



# Response of benthic macroinvertebrate community descriptors to chemical pollution in the aquatic ecosystems of Fiume Grande and Punta della Contessa saltpans (Brindisi, Italy).

Maurizio Pinna

Lorena Gravili

Alberto Basset

Department of Biological and Environmental Science and Technology, University of Salento, Italy

**Abstract** The anthropogenic activities that take place in an area are reflected in the quality of the waters in the water bodies into which the area drains; these water bodies thus represent natural tools for monitoring the state of health of the whole catchment area. The objective of this study is to evaluate the sensitivity of benthic macroinvertebrate communities in aquatic ecosystems to perturbations linked to chemical pollution. To this end a study was conducted in the aquatic ecosystems located within the Site of National Interest of Brindisi, performing seasonal samplings in 9 field sites. The analysis of the benthic macroinvertebrate communities entailed taxonomic identification, determination of biomass and evaluation of the body condition of each individual sampled. Taxonomic and non-taxonomic community indices were derived, and their models of variation were described and compared on gradients of concentration of chemical contaminants in the area.

The results indicate significant relations between the concentration of heavy metals and descriptors of the size spectra of benthic macroinvertebrates, suggesting that in aquatic ecosystems, descriptors linked to size can be a useful tool in support of bio-monitoring and assessment of ecological risk, considering their ease of application and sensitivity to perturbation pressures in an industrial area.

**Introduction** Anthropogenic activities, particularly those linked to industry, represent important potential sources of perturbation for the structure and functions of ecosystems due to the possible introduction of harmful chemical substances that are used in production processes or are derived from them.

The environmental consequences of these activities are reflected in the abiotic and biotic characteristics of water bodies receiving the run-off waters of the catchment area and the surrounding terrain. Rivers, canals and coastal

lakes thus become “key” ecosystems for the monitoring of the state of health of the entire surrounding area. Because of their capacity to respond in a predictable way to diverse pressures of both natural and anthropogenic origin (Basset *et al.*, 2004; Basset, 1995; Cattaneo *et al.*, 1995; Dadea *et al.*, 1996; Mercier *et al.*, 1999; Ravera, 2001; Solimini *et al.*, 2001; Stead *et al.*, 2005), benthic macroinvertebrate guilds have been incorporated into the regulations concerning the safeguard of waters as biological elements of quality for the evaluation of the ecological state. Although there are numerous studies available that demonstrate the role of ecological forcing factors on the taxonomic characteristics of benthic macroinvertebrate communities, we know little of these relations inside heavily polluted areas or of the dimensional descriptors of these communities.

The aim of this study is to evaluate the sensitivity of high-level descriptors to perturbations linked to chemical pollution of the aquatic ecosystems, using communities of benthic macroinvertebrates. The Site of National Interest of Brindisi was used as a model system, given that the characterization studies conducted under Italian Law DLgs 471/99 have provided us with detailed knowledge of the distribution and concentration of the pollutants present in the site.

Specifically, in this study, the models of spatial variation of taxonomic and non-taxonomic descriptors of the benthic macroinvertebrate guilds were analysed and compared in an area at high environmental risk, and the influence of environmental forcing factors on these models and the response of the taxonomic and dimensional descriptors to these variations were evaluated.

## Material and methods

The study was conducted in the aquatic ecosystems present on the land side of the Site of National Interest of Brindisi, lying to the South of the city (Puglia region, Italy). Inside the site, the industrial activities are located



Figure 1 – Area of study and location of field sites. The areas bounded by hashed lines contain the potential sources of pollution.

both in the Northern part (chemicals factories, a power station and smaller factories) and in the Southern part (a second power station), and are connected by a dedicated transport link used for the movement of coal (Fig. 1).

These two areas may be regarded as the main sources within the site of chemical pollutants. The aquatic ecosystems present in the Site are the canals known as Siedi, Foggia di Rau and Fiume Grande, characterized by the presence of moderately brackish waters, and the Punta della Contessa, a saltpan characterized by high levels of salinity. The canals intersect at various points with the transport link and also flow close to the industrial areas. For this reason they are particularly affected by surface contamination.

