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Assessing environmental health using ecological indices for soft bottom in sewage-affected rocky shores: The case of the largest seaside resort of SW Atlantic

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ABSTRACT

Efficient ecological indices can reflect the differences between impacted and nonimpacted sites, leading to significant variations at the contamination spatial scale. Here, we evaluated the spatial-temporal variability of 3 ecological indices (AMBI, M-AMBI, and BENTIX) in response to the distinct levels of sewage contamination. The indices were evaluated in two different ways: including *Brachidontes rodriguezii* (IBR) and excluding *B. rodriguezii* (EBR). The fact that mussel beds create a secondary infaunal habitat allows us to test these indices for soft bottoms in areas with rocky bottoms. The effectiveness and the level of agreement of these indices were increased when they were calculated with EBR. BENTIX and M-AMBI produced under- and overestimations of the ecological status of the studied sites. AMBI (EBR) seems to be better suited for environmental quality assessment in the study area. This index reduces the processing time of samples; thus, the AMBI (EBR) index could be used as a robust management tool for monitoring programs in areas with hard substrate.

1. Introduction

Contaminants are widespread in oceans worldwide. The human contribution to ocean contamination is incessant and global; this contribution includes urban and industrial waste, fishing, shipping, dredging, uncontrolled touristic activities, introduction of alien species, and climate change. In particular, coastal areas are the most dynamic but also the most populated areas of the world (Halpern et al., 2007). The need for reliable and accurate indicators of environmental health must therefore be in the agenda of every country with seacoasts. Ecological indices are very useful tools in decision-making processes because they describe the aggregate pressures affecting the ecosystem and can evaluate both the state of the ecosystem and the response of managers (Pintos et al., 2009).

Most of these indices are based on benthic organisms or on their assemblages (Warwick, 1993; Niemi and Mc Donalds, 2004; Simboura et al., 2005; Quintino et al., 2006; Salas et al., 2006; Devlin et al., 2007; Borja et al., 2008; Dauvin et al., 2010). Benthic communities integrate environmental conditions and changes occurred through time in a very effective manner, allowing therefore accounting for the types of disturbances that occur more often in coastal areas (i.e., organic enrichment, physical disturbance, or toxic pollution (Salas et al., 2006). The response of macrobenthic communities to several types of stress is well studied, based on multivariate analyses that consider variations in species diversity and their relative abundance between perturbed and control sites (Pearson and Rosenberg, 1978; Warwick and Clarke, 1993; Gray et al., 2002; Orfanidis et al., 2003; Ballesteros et al., 2007).

The efficiency of biotic indices or any inferences on their suitability requires some degree of congruence with criteria for degraded and undegraded sites based on non-biological measures such as chemical proxies of contamination (Benyi et al., 2009). There are a few works carried out in Latin-American environments; the studies of Muniz et al. (2005, 2011, 2012) were precursors, and similar studies were subsequently conducted for other environments (Omena et al., 2012; Albano et al., 2013; Quiroga et al., 2013; Brauko et al., 2015, 2016).

Mar del Plata city is situated at the Southwest Atlantic coast of Argentina (38° 00′S; 57° 32′W) (Fig. 1). The shoreline is characterized by many sandy open beaches alternating with abrasion platforms of consolidated loess, forming cemented sandstones (Amor et al., 1991). The coastline is influenced by a littoral current, predominantly flowing from South, and undergoes severe windstorms (from the SSE sector) mainly during autumn and winter. Tides have a semidiurnal regime, with a tidal amplitude range around 0.8 m and 1.6 m during exceptional tides. Sea surface temperature ranges between 9.3 °C in winter and 20 °C in summer (Guerrero and Piola, 1997), while seawater pH remains between 7 and 8.5 (Isla et al., 1998).

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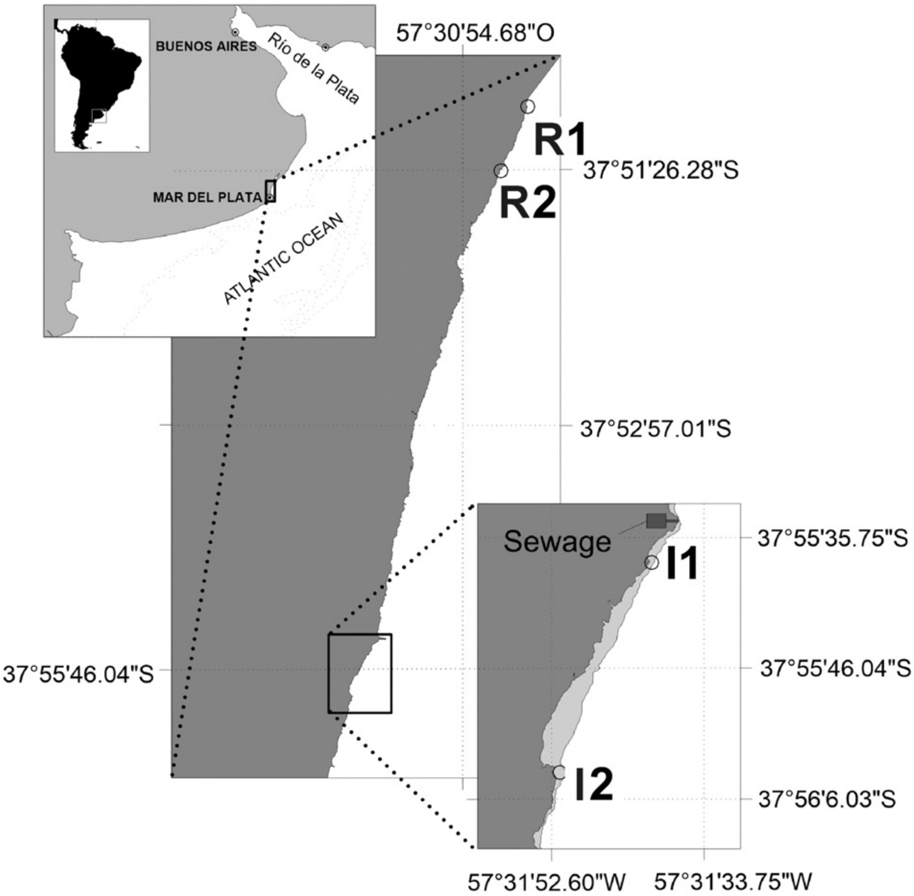

Fig. 1. Distribution of the sampling sites (I1, I2, R1, and R2) and intertidal sewage outfall location in the study area. Site I1 was located at 200 m to the south of the outfall, site I2 at 1000 m south, R1 at 9000 m north, and R2 8000 m to the north of the outfall.

