Metaphoxus</i> sp.	0.00	14.67	1.73	2.51	40.29
<i>Aonides oxycephala</i>	0.00	19.67	1.52	1.25	42.29
<i>Hediste diversicolor</i>	43.33	17.67	1.51	1.35	44.27
<i>Apeudes talpa</i>	0.00	17.33	1.47	1.23	46.20
Nematoda	0.00	24.33	1.46	0.98	48.10
<i>Ampelisca spinifer</i>	0.00	106.67	1.45	0.67	50.01
<i>Cerastoderma edule</i>	1.00	32.00	1.40	1.19	53.73
<i>Ampelisca</i> sp.	0.00	49.00	1.20	0.67	63.85
<i>Photis longicaudata</i>	0.00	43.33	1.16	0.67	65.37
<i>Cyathura carinata</i>	10.67	59.33	0.97	0.87	72.23

G1 and G2 defined by the cluster and MDS analysis (Table 2). As the stations included in G2 group were those which had lower values in diversity, richness indices and smaller number of species, it is not unexpected to find that the principal contributions to the dissimilarity come from species that are abundant in G1 group and largely or totally absent from G2. These species were the polychaetes *Paphia aurea*, *Cirriiformia tentaculata*, and *Melinna palmata*, the amphipods *Corophium multisetosum*, *Ampelisca diadema* and *Metaphoxus* sp and the bivalve *Corbula gibba*. On the other hand, only the Oligochaeta group and two polychaete species: *Prionospio cirrifera* and *Hediste diversicolor* were the most important taxa which were more frequent in group G2 than in G1. All the species present in G2 group were found in G1 group as well.

The two-dimensional MDS configuration for the abundance and biomass data that had the least stress (0.0093 and 0.0092, respectively) are given in Figure 5. These analyses confirm those of the cluster analysis although there was a slight difference between them. The abundance configuration did not consider station CB5 to be as close to the Barbate River stations as do the biomass comparison. However, in both configura-

Table 3. R-statistic values and significance levels in pairwise comparisons of community composition among stations, using the ANOSIM test: BR1–BR2: Barbate stations. CB1–CB5: stations from the Bay of Cadiz

Stations	Statistic values	Significance level
(BR1, BR2)	0.44	0.20
(CB5, BR1)	0.94	0.20
(CB5, BR2)	0.83	0.20
(CB4, BR1)	0.99	0.20
(CB4, BR2)	0.83	0.20
(CB1, BR1)	1.00	0.20
(CB1, BR2)	1.00	0.20
(CB1, CB5)	1.00	0.20
(CB1, CB4)	0.98	0.20
(CB3, BR1)	1.00	0.20
(CB3, BR2)	1.00	0.20
(CB3, CB5)	0.91	0.20
(CB3, CB4)	0.95	0.20
(CB3, CB1)	1.00	0.20

tions there is a clear difference between the stations in Barbate River and those in the Bay of Cádiz and there is none between the two Barbate River stations.

Pairwise comparisons derived from the ANOSIM test on the species abundance data (Table 3) shows that the sites are significantly different from each other ( $p < 1\%$ ).

#### Abundance/biomass comparison

Figure 6 shows the ABC curves for each station (except CB2). Stations BR1 and BR2 exhibit what Warwick (1986) defines as the ‘grossly disturbed’ configuration with the abundance curve above the biomass curve throughout its length. Stations CB3 and CB5 are of the ‘moderately disturbed’ type with the abundance and biomass curves crossing. The other two stations, CB5 and CB1 show the ‘undisturbed’ condition with the biomass curve above the numbers curve of its entire length. In the same figure, the indices SEP and DAP (McManus & Pauli, 1990) were represented with a quantification objective. In this example, Figure 7 is a scatter diagram of natural log SEP values against DAP values. The relationship between these indices is log-linear and could be used to quantify the different status of the systems in relation to the measured alteration. The link among chemical concentrations in sediments and the ABC curves and the two indices

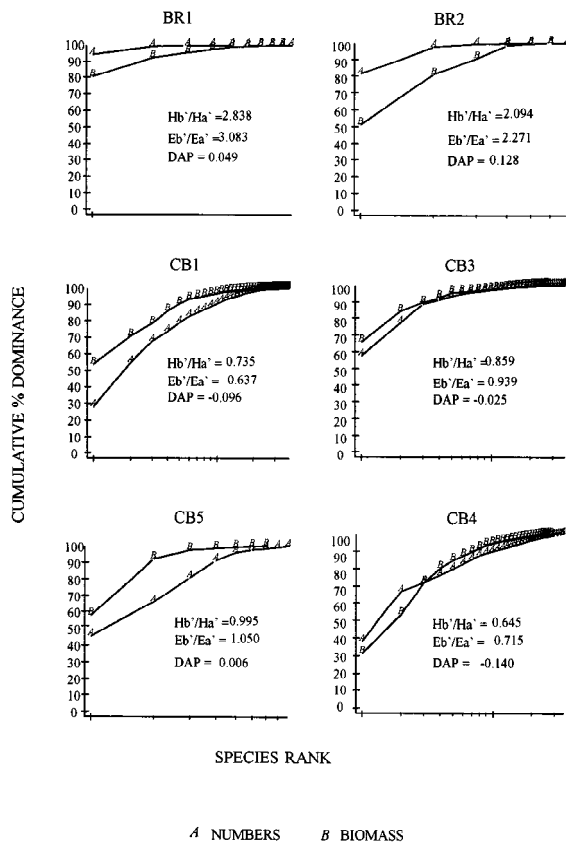


Figure 6. ABC plots for Barbate stations (BR1–BR2) and for stations from the Bay of Cadiz (CB1–CB5).