The site was the subject of characterization studies carried out to assess the state of contamination of the area; these studies provided detailed information on the concentration in the soil and in the waters of numerous pollutants (e.g. heavy metals, hydrocarbons, chloride-based solvents) linked to the industrial activities that take place in the area (Basset *et al.*, unpublished data). This study was based on seasonal samplings (2 samplings per season) carried out between September 2003 and August 2004 in 9 sampling field sites, located on the Siedi canal (field site 1), the Foggia di Rau canal (field sites 2 and 3), in the Punta della Contessa saltpans (field sites 4 and 5) and along the course of the Fiume Grande canal (field sites 6, 7, 8, 9) (Fig. 1).

At each sampling, using a multi-parametric probe, the main chemical and physical parameters, i.e. temperature, salinity, dissolved oxygen and pH, were recorded, and samples of water were gathered for analysis of content in terms of nutrients ( $\text{NO}_2$ ,  $\text{NO}_3$ ,  $\text{PO}_4$ ,  $\text{NH}_3$ ) (Tab. 1).

The benthic macroinvertebrate communities were sampled using artificial food traps made up of leaves of *Phragmites australis* (leaf-pack technique, Petersen and Cummins, 1974). At seasonal intervals, six packs of reed leaves containing 15 grams dry weight (obtained

	Temperature (°C)	Salinity (PSU)	D.O. (mg/l)	pH	NH <sub>3</sub> /NH <sub>4</sub> <sup>+</sup> (mM)	NO <sub>2</sub> <sup>-</sup> (mM)	NO <sub>3</sub> <sup>-</sup> (mM)	PO <sub>4</sub> <sup>3-</sup> (mM)	Arsenic (mg/Kg)	Cadmium (mg/Kg)	Nickel (mg/Kg)	Lead (mg/Kg)
1	17.25	3.56	11.12	7.59	7.17	11.07	204.83	0.28	5.08	0.06	46.15	3.53
2*	18.67	2.17	10.78	8.02	5.37	3.26	810.13	8.75	6.35	0.03	13.75	5.80
3*	17.27	2.26	11.04	8.08	6.69	7.65	1102.78	10.33	6.35	0.03	13.75	5.80
4#	20.84	14.09	9.69	7.97	13.40	1.58	265.25	4.75	0.50	0.003	1.30	0.50
5#	20.93	10.16	10.31	7.62	5.84	3.48	474.57	1.09	0.50	0.003	1.30	0.50
6	18.67	3.79	10.72	8.10	12.06	6.61	790.25	2.05	14.45	0.78	26.57	15.50
7	20.67	11.44	11.77	7.99	3.45	3.59	852.56	3.84	11.90	1.46	25.55	13.89
8	18.35	2.68	7.95	7.89	3.52	2.96	826.09	4.34	16.76	0.26	25.84	17.69
9	23.15	31.22	7.46	7.85	32.27	0.80	307.49	11.62	25.06	0.40	33.35	21.51

Table 1 – Values for chemical-physical, nutrient and heavy metal concentration parameters observed in each sampling site. \* # : sites 2-3 and 4-5 have similar values for contaminants since they come from the same area.

after drying at 60°C for 72 hours), held in bags made of plastic netting with a 0.5 cm mesh, were immersed in the water at each field site and recovered 30 days later. In the laboratory, the benthic macroinvertebrates were separated from the vegetable detritus and fixed in a 10% solution of formalin; subsequently, each individual was identified at the lowest taxonomic level possible, oven-dried at 60°C for 72h and weighed on a micro-analytical scales (Sartorius, ± 1µg). The individuals, grouped by taxa, were incinerated in a muffle furnace at 500°C for 6h to obtain the content in ash. All biomass values were expressed as Ash-Free Dry Weight in mg.

The quantification of the chemical pressures on the benthic macroinvertebrate communities in the aquatic ecosystems considered was based on the data for the concentration of chemical contaminants detected within a radius of 1 Km upstream of each field site. In this preliminary study, the chemical contaminants considered were the main heavy metals: Arsenic, Cadmium, Nickel and Lead (Tab. 1), since these are easily introduced into the aquatic ecosystems via mechanisms of fall-out and surface run-off, and just as easily combine with organic compounds and enter the food chain.