Mar del Plata (ca. 600.000 inhabitants) is the largest seaside resort for sun and beach tourism in Argentina, receiving visits by >3 million people during summer (Bouvet et al., 2005). The city had the worst scenario for both their environmental status and health of people, because raw sewage was directly discharged to intertidal, with a mean volume of 2.8 $m^3 \cdot s^{-1}$ (up to 3.5 $m^3 \cdot s^{-1}$ in summer) (Scagliola et al., 2006). This situation produced 15 km of beaches unfit for bathing people because there was a risk for human health (Comino et al., 2010).

The epilithic intertidal community in natural habitats is dominated by mussel beds of the ecosystem engineer *Brachidontes rodriguezii*, forming a secondary infaunal habitat for several invertebrate fauna. The response of rocky intertidal benthic community to sewage discharges has been evaluated since 1997 (Elias et al., 2015), but no attempt to assess the ecological quality has yet been made. While the assessment of ecological status plays an important role in the management of coastal zones; only a small number of ecological indices are applicable to rocky bottoms (Mangialajo et al., 2007; Juanes et al., 2008; Borja et al., 2012; Díez et al., 2012; Guinda et al., 2014). These indices are very specific for areas where they were developed. An ecologically parsimonious approach dictates that investigators should place greater emphasis on evaluating the suitability of indices that already exist prior to developing new ones (Diaz et al., 2004). Therefore, in this contribution, we apply the most common and widespread indices developed for soft-bottom environments to assess environmental health in response to distinct levels of sewage contamination. The fact that mussel beds create a secondary infaunal habitat allows us to test these indices for soft bottoms in areas with rocky bottoms.

The aim of this study was to evaluate the spatial and temporal variability of the indices in response to the distinct levels of sewage contamination at different sites on intertidal abrasion platforms of Mar del Plata, Argentina. We tested 3 ecological indices, namely AMBI, M-AMBI, and BENTIX. The indices were evaluated in two different ways: (a) including *B. rodriguezii* (IBR) and (b) excluding *B. rodriguezii* (EBR). We hypothesized that the indices should preferably: (a) be highly correlated with the indicators of contamination; (b) vary significantly among impacted and nonimpacted sites; (c) provide values for poorer environmental conditions during the second year of the study due to higher levels of organic contamination; (d) present a high percentage of similarity among responses; and (e) show increased effectiveness when they are calculated without the ecosystem engineer, *B. rodriguezii*, because the abundance of this species at all sites (impacted and reference) reduced the differences between the indices values.

2. Materials and methods

2.1. Study area

The coast of Buenos Aires Province is dominated by sandy beaches; however, around Mar del Plata city, there are quartzite outcrops and almost horizontal intertidal abrasion platforms (geological formation of consolidated loess, limestone, stony rocks, or caliche). The sewage outfall of Mar del Plata city is located 9 km towards the north of the city center (N°11 route, km 507). This intertidal urban effluent discharged 241,920 m^3 of untreated sewage daily during the winter (flow average rate of 2.8 $m^3\ s^{-1}$) and 302,400 m^3 daily during the summer (average of 3.5 $m^3\ s^{-1}$) into the coastal waters (Scagliola et al., 2006), when between 2 and 3 million people visit the city (Bouvet et al., 2005).

Table 1
Calculated indices and their ecological status threshold values.

	Ecological status				
	High	Good	Moderate	Poor	Bad
AMBI	0–1,2	1,2–3,3	3,3–4,3	4,3–5,5	5,5–7
M-AMBI	>0,82	0,82–0,62	0,61–0,41	0,40–0,20	<0,20
BENTIX	6,0–4,5	4,5–3,5	3,5–2,5	2,5–2,0	0–2

2.2. Sampling design and field and lab routines

The study was carried out from February 2008 to November 2009 at four sampling sites on intertidal abrasion platforms: Site 1 (R1), 9000 m to the north of the outfall; Site 2 (R2), 8000 m to the north; Site 3 (I1), 200 m south of the outfall; and Site 4 (I2), 1000 m to the south (Fig. 1). Sites 3 and 4 are considered impacted by sewage inputs and sites 1 and 2 are considered as reference locations (Elías et al., 2009; Jaubet et al., 2011, 2013; Sánchez et al., 2013; Garaffo et al., in press). Sampling (surveys) was conducted seasonally at each site. In each site, we selected three stations 50 m apart from each other at the high intertidal. In each station, 4 sampling units were randomly collected from independent rocks by using a 10 cm-diameter corer (78 cm^2), which was buried into the community matrix up to the basal rocky bottom. The samples were preserved in 7% neutralized formalin solution. In the laboratory, each sample was sieved through a 0.5-mm mesh and the retained organisms were identified, counted, and preserved in a 70% ethanol solution.

Selected environmental variables (turbidity and temperature of the seawater) were measured in situ with a U10 Horiba equipment. The percentage of the total organic matter (TOM) in sediments was determined by the calcination method (Byers et al., 1978).

2.3. Biotic indices and data analysis

To assess the ecological status of the impacted and reference sites, three biotic indices were used: AMBI, M-AMBI, and BENTIX (Table 1). AMBI and M-AMBI values were calculated using the software available at AZTI's webpage (http://ambi.azti.es). The AMBI index is based on the percentage of abundance of five ecological groups according to their sensitivity to organic pollution, already listed in the software (Borja et al., 2000, 2003). According to the species list included in the software package AMBI, most of the species present at Mar del Plata were assigned to a group. However, the assignment of some local species was based on local studies (Sánchez et al., 2013; Sánchez, 2014). After assignment, the mussel *B. rodriguezii* was in GIII, the nemertean *Lineus bonaerensis* in GIII, the polychaete *Protocirrineris angelicolliatio* in GIII, and the amphipods *Caprella dilatata* in GII and *Hyale grandicornis* were in GI.