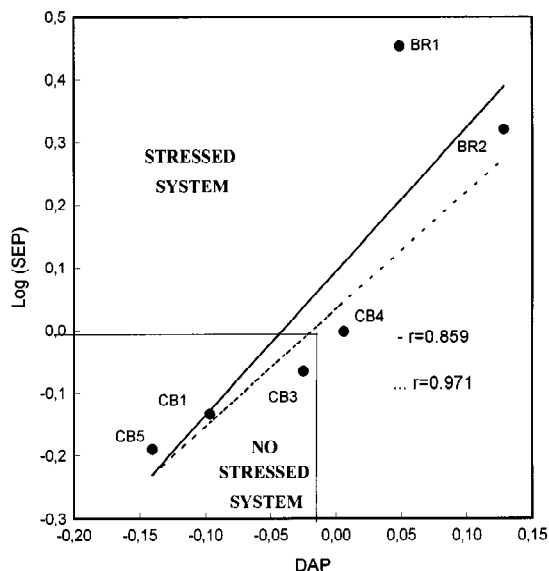


Figure 7. Scatter-diagram of natural log SEP values against DAP values (McManus & Pauli, 1990) used to quantify the ABC plots for the different stations selected in this study.

used to quantify the degree of alteration, (SEP and DAP) shows that the two stations at the saltpond of the Barbate River classified as 'grossly disturbed' and the 'moderately disturbed' CB4 station in the Bay of Cádiz, showed low levels of measured contaminants. Conversely, station CB3 presented moderate high levels of the heavy metals Sn, As, Hg (associated with a shipping) Cu, Pb and Ag (associated with the relatively close urban disposal) and the tensioactive alkylbenzenesulphate (LAS). Finally, the stations CB1 and CB5 (classified as 'undisturbed') showed different levels of chemicals in their sediments. The former, generally, had low levels of chemicals, while the latter had quite high levels of chemicals associated with industrial activities, e.g., chromium. In this sense, we have tried to find some correlations between these indices (DAP and SEP) and the chemical concentrations in sediments. Only LAS and Zn concentrations in sediments showed correlations higher than 0.9 with the DAP index, intermediate correlations were calculated for Ni concentrations ( $r = 0.824$ ), and TOC and the clay fractions ( $r \geq 0.7$ ). Nevertheless, no significant correlations were found among chemicals associated with industrial activities such as As ( $r < 0.2$ ), Hg ( $r < 0.25$ ) or Cr ( $r < 0.3$ ). Warwick (1993) reported that the disturbance status of any one station could have been ascertained without reference to the others. Also, Warwick et al. (1987), and Anderlini & Wear (1992) found that the applicability of this analysis to purely toxic pollution (i.e., without organic enrichment) has not been tested. The results obtained here show, that although the ABC plots are a very good approach for organic enrichment, this analysis is less useful when the sediments are contaminated with inorganic compounds. This assertion should be used with caution as it is based on limited data. Future studies should corroborate this theory. Also, and based on the strong salinity gradient found in the Barbate River we should be cautious when related this variability to only metals. Further and more focused studies should be done to probe differences between metals and salinity effects onto macrofauna structure in the mentioned area.

## Conclusions

This study presents the results of a chemical analysis and an analysis of macrobenthic community structure for several sediment samples. This data provides a *snapshot* of community alteration in sediments collected from two littoral ecosystems in the Gulf of

Cádiz relating the changes to different sources of measured contamination. A qualitative relationship between sources of contamination and biological effect for each station is proposed:

- (a) Station CB2 has the most stressed sediment with high levels of TOC, LAS and Pb which may be related to an urban disposal point.
- (b) Barbate River stations (BR#) have relatively highly stressed communities due to a salinity stress characteristic of estuaries areas, other non-measured contaminants or both.
- (c) Stations CB3 and CB5 have moderately stressed sediments, in the former associated with miscellaneous contamination sources: the industrial activities (associated with high levels of Hg, As and Sn) and the relatively close urban disposal point (with intermediate levels of TOC, LAS and Pb). For CB5, the contamination was related to a disused industrial discharge point (high levels of Hg and principally Cr).

(d) Stations CB1 and CB4 have unstressed sediments. Within the context of this study, we can derive a number of conclusions regarding to the quality of the sediment in the two ecosystems studied and the use of different methods to describe the link between both chemical and biological analysis. These conclusions are summarised below:

(a) As the first step in evaluating the potential alteration of the sediment, the descriptive parameters and univariate measures of the macrobenthic community have been used to establish the benthic structure in the ecosystems studied. However, neither treatment successfully linked the chemical concentrations and the biological structure apparent in the sediments. This was due mainly to the high variability in the final classification of the different stations which resulted from the use of these indices. (b) The ABC plots were used to classify the stations studied and were well correlated with those chemicals associated with an organic source of contamination. Nevertheless, poorer correlations were derived when inorganic chemicals associated with industrial inputs were used. Some indices can be applied to these ABC plots to establish the possible relationship between chemical contamination and adverse biological effect, but further studies are needed to corroborate their utility when there is inorganic enrichment in the sediments.

Finally, this paper shows that to make an overall estimation of the health of sediments there is a need to integrate different environmental measures (combination of physicochemical and biological data).

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