### Data Analysis

The study of the taxonomic structure of the benthic macroinvertebrate communities was carried out by analysing: abundance, taxonomic wealth, diversity (Shannon and Weaver, 1949) and distinctness (Warwick and Clarke, 1995), while for the dimensional structure, the

analysis encompassed the main statistical descriptors of the size spectra: average, standard deviation, skewness and kurtosis. An empirical measure of statistical width was also considered, obtained as the difference between the maximum and minimum biomass values.

The comparison between taxonomic and dimensional structure was conducted using the Bray-Curtis similarity index (Bray and Curtis, 1957).

Canonical analysis (CCA, Ter Braak 1986) was used to quantify the effect of the environmental variables (metals, nutrients and chemical-physical parameters) on the taxonomic and dimensional structure.

The analysis of the variation of the taxonomic and dimensional characteristics in relation to environmental forcing factors, and specifically to the normalized concentration of the metals, was carried out by means of correlations.

## Results **Taxonomic Structure**

The benthic macroinvertebrate communities sampled were composed of 40 taxa, considering adult and larval phases as distinct, grouped into 5 classes, with a total of 19,504 individuals (Tab. 2). The Insect class accounted for 28 taxa and constituted 31.1% of the total numerical abundance; the Gastropod class accounted for 6 taxa and was the most abundant, with 51.2% of the individuals; this was followed by the Crustacean class with 4 taxa and 15.1% of the abundance; Polychaeta and Hirudinea accounted for just one taxon each and a total abundance of 2.6%. The sampled taxa are shown in table 2 and are ordered by decreasing relative abundance; more than 90 % of the individuals belong to just 7 taxa. The taxonomic indices of the of benthic macroinvertebrate guilds do not exhibit significant variation over time; they do however exhibit significant models of spatial variation in terms of numerical abundance, wealth and taxonomic diversity (one-way ANOVA,  $F(8)= 2.643$ ,  $P<0.05$ ;  $F(8)=4.436$ ,  $P<0.05$ ;  $F(6)=5.168$ ,  $P<0.05$ ) (Fig. 2).

TAXON	A	B	TAXON	A	B
Sadleriana sp	28.235	0.266	Berosus sp (A)	0.103	2.609
Chironomus sp	14.648	0.195	Dytiscidae	0.097	1.239
Hydrobia sp	13.761	0.365	Hydrophilidae	0.087	1.073
Caenis sp	13.351	0.146	Diptera (Pupa)	0.072	0.605
Tanais sp	8.901	0.216	Stratiomyidae	0.046	5.431
Physa sp	6.665	0.216	Ephydridae	0.031	0.077
Gammarus sp	5.686	0.489	Hydraenidae (A)	0.031	0.868
Planorbis sp	2.538	0.358	Hydrobius sp	0.026	2.073
Hediste sp	1.538	4.572	Simuliidae	0.026	0.199
Erpobdella sp	1.051	2.164	Baetis sp	0.021	0.774
Lestes sp	0.754	1.273	Tipulidae	0.015	9.825
Proasellus sp	0.492	0.517	Aeshnidae	0.010	5.475
Berosus sp	0.385	1.133	Ancylidae	0.010	0.877
Tanypodinae	0.303	0.143	Ovatella sp	0.010	0.914
Ephydra sp	0.272	0.691	Tabanidae	0.010	19.121
Crocothemis sp	0.215	2.918	Ceratopogonidae	0.005	0.089
Hydrophilidae (A)	0.174	3.139	Limnephilidae	0.005	14.794
Dytiscidae (A)	0.149	0.741	Notonectidae	0.005	0.045
Syrphidae	0.144	1.306	Ochthebius sp	0.005	0.197
Haliplidae (A)	0.118	1.008	Titanethes sp	0.005	1.191

Table 2 – List of sampled taxa with relative abundance (A) and average individual biomass (AFDW mg) (B).