The M-AMBI index was calculated by the factorial analysis of AMBI, richness (as number of taxa), and Shannon–Wiener diversity values (Borja et al., 2004; Bald et al., 2005; Muxika et al., 2007).

The BENTIX is based on the same proposal as AMBI, but the taxa are categorized in three ecological groups (Simboura and Zenetos, 2002). We adapted the classification of AMBI as following (Blanchet et al., 2007): group I of AMBI is group I of BENTIX; groups II and III of AMBI correspond to II of BENTIX, and groups IV and V of AMBI are group III of BENTIX.

The indices values were calculated for each replicate, and their ecological status was therefore attributed as high, good, moderate, poor, and bad (Table 1). In addition, the indices were evaluated in two different ways: (a) including *B. rodriguezii* (IBR) and (b) excluding *B. rodriguezii* (EBR).

The spatial and temporal variability for each index was evaluated using the ANOVA model. The design incorporated two spatial scales (sites and stations) and one temporal scale (time). The factors of the mixed linear model were as follows: time (fixed) with eight levels, sites (fixed) with four levels, and stations (random) with three levels, nested in sites, with four replicates each. The analysis was not performed for the BENTIX index since the assumptions were not achieved. Principal component analysis (PCA) was carried out to explore the relationships among the biotic indices, the TOM, turbidity, and temperature.

The degree of similarity was also calculated for each possible combination of indices, as the percentage of replicates having the same ecological status. It was evaluated in two different ways: (a) for each possible combination of indices including *B. rodriguezii* (IBR) and (b) for each possible combination of indices excluding *B. rodriguezii* (EBR). Indices with a correlated response should have a high degree of similarity.

3. Results

The general patterns of environmental quality shown by mean values of indices tested indicated distinct ecological status according to the sites. The three indices (according to both IBR and EBR ways) classified the impacted sites (I1 and I2) as poor and moderate classes. The reference sites (R1 and R2) were classified from good to poor depending on the index tested. The overall environmental quality was worse in the second year (2009) (Figs. 2 and 3). The interactions times × sites and times × stations (sites) had a significant effect on the AMBI (IBR),

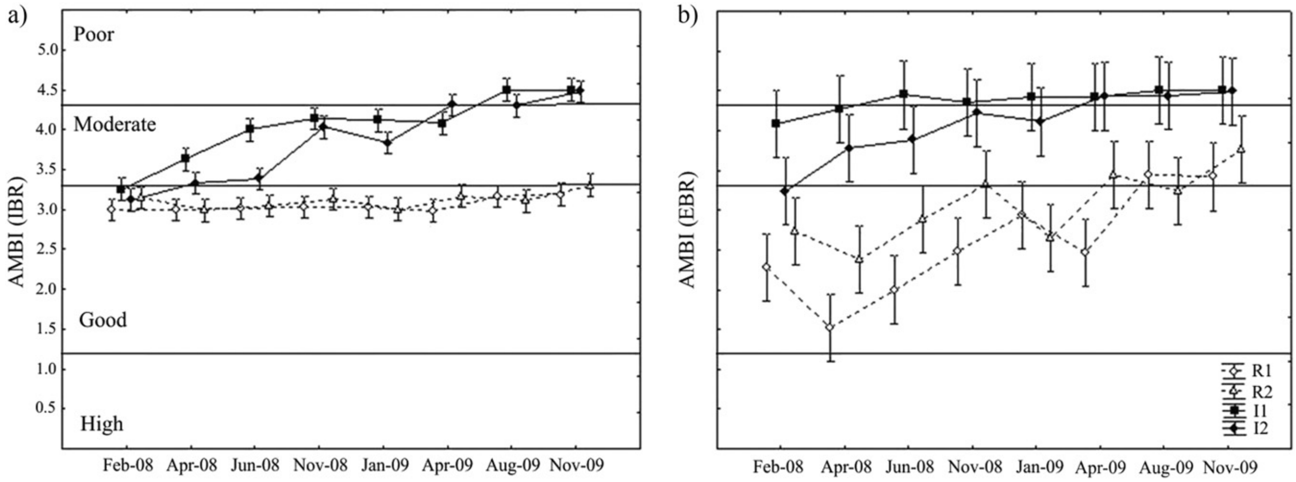

Fig. 2. Mean value (±SE) of AMBI at each site along time: a) including *Brachidontes rodriguezii* (IBR) and b) excluding *B. rodriguezii* (EBR). Lines indicate the ecological status as defined by each index.

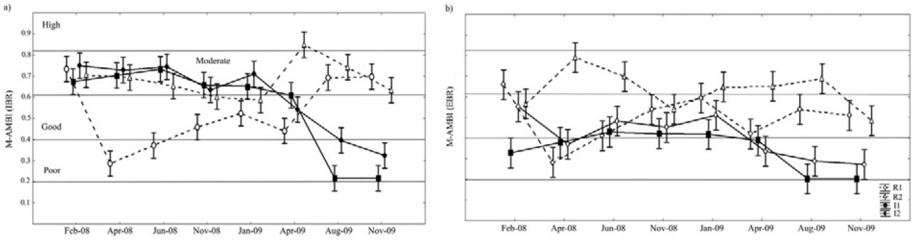

Fig. 3. Mean value (±SE) of M-AMBI at each site along time: a) including *Brachidontes rodriguezii* (IBR) and b) excluding *B. rodriguezii* (EBR). Lines indicate the ecological status as defined by each index.

AMBI (EBR), M-AMBI (IBR), and M-AMBI (EBR). The station (sites) did not have a significant effect on the evaluated indices, thus indicating no variability in this spatial scale (Table 2).

On the basis of the AMBI (IBR), reference sites were classified as good (very close to the limit of moderate status), and the index values showed low variability over time. Impacted sites were classified as moderate and from August 2009 as poor, showing an increase in the index values over time indicating a deterioration of environmental conditions (Fig. 2a)). When *B. rodriguezii* was excluded in calculating the index (AMBI (EBR)), the values in the reference sites showed an increase over time. From August 2009, these sites were classified as moderate. The same trend was observed in the impacted sites. During the last months of the study, these sites were classified as poor (Fig. 2b)). As expected, opportunistic and sensitive species dominated the impacted and reference sites, respectively (Table 3).

M-AMBI (IBR) detected a higher environmental quality status with the highest proportion of good assignments (moderate – high). Until January 2009, the indices values were higher at the impacted sites than at the reference sites, indicating better environmental quality in the former. From August 2009, the impacted sites were classified as poor and the reference sites as good (Fig. 3a)). When *B. rodriguezii* was excluded in calculating the index (M-AMBI (EBR)), the I2 site was classified as moderate until January 2009 and then as poor. On the other hand, the reference sites showed better environmental conditions based on this index (moderate and good status) (Fig. 3b)).

The BENTIX (IBR) and BENTIX (EBR) indices did not exhibit variability over time in any of the sites. The BENTIX (IBR) assigned a worse and lower range of quality classifications, and all the studied stations were classified as poor (index < 2.5). The BENTIX (EBR) showed a similar pattern for impacted sites. However, reference sites were classified from moderate to poor.

The PCA considering the biotic indices and the environmental variables showed eigenvalues of 5.775 and 1.625 for axes 1 and 2, respectively (Fig. 4). The cumulative percentage of variance explained by the first two axes accounted for 82.2% (64.17% and 18.06% for the first and second axis respectively). AMBI (IBR), AMBI (EBR), BENTIX (EBR), temperature, TOM, and turbidity were the most important variables contributing to the formation of the first principal axis. According to axis I, the impacted sites showed the highest values of TOM, temperature, turbidity, and AMBI (IBR and EBR) index, thus indicating a higher degree of organic pollution than at the remaining sites.

The degree of similarity between the evaluated indices including *B. rodriguezii* was low: only 9.4% of the samples were assigned to the same group by different indices. When *B. rodriguezii* was excluded in calculating the indices, this degree of similarity increased to 25.8%. The highest agreement was between AMBI (EBR) and M-AMBI (EBR) (53.4%), followed by AMBI (EBR) and BENTIX (EBR) (42.2%) (Table 4(a) and (b)).

4. Discussion

The present study is the first to calculate and evaluate ecological indices for soft-bottom environment (AMBI, M-AMBI, and BENTIX) in sewage-affected rocky shores of the SW Atlantic. Efficient ecological indices can reflect the differences between impacted and nonimpacted sites, consequently leading to significant variations at the spatial scale of contamination. The effectiveness of these indices and the level of agreement in determining ecological status were increased when they were calculated by excluding the ecosystem engineer *B. rodriguezii*. AMBI seems to be better suited for environmental quality assessment in the study area. Therefore, this index can be used to assess environmental quality in rocky shores, and it can be calculated by considering only the organisms present in the secondary infaunal habitat created by mussel beds. This facilitates and reduces the processing time of biological samples; thus, the AMBI (EBR) could be used as a robust management tool for future monitoring programs in the study area.

The responses of biotic indices to disturbance need to be minimally congruent with indicators of contamination (Benyi et al., 2009). The percentage of the total organic matter and the environmental variables

Table 2
ANOVA results for (a) AMBI (IBR and EBR) and (b) M-AMBI (IBR and EBR). Freedom degrees, mean square (MS), statistical value (F), and p value (p) are presented. Significant differences are shown in bold ($p < 0.01$).

a)	DF	AMBI (IBR)			AMBI (EBR)		
		MS	F	p	MS	F	p
Sites	3	24.141	195.09	<**0.001**	69.231	52,979	<**0.001**
Station (sites)	8	0.124	0.92	0.5044	1.307	2,320	0,0316
Times	7	3.142	23.45	<**0.001**	6.726	11,944	<**0.001**
Times × sites	21	0.782	5.84	<**0.001**	1.239	2,200	**0,0099**
Times × stations (sites)	56	0.134	3.14	<**0.001**	0.563	1,052	0,3856
Residual	288	0.043			0.536		
b)	DF	M-AMBI (IBR)			M-AMBI (EBR)		
		MS	F	p	MS	F	p
Sites	3	0,450	18,057	<**0.001**	1,482	68,872	<**0.001**
Station (sites)	8	0,025	0,935	0,4957	0,022	0,661	0,7234
Times	7	0,271	10,159	<**0.001**	0,157	4,814	<**0.001**
Times × sites	21	0,344	12,923	<**0.001**	0,123	3,776	<**0.001**
Times × stations (sites)	56	0,027	2,368	<**0.001**	0,033	2,011	<**0.001**
Residual	288	0,011			0,016		

Table 3
Dominant species in impacted and reference sites. The respective ecological group according to AMBI/M-AMBI (EGI, EGII, EGIII, EGIV, and EGV) and BENTIX (EGI, EGII, and EGIII) classifications are also shown.

Sites	Species	Ecological group AMBI, M-AMBI	Ecological group BENTIX
Reference sites (R1 and R2)	*Brachidontes rodriguezii*	III	II
	Syllis gracilis	II	II
	Syllis prolixa	III	II
	Siphonaria lessoni	I	I
Impacted sites (I1 and I2)	*Boccardia proboscidea*	IV	III
	Monocorophium insidiosum	III	II
	Capitella capitata	V	III