On the basis of the taxonomic indices, the field sites seem to fall into three groupings; specifically, field sites 2 and 3 have high values for taxonomic wealth, diversity and abundance; field sites 6 and 9 have consistently lower values; field sites 4 and 7 have intermediate values.

### Dimensional Structure

The average individual biomass of the sampled taxa varies between a minimum value of 0.045 mg and a maximum value of 19.120 mg (Tab. 2). On a bi-logarithmic scale, the average individual biomass of each taxon is negatively correlated with numerical abundance (Regression Analysis:  $y=-0.69x+8.23$ ;  $r=0.371$ ;  $g.l.=38$ ;  $P<0.05$ ); the slope value observed is in agreement with the expected models of energetic equivalence between coexisting species. The individuals are distributed in 11 size classes with a logarithmic width of one, and the modal classes are 5 and 6 (Tab. 3). In all the study sites, the size spectra are unimodal and mainly follow a log-normal model. The descriptors of the size structure of



Field site	Body Size Class - Ln(AFDW, $\mu\text{g}$ )												
	0	1	2	3	4	5	6	7	8	9	10	11	12
1													
2													
3													
4													
5													
6													
7													
8													
9													

Abundance
0
1-100
100-200
200-400
400-800
>800

Table 3 - Schematic representation of the distribution in size classes at each sampling site.

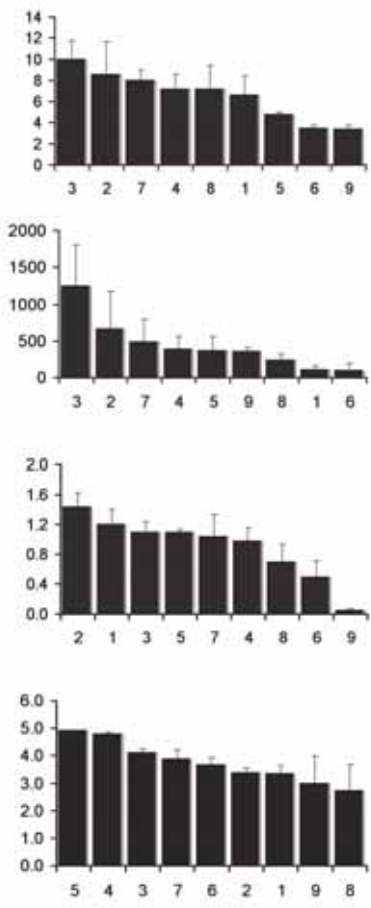


Figure 2 - Spatial variation of taxonomic descriptors. The field sites are listed in decreasing order of observed values.

the of benthic macroinvertebrate communities do not exhibit significant variation over time, while they do exhibit significant models of spatial variation reflected in the standard deviation, skewness and statistical width (one-way ANOVA:  $F(8)=2.239, P<0.05; F(8)=3.585, P<0.05; F(7)=5.134, P<0.05$  respectively). Figure 3 shows the spatial variations in the descriptors considered. On the basis of the dimensional descriptors, the field sites seem to fall into two large groupings that reflect the North-South distribution of the site; specifically, field sites 6, 7, 8 and 9 (in the Northern part) exhibit lower values than field sites 1, 2 and 4, 5 (in the Southern part), which exhibit higher values (Fig. 3).

**Community Structure and environmental forcing factors**

The comparison of the taxonomic and dimensional structure of the macro-zoological benthic communities shows that the similarity in size among the stations is significantly higher than the similarity in terms of taxonomy (Similarity (size) = 60%; Similarity (taxa) = 27%; student's t-test,  $t = 10.99; df = 70; P < 0.01$ ).

Although the variability of the dimensional structure is lower than that of the taxonomic structure, 74.4% of it is explained by the environmental forcing factors considered in the canonical analysis -chemical, physical, nutrient and metal parameters; in contrast the same environmental forcing factors explain only 52.4% of the variability of the taxonomic structure. Among the envi-

ronmental forcing factors, a significant role is played by the concentration of heavy metals. The field sites are spread out along the gradient of concentration of the metals in a spatially explicit way; sites 6, 7, 8 and 9, located along the Fiume Grande canal, have the highest concentrations; sites 1, 2 and 3 have intermediate values; finally, sites 4 and 5 have the lowest values.