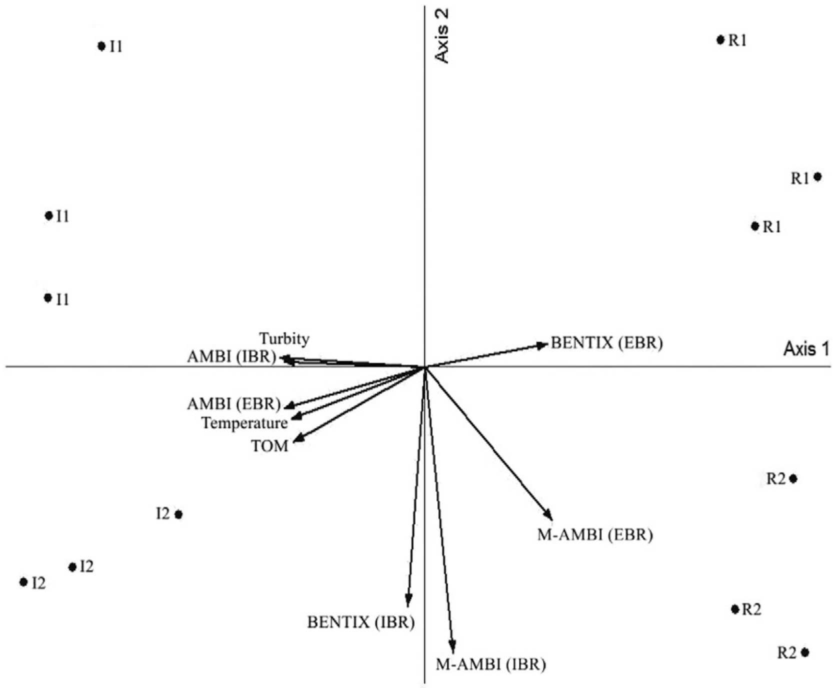

Fig. 4. PCA ordination diagram of the sampling sites. Axis I correlated positively with BENTIX (EBR) and negatively with AMBI (IBR), AMBI (EBR), temperature, TOM, and turbidity.

of turbidity and seawater temperature may be considered as proxies of sewage input. Previous studies showed significantly higher values of these proxies on the contaminated sites rather than on noncontaminated sites (Elías et al., 2009; Jaubet et al., 2011, 2013; Sánchez et al., 2013; Garaffo et al., in press). In the present study, only AMBI (IBR), AMBI (EBR), and BENTIX (EBR) were congruent with the contamination proxies. The incorporation of new proxies could be interesting for a better understanding of key ecological processes. Faecal sterols such as coprostanol have often been used as stable and source-specific molecular tracers for sewage discharges along coastal areas (Readman et al., 2005; Martins et al., 2012; Albano et al., 2013; Brauko et al., 2016). Therefore, the incorporation of this variable may have potential for future studies.

The indices applied in this study were used in various parts of the world such as the United States (Borja and Tunberg, 2011); Portugal (Sampaio et al., 2011); Spain (Borja et al., 2006); Britain (Dauvin and Ruellet, 2007); and Greece (Simboura et al., 2007). In South America, benthic indices have been applied, validated, and compared in coastal habitats subjected to multiple stressors such as urban effluents and oil spills (Muniz et al., 2005, 2011, 2012; Omena et al., 2012; Albano et al., 2013; Brauko et al., 2015, 2016). In most of these works, similar results to those of the present study were found with respect to AMBI.

BENTIX and M-AMBI produced under and overestimations of the ecological status of the studied sites. BENTIX (IBR and EBR) showed a poor discriminating power and downgraded the overall environmental health in all sites. This low level of sensitivity has been attributed to the assignment of species into only three broad ecological groups (Dauvin et al., 2007; Muniz et al., 2012). M-AMBI overestimated the assignments, as a possible reflex of the incorporation of Shannon diversity index and species richness as metrics (Simboura and Argyrou, 2010). In the study area of this contribution, an unusual phenomenon is produced on the richness and diversity of the epilithic community. The intertidal community developed on the natural area is characterized by the dominance of *B. rodriguezii* (between 82% and 91%), which results in low richness, diversity, and evenness of the associated flora and fauna (Vallarino, 2002). In contrast, areas with organic enrichment present higher richness and diversity on both temporal and spatial scale, because of organic contamination that causes a decrease in mussel dominance and an increase in the number of tolerant and opportunistic species, particularly closer to the effluent (Vallarino et al., 2002; Elías et al., 2006; Vallarino and Elías, 2006). Similar results with respect to M-AMBI were found in a subtropical estuary of Southern Brazil (Brauko et al., 2015, 2016).

Because of the low number of invertebrate taxa, together with a disturbance gradient, the simultaneous use of both flora and fauna may be more appropriate in assessing the ecological status of hard substrata (Hiscock et al., 2005; Goodsell et al., 2009). However, diversity, richness, and evenness of macroalgal assemblages in the intertidal community of Mar del Plata showed no variation between all sampling sites, and the dominance of ephemeral green algae associated with the sewage outfall was not observed (Bercherucci et al., 2016). These authors found that the sewage outfall of Mar del Plata impacts the intertidal benthic algal community by altering the specific composition. The red algae *Ceramium uruguayense* emerged as an indicator species for the nonimpacted area, and the diatom *Berkeleya* sp. as an indicator species for the sewage-impacted area. The development of a new biotic quality index that integrates the results of this work and the presence or abundance of these indicator algae could provide an integrative tool to help the environmental decision-making process. However, it would be specific to the study area. Because it is more parsimonious to use indices that already exist (Diaz et al., 2004), we recommend using the AMBI (EBR) for monitoring the study area.

Table 4
Degree of similarity between the evaluated indices: (a) for each possible combination of indices including *Brachidontes rodriguezii* (IBR) and (b) for each possible combination of indices excluding *B. rodriguezii* (EBR).

(a)	AMBI (IBR)	M-AMBI (IBR)	BENTIX (IBR)
AMBI (IBR)			
M-AMBI (IBR)	36,7		
BENTIX (IBR)	16,1	17,4	
(b)	AMBI (EBR)	M-AMBI (EBR)	BENTIX (EBR)
AMBI (EBR)			
M-AMBI (EBR)	53,4		
BENTIX (EBR)	42,2	41,4	

The capacity to distinguish human-induced and natural disturbance is a desired feature of any index for quality assessment. In this sense, temporal variability could imply that the index is more sensitive to natural variation instead of the variation attributed to human impact when the contamination source does not vary (Culhane et al., 2014). In this study, AMBI (IBR) and AMBI (EBR) indicated overall worsening trends in the benthic community health in the second year (2009). However, this variation could be attributed to human impact. From June 2008, a massive development of sandy polychaete tubes was found in a sewage-impacted area and an adjacent site located 1000 m south of the effluent. These biogenic reefs, formed by the tube-dwelling spionid polychaete *Boccardia proboscidea*, were considered as a symptom of environmental deterioration (Jaubet et al., 2011). This deterioration increased up to the setting up of a submarine outfall (2015) (unpublished data).