The taxonomic and dimensional descriptors considered were correlated with the normalized concentration values for the metallic contaminants; only the measures relating to the dimensions, especially the average, standard deviation and statistical width, exhibit significant patterns of inverse variation with the concentration of the heavy metals measured in the soils immediately surrounding the sites considered (correlation analysis in Fig. 4).

## Conclusions

The study conducted inside the Site of National Interest of Brindisi shows that the benthic macroinvertebrate community varies significantly between sites while it does not appear to be affected by seasonal variability. The spatial variation of the descriptors, used for the analysis of the benthic macroinvertebrate community, leads to the identification of groups of stations which tend to exhibit similar values. For the dimensional descriptors in particular it is possible to identify a group of stations, distributed along the Fiume Grande, that have lower values than the field sites that are further away from the industrial estate and the transport link.

The field sites are arranged in a spatially explicit way along the gradient of concentration of the heavy metals considered: the stations placed along the Fiume Grande have the highest concentrations; the field sites placed along the Foggia di Rau and Siedi, which flow through the agricultural area and intersect with the transport link have intermediate values, and the field sites inside the saltpans have the lowest values.

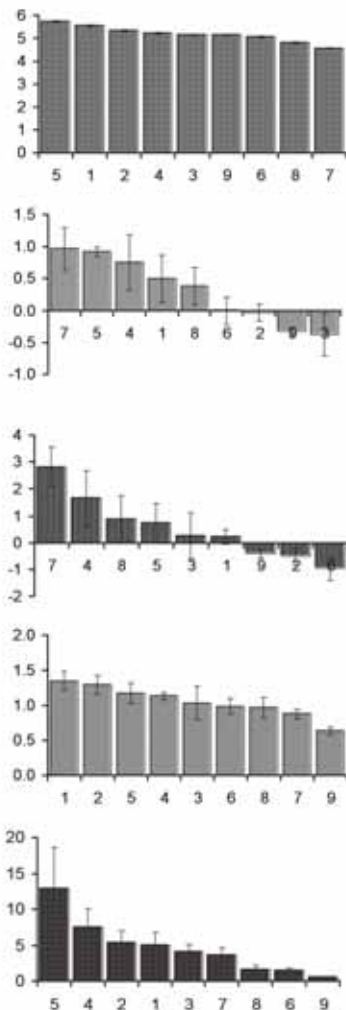


Figure 3 - Spatial variation of dimensional descriptors. The field sites are listed in decreasing order of observed values.

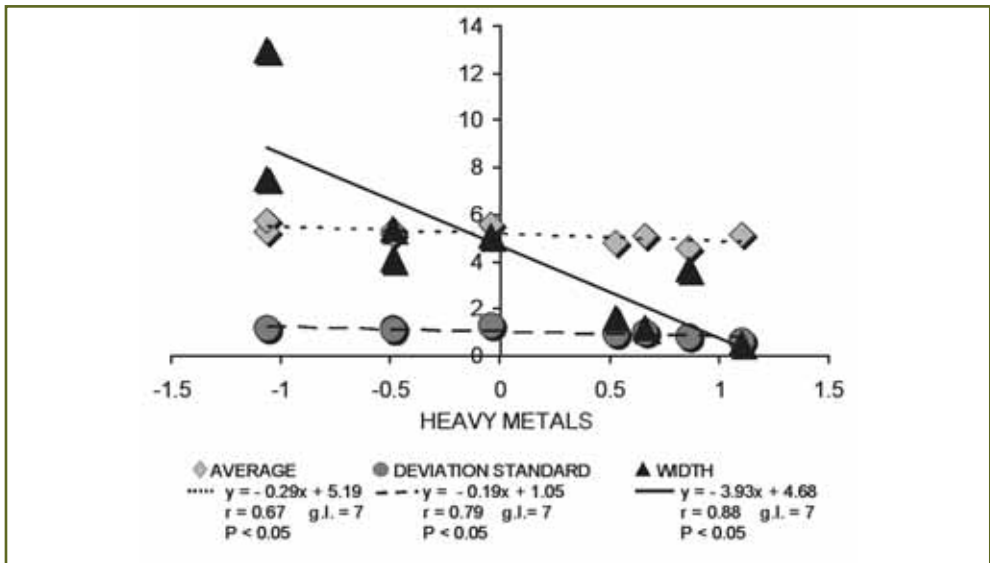


Figure 4 - Analysis of the correlation between the normalized concentration of heavy metals and the descriptors of the dimensional structure of macroinvertebrate communities.