From 2015, the sewage pre-treatment plant of Mar del Plata celebrated the setting up of a submarine outfall that furthered the discharge 3.5 km away from the coast. Futures studies focusing on the recovery of the resident communities in the area will improve the knowledge on ecological process associated with marine pollution in the coast of Mar del Plata. The above study provides an opportunity to assess whether the AMBI (EBR) reflects the changes associated with the recovery of environmental quality. If so, managers and decision-makers will have a robust tool to monitor the evolution of the coast of Mar del Plata.

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References

Albano, M.J., Bremec, C., Elías, R., Martins, C.C., Venturini, N., Muniz, P., Rivero, S., Vallarino, E.A., Obenat, S.M., 2013. Macrobenthos and multi-molecular markers as indicators of environmental contamination in a South American port (Mar del Plata, SW Atlantic). Mar. Pollut. Bull. 73 (1), 102–114.

Amor, A., López Armengol, M.F., Iñiguez Rodríguez, A.M., Traversa, L.P., 1991. Intertidal endolithic fauna and its relationship to the mineralogical, physical and chemical characteristics of the substrate. Mar. Biol. 111, 271–280.

Bald, J., Borja, A., Muxika, I., Franco, J., Valencia, V., 2005. Assessing reference conditions and physico-chemical status according to the European Water Framework Directive: a case-study from the Basque country (Northern Spain). Mar. Pollut. Bull. 50, 1508–1522.

Ballesteros, E., Torras, X., Pinedo, S., García, M., Mangialajo, L., de Torres, M., 2007. A new methodology based on littoral community cartography dominated by macroalgae for the implementation of the European Water Framework Directive. Mar. Pollut. Bull. 55, 172–180.

Benyi, S.J., Hollister, J.W., Kiddon, J.A., Walker, H.A., 2009. A process for comparing and interpreting differences in two benthic indices in New York Harbor. Mar. Pollut. Bull. 59, 65–71.

Bercherucci, M.E., Llanos, E.N., Garaffo, G.V., Vallarino, E.A., 2016. Succession in an intertidal benthic community affected by untreated sewage effluent: a case of study in the SW Atlantic shore. Mar. Pollut. Bull. 109, 95–103.

Blanchet, H., Lavesque, N., Ruellet, T., Dauvin, J.C., Sauriau, P.G., Desroy, N., Desclaux, C., Leconte, M., Bachelet, G., Janson, A.L., Bessineton, C., Duhamel, S., Jourde, J., Mayot, S., Simon, S., de Montaudouin, X., 2007. Use of biotic indices in semienclosed coastal ecosystems and transitional waters habitats – implications for the implementation of the European Water Framework Directive. Ecol. Indic. 8, 360–372.

Borja, A., Tunberg, B.G., 2011. Assessing benthic health in stressed subtropical estuaries, eastern Florida, USA using AMBI and M-AMBI. Ecol. Indic. 11, 295–303.

Borja, A., Franco, J., Pérez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within European estuarine and coastal environments. Mar. Pollut. Bull. 40, 1100–1114.

Borja, A., Muxika, I., Franco, J., 2003. The application of a marine biotic index to different impact sources affecting soft-bottom benthic communities along European coasts. Mar. Pollut. Bull. 46, 835–845.

Borja, A., Franco, J., Valencia, V., Bald, J., Muxika, I., Belzunce, M.J., Solaun, O., 2004. Implementation of the European water framework directive from the Basque country (Northern Spain): a methodological approach. Mar. Pollut. Bull. 48, 209–218.

Borja, A., Muxika, I., Franco, J., 2006. Long-term recovery of soft-bottom benthos following urban and industrial sewage treatment in the Nervión estuary (southern Bay of Biscay). Mar. Ecol. Prog. Ser. 313, 43–55.

Borja, A., Dauer, D.M., Díaz, R., Llansó, R.J., Muxika, I., Rodríguez, J.G., Schaffner, L., 2008. Assessing estuarine benthic quality conditions in Chesapeake Bay: a comparison of three indices. Ecol. Indic. 8, 395–403.

Borja, A., Basset, A., Bricker, S., Dauvin, J.C., Elliott, M., Harrison, T., Marques, J.C., Weisberg, S., West, R., 2012. Classifying ecological quality and integrity of estuaries. In: Wolanski, E., McLusky, D. (Eds.), Treatise on Estuarine and Coastal Science. Elsevier (doi: 00109 is 10.1016/B978-0-12-374711-2.00109-1).

Bouvet, Y., Desse, R.P., Morell, P., Villar, M.C., 2005. Mar del Plata (Argentina): la ciudad balnearia de los porteños en el Atlántico Suroccidental. Investigaciones Geográficas 36, 61–80.

Brauko, K.M., Souza, F.M., Muniz, P., Camargo, M.G., Lana, P.C., 2015. Spatial variability of three benthic indices for marine quality assessment in a subtropical estuary of Southern Brazil. Mar. Pollut. Bull. 91, 454–460.

Brauko, K.M., Muniz, P., de Castro Martins, C., da Cunha Lana, P., 2016. Assessing the suitability of five benthic indices for environmental health assessment in a large subtropical South American estuary. Ecol. Indic. 64, 258–265.

Byers, S., Mills, C., Stewart, P., 1978. Comparison of methods of determining organic carbon in marine sediments, with suggestions for a standard method. Hydrobiologia 58 (1), 43–47.

Comino, A.P., Scagliola, M., Ge, Z., Frick, W., 2010. Integrated Coastal Management Strategy of Mar del Plata City and the Use of Virtual Beach as a Management Tool. Proceedings of MWWD 2010, 6° International Conference on Marine Waste Water Disposal and Marine Environment ((www.mwwd.org) 25–29 Octubre, Langkawi, Malaysia).

Culhane, F.E., Briers, R.A., Tett, P., Fernandes, T.F., 2014. Structural and functional indices show similar performance in marine ecosystem quality assessment. Ecol. Indic. 43, 271–280.

Dauvin, J.C., Ruellet, T., 2007. Polychaete/amphipod ratio revisited. Mar. Pollut. Bull. 55, 215–224.

Dauvin, J.C., Ruellet, T., Desroy, N., Janson, A.L., 2007. The ecological quality status of the Bay of Seine and the Seine estuary: use of biotic indices. Mar. Pollut. Bull. 55, 241–257.

Dauvin, J.C., Bellan, G., Bellan-Santini, D., 2010. Benthic indicators: from subjectivity to objectivity-where is the line? Mar. Pollut. Bull. 60, 947–953.

Devlin, M., Best, M., Coates, D., Bresnan, E., O'Boyle, S., Park, R., Silke, J., Cusack, C., Skeats, J., 2007. Establishing boundary classes for the classification of UK marine waters using phytoplankton communities. Mar. Pollut. Bull. 55, 91–103.

Diaz, R.J., Solan, M., Valente, R.M., 2004. A review of approaches for clarifying benthic habitats and evaluating habitat quality. J. Environ. Manag. 73, 165–181.

Díez, I., Bustamante, M., Santolaria, A., Tajadura, J., Muguerza, N., Borja, A., Muxika, I., Saiz-Salinas, J.I., Gorostiaga, J.M., 2012. Development of a tool for assessing the ecological quality status of intertidal coastal rocky assemblages, within Atlantic Iberian coasts. Ecol. Indic. 12, 58–71.

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 (Southwestern Atlantic, Argentina). Sci. Mar. 70 (3), 187–196.

Elías, R., Sánchez, M.A., Jaubet, M.L., Rivero, M.S., Vallarino, E.A., 2009. Do treatment sewage plants really work? The intertidal mussels' community of the southwestern Atlantic shore (38°S, 57°W) as a case study. Rev. Biol. Mar. Oceanogr. 44, 357–368.

Elias, R., Jaubet, M.L., Llanos, E.N., Sánchez, M.A., Rivero, M.S., Garaffo, G.V., Sandrini-Neto, L., 2015. Effect of the invader *Boccardia proboscidea* (Polychaeta: Spionidae) on richness, diversity and structure of SW Atlantic epilithic intertidal community. Mar. Pollut. Bull. 91, 530–536.

Garaffo, G.V., Jaubet, M.L., Sánchez, M.A., Llanos, E.N., Vallarino, E.A., Elías, R., 2016. Modelling the influence of environmental and weather factors on the density of the invasive polychaete *Boccardia proboscidea*. Mar. Ecol. 37 (in press).

Goodsell, P.J., Underwood, A.J., Chapmann, M.G., 2009. Evidence necessary for taxa to be reliable indicators of environmental conditions or impacts. Mar. Pollut. Bull. 58, 323–331.

Gray, J.S., Wu, R.S., Or, Y.Y., 2002. Effects of hypoxia and organic enrichment on the coastal marine environment. Mar. Ecol. Prog. Ser. 238, 249–279.

Guerrero, R.A., Piola, A.R., 1997. Masas de agua en la plataforma continental. In: Boschi, E.E. (Ed.), El Mar Argentina y sus recursos pesqueros 1. Instituto nacional de investigación y desarrollo pesquero, Mar del Plata. Argentina, pp. 107–118.

Guinda, X., Juanes, J.A., Puente, A., 2014. The quality of rocky bottoms index (CFR): a validated method for the assessment of macroalgae according to the European Water Framework Directive. Mar. Environ. Res. 102, 3–10.

Halpern, B.S., Selkoe, K.A., Micheli, F., Kappel, C.V., 2007. Evaluating and ranking the vulnerability of global marine ecosystems to anthropogenic threats. Conserv. Biol. 21, 1301–1315.

Hiscock, K., Smith, A., Jenkins, S., Sewell, J., Hawkins, S., 2005. Development of a hard substratum Benthic invertebrate Water Framework Directive compliant classification tool. Report to the Environment Agency and the Joint Nature Conservation Committee from the Marine Biological Association. Marine Biological Association. Inc., Plymouth (Contract F90-01-790, 54 pp).

Isla, F.I., Pérez Guzzi, J., Folabella, A., 1998. Aportes de coliformes a la costa de Mar del Plata (Argentina) por vías naturales e inducidas. Thalassas 14, 63–70.

Jaubet, M.L., Sánchez, M.A., Rivero, M.S., Garaffo, G.V., Vallarino, E.A., Elías, R., 2011. Intertidal biogenic reefs build by invasive polychaete in sewage-impacted areas of Argentina, SW Atlantic. Mar. Ecol. 32, 1–10.

Jaubet, M.L., Garaffo, G.V., Sánchez, M.A., Elías, R., 2013. Reef-forming polychaetes outcompetes ecosystem engineering mussels. Mar. Pollut. Bull. 71, 216–221.

Juanes, J.A., Guinda, X., Puente, A., Revilla, J.A., 2008. Macroalgae, a suitable indicator of the ecological status of coastal rocky communities in the NE Atlantic. Ecol. Indic. 8, 351–359.

Mangialajo, L., Ruggieri, N., Asnaghi, V., Chiantore, M., Povero, P., Cattaneo-Vietti, R., 2007. Ecological status in the Ligurian Sea: the effect of coastline urbanization and the importance of proper reference sites. Mar. Pollut. Bull. 55, 30–41.

Martins, C.C., Aguiar, S.N., Bícego, M.C., Montone, R.C., 2012. Sewage organic markers in surface sediments around the Brazilian Antarctic station: results from the 2009/10 austral summer and historical tendencies. Mar. Pollut. Bull. 64, 2867–2870.

Muniz, P., Venturini, N., Pires-Vanin, A.M.S., Tommasi, L.R., Borja, A., 2005. Testing the applicability of a Marine Biotic Index (AMBI) to assessing the ecological quality of soft-bottom benthic communities, in the South America Atlantic region. Mar. Pollut. Bull. 50, 624–637.