The comparison of the taxonomic and dimensional structures, performed by using the similarity values, shows that the dimensional structure is less variable than the taxonomic structure but that this variability is explained to a greater extent by environmental forcing factors. As already shown by other authors (Basset, 1995; Pinna and Basset, 1997, Basset *et al.*, 2004), in the aquatic ecosystems that are the object of this study the dimensional structure of the benthic macroinvertebrate community is more conservative than the taxonomic structure, showing the importance of body size in the mechanisms of community organization.

The relationship between contaminants and descriptors of the dimensional structure of the benthic macroinvertebrate communities shows that the analysis of body size characteristics, being on a high hierarchical level and not requiring taxonomic identification, make it possible, in a simple, fast and inexpensive way, to measure the response of communities to perturbations induced by anthropogenic activities carried out in an industrial area. Therefore, it is suggested as a useful tool for bio-monitoring and the evaluation of ecological risk.

## References

- Basset A., Sangiorgio F., and Pinna M., 2004. Monitoring with benthic macroinvertebrates: advantages and disadvantages of body size descriptors. *Aquatic Conservation-Marine and Freshwater Ecosystems*, 14: S43-S58.
- Basset A., 1995. Mole corporea ed organizzazione delle guild a base detrito all'interfaccia terra-acqua dolce. *S.It.E. Atti*, 16, 155-160.
- Bray J.R., and Curtis J.T., 1957. An ordination of the upland forest communities of southern Wisconsin. *Ecological Monographs*, 27, 325-349.
- Cattaneo A., Méthot G., Pinel-Alloul B., and Niyonsenga T., 1995. Epiphyte size and taxonomy as biological indicators of ecological and toxicological factors in lake Saint-François (Québec). *Environmental Pollution*, 87, 357-372.
- Dadea C., Ponti A., Basset A., and Caboi R., 1996. Effects of tailing leaching on water chemistry, structure and functions of detritus-based communities. *S.It.E. Atti*, 17, 769-773.
- Mercier V., Vis C., Morin A., and Hudon C., 1999. Patterns in invertebrate and periphyton size distributions from navigation buoys in the St. Lawrence River. *Hydrobiologia*, 394, 83-91.
- Petersen R.C., and Cummins K.W., 1974. Leaf processing in a woodland stream. *Freshwater Biology*, 4, 343-368.
- Pinna M., and Basset A., 1997. Analisi delle distribuzioni taglia-abbondanza in comunità a base detrito nel bacino del fiume Tirso (Sardegna), *S.It.E. Atti* 18, 179-180.
- Ravera O., 2001. A comparison between diversity, similarity and biotic indices applied to the macroinvertebrate community of a small stream: the Ravella river (Como Province, Northern Italy). *Aquatic Ecology*, 35, 97-107.
- Solimini A.G., Benvenuti A., D'Olimpo R., De Cicco M. and Carchini G., 2001. Size structure of benthic invertebrate assemblages in a Mediterranean river. *Journal of the North American Benthological Society*, 20 (3), 421-431.
- Stead T.K., Schmid-Araya J.M., Schmid P.E. and Hildrew A.G., 2005. The distribution of body size in a stream community: one system, many patterns. *Journal of Animal Ecology* 74: 475-487.
- Ter Braak C.J.F. 1986. Canonical Correspondence Analysis: a new eigenvector technique for multivariate direct gradient analysis. *Ecology*, 67(5), 1167-1179.
- Warwick R.M. and Clarke K.R. 1995. New 'biodiversity' measures reveal a decrease in taxonomic distinctness with increasing stress. *Marine Ecology Progress Series*, 129, 301-305.