Muniz, P., Hutton, M., Kandratavicius, N., Lanfranconi, A., Brugnoli, E., Venturini, N., Giménez, L., 2011. Performance of biotic indices in naturally stressed estuarine environments on the Southwestern Atlantic coast (Uruguay): a multiple scale approach. Ecol. Indic. 19, 89–97.

Muniz, P., Hutton, M., Kandratavicius, N., Lanfranconi, A., Brugnoli, E., Venturini, N., Giménez, L., 2012. Performance of biotic indices in naturally stressed estuarine environments on the Southwestern Atlantic coast (Uruguay): a multiple scale approach. Ecol. Indic. 19, 89–97.

Muxika, I., Borja, Á., Bald, J., 2007. Using historical data, expert judgement and multivariate analysis in assessing reference conditions and benthic ecological status, according to the European Water Framework Directive. Mar. Pollut. Bull. 55, 16–29.

Niemi, G.J., Mc Donalds, M.E., 2004. Application of ecological indicators. Annu. Rev. Ecol. Evol. Syst. 35, 89–111.

Omena, E.P., Lavrado, H.P., Paranhos, R., Silva, T.A., 2012. Spatial distribution of intertidal sandy beach polychaeta along an estuarine and morphodynamic gradient in an eutrophic tropical bay. Mar. Pollut. Bull. 64, 1861–1873.

Orfanidis, S., Panayotidis, P., Stamatis, N., 2003. An insight to the ecological evaluation index (EEI). Ecol. Indic. 3, 27–33.

Pearson, T.H., Rosenberg, R., 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. Oceanogr. Mar. Biol. 16, 229–311.

Pintos, R., Patrício, J., Baeta, A., Fath, B.D., Neto, J.M., Marques, J.C., 2009. Review and evaluation of estuarine biotic indices to assess benthic condition. Ecol. Indic. 9, 1–25.

Quintino, V., Elliot, M., Rodrigues, A.M., 2006. The derivation, performance and role of univariate and multivariate indicators of benthic change: case studies of differing spatial scales. J. Exp. Mar. Biol. Ecol. 330, 368–382.

Quiroga, E., Ortiz, P., Reid, B., Gerdes, D., 2013. Classification of the ecological quality of the Aysen and Baker Fjords (Patagonia, Chile) using biotic indices. Mar. Pollut. Bull. 68 (1–2), 117–126.

Readman, J.W., Fillmann, G., Tolosa, I., Bartocci, J., Mee, L.D., 2005. The use of steroid markers to assess sewage contamination of the Black Sea. Mar. Pollut. Bull. 50, 310–318.

Salas, F., Marcos, C., Neto, J.M., Patrício, J.A., Pérez-Ruzafa, A., Marques, J.C., 2006. User-friendly guide for using benthic ecological indicators in coastal and marine quality assessment. Ocean Coast. Manag. 49, 308–331.

Sampaio, L., Rodrigues, A.M., Quintino, V., 2011. Can biotic indices detect mild organic enrichment of the seafloor. Ecol. Indic. 11, 1235–1244.

Sánchez, M.A., 2014. Aplicación de un diseño BACI para la evaluación de impacto antrópico mediante poliquetos indicadores y la estructura de la comunidad intermareal de *Brachidontes rodriguezii*. Universidad Nacional de Mar del Plata (Doctoral Thesis. 206 pp).

Sánchez, M.A., Jaubet, M.L., Garaffo, G.V., Elías, R., 2013. Spatial and long-term analyses of reference and sewage-impacted sites in the SW Atlantic (38°S, 57°W) for the assessment of sensitive and tolerant polychaetes. Mar. Pollut. Bull. 74, 325–333.

Scagliola, M., Furchi, P., Von Haeften, G., Comino, A.P., Moschione, E., Gonzales, R., Gayoso, G., Caldararo, A., Cerda, G., Vergara, S., Genga, G., Elías, R., Vallarino, E.A., 2006. Sewage outfall project of Mar del Plata city (Argentina): an effective intervention to achieve quality objectives on the marine environment. Annals of the 4th International Conference on Marine Waste Water Disposal and Marine Environment (Antalya (Turquía), 6–10 November, 22 pp).

Simboura, N., Argyrou, M., 2010. An insight into the performance of benthic classification indices tested in Eastern Mediterranean coastal waters. Mar. Pollut. Bull. 60, 701–709.

Simboura, N., Zenetos, A., 2002. Benthic indicators to use in ecological quality classification of Mediterranean soft bottom marine ecosystems, including a new biotic index. Mediterr. Mar. Sci. 3 (2), 77–111.

Simboura, N., Panayotidis, P., Papathanassiou, E., 2005. A synthesis of the biological quality elements for the implementation of the European Water Framework Directive in the Mediterranean ecoregion: the case of Saronikos Gulf. Ecol. Indic. 5, 253–266.

Simboura, N., Papathanassiou, E., Sakellariou, D., 2007. The use of biotic index (Bentix) in assessing long-term effects of dumping coarse metalliferous waste on soft bottom benthic communities. Ecol. Indic. 7, 164–180.

Vallarino, E.A., 2002. La comunidad bentónica intermareal de Brachidontes rodriguezii (D'Orb) y su relación con el efluente cloacal de la ciudad de Mar del Plata (38°S). Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Mar del Plata (Tesis Doctoral de Biología, 188 pp).

Vallarino, E.A., Elías, R., 2006. High-diverse lowly variable sewage-impacted community, low-diverse highly variable natural community: the paradox of the intertidal mussel beds of temperate areas of the SW Atlantic (38°S, 57°W). Current Trends Ecology. 1, pp. 77–91.

Vallarino, E.A., Rivero, M.S., Gravina, M.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. Rev. Biol. Mar. Oceanogr. 37, 25–33.

Warwick, R.M., 1993. Environmental impact studies on marine communities: pragmatical considerations. Aust. J. Ecol. 18, 63–80.

Warwick, R.M., Clarke, K.R., 1993. Comparing the severity of disturbance: a meta-analysis of marine macrobenthic community data. Mar. Ecol. Prog. Ser. 92, 221–231.